



AGRICULTURE 3.0

INNOVATIONS IN MODERN FARMING



Editors:

Dr. D. P. S. Badwal

Dr. Sushila Hooda

JUST AGRICULTURE PUBLICATIONS

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Preface

Agriculture today stands at a pivotal crossroads. As global populations rise, climate uncertainties intensify, natural resources become increasingly strained, and market dynamics evolve, the need for a more resilient, intelligent and sustainable farming system has never been greater. It is in this transformative landscape that the concept of Agriculture 3.0 has emerged an era defined by innovation, digitization, precision and a renewed commitment to ecological balance.

Agriculture 3.0: Innovations in Modern Farming brings together contemporary research, technological advancements and field-level practical insights that are reshaping modern agriculture. This volume explores the integration of smart farming tools, sensor-based monitoring, automation, biotechnology, nanotechnology, renewable energy integration, climate-smart agricultural practices and data-driven decision systems. Each chapter aims to bridge the gap between theory and practice, offering readers a comprehensive understanding of how modern innovations can enhance productivity, profitability and sustainability.

The book is designed for students, researchers, academicians, policymakers and farming professionals who seek to understand emerging agricultural paradigms. By presenting diverse perspectives from experts across disciplines, this compilation encourages knowledge exchange and inspires future innovations.

We express our sincere gratitude to the contributing authors for their valuable insights, to the reviewers for their meticulous evaluation, and to Just Agriculture Publications for supporting the dissemination of this important work. It is our hope that this book serves as a meaningful resource, guiding the next generation of researchers, innovators and practitioners toward a future-ready agricultural ecosystem.

Editors

Agriculture 3.0: Innovations in Modern Farming

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Global public health concern on hypovitaminosis D: Facts and Fallacies

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Hypovitaminosis D, or vitamin D deficiency, is a major yet often overlooked global public health concern, affecting populations even in sunlight-abundant regions like India. Despite year-round sun exposure, 50–90% of Indians exhibit insufficient serum 25-hydroxyvitamin D levels due to limited outdoor activity, darker skin pigmentation, air pollution, and poor intake of vitamin D-rich foods. Beyond bone health, vitamin D plays key roles in immune regulation, cardiovascular stability, and neuroprotection, with deficiency linked to disorders such as osteoporosis, diabetes, cancer, and cognitive decline. This review discusses vitamin D synthesis, global prevalence, and strategies for prevention through supplementation and food fortification. Edible mushrooms, when UV-treated, emerge as an effective, sustainable, and plant-based source of vitamin D₂ for vegetarians and vegans. Addressing hypovitaminosis D through integrated public health initiatives, fortified foods, and standardized UV treated mushroom technologies offers a practical path to mitigate this widespread deficiency.

Keyword : Hypovitaminosis, Vitamin D, Deficiency, Supplement, Fortification

Introduction

Hypovitaminosis D is the medical name for vitamin D deficiency. Vitamin D, a fat-soluble vitamin acts like a steroid hormone, helps the body keep key systems running smoothly from building strong bones to supporting steady muscle movement under warm sunlight.

The main vitamin D derivatives are calcitriol, tacalcitol, calcipotriol, maxacalcitol, and hexafluoro-1,25 (OH)₂D, each linking back to that familiar sunshine-sparked nutrient. Vitamin D deficiency is common globally, in both sunlight-deprived and sunlight-rich countries, but it remains one of the most under-diagnosed and under-treated diet deficits. Because sunlight is our main source of vitamin D, people long assumed that a sun-soaked country like India with bright skies blazing nearly all year couldn't possibly face a vitamin D shortage. Meanwhile, studies of the Indian population showed a mixed picture like threads of many colours woven through a single fabric. By the Institute of Medicine's current guideline for normal vitamin D levels, many otherwise healthy Indians across the country still show a deficiency even those who spend hours in bright afternoon sun (Selvarajan *et al.*, 2017). Although sunny regions offer plenty of vitamin D, there's still a mix of truth and myth about how much we actually need. This chapter explores why vitamin D absorption often falls short, how the body processes it, and what foods like oily fish or fortified milk can help reverse a deficiency.

Hypovitaminosis D: Global public health concern

A lack of vitamin D can hit anyone, young or old, even the kid who never sees the sun. Even though sunlight's plentiful year-round, our study found most patients about 80% still had low vitamin D levels, and women showed a definite tendency toward the deficiency. Various other studies and results on the South Asian population show the same type of vitamin deficiency, even when exposed to sunlight (Achakzai *et al.*, 2016). People have found vitamin D deficiency in countries that see little sun and in places where sunlight pours down almost every day. India officially recognizes it as a public health issue, a concern that touches crowded clinics and busy city streets alike. Although India basks in sunlight most of the year, vitamin D deficiency is still becoming a serious concern. Vitamin D insufficiency is frequent (50-90%) in the Indian population; insufficient dietary calcium intake (Kamboj *et al.*, 2018). Lips *et al.* (2021) have extensively reviewed recent global vitamin D status assessments, which suggest that North and Latin America, as well as Australia, have higher vitamin D levels than Europe; South Eastern Asia has higher levels than India and Northern Asia; and Central Africa has higher levels than Northern and Southern Africa. Overall, people in the Middle East showed the lowest vitamin D levels; many rarely saw the sun except through the heat shimmer on dusty streets. People in Northern Europe generally have higher vitamin D levels than those living in the sunnier south or east, even on gray mornings when sunlight barely filters through the clouds. A worldwide study on vitamin D insufficiency revealed striking numbers of severely deficient infants: 61% in India, 86% in Iran, and 51% in Turkey, even though overall deficiency rates topped 90% in each country (Gupta, 2016).

Aparna *et al.* (2018) have elaborated that regardless of age, gender, menopausal status, or whether they live in an urban or rural region, insufficient vitamin D nourishment is frequently observed among Indians. In Indians, vitamin D deficiency often stems from lower vitamin D synthesis due to darker skin, sunscreen use, spending long hours indoors, and eating fewer vitamin D-rich foods like fish or fortified milk. It is uncertain why vitamin D contributes to organ damage and etiopathogenesis in the body's major organs, aside from the well-known skeletal and bone issues. Vitamin D is crucial for strong bones. It keeps calcium in balance across the kidneys, bones, parathyroid gland, and gut, steady as chalk on a slate. In India, vitamin D deficiency runs high anywhere from 40% to 99% with most studies pointing to about 80–90%; in crowded city clinics, it's almost everyone under the bright sun yet low on the “sunshine” vitamin. Vitamin D insufficiency causes autoimmune illnesses, cardiovascular problems, cancer, and TB. Both the public and healthcare providers need to understand how crucial vitamin D is to the quiet epidemic behind so many tired faces, and it must be tackled with real, measurable public health action.

Plausible reasons of Vitamin D deficiency

- In many places, vegetarian and lacto-vegetarian diets, where people eat dairy like milk, cheese, yogurt, or paneer, rarely emphasize traditional foods rich in vitamin D, such as eggs, mushrooms, or a piece of buttery salmon sizzling in a pan. People still drink skim or low-fat milk, even though it's short on vitamin D and looks almost watery in the glass.
- Fatty fish and seafood, among the best natural sources of vitamin D, don't show up often in Indian cooking, where dishes lean more on lentils, spices, and vegetables. Sufficient consumption of seafood is hampered by social restrictions on its consumption, exorbitant costs, the inaccessibility of fish oil capsules, and a lack of knowledge about its advantages.
- Indian food relies heavily on lentils and pulses, which are low in vitamin D.
- Consuming basics like rice and wheat without extra foods can reduce vitamin D absorption.
- Cod liver oil, beef, eggs, milk products, and mushrooms are minor sources of vitamin D in the diet.
- Increased indoor activities limits proper exposure to sunshine.
- Polluted air can block the Sun's UV rays, keeping your skin from making vitamin D, like a cloudy haze dulling the warmth on your arm.
- Changes in food habits relate to decreased dietary calcium and vitamin D consumption.
- Eating a fiber-rich diet loaded with phytates and phosphates can drain your vitamin D stores and make your body crave more calcium, much like chalk dust drying on your hands. Harinarayan *et al.* (2007) found that low calcium intake and reduced 25(OH)D levels disrupted bone mineral balance, leading to vitamin D deficiency.

- Darker and increased skin pigmentation and use of sunscreen lotions.
- Unspaced and unexpected births in women with nutritional deficiencies can lead to a decline in vitamin D status in both mother and child. Darker, more noticeable skin tone often appears, especially if you're using sunscreen lotion.
- The presence of cancer, namely lymphoma, celiac disease, cystic fibrosis, Crohn's disease, inflammatory bowel disease, or exocrine pancreatic insufficiency, reduces vitamin D absorption into the body.

Vitamin D Dosages

Vitamin D levels differ from one continent to another, and even between neighbouring countries; sunlight in Sydney isn't the same as in Stockholm. Vitamin D levels are usually fine in Latin America and Australia, but they drop sharply in the Middle East and in parts of Asia, where sunlight hardly touches the skin. Changes in vitamin D levels rising or falling carry serious weight, shaping how we understand long-term health. Though trends are usually small, they can hint at bigger shifts and reveal how healthy things really are, like a steady pulse showing the heart's rhythm. Older adults in the U.S. now show higher vitamin D levels, a shift tied to more dairy on their tables and regular use of vitamin D supplements. In contrast, the Inuit of Canada have seen their vitamin D levels fall after shifting from meals rich in fresh-caught fish to a Western-style diet. In contrast, the Inuit of Canada have seen their vitamin D levels fall after shifting from meals rich in fresh-caught fish to a Western-style diet. Among African Americans, vitamin D deficiency shows up far more often than in other Americans, and throughout the year, most young, healthy Black people in North America never reach enough 25-hydroxyvitamin D [25(OH)D] even after long hours in the sun. African Americans face higher risks than European Americans for many health problems, and they're more likely to die from them, whether it's heart disease, diabetes, or another illness that steals breath too soon. They're also 15 to 20 times more likely to suffer from severe vitamin D deficiency, the kind that leaves skin pale and energy drained. That's mainly because skin pigment blocks the process that makes vitamin D like a faint shadow dulling sunlight on your arm. Ames *et al.* (2021) found the same thing in their study, that this health disparity is partly due to insufficient vitamin D production, which is caused by (i) Melanin in the skin blocking the UVB solar radiation required for its synthesis; (ii) vitamin D insufficiency is exacerbated at high latitudes because of the combination of dark skin color with lower UVB radiation levels; and (iii) individuals with dark skin can benefit significantly from correcting deficiency and obtaining optimal vitamin D status, which can be accomplished through supplementation and/or fortification. Solid evidence suggests that higher 25-hydroxyvitamin D levels or taking vitamin D supplements can lower the risk of a wide range of health problems,

from all-cause mortality and pregnancy complications to cancer, diabetes, Alzheimer's and other dementias, multiple sclerosis, and even respiratory infections like COVID-19 or asthma flare-ups; it also helps prevent bone diseases such as rickets and osteomalacia. People with darker skin who live in northern regions and often have low vitamin D should talk with their doctor about taking vitamin D3 supplements to raise their blood 25-hydroxyvitamin D to at least 30 ng/ml (75 nmol/l); even a quick walk in weak winter sunlight may not be enough. Since there's no single agreed-upon way to define vitamin D status, each study set its own cutoff for deficiency one might call it low at 20 ng/ml, another at 30 Most studies defined vitamin D insufficiency as serum 25(OH)D levels below 20 ng/ml, about what you might see in a blood test drawn on a cloudy winter morning. Footnotes show which studies used different cutoffs; for instance, some marked results at a lower threshold. Over the past decade, community studies in India on people who appeared healthy found prevalence rates from about 50% up to 94%, except for one study that showed only 34.5%, likely because its cutoff point was set too low. These studies, spanning every age group from kids to retirees, reveal just how vast the problem really is. Cases cropped up all over the country, from crowded city streets to quiet rural towns. According to the Endocrine Society's guidelines, vitamin D levels are grouped as deficient, insufficient, or sufficient, based on serum 25-OH vitamin D values of less than 20 ng/ml (50 nmol/l), 21–29 ng/ml (52.5–72.5 nmol/l), and 30–100 ng/ml (75–250 nmol/l), respectively (Holick *et al.*, 2011); a sample reading of 18 ng/ml, for instance, would fall squarely in the deficient range. Forty-one endocrinologists from across India came together to form an expert panel, using the DELPHI method to reach consensus, and they ended up agreeing on more than 90% of the recommendations. The consensus outlines what counts as vitamin D deficiency, severe deficiency, and insufficiency, and it recommends supplements to keep 25(OH)D levels steady around 40 to 60 ng/ml (100–150 nmol/l), about where your body runs best. In India, customized treatments are offered for every stage of life, from fragile newborns and restless toddlers to teens, adults, older adults, pregnant or nursing women, and those managing other conditions, to help each person reach their best health (Kalra *et al.*, 2025).

Vitamin D Synthesis and metabolism

The two key inactive forms of vitamin D are D3 and D2 think of them as the quiet versions waiting for sunlight or the body's enzymes to switch them on. Your skin makes its own vitamin D3 when sunlight in the 290–310 nm range touches it, like a pale shimmer warming your arm on a bright noon. With this approach, the more UV light your skin soaks up, the more vitamin D3 it makes, and darker pigmentation slows that process down. Plus, where you live, the weather, your age, how much sunscreen you wear, and the hours you spend in the sun all shape how much vitamin D3 your body makes, whether it's a bright morning walk or a cloudy afternoon indoors.

Plants make vitamin D₂ when sunlight touches their leaves, and your body takes it in through foods like mushrooms or fortified grains. You'll find only traces of vitamin D₂ in natural foods bits hiding in meat or the rich yellow of an egg yolk, and most of it shows up in fish. In plants, ergosterol serves as the starting point for vitamin D, while in animals, it's 7-dehydrocholesterol that plays that role, like sunlight flipping a chemical switch inside the cells. We call both vitamin D₂ and D₃ simply "vitamin D" since the body processes them in much the same way, giving each the same potential for activity (Cicek *et al.*, 2023).

Table 1: Natural Sources of vitamin D Nutrients

Sources of vitamin D	Vit D (IU)
Cod Liver Oil 1 tbsp	1360
Salmon Fish (100 g)	360
Sardines (100 g)	250
Tuna (100 g)	200
Mackerel (100 g)	345
Egg (yolk)	20
Beef Liver (cooked 100 g)	15
Swiss Cheese (30 g)	12

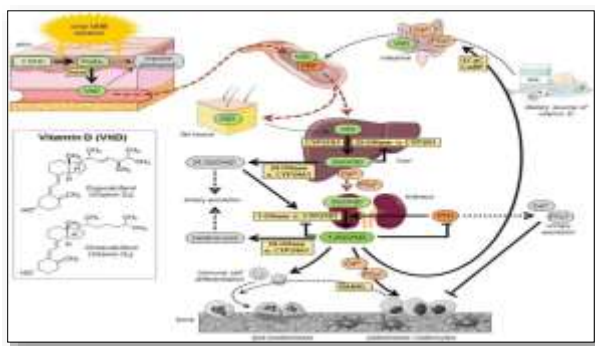
Most vertebrates make enough vitamin D when sunlight those warm UVB rays touches their skin. Most vertebrates need enough vitamin D, getting it from their food or from sunlight warming their skin. The term "vitamin D" actually points to two specific compounds, cholecalciferol and ergocalciferol, both vital for keeping bones strong, like sunlight bottled in a pill. Sunlight hits your skin, and that spark of warmth helps it make vitamin D₃. When sunlight hits your skin, ultraviolet rays flip 7-dehydrocholesterol into vitamin D₃ like a quiet spark beneath the surface. Foods rich in vitamin D₃ come from animals; think of oily fish like salmon, mackerel, or tuna sizzling in a pan, along with cod liver oil, milk, and other similar sources. Sunlit mushrooms and yeast naturally hold vitamin D₂, soaking it up the way they do warm afternoon light. Most foods don't have much vitamin D; think of a slice of bread or a bowl of rice, both nearly empty of it. Vitamin D, whether D₃ or D₂, acts as a prohormone, needing two hydroxylation steps before it changes into its active form, 1,25(OH)₂D, the compound that really gets things moving in your cells like sunlight warming bare skin.

Vitamin D gets hydroxylated first in the liver, then in the kidneys, and the finished form travels through the blood to tissues packed with vitamin D receptors. The liver performs the first hydroxylation at the C₂₅ position, forming 25-hydroxyvitamin D, known as 25(OH)D or calcidiol, a

reaction as precise as a drop of clear liquid meeting its enzyme target (Ramasamy, 2020). The main form of vitamin D moving through your bloodstream is $25(\text{OH})\text{D}$, the type doctors measure when they check your levels. The second hydroxylation takes place at position $\text{C}1\alpha$, producing $1,25(\text{OH})_2\text{D}$ better known as calcitriol, the active form of vitamin D that flickers into life like sunlight on skin. The kidneys mainly produce $1,25(\text{OH})_2\text{D}$, though they're not the only source other tissues lend a hand, quietly working behind the scenes like small engines humming. $1,25(\text{OH})_2\text{D}$ enters the bloodstream and quickly binds to the vitamin D-binding protein (DBP), drifting through like a drop of oil on water until it reaches its target tissues, where it activates the vitamin D receptor (VDR) to carry out its endocrine tasks. A few tissues outside the kidneys also make $1,25(\text{OH})_2\text{D}$, using it to signal nearby cells and even themselves like a quiet message passed from one neighbour to the next. Most of the body's cells have VDRs, from skin to muscle, each one quietly responding to light and touch. Several kinds of cells can produce $1,25(\text{OH})_2\text{D}$, like tiny factories turning raw material into the active form of vitamin D. $1,25(\text{OH})_2\text{D}$ controls a wide array of genes that guide how cells grow and take shape, much like light nudging a seedling toward its first green leaf.

Pleomorphic role of vitamin D

Beyond keeping bone minerals in balance, vitamin D also acts in many ways: it calms inflammation, helps cells survive instead of breaking down, and protects tissues from scarring, much like sunlight warming skin after a long winter. It also plays a key part in keeping the heart steady, the kidneys balanced, and the immune system alert, like the quick pulse you feel after a sprint. Low vitamin D levels are linked to a higher risk of diseases like diabetes, schizophrenia, and certain cancers a connection as clear as sunlight warming your skin. When people don't get enough vitamin D, kids can develop rickets, while adults may end up with soft bones osteomalacia or brittle ones, known as osteoporosis. In children, an uneven balance of calcium and phosphorus weakens bone mineralization, leading to rickets and visible skeletal deformities, like legs that bow slightly outward.



Vitamin D Synthesis and metabolic pathway

(Courtesy: *Nutrients*, 2016, 8: 319)

A single drop of rain slid down the window. It can leave your muscles feeling weak and your bones aching, like a dull throb after a long day. When adults don't get enough vitamin D from food, their bodies absorb less calcium from meals, pull more of it from bones and kidneys, and gradually lose bone strength leaving bones brittle, muscles weak, and falls more likely. Plenty of studies link low vitamin D levels to a higher chance of fractures and falls in older adults, like slipping on a damp kitchen tile after dusk. Vitamin D helps safeguard certain tissues by triggering cell death and slowing the growth of new blood vessels, much like dimming the light to keep runaway cells in check. Low vitamin D levels in tissues like the lungs, breast, colon, prostate, ovaries, pancreas, and esophagus are linked to cancer, much like a dim light signaling something's wrong deep inside. Vitamin D slows cell growth and helps cells mature, like sunlight guiding buds to bloom. It blocks new blood vessels from forming and powerfully calms inflammation, like cooling a burn with a splash of cold water. Patients suffering a heart attack often show a marked vitamin D shortage, sometimes as low as a pale winter sun. Low levels of vitamin D have been linked to a higher risk of type 2 diabetes, insulin resistance, reduced insulin output, and metabolic syndrome the body struggling to handle sugar, like honey left too long in the sun. Vitamin D regulates the cellular processes like oxidative stress, DNA repair, mitochondrial function, inflammation, all these contribute to age related decline leading to clinical conditions like osteoporosis and cognitive decline. Ageing shows both phenotypic and genotypic changes in expression range.

One proof is that what we see our phenotypic expression. As ageing proceeds skin shrinkage occurs and is visible through naked eyes. Thus we can guess a person's age by seeing appearance. Thus ageing affects each and every part of human body such as organ, tissues and cells of different types. Thus at birth some proteins are at high level and as ageing proceeds from birth there occurs both phenotypic and genotypic changes. The vitamin D has its role in preventing photoaging *via* much process involving many proteins such as TGF-beta, IL-4, IL-10, etc. It attributes to protection against oxidative stress, DNA damage, inflammation of skin and other organs. vitamin D deficiency leads to various cutaneous disorders as follows Psoriasis, Rosacea, Seborrheic dermatitis, atopic dermatitis, congenital ichthyosis, vitiligo, alopecia areata, atopic eczema and hidradenitis suppurativa, epidermolytic acanthoma, clear cell acanthoma, circumscribed plantar hypokeratosis, dystrophic epidermolysis bullosa (DEB), Sorafenib-associated hand-foot syndrome, linear atrophoderma of mouli and warts. It is more than 20 years ago that the vitamin D has the function of anti-oxidant proven by Wiseman, in liposomal lipid peroxidation. This is due to the similarity of its structure with cholesterol and ergosterol.

Through its receptors in the cytosol and nucleus, vitamin D switches on several genes vital to the body's antioxidant defense, including ones for superoxide dismutase and glutathione peroxidase, the enzymes that quietly mop up harmful oxygen radicals. The vitamin D supplementation decreases the ROS biomarkers. Parkinson's disease, a neurodegenerative disorder that often affects older adults, stems from chronic vitamin D deficiency, which gradually destroys dopaminergic neurons in the brain's dark, compact region called the substantia nigra. When vitamin D levels drop too low in the blood, the risk of dementia and Alzheimer's rises, especially among older adults whose skin no longer soaks up much sunlight. Vitamin D also plays a role in shaping the gut's microbiota and helping the body make serotonin, the well-known "daylight" hormone, so when it's lacking, mental health can suffer, leading to conditions like schizophrenia or depression.

Vitamin D supplements and fortification

Adding vitamin D to staple foods, pairing that vitamin D with calcium, and folding fortified local foods into government nutrition programs could all help tackle hypovitaminosis D, like sunlight strengthening a field after rain. More and more experts agree that most older adults don't get nearly enough vitamin D each day, and everyone should take whatever amount it takes to bring their levels up to about 32–40 ng/ml (80–100 nmol/l), enough to turn a gray morning into one that feels a little brighter. Many older adults often need a vitamin D supplement because diet and sunlight don't provide enough, especially on short, gray winter days. According to Cherniack *et al.* (2008), keeping 25(OH) vitamin D levels at or above 32 ng/ml (80 nmol/l) usually means older adults need about 2,000 IU of cholecalciferol each day, roughly what you'd find in two small supplement tablets. Because modern habits limit sun exposure and diet alone can't supply enough vitamin D, scientists need to dig deeper into how this vitamin works and run new clinical trials to see if higher supplement levels beyond those proven to prevent falls and fractures might also ease other medical problems, offering an affordable way to boost health, extend life, and strengthen public well-being.

Vitamin D in Edible Mushrooms

Edible mushrooms pack plenty of vitamin D, especially D₂ (ergocalciferol), because their ergosterol turns into D₂ under UV light, much like how sun on a forest floor coaxes them to grow. When mushroom cell membranes absorb UV light, whether sunlight or a lamp's glow, the ergosterol in them changes into vitamin D₂ (Sharma *et al.*, 2024; Cardwell *et al.*, 2018). They supply bioavailable vitamin D that may enhance insufficient vitamin D levels, promote bone health, and potentially confer supplementary health advantages.

For vegetarians, vegans, and others who avoid animal products, edible mushrooms are a good source of vitamin D (Kaur *et al.*, 2025). This makes mushrooms naturally "fortified." Vitamin D helps

bones, calcium metabolism, and immunity. It may also reduce inflammation and metabolism. People worldwide don't receive enough vitamin D because they don't get enough sun, protect their skin, live in a certain area, or consume enough. When mushrooms sit under UV light sunshine or a lamp they turn their ergosterol into vitamin D₂. Edible mushrooms are a great choice they naturally contain ergosterol, a compound that turns into vitamin D₂ (ergocalciferol) when exposed to UV-B or UV-C light, like sunbeams warming a kitchen counter. People are increasingly interested in eating mushrooms to boost their vitamin D levels, especially after a study by Judprasong *et al.* (2023) highlighted their potential benefits. Thus, UV-treated mushrooms are being considered as a means to gain vitamin D from food, especially for vegans, vegetarians, and others who don't get much sunlight.

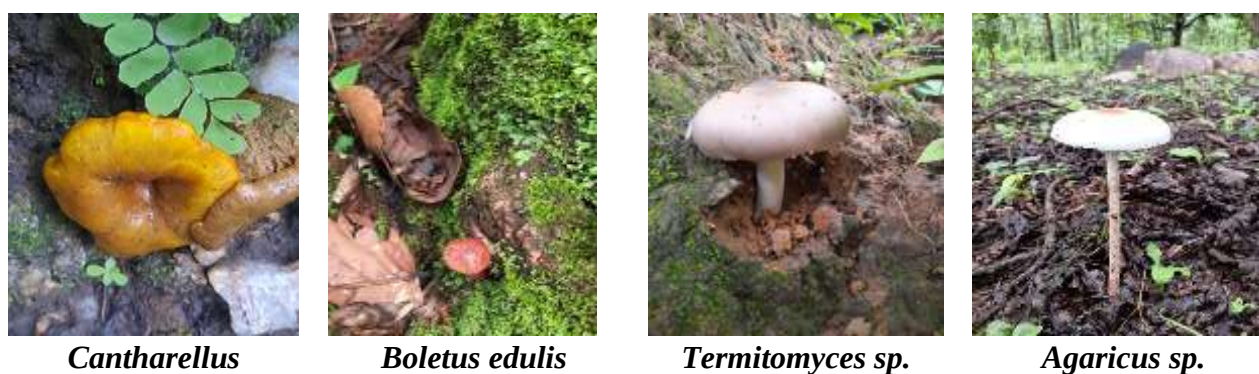


Fig. 1. Mushrooms in natural forest ecosystem, Tropical Forest Research Institute (TFRI), Jabalpur, Madhya Pradesh, India



Fig. 2. Marketing of local edible mushrooms by tribals at Mandla, Madhya Pradesh, India

Rondanelli *et al.* (2023) posited that the consumption of UV-irradiated mushrooms may serve as an effective dietary source of vitamin D. In 2023, researchers suggested that eating mushrooms exposed to UV light could be a practical way to boost vitamin D. Think of those pale caps warmed briefly under a lamp. The review, however, stressed that differences in mushroom species, UV treatment methods, storage conditions, cooking methods, participants' baseline vitamin D levels, and the length of the

studies make it hard to compare the results (Rondanelli *et al.*, 2023). An article in *Frontiers in Nutrition* said that we need stability data and consistency in mushroom UV treatments. It also said that we need lengthier human studies to show that they work (Starck *et al.* 2024). Stephensen *et al.* (2012) found that UV-treated white button mushrooms increased serum 25(OH)D₂ levels in healthy persons over six weeks but had negligible impact on vitamin D status. In a randomized controlled trial, people with pre-diabetes and low vitamin D who ate 100 grams of UVB-treated mushrooms each day for 16 weeks improved their vitamin D levels (Mehrotra *et al.*, 2014).

In a human bioassay experiment, wild edible mushrooms (*Cantharellus tubaeformis*) yielding ~14 µg ergocalciferol/day boosted serum 25(OH)D levels similarly to supplemented D₂ (Outila *et al.*, 1999). Recent clinical trials and controlled feeding studies have examined vitamin D status after daily or weekly consumption of UV-treated mushrooms or mushroom powders. Participants who took part in trials and ate UV-treated mushroom products for weeks or months had higher levels of 25(OH)D₂. Some studies demonstrate that 25(OH)D₂ goes up while 25(OH)D₃ goes down. This means that the overall amount of 25(OH)D doesn't vary as much as expected when the same amount of vitamin D₃ is given. Mushroom-derived vitamin D₂ is bioavailable, although it may not be as effective as equal doses of D₃ at raising total vitamin D levels. Recent research corroborates the notion that both D₂ and D₃ can elevate blood levels, with D₃ exhibiting greater potency and stability over time (van den Heuvel *et al.*, 2024). Animal studies show that eating mushrooms with vitamin D₂ makes bones stronger, better shaped, and more mineralized. Experiments on humans are uncommon but promising. Human trials are infrequent, however animal studies offer mechanistic insights. In animal models (rats, mice), meals containing UV-treated mushrooms or vitamin D-enriched powdered extracts elevate serum 25(OH)D levels, improve bone mineralization and strength, and modify gene expression related to vitamin D metabolism (CYP2R1, CYP27B1, and VDR) (Malik *et al.*, 2022; Rondanelli *et al.*, 2023). Some studies indicate anti-inflammatory or hepato-protective advantages in models of induced liver injury.

A major practical issue is how much of the synthesized vitamin D₂ stays intact after normal cooking, drying, and rehydration, as well as during storage (refrigeration and shelf life). Recent food chemistry research, especially in Thailand and a few other places, has measured how much vitamin D₂ mushrooms actually keep after everyday cooking like when they're lightly sautéed in a pan. Studies show that everyday ways of cooking mushrooms like sautéing or roasting keep about 60% to 100% of their vitamin D₂ (Sridonpai *et al.*, 2025; Kapcum *et al.*, 2025; Taofiq *et al.*, 2017). Industrial UV-B irradiation procedures have been enhanced to ensure consistent vitamin D₂ production in mushrooms like shiitake, oyster, and white button. Hu *et al.* (2020) and other research provide process-optimization parameters (exposure length, lamp intensity, cap orientation) for commercial

scaling. Ergosterol baseline levels vary by species, UV penetration by thickness, and orientation, making standardization necessary for consistent output. Over the past five years, research has focused on Kapcum *et al.* (2025); Malik *et al.* (2022); Rondanelli *et al.* (2023) have been focused on the following

1. Mushroom vitamin D production through UV treatment
2. Human 25(OH) D levels.
3. Cooking and storage effects
4. Safety and regulatory concerns for sold UV-treated mushroom products.

As commercial UV-treated mushroom products (powders, entire mushrooms, and fortified foods) grow increasingly popular, they must be evaluated properly. EFSA issued its full conclusion on the safety of vitamin D₂ mushroom powder made from *Agaricus bisporus* exposed to UV light that gives the caps a faint golden tint. EFSA assessed that this mushroom powder is safe at specific doses if produced and labelled properly (EFSA, 2024). This is an important step towards European authorities approving UV-treated mushroom products for sale. This position mimics safety evaluation and approved applications, although US, Indian, *etc.* regulations may differ.

Conclusion

Vitamin D deficiency is common across India; millions go without enough sunlight warming their skin. Still, the diagnosed cases are just the tip of the iceberg, like the small white peak you see above dark, freezing water. Knowing the many ripple effects it could trigger, we can picture the heavy burden, like a weight pressing on every corner, that this silent epidemic would place on the country's growth. We need to tackle vitamin D deficiency with real focus and firm action, like stepping out into the morning sun instead of ignoring the problem. Mushrooms are a key food in public health efforts to combat vitamin D deficiency; they hold onto plenty of the vitamin even after cooking and fit easily into a plant-based meal, whether tossed in a quick stir-fry or layered on toast. It is imperative that researchers focus on standardized UV procedures, long-term human studies, and models of mushroom intake to impact public health in the future. Adding vitamin D to foods and taking supplements both fuel growth like a steady light that helps young bones strengthen. Adding nutrients to everyday foods can boost vitamin D levels for everyone like milk fortified to catch a bit of sunshine in each glass. The use of bone minerals and vitamin D as fortifiers in food or dietary supplements for older infants and toddlers is likely to be both useful and safe in many countries. Hence the fact that sunlight exposure along with vitamin D fortified food supplements paves way for curing hypovitaminosis D on the whole.

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TRANSGENIC MODELS IN TOXICOLOGY

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Transgenic animal models are powerful tools for developing a more detailed understanding on the roles of specific genes in biological pathways and systems. Applications of these models have been made within the field of toxicology, most notably for the screening of mutagenic and carcinogenic potential and for the characterization of toxic mechanisms of action. It has long been a goal of research toxicologists to use the data from these models to refine hazard identification and characterization to better inform human health risk assessments. This review provides an overview on the applications of transgenic animal models in the assessment of mutagenicity and carcinogenicity, their use as reporter systems, and as tools for understanding the roles of xenobiotic-metabolizing enzymes and biological receptors in the etiology of chemical toxicity. Perspectives are also shared on the future outlook for these models in toxicology and risk assessment and how transgenic technologies are likely to be an integral tool for toxicity testing in the 21st century.

Key Words: Transgenic models; Knock-out models; Humanized models; Risk assessment.

INTRODUCTION-

The development of transgenic animals has already had a significant impact in biomedical research. Non-human models of human diseases (i.e., sickle cell disease, cystic fibrosis and AIDS) have been produced in mice and are being used to screen potential therapies. Domesticated animals genetically engineered to: (a) resist disease, (b) produce meat or eggs with less cholesterol, or (c) accelerate growth may be introduced into agriculture in the near future. Transgenic animals have also been proposed as living bioreactors for the commercial production of pharmaceuticals and other biologicals not easily synthesized by other method. Transgenic models which fit into chemical safety assessment schemes consist of those used to screen for toxicity (i.e., detection of carcinogens and mutagens) and those used to elucidate mechanisms of toxicity.

CURRENT ROLES FOR TRANSGENIC ANIMAL MODELS IN TOXICOLOGICAL ASSESSMENT-

Role	Process	Example
Screening	Development of susceptible animals which can quickly detect toxic agents	OncoMouse MutaMouse™ PIM®Mouse
Elucidation of Mechanisms	Use of gene targeting to study toxicity response mechanisms	Ahcomplex Hoxcomplex

To date, all commercial models are limited to screens for mutation (Gossen and Vijg, 1993) or tumors (Sullivan et al., 1993) and have been constructed in mice. The use of the mouse as the host animal for the transgenes in toxicology screening models is the consequence of several critical factors:

1. The mouse pronucleus is more accessible than the rat pronucleus.
2. Embryonic stem cell cultures can only be produced in mice.
3. Large gene libraries are available for mice, but not for rats.
4. Short gestation time and large litter size facilitates the use of mice.

From a toxicological perspective, rat models would be substantially more useful, as a larger body of toxicity data is available in the rat. Comparative risk assessments and mechanistic extrapolations could be more informative if results from rat transgenic models of carcinogenesis and mutagenesis were also available. A rat model of Stratagene's Big Blue™ transgenic model for mutation detection is reportedly close to commercialization (Sullivan et al 1993).

XENOBIOTIC METABOLISM MODELS-

The cellular constitution of xenobiotic enzymes is vital for determining whether a chemical will be metabolized to induce a toxic response. In order to determine the role of metabolizing enzymes in mammalian development, physiological homeostasis, and xenobiotic metabolism, several mouse lines lacking xenobiotic-metabolizing enzymes, such as the P450s Cyp1a1, Cyp1a2, Cyp2e1, microsomal epoxide hydrolase, glutathione S-transferase, or NADPH-quinone oxidoreductase, have been developed (Table 3). In general, these models are viable and show no developmental abnormalities which generally suggests a redundancy in xenobiotic-metabolizing enzymes and that their roles are primarily involved in metabolism of foreign compounds. However, the Cyp4a10 and Cyp4a14 P450s metabolize fatty acids and have a role in control of blood pressure because of their arachidonate monooxygenases activities as revealed with the knockout mice (Holla et al., 2001; Nakagawa et al., 2006). The various P450-null mouse lines have firmly established that these

enzymes influence the metabolism, toxicity, and carcinogenicity of xenobiotic in-vivo. The use of xenobiotic enzyme-null transgenic mice has provided great insights in toxicology by enabling the identification of the major metabolic enzymes involved in toxicant biotransformation in vivo. In addition, new metabolic pathways important for toxicant biotransformation have also been elucidated through the use of these models. Mice lacking expression of P450s known to be involved in metabolism of carcinogens and toxins show marked resistance to cancer and toxicity upon chemical treatment. For example, Cyp1b1-null mice are resistant to 7,12-dimethylbenz[a]anthracene-induced ovarian cancers (Buters et al., 2003). Mice lacking expression of Cyp2e1 are resistant to acetaminophen (APAP) induced hepatotoxicity (Lee et al., 1996) and benzene-induced myelotoxicity (Valentine *et al.*, 1996). These protective effects are compatible with the known role of Cyp2e1 in the conversion of APAP to the electrophilic derivative N-acetyl-p-benzo-quinone-imine and production of the reactive benzene epoxide, respectively. In contrast, Cyp1a1-null mice are more susceptible to benzo[a]pyrene (BaP)-induced toxicities indicating inducible Cyp1a1 functions in the detoxification and protection against oral BaP (Uno et al., 2004). This result was not predictable because Cyp1a1 can also metabolically activate BaP as revealed in vitro. Thus, in the gut, after oral administration, Cyp1a1 primarily functions to inactivate this procarcinogen through ring hydroxylation. There are numerous other examples of how the in-vivo roles of P450s in the metabolism, toxicity, carcinogenicity of xenobiotics has been determined through the use of gene knockout mice. Sensitivity and Positive Predictivity of Transgenic Rodent Assays in Comparison with Other Short-Term In-Vivo Assays for Chemicals Listed in the Database (data derived from the analysis by Lambert et al., 2005).

METHODS FOR THE PRODUCTION OF TRANSGENIC ANIMALS-

Transgenic animals are produced by the introduction of foreign DNA (genes) into their genomes using three different approaches. Depending upon the method used, the transgene will become stably integrated into the host genome at selective sites or at random. Three methods used in the production of transgenic models are summarized in Table 2. Engineered animals can be made homozygous for the integrated DNA transgene through selected breeding, with the result that the transgene is transmitted to future generations. Integration of the foreign DNA can be confirmed through the use of probes specific for the transgene DNA. The first method uses viral shuttle vectors for the production of transgenes, and is limited to the introduction of relatively small segments of DNA (8-10 kilobases). This method has been virtually replaced by the introduction of direct pronuclear injection and recombinational insertion into embryonic stem cell (ES) cultures, which can introduce large segments of DNA. In the second method, the DNA construct is injected into either the male or female pronucleus in fertilized ova (Figure I). Single or multiple tandem concatemers of the

transgene are integrated at sites located randomly along the mouse chromosome. (Figure 2). Expression of the transgene along the chromosome (active or inactive) is dependent upon either the insertion site or the presence of regulatory elements (tissue specific or general). Selection of a tissue specific controlling element for combination with the transgene (fusion genes) will determine the specific tissue or organ in which the transgene is expressed. The third method involves the use of embryonic stem (ES) cells. ES cells are pluripotent embryonic cultures that can be reintroduced into a normal mouse blastocyst resulting in chimeric mice with ES cells forming with somatic and germinal tissues (Figure 3). This property of ES cells makes them ideal for producing transgenic mice. ES cell cultures can be genetically modified by transfection of the cells with transgene.

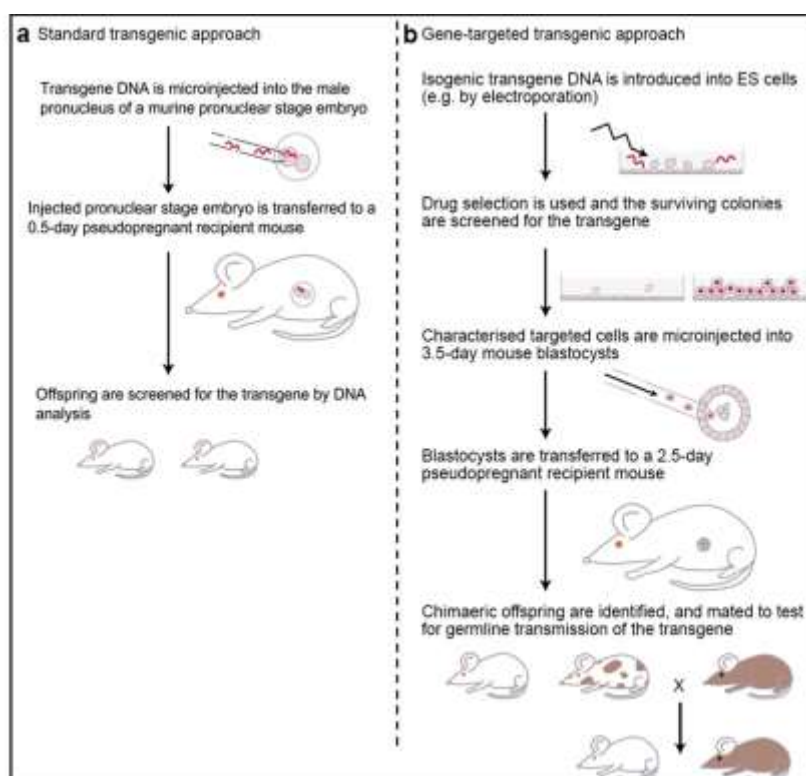


Fig- Classical methods to produce transgenic mice (a) standard transgenic mice (b) Gene targeted transgenic approach

TRANSGENIC RECEPTOR MODELS

A selection of ligand-activated receptor transcription factors that respond to xenobiotics include the AhR, constitutive androstane receptor (CAR, NR1I3), farnesoid X receptor (FXR, NR1H4), PXR (NR1I2), and peroxisome proliferator– activated receptor (PPAR) α (NR1C1). The latter receptor is part of the PPAR family trio, including PPAR β (NR1C2) and PPAR γ (NR1C3), which are critical for metabolic control of fatty acid and glucose homeostasis. CAR, FXR, PXR, and PPAR α all belong to the metabolic sensor class of nuclear receptors that alter metabolism of xenobiotics and endogenous

chemicals such as fatty acids, cholesterol, and bile acids (Gonzalez and Shah, 2008; Modica et al., 2009). They contain the ligand-binding, DNA-binding, transcription activator, and coactivator/corepressor-binding domains found in the nuclear receptor superfamily. Typical of all type 2 nuclear receptors, CAR, FXR, PXR, and PPARα require the retinoid X receptor as an obligate heterodimerization partner for activation of gene expression. The heterodimers can bind to specific direct repeat (DR) elements located upstream of target genes and, through interaction with coactivators and other components of the transcriptional machinery, activate target gene expression. Unlike the relatively specific binding of the type 1 steroid hormone receptors (i.e., ER, progesterone receptor, glucocorticoid receptor), CAR, FXR, PXR, and PPARα are capable of being activated by structurally diverse ligands including xenobiotics. These receptors, once activated by ligand, induce the expression of a battery of target genes, including many P450s, which have effects on the metabolism of both endogenous and exogenous chemicals.

OUTLOOK ON TRANSGENIC MODELS IN TOXICOLOGY AND RISK ASSESSMENT-

Transgenic models have certainly had an important impact on the field of toxicology, and there are many factors that are likely to influence and expand the use of these models in both basic toxicology research and risk assessment. In the past, the mouse has been the predominant animal model for generation and subsequent research with transgenic models, owing largely to the success of ESC-based targeting technology (Capecchi *et al.* 2005). Such an approach has not been amenable to application in other species such as the rat; however, recent approaches have been described that will open the door for the creation of a more diverse set of transgenic models. For example, the microinjection of specifically designed zinc finger nucleases into embryos have been used to introduce nonhomologous endjoining deletions or insertions at a target gene site to generate knockout rats and mice (Carbery *et al.*, 2010). More recently, this approach has been extended in both species to introduce sequence-specific modifications through homologous recombination. The approach enables precise alteration to the genome to generate insertions, deletions, and even point mutations in a whole animal system. Such approaches will clearly pave the way for more expanded research using transgenic models that will include extension into species beyond the mouse and allow for alterations that may more accurately mimic the genetic makeup of the human population. Conceivably, this could also be applied to understand and compare human susceptibilities based on known polymorphisms in the human population. Although this review has focused solely on rodent transgenic models, another expanding and promising area of research surrounds the use of alternative model systems such as zebrafish and *Caenorhabditis elegans*, both as screening tools for toxicity assessment and in MOA-based research. In zebrafish, various approaches to manipulate the genome have been used to generate both knockout and knock-in models that have been applied extensively to

characterize vertebrate development and have served as models for research on human diseases (Yang *et al.*, 2009). Transgenic zebrafish have also been engineered to express fluorescent reporter genes in response to tissue damage, oxidative stress activation, or other indicators of toxicity that could be used in a toxicity screening approach or MOA research (Yang *et al.*, 2009). Similar applications have been published for *C. elegans* including transgenic reporter systems for biomonitoring and model systems for researching neurodegenerative disease. There have been widespread discussions in recent years on the need to overhaul our approaches to toxicity testing and risk assessment with the application of new technologies that will help to streamline and increase the human relevance of the process. One of the key drivers for these discussions has been the U.S. National Research Council (NRC) report—Toxicity Testing in the 21st Century: A Vision and a Strategy (NRC, 2007). The United States Environmental Protection Agency’s MOA framework and Strategic Plan for Evaluating the Toxicity of Chemicals is also helping to facilitate change in the risk assessment process for environmental chemicals (U.S.EPA, 2009). According to EPA, the purpose of these plans is to serve as a blueprint for incorporating advances in molecular biology and computational sciences into toxicity testing and risk assessment practices across the EPA. The NRC report and the EPA strategy both outline the promise that the new technologies could dramatically increase both the numbers of chemicals comprehensively evaluated while also broadening and improving the human relevance of the assessed toxicity endpoints. According to the NRC report, this would be accomplished in the not-so-distant future through the use of human cells and cell lines by evaluating cellular “toxicity pathways” using high-throughput testing approaches. Risk assessments would be based on the results of these assays and focus on avoiding perturbations in key cellular pathways as assessed through comprehensive dose-response analysis and extrapolation to human-relevant exposure conditions. Such outcomes, if successfully achieved, would significantly improve and inform science-based health assessments on a broad range of chemicals. However, further reflection has indicated that such a change will require a strategic and systematic transition with a continued reliance on whole animal testing in the near-term (Andersen and Krewski, 2010). Such a transition could certainly benefit from the strategic implementation of transgenic models as part of an integrated testing strategy. Transgenic models have the potential to serve as important tools in this process including, but not limited to, characterization of the MOA, decreasing uncertainty in cross-species extrapolations, and defining the shape of the dose-response and the existence of thresholds, all with the ultimate goal of establishing more relevant measures of human risk. With the ever increasing ability to create transgenic models with greater ease and within a wider range of research species (both rodent and nonrodent), our ability to characterize the biological pathway perturbations that lead to toxicity will certainly increase. This in turn will foster better mechanistic understandings

within the field of toxicology which will enable the subsequent development of more relevant and specific in vitro assays for predicting toxicity that will help to deliver on the vision of the NRC report, the EPA Strategic plan, and other international initiatives focused on streamlining the risk assessment process while improving human relevance and decreasing animal use. The application of these models to human risk assessment will require appropriate and consistent interpretation of the data, recognition of the limitations of these models, and acceptance of the resultant data and its relevance to humans. Such progress will not be achieved without concerted efforts and focused communication from all stakeholders (Andersen and Krewski, 2010), most notably between research toxicologists and regulatory toxicologists to ensure that these new technologies can be applied and accepted within a regulatory context. The field of toxicology and risk assessment has entered an exciting period wherein the biological tools to streamline and advance the human relevance of toxicology testing are available. What is now required are creative and consensus approaches for their successful application to risk assessment.

CONCLUSION

A great deal of effort is underway to characterize genes associated with disease processes. In order to establish the function(s) of individual genes, much of the research involves the biological consequences of gene mutation and subsequent alterations in gene expression. The ability to alter the genome by transgenic mouse models provides a powerful approach to examine the contribution of specific genes in a variety of toxicological endpoints. These mouse models may prove to be excellent systems for studying normal gene function and the consequence of alterations, since they are easily experimentally manipulated and can be studied with classical genetic techniques. Transgenic animals are constructed using molecular genetics techniques to produce animals that have functional genes added to the genome, or contain marker genes to assess in vivo effects, or have alterations of specific genes believed to be involved in disease processes. The availability of transgenic mice permits the in vivo assessment of the role of specific genes in chemically induced genetic or nongenetic alterations. A large number of transgenic animal models have been developed for studying cancer processes. At present, many of these animals are used for studies of carcinogenesis, since they permit the in vivo assessment of gene mutation, contain activated oncogenes, or contain inactivated tumour suppressor genes that are believed to be involved in tumour development. Ultimately, transgenic animals may aid in identifying critical genetic alterations required for specific tumour formation and how chemical carcinogens can act to modulate these steps. Applications for transgenic mice in the field of toxicology and chemical carcinogenesis have been developed. For example, transgenic mouse systems have demonstrated the relationship between a specific gene alteration and the appearance of specific neoplasias. Such mice show a high susceptibility to the development of spontaneous tumours

at various tissue sites. However, it is clear that single gene alterations on their own are not sufficient to cause tumour development. Identification of secondary events, such as induction or activation of cooperating genes, is an area for future research. There are several important considerations in the use of transgenic animal models for toxicology research that are raised in this report. A major question is to define what can be learned through the use of transgenic models that cannot already be learned from existing rodent systems. There is no doubt that the ability to identify some carcinogens will be improved and the unique sensitivity of transgenic mice will provide insight on dosage and tissue specificity. However, it is clear that data with transgenic mice have been obtained only recently and a number of questions concerning transgenic mouse models need to be answered so that such data can be placed into proper context. Certainly transgenic animals have provided a powerful toxicological tool for studying *in vivo* chemical effects and have increased our understanding of the role of specific genetic alterations as predisposing factors for chemical carcinogenesis. As advances are made in the characterization of a number of transgenic models, the future challenge for toxicology is to appropriately incorporate the data for improving estimations of human health risks.

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Principles and Importance of Clean Milk Production

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1. Introduction: Milk Quality and Global Public Health

1.1. Milk as a Perfect Medium: Milk is arguably nature's most complete food, providing a rich array of proteins (casein and whey), fats, essential vitamins (A, B₂, B₁₂ and D) and minerals (calcium and phosphorus). This nutritional completeness, however, is precisely why milk is also an ideal culture medium for a wide range of microorganisms, including pathogenic bacteria. Consequently, milk quality and safety are intrinsically linked to public health, global trade, and the sustainability of the dairy sector.

1.2. Defining Clean Milk Production: Clean milk production (CMP) refers to a systematic approach encompassing all stages of the dairy chain—from the health of the dairy animal and the hygiene of the milking environment to the rapid cooling and storage of the final product—to ensure the milk produced is safe, wholesome, and of high quality. The goal is to minimize contamination by bacteria, physical debris, and chemical residues, ensuring the product retains its nutritional value, extends its shelf life, and meets both regulatory and consumer standards (Hayes et al., 2018).

1.3. Importance: A Three-Pillar Framework: The importance of CMP transcends mere compliance; it rests on three pillars:

1. **Public Health:** Preventing foodborne diseases and zoonoses.
2. **Economic Viability:** Ensuring product longevity, reducing waste, and facilitating market access.
3. **Dairy Sustainability:** Reducing antibiotic usage and promoting animal welfare.

This chapter details the foundational principles of clean milk production, outlines the primary sources of contamination, provides a comprehensive step-by-step guide to implementation, and explores the economic and public health implications of maintaining high standards.

2. The Science of Contamination: Sources and Risks: To achieve clean milk, one must understand the origin and nature of potential contaminants, which can be biological, chemical, or physical.

2.1. Biological Contamination: The Bacterial Load: Milk, as secreted from a healthy udder, is virtually sterile. Contamination begins immediately upon secretion and primarily determines the milk's quality grading.

2.1.1. Primary Source: The Udder and Mastitis: The most significant bacterial contamination source is the infected udder, primarily through mastitis. Mastitis, an inflammation of the mammary gland, is usually caused by bacteria such as *Staphylococcus aureus*, *Streptococcus agalactiae*, and *Escherichia coli*.

- **Significance:** Mastitic milk contains high concentrations of pathogenic bacteria and somatic cells (leukocytes or white blood cells). The Somatic Cell Count (SCC) is the gold standard indicator for udder health and milk quality. High SCC is associated with lower milk yield, altered protein/fat ratios, and reduced processing efficiency (Ruegg, 2017).

2.1.2. Secondary Sources: Environment and Equipment: Once milk leaves the teat, it is exposed to secondary contamination:

- **Milking Environment:** Dust, soil, and manure are sources of environmental bacteria (e.g., *E. coli*, *Klebsiella*).
- **Personnel and Equipment:** Poorly sanitized milking machines (e.g., hoses, liners, bulk tanks) provide surfaces for biofilm formation, creating reservoirs for thermotolerant (heat-resistant) and psychrotrophic (cold-loving) bacteria (Hayes et al., 2018). Psychrotrophic bacteria (e.g., *Pseudomonas* species) are critical; they grow slowly even at refrigerated temperatures and produce heat-stable enzymes (lipases and proteases) that break down fat and protein, leading to off-flavors and spoilage even after pasteurization.

2.2. Chemical Contamination: Residues and Adulterants: Chemical residues pose a direct threat to public health and international trade.

- **Antibiotic Residues:** These result from inadequate withdrawal periods following treatment for mastitis or other infections. Antibiotics in milk pose two major risks: they cause allergic reactions in sensitive consumers, and, more critically, they inhibit the starter cultures used in dairy processing (cheese, yogurt), and contribute to the global crisis of Antimicrobial Resistance (AMR) in human populations (WHO, 2017).
- **Pesticide and Heavy Metal Residues:** Contamination can occur through contaminated feed or water.
- **Adulterants:** Deliberate addition of substances (e.g., urea, melamine) to increase volume or falsely enhance apparent protein content is a severe, though geographically variable, public health hazard (Food Safety Authority of India, 2019).

3. Principles of Clean Milk Production: The Five Pillars of Hygiene: Clean milk production is achieved by adhering to five core principles that target and control the primary sources of contamination.

3.1. Healthy Animal Management (The Source): The health of the animal dictates the initial bacterial load.

- **Mastitis Prevention and Control:** Implementation of the Five-Point Mastitis Control Plan (Post-milking teat dipping, dry cow therapy, prompt treatment, culling chronic cases, proper milking machine function) is essential. Regular screening using the California Mastitis Test (CMT) or bulk tank SCC monitoring is necessary to identify subclinical cases (Ruegg, 2017).
- **Vaccination and Biosecurity:** Regular vaccination against common diseases and strict biosecurity protocols (e.g., restricted access, quarantine of new animals) minimize the overall disease burden.

3.2. Hygienic Milking Environment (The Prevention): The milking parlor or shed must be designed and maintained to prevent fecal and soil contamination.

- **Cleanliness:** Floors and walls should be non-porous and easy to clean. Adequate bedding and regular scraping prevent the build-up of manure, which harbors high levels of *E. coli* and other environmental mastitis agents.
- **Ventilation:** Good air circulation minimizes dust and controls humidity, inhibiting microbial growth.

3.3. Milking Hygiene (The Act): Procedures immediately before and during milking are critical to preventing external bacteria from entering the milk stream.

- **Pre-milking Udder Preparation:** The process must be standardized:
 1. **Fore-stripping:** Drawing a few squirts of milk into a strip cup to check for clinical mastitis and flush high-bacterial-load milk from the teat canal.
 2. **Cleaning and Drying:** Washing the udder with a sanitizer solution and, crucially, thoroughly drying the teats with a single-use towel. Wet teats are major sources of contamination (Hayes et al., 2018).
 3. **Attachment:** Attaching the milking machine within 60-90 seconds of stimulation to maximize oxytocin release and ensure complete milk let-down.

3.4. Equipment Cleanliness (The Vehicle): The milking equipment (machines, buckets, tanks) acts as the primary vehicle for microbial transfer.

- **Washing Protocol:** A strict, multi-step Clean-in-Place (CIP) protocol for pipeline systems or manual washing for bucket systems is mandatory:
 1. **Pre-rinse:** Cold or lukewarm water removes residual milk solids (prevents protein denaturing).
 2. **Detergent Wash:** Hot water (60-70 °C) with an alkali detergent (to break down fat and protein) followed by an acid detergent (to remove mineral deposits and scale).
 3. **Sanitization/Disinfection:** A chemical sanitizer (e.g., hypochlorite or iodophor solution) immediately before the next milking to kill residual bacteria.
- **Maintenance:** Regular inspection and replacement of rubber parts (liners, hoses) are vital, as worn rubber harbors bacteria and affects milking efficiency.

3.5. Rapid Cooling and Storage (The Preservation): The rate of bacterial multiplication is a function of temperature. CMP dictates the rapid inhibition of microbial growth.

- **Immediate Cooling:** Milk must be cooled to 4°C or below as quickly as possible (ideally within two hours of milking). Failure to cool drastically increases the total bacterial count (TBC) (Ruegg, 2017).
- **Storage:** Milk must be stored in a clean, insulated, and agitated bulk tank to maintain temperature and ensure homogeneity until collection.

4. Importance of Clean Milk: Public Health and Economic Imperatives: The strict adherence to CMP principles yields quantifiable benefits across public health and economic spheres.

4.1. Public Health Protection

4.1.1. Prevention of Foodborne Illnesses: Milk contaminated with pathogens is a significant vehicle for foodborne diseases. Clean milk production prevents the transmission of critical pathogens, including:

- ***Salmonella* and *E. coli* O157:H7:** Often introduced via fecal contamination.
- ***Listeria monocytogenes:*** A psychrotroph that can grow at refrigeration temperatures and is particularly dangerous to pregnant women, infants, and the immunocompromised.
- ***Brucella abortus* and *Mycobacterium bovis* (TB):** Zoonotic pathogens primarily controlled through animal health management (Principle 3.1). Although pasteurization eliminates these risks, CMP minimizes the initial contamination burden.

4.1.2. Combating Antimicrobial Resistance (AMR): By reducing the incidence of mastitis (Principle 3.1), CMP directly reduces the reliance on antibiotics. Fewer treatments mean less antibiotic residue in the milk and, more importantly, less selection pressure for resistant bacteria, making CMP a critical strategy in the global fight against AMR (WHO, 2017).

4.2. Economic Sustainability and Market Access: Milk quality is the key determinant of its market value and utility.

4.2.1. Increased Shelf Life and Reduced Spoilage: Low bacterial counts (low TBC) significantly extend the storage life of both raw and pasteurized milk. Psychrotrophic bacteria are especially critical here; controlling them prevents the enzymatic breakdown of milk components, thus preserving flavour and stability (Hayes et al., 2018). This reduces post-harvest losses, which is vital for profitability.

4.2.2. Enhanced Processing Efficiency: High-quality milk is essential for manufacturing dairy products.

- **Cheese Yield:** High SCC and high TBC milk often have altered protein structures, leading to reduced curd tension and lower cheese yield.

- **Shelf Stability:** Off-flavors and reduced stability in products like yogurt, UHT milk, and dried milk powder are often traceable to the heat-stable enzymes produced by psychrotrophs in poor quality raw milk.

4.2.3. Facilitating Trade and Premium Pricing: Dairy exports are heavily regulated, with international trade barriers often based on stringent quality parameters (e.g., maximum limits for SCC, TBC, and antibiotic residues). Countries or producers that adhere to these standards gain access to high-value export markets, often receiving premium prices for their milk (Ruegg, 2017).

5. Quantitative Measures and Regulatory Standards: Milk quality is a measured science, relying on laboratory testing to ensure compliance with standards.

5.1. Key Quality Parameters

Parameter	Unit	Significance	Typical EU/US Standard (Bulk Tank)
Total Bacterial Count (TBC)	cfu/ml	Overall hygiene of milking and cooling.	<100,000 cfu/ml
Somatic Cell Count (SCC)	cells/ml	Udder health and mastitis incidence.	<400,000 cells/ml
Inhibitory Substances (Antibiotics)	ppb	Presence of antibiotic residues.	Zero tolerance (Negative test)
Freezing Point	°C	Indicator of water adulteration.	≤ -0.525°C

5.2. Regulatory Frameworks: Different regulatory bodies (e.g., FDA in the US, CFIA in Canada, and the EU's Council Directives) set stringent standards. These standards are not static; they have trended toward lower SCC and TBC limits over the last three decades, driving continuous quality improvement globally. For example, the EU standard for raw milk is exceptionally strict, reflecting a zero-tolerance policy for substandard inputs (Hayes et al., 2018).

6. Challenges and Future Directions in Clean Milk Production: While the principles of CMP are universal, implementation faces significant challenges, particularly in smallholder and low-resource settings.

6.1. Technical and Infrastructure Gaps: The most significant barrier is the lack of a cold chain (Principle 3.5). In many developing regions, unreliable electricity and high costs prevent rapid cooling. Future solutions require innovation in solar-powered bulk coolers and the development of local chilling centres.

6.2. Knowledge and Behavioural Change: A critical element is the human factor. Even with proper equipment, lapses in hygiene protocols (e.g., using a dirty cloth to dry teats, insufficient CIP time, failure to observe withdrawal periods) undermine quality. Effective extension services and farmer training programs are essential to foster the necessary behavioural change (Food Safety Authority of India, 2019).

6.3. Technology: Rapid Testing and Traceability: The future of CMP lies in rapid, on-farm testing technologies (e.g., quick enzyme-based tests for antibiotics) and blockchain-based traceability systems. These technologies can provide real-time assurance of quality, empowering farmers and providing irrefutable proof of compliance to consumers and regulators.

7. Conclusion: Clean milk production is a non-negotiable prerequisite for a safe, profitable, and sustainable dairy industry. It is a comprehensive system built upon five critical pillars: healthy animals, hygienic environment, strict milking hygiene, rigorous equipment sanitation, and rapid cooling. Adherence to these principles directly safeguards public health by minimizing pathogenic and chemical risks, while simultaneously driving economic efficiency through reduced spoilage and enhanced market access. As the global demand for dairy continues to rise, the commitment to the principles and implementation of Clean Milk Production will remain the ultimate benchmark for industry excellence.

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Gut Health as a Keystone of Animal Productivity: Nutritional Insights and Research Directions

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1. Introduction

In modern livestock- and animal-production systems, the gastrointestinal tract (GIT) has emerged as far more than merely a conduit for feed. Rather, it sits at the interface of nutrition, metabolism, immunity, behaviour and welfare. Gut health is central to animal performance, welfare and environmental sustainability. It integrates digestion and nutrient absorption, immune competence, barrier integrity, and the complex ecosystem of resident microbes. Nutrition is the primary modifiable factor that shapes gut structure and function — through substrate supply to the host and to gut microbes, by affecting gut luminal environment, and by providing bioactive compounds that modulate host physiology.

Gut function is closely related to the effectiveness of feed usage, the strength of immune defenses, the ability of animals raised for meat, milk, eggs, or other by-products to withstand environmental, nutritional, or managerial stress, and ultimately the product's quality. From the perspective of nutrition, optimizing gut health means not only providing the right nutrients but also enabling the gut ecosystem to perform, adapt and buffer challenges. In doing so, production systems can achieve greater output with fewer inputs, better animal welfare, and lower environmental footprint.

2. The Gut Ecosystem: Structure, Function and Microbiome

2.1 Anatomy and Physiology of the Gastro-intestinal Tract

Animal digestive tracts differ greatly between species; ruminants (cattle, sheep, and goats) have a foregut fermentation chamber (rumen) followed by small and large intestines, whereas monogastric species (pigs, poultry, and rabbits) depend more on enzymatic digestion and, to varying degrees, hindgut fermentation. Because of these physiological and anatomical differences, the idea of gut health needs to be modified for the particular species.

In ruminants, the rumen microbial fermentation of plant-based feeds produces volatile fatty acids (VFAs) which provide a large portion of energy to the animal; the downstream intestines then absorb amino acids and remaining nutrients. In monogastrics, the small intestine is the primary site of nutrient absorption, while the large intestine or cecum may house important fermentation processes, especially of fibres or resistant starches.

Beyond simple digestion, the GIT creates a barrier consisting of a mucus layer, tight-junctional epithelium, an immune cell network (gut-associated lymphoid tissue, or GALT), a mucosal lining, and a consortium of microorganisms. In addition to controlling nutrition intake, this barrier also controls immunological signaling, allergen exposure, pathogen exclusion, and even neuroendocrine reactions.

2.2 The Gut Microbial Community: Composition, Functions and Development

The gut is colonized by a complex microbial ecosystem comprising bacteria, archaea, fungi, protozoa and viruses. In livestock, this community is increasingly recognized as a key determinant of health and productivity. Differences in gut microbial composition are associated with differences in feed efficiency, disease susceptibility, immune status and behaviour.

The phyla Firmicutes and Bacteroidetes dominate the rumen microbiome in ruminants, and fermentation is driven by specialized functional guilds (cellulolytic, proteolytic, and methanogenic). Although the microbial load and composition of digestive tract segments vary considerably in monogastrics, the large intestine microbiota frequently plays important roles in the fermentation of non-digestible carbohydrates and the production of short-chain fatty acids (SCFAs).

Crucially, the development of the microbial community is dynamic: the gut's morphological development (villus length, crypt depth, gut motility), immune maturation, and subsequent productivity are all influenced by the early life microbiota. Neonates are essentially sterile before being colonized by maternal, environmental, and dietary inputs. Animal performance and intestinal health may suffer long-term effects from early life disturbances (nutrition, antibiotic use, stress). Functions of the gut microbiota relevant to productivity include:

- Fermentation of dietary fibre/resistant starch to SCFAs (acetate, propionate, butyrate), which serve as energy substrates and modulate host physiology.
- Synthesis of microbial protein, vitamins (especially B-vitamins) and other metabolites.

- Regulation of gut barrier integrity and immune signaling
- Competitive exclusion of pathogens, modulation of gut pH, production of antimicrobials, regulation of gut motility and transit.
- Communication with host neuro-endocrine systems via metabolites, hormones and neurotransmitters. For instance, SCFAs and peptide hormones link gut microbes to feed intake in farm animals.

Thus, the gut microbiome acts as an integral functional component — sometimes called the “last organ” of the animal — bridging feed, host, and environment.

3. Nutrition–Microbiome Interactions

3.1 Diet Composition and Its Impact on Microbial Ecology

The diet is the principal modifiable factor influencing the gut ecosystem. Feed ingredients, particle size, processing, nutrient balance, presence of anti-nutritional factors or feed additives all impact gut structure, function and microbiome. Key aspects include:

- **Carbohydrates and fibres:** Especially non-starch polysaccharides or resistant starches which escape enzymatic digestion and reach the large intestine or rumen for microbial fermentation. A diet rich in fermentable fibre promotes SCFA production, supports beneficial microbes and can enhance gut barrier integrity. Conversely, low-fibre/high-starch diets may reduce microbial diversity, increase acid loads (in ruminants: sub-acute ruminal acidosis) and impair gut health.
- **Proteins and amino acids:** Excess protein or poorly digested protein entering the hindgut may promote proteolytic fermentation, producing harmful metabolites (ammonia, amines, phenols) that compromise gut function. Balanced dietary protein and amino acid supply are thus important for gut health.
- **Lipids:** Though less often emphasized, dietary fats (type, saturation, oxidised lipids) can influence gut microbiota composition and the gut barrier.
- **Feed processing and particle size:** Processing that overly reduces particle size or increases fermentable sugar may accelerate rumen or hindgut fermentation beyond safe limits, leading to dysbiosis.
- **Feed additives and secondary metabolites:** The inclusion of prebiotics, probiotics, synbiotics, postbiotics (microbial metabolites) and phytobiotics is increasingly used to modulate the gut microbiome in favour of productivity and health. For example, the use of probiotics, prebiotics and synbiotics in various species has been shown to improve weight gain, milk yield, feed efficiency, immune responses and gut barrier function.

3.2 Microbial-Mediated Nutrient Utilisation and Feed Efficiency

Beyond diet influencing microbes, the microbiome in turn modulates how effectively an animal uses its diet. Feed efficiency is a major economic trait in livestock production. Recent research indicates that gut microbiota and their metabolic outputs (especially SCFAs, microbial protein, vitamins and secondary metabolites) are key contributors to feed efficiency.

For instance, in ruminants, rumen microbes break down fibrous feeds, generate VFAs and microbial protein which the host absorbs; inefficient fermentation or dysbiosis reduces nutrient capture and may increase waste in term of methane, heat and urea. In monogastrics, hindgut microbial metabolism of fibre/resistant starch likewise contributes to energy supply; a poorly performing microbiome may leave potential energy unrecovered, increase pathogen load, and reduce growth. Thus, nutritional strategies that support beneficial microbial communities may improve nutrient recovery, lower feed costs and improve sustainability.

3.3 Gut Barrier, Immune Function and Nutrition

Nutrition–microbiome interactions also affect the gut barrier, immune system development and systemic immune-metabolism. A healthy gut barrier prevents entry of pathogens, controls inflammation and supports steady nutrient absorption. Dietary elements such as fermentable fibres, specific fatty acids, antioxidants and targeted additives can enhance barrier function.

The gut microbiome maintains a dynamic and reciprocal relationship with the host immune system. Commensal microorganisms are integral to the development and function of the gut-associated lymphoid tissue (GALT), where they help establish immune tolerance and defense mechanisms. These microbes modulate the differentiation and activity of various T-cell subsets, influence cytokine secretion patterns, and regulate oxidative stress pathways that contribute to intestinal homeostasis. Through these interactions, the microbiota educates and fine-tunes both innate and adaptive immune responses. Conversely, dietary interventions such as the inclusion of probiotics, prebiotics, or synbiotics can beneficially alter the gut microbial ecosystem. Such modulation strengthens immune responsiveness, mitigates inflammatory reactions, reduces the incidence of infectious and metabolic diseases, and consequently enhances animal health and productivity.

3.4 Feed Intake, Behaviour and Microbiome Interplay

An interesting emerging dimension is the role of the gut microbiome in regulating feed intake, appetite, and behaviour. As noted in recent reviews, microbial metabolites such as SCFAs, tryptophan derivatives, GABA, serotonin precursors may interact with host endocrine and neural pathways, altering appetite, feed motivation and perhaps even feed preference.

Higher microbial diversity and favorable microbiota in the industrial setting may result in more consistent feed intake, fewer digestive disturbances, improved welfare, and ultimately higher

productivity. On the other hand, feed refusal, inadequate intake, digestive problems, and production losses might result from dysbiosis.

4. Indicators and Assessment of Gut Health

To connect gut health to nutrition and productivity, we must comprehend and measure it. A number of tools and markers have been developed.

4.1 Morphological and Functional Indicators

Classic gut health measures include gut morphology (villus height, crypt depth, intestinal mucosa thickness), epithelial cell turnover, gut enzyme activities (maltase, sucrase, alkaline phosphatase), and measures of permeability or leakage (e.g., tight junction proteins, plasma endotoxin, FITC-dextran assays). Changes in these may reflect positive or negative shifts in gut health.

4.2 Microbial and Metabolite Indicators

Comprehensive microbiome analyses—including 16S rRNA gene sequencing, metagenomics, and metatranscriptomics—offer valuable insights into the structure and function of the gut microbial community. These approaches reveal patterns of microbial diversity (alpha and beta diversity), taxonomic composition, and functional potential, such as genes associated with fibre degradation, methanogenesis, and proteolysis. Furthermore, metabolomic profiling of gut contents, feces, or blood provides complementary information on metabolic outputs and host–microbe interactions. Quantification of short-chain fatty acids, bile acids, microbial metabolites (e.g., phenols, amines), endotoxins, ammonia, and host-derived biomarkers such as cytokines and immunoglobulins reflects the integrated immune and metabolic status of the gut ecosystem.

4.3 Performance-Linked Indicators

Ultimately, gut health must link to production traits: feed intake, feed conversion ratio, average daily gain, milk yield and quality, egg production, carcass traits, reproductive performance and disease incidence. Correlating gut health markers to performance allows one to validate gut-health interventions.

4.4 New-Era Tools and Big Data Integration

Advances in omics technologies (genomics, transcriptomics, proteomics, metabolomics) bioinformatics and precision livestock management allow for holistic gut-health profiling. Projects

are underway to integrate microbiome profiling with feed-input data, environmental sensors and performance monitoring to build predictive models of gut health and productivity.

5. Impact of Gut Health on Animal Productivity

The connection between gut health and animal productivity is now well established. A healthy gut promotes efficient digestion and nutrient absorption, maintains immune stability, reduces pathogen load, and minimizes metabolic stress—all of which collectively enhance growth performance, feed efficiency, and overall production outcomes.

5.1 Feed Efficiency and Growth

Animals with excellent gut ecosystems may extract more energy and nutrients from the same meal than their less efficient counterparts due to microbial contributions to nutrient extraction, as was previously mentioned. Research on ruminant and monogastric animals demonstrates links between feed efficiency and microbial diversity and composition. Reduced digestive losses, fewer metabolic byproducts, and improved growth or production performance are all associated with improved gut health.

5.2 Milk, Egg and Meat Production

In dairy cows, the rumen microbiota regulates the supply of VFAs and microbial protein that underlie milk synthesis. Recent work indicates that the gastrointestinal microbiome significantly influences production traits in dairy cows. In poultry, gut health is a major determinant of egg production, shell quality, growth rate and meat quality. In pigs, gut-health status impacts lean growth, backfat, meat quality and reproductive performance.

5.3 Health, Welfare and Disease Resistance

Digestive diseases, subclinical inflammation, decreased immunological competence, and more frequent veterinary treatments are all more common in animals with poor gut health. Reduced growth, higher mortality, poorer product quality, and higher prescription expenses are all associated with costs. Reviews highlight how gut microbiota affects wellbeing, behavior, and stress reactions, all of which have an impact on output.

5.4 Environmental and Sustainability Implications

Efficient gut function means less feed wasted, less excreted nitrogen and phosphorus, lower enteric methane (in ruminants) and lower emissions per unit of product. Thus, improving gut health contributes to sustainable production. For example, microbial and feed additive strategies to optimize rumen fermentation are being explored as ways to simultaneously enhance productivity and mitigate methane.

6. Nutritional Strategies for Enhancing Gut Health

What nutritional levers can we apply, considering the importance of gut health? Below are several key strategies.

6.1 Fibre and Fermentable Substrates

Non-starch polysaccharides, such as sufficient dietary fiber or fermentable substrates like resistant starch, can promote beneficial microbial fermentation, generate SCFAs (particularly butyrate), which sustain beneficial microbial diversity and nourish gut epithelial cells and regulate immunity. Maintaining adequate rumen fiber in ruminants helps to maintain steady fermentation and avoid sub-acute ruminal acidosis. Appropriate fiber, fructo-, and galacto-oligosaccharide inclusion promotes hindgut health in monogastrics.

6.2 Functional Feeds, Prebiotics, Probiotics and Synbiotics

As previously mentioned, it has been shown that adding direct-fed microbials, non-digestible feed ingredients that promote beneficial microbes and combinations of these (synbiotics) improves gut health metrics in terms of microbiota composition, barrier integrity, immune markers and production responses. For example:

- Cultures of *Lactobacillus*, *Bifidobacterium*, *Bacillus*, *Enterococcus* have been used.
- Prebiotics such as fructo-oligosaccharides (FOS), galacto-oligosaccharides (GOS), mannan-oligosaccharides (MOS) support beneficial bacteria.
- Synbiotics combine both for synergistic effect.
- Selecting appropriate strains, doses and carrier substrates remains a challenge and an active research area.

6.3 Phytochemicals and Plant Secondary Metabolites: Plant-derived extracts and essential oils (e.g., oregano oil, thyme, rosemary, moringa leaf extract) may modulate microbial fermentation, promote

beneficial microbial populations, reduce pathogen load, mitigate oxidative stress, and support gut barrier function.

6.4 Precision Nutrition and Microbiome-Targeted Feeding

As microbiome profiling becomes more accessible, there is potential to move from “one-size-fits-all” feeding programs to microbiome-informed or precision diets. For instance, animals might be grouped by microbial phenotype and fed differently to optimize gut function and nutrient recovery. Predictive modeling of feed-additive efficacy based on rumen microbiome data is an emerging frontier.

6.5 Management and Early-Life Interventions

Nutrition during early life (pre-weaning, weaning transition) is critical: establishing a healthy microbiome, avoiding dysbiosis, supporting gut development and immune maturation. Strategies include colostrum management, early feed introduction, minimal antibiotic exposure, and targeted probiotic/prebiotic supplementation. Early life gut health sets trajectories for lifetime productivity.

7. Challenges, Emerging Concepts and Future Horizons

7.1 Gut Health under Stress and Environmental Pressures

Animals in production systems routinely face stressors: heat stress, crowding, dietary transitions, pathogen exposure, mycotoxins, high-grain diets (in ruminants) and environmental change. These stressors often manifest first in the gut via changes in microbial communities, barrier dysfunction, increased endotoxin translocation and inflammation. Nutrition and gut-health strategies must account for these dynamic challenges.

7.2 Antibiotic Reduction, Gut Microbiome and Resistance

With increasing regulatory and societal pressure to reduce antibiotic use in livestock, gut-health management becomes even more important. Probiotics, prebiotics, synbiotics and microbial-based additives are seen as alternatives to antibiotic growth promoters. However, the complexity of the gut ecosystem, variable responses and incomplete mechanistic understanding remain obstacles.

7.3 Gut –Brain and Gut–Immune Axes

Emerging research in humans and animals points to interactions between the gut microbiota and both immune and neuro-endocrine systems. In production animals this may manifest as altered feed intake, stress resilience, behaviour and welfare. Harnessing these links could open new nutritional or microbial interventions.

7.4 Sustainability, Carbon Footprint and Microbiome Manipulation

Improving gut health offers pathways to reduce environmental footprint: better feed efficiency, lower excreted nutrients, reduced methane in ruminants, less reliance on pharmaceuticals. However, large-scale adoption demands cost-effective, reproducible strategies, full life-cycle analyses and

consideration of unintended consequences (e.g., microbial resistance, unintended shifts in emissions).

7.5 Omics, Big Data and Predictive Gut Health

The explosion of sequencing technologies, bioinformatics, metabolomics and AI offers unprecedented opportunity to characterize gut ecosystems, link microbial data to animal phenotypes and build predictive models. The challenge will be to translate complex omics data into actionable feeding recommendations and management tools, and to integrate across livestock systems, breeds and environments.

8 Research Priorities in Animal Science

Based on the foregoing narrative, the following research priorities are proposed:

1. **Establishing Standardised Gut Health Metrics:** Develop validated, species-specific indicators (microbial, functional, morphological) of gut health that reliably link to productivity in different production systems.
2. **Microbiome-Phenotype Mapping:** Large-scale studies correlating individual animal microbiota composition and function with feed efficiency, growth, health outcomes and resilience to stress.
3. **Mechanistic Understanding of Microbe–Host–Feed Interactions:** Research to elucidate causal pathways (microbial metabolites → gut barrier/immune function → nutrient absorption) rather than only associative studies.
4. **Early Life Microbiome Programming:** Investigate interventions that shape early gut microbial colonisation and long-term productivity.
5. **Precision Nutrition and Microbiome-Informed Feeding:** Develop feeding regimens tailored to microbial phenotypes or targeted for microbial modulation via feed ingredients, additives and validate economically in commercial settings.
6. **Feed Additive Development and Efficacy Testing:** Rigorous evaluation of probiotics, prebiotics, synbiotics, phytobiotics, postbiotics in production settings, including dose, timing, strain specificity, cost-benefit, and regulatory feasibility.
7. **Stress, Climate Change and Gut Health:** Research on how heat stress, dietary transitions, social stress, toxin exposure affect the gut microbiome and gut health, and how nutritional strategies can build resilience.
8. **Sustainability and Environmental Impact Studies:** Quantify how gut-health interventions affect feed conversion, emissions of methane, nitrogen and phosphorus, manure quality and overall environmental footprint.

9. **Integration of Omics and Predictive Tools:** Build data-analytics platforms linking microbiome, metabolome, animal performance, and environmental data; develop decision-support tools for producers.
10. **Translational Research and Knowledge Transfer:** Ensure that findings from controlled experiments are translated into field-scale recommendations, extension programmes and integrated into feed-industry practice.

Conclusion

In conclusion, gut health lies at the intersection of nutrition, microbiology, immunology, physiology, behaviour, and sustainability in animal production. The efficient animal of the future will not only receive optimal nutrition but will also maintain a balanced and resilient gut ecosystem capable of maximizing nutrient utilization, resisting pathogens, and converting feed into high-quality products with minimal waste. From a nutritional perspective, this requires recognizing the gut as a dynamic ecosystem—one that thrives on substrates supporting beneficial microbes, stable feeding practices, robust gut barrier function, and targeted use of functional additives. However, many challenges remain. The gut microbiome is highly complex, dynamic, and influenced by genetics, environment, and management practices. Future research must explore these interactions across diverse breeds, diets, and production systems. By placing gut health at the core of nutrition and management, the livestock sector can advance toward a more productive, resilient, and sustainable future.

Molecular Biomarkers in Veterinary Diagnostics

Pavithra P

Molecular biomarkers offer earlier and more accurate identification of diseases in animals than conventional clinical or serological indices, as it improves surveillance, prognosis and treatment monitoring across species. Expansion of omics technology has accelerated biomarker discovery while revealing persistent gaps in validation and field level application in veterinary settings. Adopting reported standards such as Minimum Information for Publication of Quantitative Real-Time PCR Experiments (MIQE) for quantitative Real time PCR (qRT-PCR) and Standards for Reporting Diagnostic Accuracy Studies (STARD) for diagnostic accuracy strengthens analytical standards and supports biomarkers to come into routine. Current clinical use spans acute-phase proteins for inflammation, β -hydroxybutyrate for subclinical ketosis and cardiac troponins for myocardial in companion animals. Emerging directions include point-of-care biosensors, MALDI-TOF mass spectrometry for rapid microbial identification and CRISPR-based nucleic acid assays that align with One Health priorities.

Keywords: biomarkers; veterinary diagnostics; MIQE; STARD; One Health

Introduction

Biomarkers are objectively measured characteristics that reflect normal biology or pathology, and they assess risk, diagnosis, prognosis and monitoring in veterinary medicine (Strimbu and Tavel, 2010). The shift from phenotype-based diagnosis to molecular approach can be attributed to high-throughput sequencing, targeted amplification, proteomic profiling and metabolomic fingerprints that capture pathology at an earlier stage (Goldansaz *et al.*, 2017). Because animal, human and environmental health are interdependent, biomarker-guided diagnostics fit the One Health framework for integrated surveillance of zoonoses and production based diseases (Garzón *et al.*, 2018).

Classes of Molecular Biomarkers

Genomic biomarkers comprises of single-nucleotide polymorphisms, copy number variants and pathogen gene targets measured by PCR or sequencing to confirm etiology or host susceptibility (Pareek *et al.*, 2011). Transcriptomic biomarkers include mRNAs, microRNAs and long non-coding RNAs whose expression reveal host response to certain treatment or disease stage across infectious, metabolic and reproductive conditions (Do *et al.*, 2021). Metabolomic biomarkers track pathway-level disturbances captured by Nuclear Magnetic Resonance (NMR) or Liquid Chromatography – Mass Spectrometry (LC-MS) and support early detection of metabolic diseases in livestock (Goldansaz *et al.*, 2017).

Discovery and Validation Technologies

QRT-PCR and digital PCR remain as foundation for targeted assays, and strict adherence to MIQE improves design quality and reproducibility across laboratories (Bustin *et al.*, 2009). Diagnostic accuracy research should follow STARD 2015 so that reference standards, blinding and precision are reported in a way that enables critical appraisal and adoption (Bossuyt *et al.*, 2015). Metabolomics utilises spectral deconvolution and multivariate modeling to derive notable discriminators from spontaneous field cases rather than only experimental disease models (Goldansaz *et al.*, 2017). Bioinformatics workflows that integrate multi-omics data and perform disciplined feature selection increase the chances of discovery signatures that rise into clinically useful tests (Perera *et al.*, 2022). Workflow for discovering, validating and deploying veterinary molecular biomarkers covering pre-analytics, platforms, QC, selection, validation and translation are given in Figure 1.

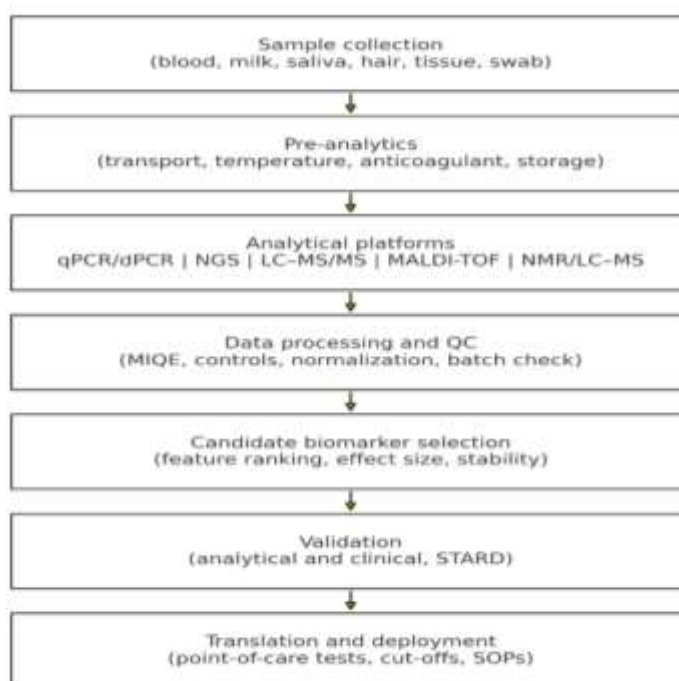


Figure1. Workflow for discovering, validating and deploying veterinary molecular biomarkers covering pre-analytics, platforms, QC, selection, validation and translation.

Established Biomarker Applications

Acute-phase proteins and inflammation

- a. Haptoglobin and serum amyloid A are the pivotal acute-phase proteins in cattle serving as reliable signals of systemic inflammation and tissue injury (Petersen *et al.*, 2004). These milk-derived markers complement somatic cell count and culture for mastitis surveillance under field conditions (Trela *et al.*, 2022).

Energy metabolism and subclinical ketosis in dairy cattle

- b. β - Hydroxybutyrate is the ideal biomarker for subclinical ketosis with decision limits commonly set near 1.2–1.4 mmol/L in early lactation cows (Duffield, 2000). Validated manual and cow-side devices can support herd-level testing when standardized against serum BHBA assays (Iwersen *et al.*, 2009).

Cardiac injury biomarkers in companion animals

- c. Cardiac troponin I is a fine indicator of cardiomyocyte injury and aligns with disease severity and impacts in dogs (Langhorn and Willesen, 2016). High-sensitivity cardiac troponin I assay validation demonstrates strong precision and discrimination between health and disease, which supports clinical integration in veterinary cardiology (Wesselowski *et al.*, 2023).

Stress and welfare biomarkers

- d. Heat-shock protein 70 (*HSP 70*) expression is widely used as a molecular indicator of thermal load and thermotolerance in ruminants exposed to hot climates (Sejian *et al.*, 2018).

Enabling Platforms for Rapid and Field-Ready Diagnostics

- e. MALDI-TOF mass spectrometry shortens time to identification for veterinary pathogens and complements culture by matching spectral fingerprints against curated libraries (Singhal *et al.*, 2015). Bedside biosensors and chip-based diagnostic devices support rapid sorting, herd screening and remote case management in farm and companion animal practice (Manassis *et al.*, 2022). CRISPR-based diagnostics pair isothermal amplification with Cas nucleases to deliver rapid and specific nucleic acid detection suitable for pen-side testing of veterinary pathogens (Chen *et al.*, 2018).

Case Studies

Milk acute-phase proteins for screening subclinical mastitis in dairy cows

Quarter-wise measurements of milk serum amyloid A (SSA) and haptoglobin differentiated chronically high somatic cell count quarters from healthy controls in a field population of Swedish dairy cows, supporting their use as indicators of subclinical intramammary inflammation (Gronlund *et al.*, 2005). A Central European study reported strong positive correlations between milk haptoglobin and milk serum Amyloid with somatic cell count having $r \approx 0.83$ and $r \approx 0.81$ respectively, and demonstrated concordance between serum and milk acute-phase responses, highlighting biological plausibility for quarter-level diagnostics (Kováč *et al.*, 2007).

Cowside β -hydroxybutyrate testing to detect subclinical ketosis

In early lactation herds, subclinical ketosis is defined at β -hydroxybutyrate levels around 1.2–1.4 mmol / L, which is in concordance with increased risk of displaced abomasum and reduced performance, making BHBA a practical metabolic biomarker for on-farm screening (Suthar *et al.*, 2013). Validation of manual whole-blood BHBA meter reported sensitivities of 88% and 96% at 1.2

and 1.4 mmol / L cut-offs with specificities of 96% and 97% respectively, indicating clinically useful accuracy for pen-side decisions (Iwersen *et al.*, 2009).

Cardiac troponin I as a prognostic marker in canine emergency patients

In dogs presenting to emergency care, elevated cardiac troponin I measured at admission and on serial sampling associated with worse short-term outcomes, indicating that cTnI adds prognostic information beyond clinical examination and routine biochemistry (Porter *et al.*, 2016).

Serum amyloid A to assess severity and outcome in equine colic

At admission, horses with inflammatory gastrointestinal disease show markedly higher serum amyloid A than healthy horses, and SAA dynamics help distinguish medical from surgical cases during clinical follow-up (Jacobsen and Andersen, 2007). Past - record based and follow up based cohorts demonstrate that SAA and related acute-phase proteins differentiate clinically normal horses from colic cases and informs the prognosis (Dondi *et al.*, 2015). A large multi- centre analysis encompassing more than 700 horses reported that admission SAA concentrations carry prognostic value, supporting routine inclusion of SAA in equine colic diagnostic panels (Witkowska-Piłaszewicz *et al.*, 2019).

Future Prospects

Integrating multi-omics panels with machine learning is expected to raise diagnostic yield while capturing host–pathogen interactions that inform surveillance and treatment (Perera *et al.*, 2022). As pipelines and reference datasets advance, metabolomics will deliver applicable biomarkers for production traits and reproductive management in cattle (Goldansaz *et al.*, 2017). Practical uptake will rely on calibrated on-site formats, validated decision limits and strict adherence to reporting standards to maintain reproducibility and clinical utility in the field (Bossuyt *et al.*, 2015). Embedding biomarker-based diagnostics within One Health programs should advance early warning for zoonoses and improve outcomes for animals and communities (Garzón *et al.*, 2018).

Conclusions

- a. Molecular biomarkers are now established for inflammation, metabolic imbalance, reproduction, and continuing advances in proteomics and metabolomics will broaden their clinical utility across species. Methodological discipline grounded in MIQE and STARD is essential to deliver reproducible results that veterinarians can trust in routine practice and field conditions. Rapid biosensors, MALDI-TOF workflows and CRISPR platforms are poised to support fast, cost-effective decisions that align with One Health goals in livestock and companion animals.

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Effect of abiotic stress on plant under the current scenario of climate change

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Abiotic stress causes serious threats to the environment and agriculture which have been intensified in the current century due to global warming and industrialization. Continuous increase in population along with variation in weather patterns due to climate changes has hampered global crop productivity. Unfavorable climate causes abiotic stresses and leads to changes in agro-ecological conditions which ultimately influence growth and distribution of income leading to increase in demand for agricultural production. Abiotic stress refers to the effect of non-living factors such as temperature, rainfall, drought etc. on living organisms. Abiotic stresses include cold, salinity, drought and heat. Several morphological, biochemical, physiological and molecular changes occur in plants causing reduction in growth and productivity due to abiotic stress. Due to Stress biosynthesis, concentration, transport, and storage of primary and secondary metabolites is also affected. Utilization of selection, hybridization, germplasm conservation, transgenic technology and marker assisted selection will enable to develop better genotype which will be tolerant or resistant for different types of abiotic stress.

Introduction:

Any external condition that adversely affects growth, development and productivity of the plant is called stress. [9]. Sudden change in environmental condition causes stress in plant which may lead to changes in cellular metabolism, altered gene expression, changes in growth rates, crop yields, etc. There are mainly two types of stress biotic and abiotic stress. Biotic stresses are imposed by living organism such as pathogens specially viruses, bacteria, fungi, nematodes, insects, arachnids, weeds and insects while, abiotic stresses are imposed on plants by non-living factors like sunlight, temperature, wind, salinity etc. Plant can easily recover from mild or short term stress. Abiotic stresses such as water logging, drought, extreme,

Temperature such as cold, frost & heat, salinity and mineral toxicity hampers growth & development leading to reduction in yield and deterioration of seed quality [7]. These abiotic stresses may lead to osmotic stress, failure of proper ion distribution and cell homeostasis leading to reduced growth and productivity. Hence it becomes important to understand the response of plant towards abiotic stress and to identify the mechanism of genes associated with avoidance or tolerance of abiotic stress.

Cold stress

The type of stress induced due to low or non-freezing temperatures is known as cold stress. This stress may injure or kill crop and may affect its survival, productivity & ecological distribution. Cold acclimation refers to the phenomenon which gives capacity to tolerate such adverse low temperature. The Temperatures for cold stress varies from species to species. There are two types of tolerance associated with low temperature namely cold tolerance and freeze tolerance. Cold tolerance refers to the ability of plants to withstand non-freezing temperatures (0–15 °C) while freeze tolerance refers to the capacity of plants to survive at subzero (< 0 °C) temperature [5]. In order to mitigate these cold stresses, so many signal transduction pathways such as ROS, protein phosphate, protein kinase, Ca^{2+} and ABA etc. There and among all ABA (abscisic acid) pathway provides the best cold tolerance mechanism in plants.

Salinity stress

Salinity refers to the condition of soil having higher concentration of soluble salts. It hampers plant growth and also limits the use of land leading to reduction in agricultural productivity. When osmotic pressure produced by ion(s) are equivalent to that generated by 40 mM NaCl i.e. 0.2 MPa or more (USDA-ARS 2008), those Soils are considered as saline. Presence of ions in root zone creates osmotic stress which limits water absorption by roots and results in stunted plant growth. It also causes ionic disequilibrium which may result in inactivation of several enzymes, starvation due to less nutrient uptake, toxicity in tissues due to ions and oxidative stress [8]. Due to oxidative stress, Reactive oxygen species (ROS) is produced which damage plants by photosynthesis inhibition, increase in lipid peroxidation and DNA damage. Plants adopt certain mechanism to combat salinity stress such as salt exudation or tissue compartmentalization ie. accumulation of ions into different tissue compartments, vacuoles or old leaves. In most of the plants, uptake of Na^+ and Cl^- are excluded by roots during water uptake from the soil. Several osmolytes are produced by plants such as glycine betaine, trehalose or proline are produced during osmotic stress, to protect them from dehydration or protein denaturation. However, oxidative stress produces different enzymatic or non-enzymatic antioxidants, which are responsible for protection of plants from harmful effects of reactive oxygen species.

Drought

Drought refers to the condition in which soil moisture is depleted which causes dehydration losses in plant tissues. If more water loss occurs from the plant as compared to those absorbed by roots then there is reduction in the plant's water content which leads to reduction in normal plant processes is termed as dehydration stress. To cope up with dehydration stress, plant halt growth and reduce photosynthesis, change in foliage colour, wilting of foliage, accumulation of abscisic acid which closes stomata and reduction of leaf relative water content [1,4]. There are several mechanism developed in plant to combat with the situation of drought such as avoidance, tolerance, escape and resistance.

Avoidance: Plant develops certain physiological changes such as leaf rolling, thick epicuticular wax on leaf, leaf posture or development of deep root (water spenders), to cope with the drought situation without much reduction in yield is called drought avoidance. In case of dehydration avoidance plant production is normal under dehydration stress without much reduction in yield. Its role is to maintain water supply in plant in order to sustain leaf hydration and turgidity to maintain stomatal opening and transpiration under water deficit condition. This mechanism is essential for photosynthesis, gaseous exchange in leaves and carbon assimilation.

Tolerance: It refers to the plant ability to withstand drought situation. A plant is said to be dehydration tolerant when environment factors attacks plant tissue but still it survives either by osmotic adjustments or by maintaining cell's turgidity. In case of osmotic adjustment, plant is able to maintain cell turgor pressure under reduced soil water potential. When dehydration stress occurs, a response is triggered in which certain adaptive mechanism are triggered which provides dehydration tolerance to plants.

Escape: Dehydration escape is the mechanism in which plant uses previous optimal conditions to develop vigor. It may involve characteristics such as induction of dormancy at the time of stress or cultivation of early maturing genotypes. There are certain genotypes which are able to recover after exposure to a brief period of drought stress. Dehydration escape strategy involves timing of life cycle of a plant in which most critical developmental stages are completed when water is abundant before occurrence of stress. Use of short durational variety in case of predictable terminal stress is often preferred to minimize yield losses.

Resistance: It is the mechanism through which plant can sustain even under drought situation without reduction in yield. Drought resistance mechanisms are controlled by genes and several QTLs are reported for the same. There are some traits which are associated with drought resistance and are mentioned as follows:

1. **Osmotic adjustment:** Certain osmotic active compounds such as glycine betaine and proline gets accumulated when a plant experiences water deficiency, which results in reduction of water potential. This phenomenon is called osmotic adjustment which enables the plant to take up water and maintain turgor under stress condition and can survive for a longer period.
2. **Cell membrane stability:** It is complementary to osmotic adjustment as it enables plant to survive at reduced water potential. The capacity of cell membrane at reduced moisture to prevent electrolyte leakage is termed as cell membrane stability which varies for different crops .
3. **Epicuticular wax: In certain crops such as sorghum and rice,** drought resistance traits are found to be highly correlated with the epicuticular wax layer thickness. Those plants having thick layer of epicuticular wax remain turgid for a longer duration in case of dehydration stress.
4. **Accumulation of ABC:** It helps plants in mitigating drought stress by decreasing rate of cell division, stomata conductance, size of organs and increasing the rate of development.
5. **Leaf anatomy: Pubescence** Waxiness, thickness, rolling and posture are the traits that are associated with leaf which leads to decrease in radiation absorbance by the surface of leaf which results in reduction in rate of evapotranspiration and risk of irreversible photo-inhibition.
6. **High tiller survival:** Fewer tillers are produced by drought resistant lines and most of them can survive under drought condition in comparison to the plants that produces more tillers and on the onset of drought they fail in proper grain filling.
7. **Chlorophyll content:** Higher content of chlorophyll at all developmental stages improves yield and transpiration efficiency under dehydration stress condition.

Heat

When increase in temperature causes irreversible damage to plant growth and development is termed as heat stress. If the temperature rises above 10 to 15 °C as compared to ambient, it may lead to heat shock or heat stress. Increased level of greenhouse gases such as CO₂, methane, chlorofluorocarbons and nitrous oxides has led to global warming. According to the Inter governmental Panel on Climatic Change (IPCC), there has been rise of 0.3 °C per decade which has reached to approximately 1 and 3 °C above the present value by 2025 and 2100 AD, respectively. Higher temperature exposure for longer duration may cause severe cellular injury causing death of cell, protein denaturation and aggregation, increased lipid membrane fluidity, enzymes inactivation, protein degradation, inhibition of protein synthesis, and loss of membrane integrity. Heat stress that are associated with injuries lead to starvation, inhibition of growth, reduced ion flux, production of toxic compounds and production of reactive oxygen species (ROS) ultimately [3, 6].

Heat tolerance: Heat tolerance is the ability of the plant to tolerate heat without much reduction in yield at high temperatures. In this mechanism, heat shock proteins (HSPs) gets expressed immediately after exposure to high temperature stress as stress defensive strategy of the cell. Expression of HSPs (10 to 200 kDa) gets involved during heat stress for signal transduction. As a result, it improves several physiological phenomena such as photosynthesis, water and nutrient use efficiency, assimilate partitioning and membrane stability. Several traits are found to be associated with heat stress tolerance such as canopy temperature depression, stomatal conductance, Membrane thermostability, Chlorophyll fluorescence, Chlorophyll content & stay green and Photosynthesis.

(a) Canopy temperature depression (CTD): Canopy temperature depression helps to select physiologically superior lines in case of warm & low relative humidity environments, which can lead to leaf cooling up to 10 °C below ambient temperatures. It is highly heritable trait that shows significant correlation with seed yield.

(b) Stomatal conductance: The stomatal conductance is the highly heritable trait ranging from 0.5 to 0.8. Viscous flow porometer is used to assess leaf conductance of plants and can give a relative measure of stomatal conductance in a few seconds.

(C) Membrane thermostability: Membrane thermostability is reasonably heritable and shows high genetic correlation with yield under heat stress.

(D) Chlorophyll fluorescence: Chlorophyll fluorescence represents the fate of excitation energy in the photosynthetic apparatus, which can be used to indicate heat stress tolerance.

(E) Chlorophyll content & stay green: Chlorophyll content and stay green traits are associated with heat stress tolerance. QTL associated with chlorophyll content are reported to contain the genes for photosynthetic enzymes, heat shock proteins, and abscisic acid (ABA) responsive gene.

(F) Photosynthesis: Declined photosynthesis is suggested as measure of heat stress sensitivity in plants.

Breeding approaches for abiotic stress

The breeding approaches which involves selection of physiological trait has advantage over selection based on breeding for yield. The phenomics concept can be used for selection of physiological traits because that two or more stress have common physiological traits, which indicates the entire plant health. Selection for the traits associated with tolerance or resistance characteristics of the respective stress must have high heritability along with high genetic advance. Conservation and characterization of germplasm will act as a useful source of resistance gene for different types of abiotic stress. Identification of QTL will also give an insight about the region of genome associated directly or indirectly with abiotic stress resistance breeding. Utilization of marker along with the marker assisted as well as transgenic technology, resistant genotypes may be developed.

Conclusion

Abiotic stresses will always remain a challenge to the agriculture. The early evolution of landraces or primitive cultivars occurred under dry conditions having extremes of temperature, while later on crop cultivars were domesticated under favorable environments. Natural variations present in crops are the genetic reservoirs of traits associated with abiotic stress adaptation. Subsequently, selection of plants for yield and its attributes does not always result in production of stress tolerant productive crops. It is expected that in the coming 50–100 years there will be rise of 3–5°C temperature on earth. And if there is continuous increase in temperature, then there are chances of uneven rainfall leading to flood or drought. The human activities like use of excessive fertilizers, inappropriate irrigation and exploitation of metal resources may lead to salinity stress upto a large extent. So, Under these circumstances there are chances that plant will encounter both biotic and abiotic stresses concurrently and more frequently. So it becomes the responsibility of plant breeders to develop stress tolerant cultivars in order to secure food security and to ensure safety to the farmers. Molecular breeding must be done at genetic level to develop better insights against stress responsive mechanisms in plants which will prevent them from different types of stress conditions. Unless responsive mechanisms are not developed against biotic and abiotic stresses, the plants will continuously subjected to such stresses and ultimately will prove a great threat to world agriculture.

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Artificial Intelligence & Agricultural Extension Education

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Artificial Intelligence (AI) is rapidly emerging as a transformative force in agriculture. With the global population exploding exponentially which thrusts on the importance of nutritional security of the rising demographics. Traditional agricultural extension systems, which are essential for guiding farmers, are strained, particularly in developing countries with numerous smallholder farms. AI offers a solution by providing data-driven support to farmers and extension services. It enables precision farming through technologies like drones and sensors, optimizing resource utilization and crop yields.

AI-powered crop surveillance detects diseases, pests, and nutrient deficiencies, offering timely recommendations. Chatbots and virtual assistants provide instant access to agricultural information, overcoming barriers like limited extension workers and remote locations. Data analytics, language processing, and market analysis harness AI's power to make informed decisions, adapt to climate change, and improve farm management. AI supports training and education, ensuring farmers stay updated on best practices. However, challenges include the high cost of AI installation, resistance from farmers, the need for digital education, potential technical glitches, and the irreplaceable human touch in extension services. Real-life examples like the M-Velanmai initiative in India, the Plantix app from Germany, and an early warning system for pest management demonstrate AI's impact in agriculture. These innovations empower farmers, enhance productivity, and contribute to global food security. AI's potential to reshape agriculture is undeniable, making it a crucial tool for the future of farming.



Artificial Intelligence in Agriculture

Artificial intelligence (AI) is transforming agricultural extension education by providing farmers with real-time, data-driven insights and personalized recommendations that enhance productivity, optimize resource use, and support sustainable farming practices.

AI's role in revolutionizing agricultural extension

Real-time Information Dissemination: AI-powered chatbots, virtual assistants, and mobile apps provide farmers instant access to information on best agricultural practices, weather forecasts, and market prices, overcoming barriers like limited extension workers in remote areas.

Precision Farming Guidance: By analyzing data from IoT sensors, drones, and satellites, AI systems offer precise recommendations for planting, irrigation, fertilization, and pest control, which helps farmers optimize resource usage (water, pesticides, fertilizers) and minimize environmental impact.

Resource efficiency: AI helps optimize resource use, such as by using smart irrigation systems to water crops only when necessary, leading to water savings and reduced costs.

Early Detection and Management: Image recognition and machine learning models enable the early detection and diagnosis of crop diseases, pests, and nutrient deficiencies from photos taken by farmers' smartphones, allowing for timely intervention and reduced crop losses.

Predictive Analytics and Risk Management: AI models process historical data and real-time conditions to forecast disease outbreaks, yield potential, and climate risks, empowering farmers to make proactive, informed decisions and adapt to changing conditions.

Climate adaptation: AI models can help farmers adapt to climate change by providing insights into climate trends and helping them make informed choices about what crops to plant and how to manage their fields in a changing environment.

Localized and Inclusive Content: Natural Language Processing (NLP) tools can translate complex agricultural research and extension materials into various local languages and voice-based interfaces, making information more accessible to a broader range of farmers, including those with low literacy levels.

Automated monitoring: Through drones and satellite imaging, AI-powered systems can constantly monitor crops for diseases, pests, and nutrient deficiencies, allowing for early intervention.

Capacity Building for Extension Agents: AI tools are used to train extension professionals, equipping them with the necessary skills to leverage cutting-edge technologies and provide more effective guidance to farmers.

Early warnings and risk management: Predictive models can alert farmers to potential pest outbreaks, diseases, and weather events, allowing for proactive planning and mitigating potential losses.

Empowering farmers with knowledge: AI can improve farmer literacy by providing access to new technologies and best practices, helping them adapt to climate change and improve their livelihoods.

Bridging knowledge gaps: AI can act as a bridge between farmers, extension agents, researchers, and policymakers, creating a feedback loop that leads to better-targeted research and policy recommendations.

Improving sustainability: By optimizing the use of water, fertilizers, and pesticides, AI helps promote more sustainable and environmentally friendly agricultural practices.

Challenges and Considerations

Cost and Accessibility: High initial investment in AI infrastructure (sensors, drones, software) and a lack of reliable internet access in many rural areas pose significant barriers to adoption, especially for smallholder farmers.

Digital Literacy: A lack of technical skills and digital education among both farmers and extension agents can hinder the effective use of AI tools, requiring targeted training programs.

Data Quality and Privacy: The effectiveness of AI models relies on high-quality data, and there are concerns regarding data privacy, security, and the ethical use of farmer data.

Need for Human Touch: While AI provides valuable data and insights, it cannot entirely replace the nuanced, on-the-ground human interaction and local knowledge provided by traditional extension agents.

High Costs: The initial investment required for AI infrastructure, such as sensors, drones, and software, can be a significant barrier for small-scale farmers.

Digital Divide and Skill Gaps: Many rural areas lack the necessary internet connectivity and reliable power supply. Additionally, both farmers and extension agents may lack the technical skills and digital literacy needed to effectively use AI tools.

Data Concerns: Issues related to data quality, availability, privacy, and security are major hurdles. There are also concerns about who owns the data collected from farms and how it is used.

Resistance to Change: Some farmers may be skeptical of new technologies and prefer traditional, experience-based farming methods.

Key Takeaway

The integration of AI into agricultural extension is a powerful tool for modernizing farming and working towards global food security and sustainability. However, its successful implementation requires collaborative efforts from governments, researchers, and the private sector to address the challenges of accessibility, cost, and digital literacy, ensuring that all farmers can benefit from these technological advancements.

AI boosts crop productivity with precision farming and IoT. AI reduces resource use, enhancing agricultural sustainability. Identifies key barriers to AI adoption and offers solutions.. “Artificial Intelligence in Agriculture” focuses on fundamental and applied research targeting AI in all stages of agriculture, from soil preparation to the sowing of seeds, addition of fertilizers, irrigation, weed protection, harvesting, storage, packing, and transportation. Agriculture will drive more efficient, productive, and sustainable farming through applications like precision farming, which optimizes resource use, and automation, which uses robots and drones for tasks like planting, monitoring, and harvesting. AI will also improve predictive analytics for weather, crop diseases, and pest outbreaks, and enhance supply chain management to reduce food waste and improve logistics. This will lead to increased yields, lower costs, and a reduced environmental impact.



Artificial Intelligence with Drones

Key applications of AI in the future of agriculture

Precision Farming: AI analyzes data from sensors, drones, and satellites to optimize the use of water, fertilizers, and pesticides, leading to increased yields and reduced waste.

Automation: AI-powered robots and self-driving tractors can automate labor-intensive tasks like planting, harvesting, and weeding with greater precision and efficiency.

Predictive Analytics: AI algorithms analyze weather patterns, soil data, and historical information to predict weather events, identify disease and pest outbreaks before they become widespread, and suggest the optimal time for planting and harvesting.

Crop and Soil Monitoring: AI-powered computer vision can analyze images of crops to detect diseases, pests, and nutrient deficiencies at an early stage, allowing for timely and targeted interventions. AI can also analyze soil composition to ensure the right nutrient mix.

Supply Chain Optimization: AI can improve logistics by optimizing transportation routes, monitoring storage conditions, and forecasting demand to reduce spoilage and food waste from farm to consumer.

Sustainable Practices: By optimizing resource use and minimizing chemical inputs, AI promotes sustainable farming, improves soil health, and can reduce fuel consumption and emissions.

Smart Indoor Farming: In controlled environments like vertical farms, AI optimizes light, temperature, and nutrients to increase production and efficiency while using significantly less water.

Potential impacts and considerations

Increased efficiency and yield: AI can significantly boost crop yields and reduce operational costs through optimized resource management and automation.

Improved sustainability: AI helps reduce the environmental footprint of farming by minimizing water and chemical use and improving soil health.

Labor market changes: While AI automation may reduce certain manual labor jobs, it will also create new jobs in technology, data analysis, and AI system maintenance.

Enhanced decision-making: AI provides farmers with data-driven insights to make more informed decisions, from planting to harvesting.

Other forms of AI have been used to analyze data to provide support for management decisions, such as in livestock monitoring, food traceability, genetic studies, and predicting weather and crop yield. Furthermore, AI is particularly adept at image analysis and can identify insects, weeds, and diseases. Smart farming, also known as smart agriculture, is the adoption of advanced technologies and data-driven farm operations to optimize and improve sustainability in agricultural production.

In 2025, the evolving needs of food security, sustainability, and climate resilience are driving continuous innovation across India's vast agricultural landscape. With the emergence of dedicated agriculture innovation funds and the establishment of innovation centers, the sector is undergoing a paradigm shift.

Artificial intelligence (AI) is transforming agricultural extension education by providing farmers with real-time, data-driven insights and personalized recommendations that enhance productivity, optimize resource use, and promote sustainable practices. To ensure successful integration, a collaborative effort is needed among governments, research institutions, and private companies to develop supportive policies, provide financial incentives, and establish robust training and education programs that build the capacity of all stakeholders in the agricultural sector. AI is important to agriculture extension because it personalizes and automates advice, monitor crops with high precision, and makes information more accessible to farmers, even in remote areas. It enables data-driven decision-making for issues like pest and disease management, resource allocation, yield prediction, and market analysis, ultimately increasing productivity and sustainability.

The scope of artificial intelligence (AI) in agriculture extension education is to transform traditional farming support systems into data-driven, personalized, and efficient services by using AI tools to enhance farmer education and advisory services. This includes providing customized, localized advice via chatbots and virtual assistants, enabling extension agents to reach more farmers with the

right information, and using AI-powered data analytics for predictive modelling, resource management, and pest/disease monitoring. The goal is to boost farmer productivity and income, improve sustainability, and address challenges like climate change through a more accessible and effective extension system.

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Climate Change Adaptation and Mitigation in Livestock Production: Building Resilience and Reducing Emissions

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1. Introduction: The Dual Challenge of Livestock in a Changing Climate: Livestock production stands at a critical intersection in the global climate challenge. Providing approximately 33% of global protein and supporting the livelihoods of over a billion people, the sector is fundamental to food security and poverty alleviation, particularly in low- and middle-income countries (LMICs) (Gerber et al., 2013; Rojas-Downing et al., 2017). Simultaneously, the sector is a significant contributor to anthropogenic greenhouse gas (GHG) emissions, primarily through enteric methane (CH₄) from ruminants, and nitrous oxide (N₂O) from manure and fertilizer use. The challenge is therefore dual: the sector must adapt to the inevitable impacts of a warmer, more volatile climate to ensure continuous production and protect vulnerable livelihoods, while also aggressively mitigating its own contribution to global warming. Simple solutions are insufficient; a transition towards Climate-Smart Livestock (CSL) systems—which integrate adaptation, mitigation, and productivity enhancement—is imperative. This chapter explores this dual challenge by detailing the physical, biological, and socio-economic impacts of climate change on livestock, followed by an in-depth analysis of the leading adaptation and mitigation strategies, emphasizing the synergy and potential trade-offs between them. The final section addresses the policy and economic frameworks required to scale these solutions globally.

2. Climate Change Impacts on Livestock Systems: The impacts of climate change on livestock production are diverse, affecting animals directly through physiological stress and indirectly through disruptions to feed, water, and health ecology (Thornton & Herrero, 2015).

2.1. Direct Physiological Stress: The Threat of Heat: The most immediate and pervasive direct impact is thermal stress, particularly heat stress. When the ambient temperature and humidity exceed an animal's Thermal Comfort Zone (TCZ), physiological mechanisms shift from production to costly cooling efforts, such as increased respiration (panting) and peripheral blood flow. This stress is commonly quantified using the Temperature-Humidity Index (THI).

Productivity Losses: Heat stress significantly reduces feed intake across all species (ruminants, poultry, pigs), resulting in lower body weight gain, decreased milk yield (up to 30% reduction in severe cases for dairy cattle), and smaller egg sizes (Mader et al., 2010).

- **Reproductive Failure:** Elevated body temperatures are highly detrimental to reproduction, leading to decreased sperm quality, reduced estrous expression, and significantly lower conception rates due to damage to oocytes and early embryos.
- **Immunity and Health:** Chronic heat stress compromises the immune system, making animals more susceptible to infectious diseases.

2.2. Indirect Impacts: Feed, Water, and Disease Ecology: The majority of long-term climate risk to livestock stems from indirect effects on the wider ecosystem.

- **Feed and Fodder Scarcity:** Changes in precipitation patterns and increased temperatures directly impact forage quality and quantity. Droughts degrade rangelands, while elevated temperatures accelerate the lignification of plants (reducing digestibility and nutritional value) and lower the overall yield of key feed crops like maize and soy (Gerber et al., 2013). This places immense pressure on grazing systems, forcing premature culling and market instability.
- **Water Insecurity:** Increased frequency and intensity of droughts deplete natural water sources crucial for both animal consumption and the production of feed. In water-scarce regions, competition between human, crop, and livestock water needs intensifies conflict and vulnerability.
- **Disease Range Expansion:** Warmer average temperatures allow disease vectors (ticks, mosquitoes, tsetse flies) to survive in areas previously too cold, expanding the geographical range and increasing the seasonal duration of vector-borne diseases such as Rift Valley Fever and Blue Tongue Virus (Grace et al., 2020). Changes in humidity and rainfall also affect the survival and infectivity of internal parasites.

3. Building Resilience: Key Adaptation Strategies: Effective adaptation builds the capacity of livestock systems to absorb climatic shocks and recover quickly, maintaining productivity and safeguarding livelihoods.

3.1. Genetic Improvement and Species Diversification: Genetic strategies are foundational for long-term resilience.

- **Breeding for Thermo-tolerance:** Instead of solely relying on high-yielding but heat-sensitive European breeds (*Bos taurus*), breeding programs are increasingly focusing on incorporating traits from indigenous, thermo-tolerant breeds (*Bos indicus* or locally adapted breeds). Key genetic markers include the "slick-hair gene" in dairy cattle, which allows for better heat dissipation and results in higher productivity under heat stress.
- **Species Shift:** In highly vulnerable areas, a strategic shift to more resilient species may be necessary. For instance, moving from cattle to small ruminants (goats and sheep) or camels in increasingly arid regions, as these species are inherently more water- and heat-efficient (UNFCCC, 2020).

3.2. Feed and Water Security Enhancements: Securing the resource base is central to adaptation.

- **Climate-Smart Forages:** Promoting the cultivation of drought-resistant and deep-rooted forage species, such as specific legumes (e.g., *Stylosanthes* species) and perennial grasses. The use of feed conservation techniques (e.g., silage and haymaking) allows farmers to stockpile feed during good years to buffer against drought years.
- **Strategic Water Management:** Implementing local water harvesting techniques (e.g., check dams, trenches, or small storage tanks) to replenish groundwater and ensure year-round access to drinking water. Borehole development and efficient water delivery systems are essential infrastructure investments.
- **Rangeland Management:** Implementing rotational or deferred grazing systems to prevent overgrazing, allowing rangeland vegetation to recover and increasing the long-term productivity and carbon-storage capacity of the soil.

3.3. Infrastructure and Management Practices: Technological and management adjustments provide immediate relief.

- **Shelter and Cooling:** In intensive systems, this involves constructing well-ventilated barns with high ceilings, insulation, and the use of passive cooling (e.g., proper orientation, natural ventilation) or active cooling (e.g., fogging/sprinkling systems, fans). For grazing animals, establishing silvopastoral systems (integrating trees with pasture) provides natural shade and additional feed, an excellent example of a co-benefit strategy.
- **Precision Livestock Farming (PLF):** Utilizing affordable, data-enabled technologies like wearable sensors to monitor real-time physiological indicators (e.g., body temperature, respiration rate, and rumination activity). This allows for early detection of heat stress or disease, enabling immediate, individualized intervention and reducing overall herd losses (Bashiru & Oseni, 2025).

4. Reducing Emissions: Mitigation Strategies: Mitigation efforts in the livestock sector focus overwhelmingly on reducing methane emissions from enteric fermentation and optimizing nutrient cycling to reduce N₂O losses. Globally, the most cost-effective mitigation path often involves improving efficiency—producing the same or more output (milk/meat) from fewer animals, thereby reducing the GHG intensity (Gerber et al., 2013).

4.1. The Methane Challenge: Enteric Fermentation: Methane (CH₄) from the digestive process of ruminants accounts for over 70% of the sector's total emissions. The mitigation focus is on disrupting the methanogenesis process in the rumen.

- **Nutritional Strategy:** Improving diet digestibility means less energy is wasted as methane (methane production represents a loss of up to 12% of feed energy). This includes using higher quality forages, strategically supplementing with concentrates, and improving the balance of energy and protein in the diet.

- **Novel Feed Additives:** This is the most rapidly evolving and promising area of research.
- **3-Nitrooxypropanol (3-NOP):** This synthetic compound (marketed as Bovaer®) works by specifically inhibiting the methyl-coenzyme M reductase (MCR) enzyme, the final step in methane production by *archaea* (Hristov et al., 2018). Studies consistently show CH₄ reductions of 20-40% in intensive systems without affecting productivity (Knapp et al., 2014; Teagasc, 2025).
- **Red Seaweed (*Asparagopsis taxiformis*):** This natural additive contains bromoform, a compound that acts as a potent methane inhibitor. Low inclusion rates (~0.2-0.5% of feed) have demonstrated reductions in enteric CH₄ of up to 80-90% in *in vivo* trials (Frontiers, 2020). Large-scale sustainable production and consistency of the active compound are the current challenges (Frontiers, 2020).

4.2. Manure Management and Nutrient Cycling: The handling of manure is a significant source of both CH₄ (under anaerobic storage) and N₂O (from poor soil application).

- **Anaerobic Digestion (Biogas):** In intensive systems, storing manure in sealed tanks and using **anaerobic digesters** captures CH₄ and converts it into renewable energy (biogas/biomethane). This is a powerful mitigation tool that also provides a clean energy source and a nutrient-rich digestate that can be used as fertilizer, replacing high-GHG synthetic fertilizers.
- **Composting and Solid Separation:** In grazing systems, rapid aeration of manure through composting or drying reduces the anaerobic conditions necessary for CH₄ production.
- **Optimizing Nitrogen Use:** Reducing the amount of excess nitrogen excreted by animals (via precise diet formulation) and applying manure and fertilizers based on crop needs minimizes the conditions that lead to N₂O volatilization from the soil.

5. Synthesis: Integrating Co-benefits, Trade-offs, and Policy

5.1. Co-benefits and Trade-offs: The most effective CSL strategies deliver both adaptation and mitigation benefits, often resulting in increased productivity—a crucial third objective.

Strategy	Primary Benefit	Secondary Benefit (Co-benefit)	Potential Trade-off
Improved Diet Quality	CH ₄ Mitigation (Efficiency)	Increase Milk/Meat Yield (Productivity)	Higher cost of high-quality feed, potential land-use change risk.
Silvopastoral Systems	Heat Stress Adaptation (Shade)	CO ₂ Sequestration (Mitigation)	Slower animal movement, initial planting cost, reduced immediate forage area.
Genetic Selection	Heat/Disease Tolerance (Adaptation)	Decrease CH ₄ Intensity (Mitigation)	Initial high cost, potential loss of local genetic diversity if not managed carefully.

A significant trade-off often exists between maximizing short-term productivity and long-term resilience. For instance, selecting solely for high-input, high-output productivity (lowering CH₄

intensity) can increase an animal's vulnerability to feed and climate shocks (Bashiru & Oseni, 2025). Sustainable solutions must prioritize resilience alongside mitigation.

5.2. Policy and Economic Imperatives: Scaling CSL requires supportive policies that address financial and institutional barriers.

- **Policy Integration:** Adaptation and mitigation must be integrated into national agricultural policies, Nationally Determined Contributions (NDCs) under the Paris Agreement, and national development plans.³⁹ Incentives, such as carbon credit schemes for methane reduction or subsidies for feed additives and PLF technology, are vital for overcoming the initial high investment costs (World Bank, 2017).
- **Inclusive Finance:** Financial mechanisms, such as Index-Based Livestock Insurance (IBLI), provide economic safety nets that allow vulnerable pastoralists to recover from climate-induced losses, fostering the long-term adoption of resilient practices.
- **Research and Extension:** Significant investment is still required to develop low-cost, effective solutions for grazing and pastoral systems, where feed additives are harder to deliver and genetic improvement is slower. Robust extension services are needed to transfer this knowledge to smallholder farmers (UNFCCC, 2020).

6. Conclusion: The Path to a Resilient and Low-Carbon Livestock Future

The future of global livestock production hinges on the sector's ability to navigate the complex interactions between climate change, food security, and environmental stewardship. Adaptation and mitigation are not discrete goals but intertwined elements of a single, necessary transformation toward Climate-Smart Livestock. By combining novel, high-impact mitigation technologies (like 3-NOP and seaweed) with fundamental adaptation strategies (like genetic selection for resilience and climate-smart water/feed management), the sector can meet rising global demand while reducing its environmental footprint and safeguarding the livelihoods of its most vulnerable producers. Achieving this will require committed political will, equitable financing, and a sustained focus on local, context-specific, and inclusive innovation.

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Greening the Desert: Extension Innovations Driving Organic Agriculture in Rajasthan

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Organic agriculture has emerged as a critical pathway toward sustainability, particularly in resource-poor ecosystems like Rajasthan, India. It offers solutions to soil degradation, biodiversity loss, and declining water resources. Yet, despite growing policy and market support, adoption remains uneven. Agricultural extension plays a transformative role in bridging this adoption gap linking research, policy, and practice through awareness, capacity-building, innovation diffusion, and institutional facilitation. This chapter examines how agricultural extension contributes to promoting organic farming in the state of Rajasthan, drawing upon empirical data, policy frameworks, and field-level experiences. It analyses extension strategies, innovations, and challenges in arid and semi-arid regions. The findings reveal that extension-led interventions such as farmer field schools, cluster approaches, digital platforms, and participatory learning have significantly improved awareness, soil health, and farmer incomes. The paper concludes with strategic recommendations to strengthen extension systems, integrate policy support, and scale organic transitions sustainably in dryland ecosystems.

Keywords: Agricultural Extension, Organic Farming, Innovations, Farmers, Rajasthan

Introduction

Global agriculture is experiencing a paradigm shift from chemical-intensive systems toward ecologically responsible, low-input, and resilient models. Organic farming defined by the FAO as a holistic production system that sustains soil, ecosystem, and human health has become a cornerstone of this transformation. In India, the government and several states have adopted ambitious organic farming programmes, recognising its potential for environmental protection, food safety, and livelihood resilience. Rajasthan, the largest state of India, exemplifies both the challenges and opportunities of this transition. Despite its arid climate, limited water, and fragile soils, it has shown promising results in expanding organic cultivation through innovative extension mechanisms.

Agricultural extension services whether public, private, or community-based are pivotal in translating scientific knowledge into practice. They help farmers adopt improved organic methods, link them to certification and markets, and support collective learning. This chapter explores how extension innovations are driving organic transformation in Rajasthan, focusing on data-driven evidence, farmer experiences, and institutional practices.

According to the International Federation of Organic Agriculture Movements (IFOAM) organic farming rests on four core principles *i.e* health, ecology, fairness, and care. These guide sustainable production practices such as composting, green manuring, crop rotation, biological pest control, and biodiversity conservation. In Rajasthan's context, where soils are fragile and inputs costly, organic practices can improve long-term fertility and resilience. However, transitioning requires strong extension support for knowledge transfer, demonstration, and risk mitigation. Agricultural extension is the process through which knowledge and technologies are shared with farmers to improve productivity, income, and sustainability. In the organic domain, extension performs six major functions:

- Awareness creation and motivation for adoption
- Demonstration of organic techniques such as composting, bio-fertilizer use, and crop diversification
- Capacity building through structured training and field schools
- Facilitation of certification, documentation, and market linkages
- Strengthening of farmer cooperatives and producer groups
- Feedback and evaluation for continuous improvement

Organic Farming in India and Rajasthan: An Overview

India is one of the world's largest producers of organic products, covering nearly 2.76 million hectares of cultivated organic area and over 11 lakh farmers under certification (APEDA, 2024). Major states include Madhya Pradesh, Rajasthan, Maharashtra, and Uttarakhand. The country produced over 3.6 million tonnes of certified organic products in 2023-24. India's dual certification frameworks such as NPOP (National Programme for Organic Production) and PGS-India (Participatory Guarantee System) enable both export-oriented and domestic market participation. Rajasthan accounts for approximately 15 per cent of India's total organic farming area, with over 1.5 lakh hectares under certified cultivation (GoR, 2023). The Rajasthan Organic Farming Policy (2017) identified organic farming as a strategic priority to enhance soil fertility and reduce input dependency. Crops such as bajra, wheat, pulses, oilseeds, and spices (cumin, coriander) dominate organic systems. The arid and semi-arid zones of western Rajasthan, once considered unsuitable for

intensive agriculture, are now emerging as centres of organic innovation. Organic farms in southern Rajasthan recorded 20-30 per cent higher net returns than conventional systems due to reduced input costs and premium pricing (Bhattacharyya and Chakraborty, 2015). Organic methods improved soil organic carbon by 19-20 per cent, with better moisture retention and water-use efficiency (Anonymous, 2025). Wild-collection organic produce from Rajasthan reached 10,655 metric tonnes in 2023–24. These statistics highlight both progress and potential largely catalysed by extension-led initiatives.

Extension Innovations and Practices in Rajasthan

Awareness and Motivation: The first step in organic adoption is behavioural change. Rajasthan’s Department of Agriculture, Krishi Vigyan Kendras (KVKs), and NGOs have organised awareness campaigns, field demonstrations, and farmer fairs. Such fertilizer use and soil health benefits, translating abstract ecological benefits into tangible economic language

- **Farmer Capacity Building:** Extension agents conduct training on vermicomposting, integrated pest management (IPM), and bio-input production. Programmes like “Cluster Organic Farming Model” supported under Paramparagat Krishi Vikas Yojana (PKVY) have trained thousands of farmers. Studies show that farmers who attended structured training achieved significant improvements in yield and income, reflecting the value of targeted skill development.
- **Participatory Learning and Farmer Field Schools:** Rajasthan has increasingly embraced Farmer Field Schools (FFS) and Participatory Rural Appraisal (PRA) approaches. These methods treat farmers as co-learners rather than recipients, encouraging experimentation and adaptation to local soil and water conditions. Clusters of farmers jointly manage demonstration plots, exchange inputs, and share observations, which fosters community ownership and social learning.
- **Demonstration Plots and On-Farm Trials:** Hands-on learning through demonstration plots has proven highly effective. In Jodhpur, Barmer, and Sirohi districts, extension officers established model farms showcasing entire organic packages from composting to pest control. Such demonstrations reduce perceived risk and build farmer confidence in yield stability.
- **Digital and ICT-based Extension:** The integration of ICT tools (mobile-based advisories, WhatsApp farmer groups, video tutorials) has broadened outreach. Rajasthan’s arid geography poses challenges of connectivity, but hybrid approaches combining field visits with digital support are showing promises
Farmers can now access pest alerts, price updates, and training materials in local language.
- **Certification and Market Linkages:** Extension officers increasingly assist farmers in navigating certification helping maintain records, coordinate group audits, and liaise with certifying bodies. Cluster certification has reduced individual costs by over 40 per cent. Linkages with organisations

like Morarka Organic have connected over 70,000 farmers across six districts, demonstrating how extension can integrate production, certification, and marketing within a single support ecosystem.

Empirical Insights from Rajasthan

The data-driven observations and field-level experiences from different districts of Rajasthan, highlighting the following economic, environmental, and social outcomes of organic farming as supported by extension services.

- **Economic and Social Impact:** Field surveys show that organic farmers in Rajasthan earned 20-30 per cent higher net returns than conventional farmers due to input savings and price premiums. Labour employment increased by 15-20 per cent, strengthening rural livelihoods. Women's participation also rose as organic farming relies more on composting, seed cleaning, and manual operations.
- **Soil and Environmental Impact:** Soil organic carbon levels improved from 13.8 g/kg to 16.5 g/kg, and moisture retention rose by 20 per cent, enhancing drought resilience. Organic farming also reduced pesticide residues in soil and groundwater, improving ecological health in water-scarce regions.
- **Farmer Innovators and Local Champions:** Rajasthan's organic movement has been powered by local champions-farmers who experiment, succeed, and share. In Jhalawar district, an award-winning farmer trained dozens of others with support from local extension officers. These "lead farmers" act as grassroots extension multipliers.

Constraints and Emerging Challenges

Despite promising outcomes, organic farming in Rajasthan faces several structural, institutional, and ecological challenges. The following are the major barriers that must be addressed for wider adoption:

- Limited extension manpower with organic expertise
- Inadequate supply of organic inputs (bio-fertilizers, seeds etc.)
- High certification costs and documentation burdens
- Weak marketing infrastructure for organic produce
- Pest and disease pressure in arid zones
- Lack of systematic monitoring and evaluation frameworks

Strategies for Strengthening Extension Support

The following are practical, evidence-based strategies to enhance the effectiveness of extension services in supporting organic agriculture across the state:

- **Capacity Enhancement:** To enhance capacity in organic agriculture, organic farming modules should be incorporated into agricultural extension training curricula to strengthen the technical

competence of extension personnel. Establishing district-level Organic Resource Centres will provide practical, hands-on learning opportunities and serve as hubs for demonstrations and knowledge sharing. Additionally, recognising and rewarding extension agents for their efforts in promoting organic practices will motivate greater engagement and encourage wider adoption of organic farming among farmers.

- **ICT-Driven Advisory Systems:** To strengthen ICT-driven advisory systems, mobile applications should be developed in local languages to provide farmers with real-time guidance tailored to their needs. Short instructional
- Videos and social media platforms can be leveraged to showcase farmer success stories, helping to inspire wider adoption of improved practices. Additionally, promoting digital certification tracking will enhance transparency and build trust in organic value chains.
- **Farmer Organisation and Cluster Models:** To promote farmer organisation and cluster-based models, dedicated Farmer Producer Organisations (FPOs) should be formed and strengthened to support organic value chains. Facilitating group certification and establishing shared marketing platforms will help reduce costs and improve market access for smallholders. Additionally, integrating women's self-help groups (SHGs) into organic input production and value-addition activities will enhance livelihood opportunities and ensure more inclusive participation in organic agriculture.
- **Research-Extension-Market Linkages:** To support research-extension- market linkages, collaboration among agricultural universities, KVKs, NGOs, and private sector stakeholders should be actively promoted to ensure seamless knowledge flow and innovation. Extension efforts must be aligned with the policy objectives outlined in the Rajasthan Organic Farming Policy (2017) to maintain coherence and direction. Furthermore, supporting participatory research will help tailor organic farming technologies to local agro-ecological conditions, making them more practical and adoptable for farming communities.

The Rajasthan experience illustrates that effective extension services form the foundation of a successful shift toward organic farming. When farmers are provided with regular training, practical demonstrations, and strong institutional backing, the adoption of organic practices rises significantly.

Yet, extension strategies must remain flexible and responsive to emerging challenges, including water stress, fluctuating markets, and evolving policy frameworks. Ultimately, long-term progress will depend on strengthening institutional capacities, fostering farmer-led initiatives, and establishing continuous feedback mechanisms.

Conclusion

Organic farming represents not just a production shift but a paradigm of ecological responsibility and livelihood resilience. In Rajasthan's fragile desert landscapes, innovative extension approaches such as awareness drives, digital advisory tools, farmer collectives, and participatory learning are gradually transforming the terrain and revitalising local agriculture. To expand this progress, extension system reforms must be closely aligned with organic farming policies, supported by stronger market connections, and backed through continuous capacity building initiatives. With steady investment and coordinated efforts, Rajasthan can evolve into a leading example of dry land organic farming globally. Ultimately, advancing toward a chemical-free and climate-resilient future relies on empowering farming communities, reinforcing institutional systems, and ensuring sustained support across the entire organic value chain.

Role of Veterinarian in Meeting the Challenges of Health and Welfare of Livestock and Food Security

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This chapter highlighted the veterinarian's vital role in modern farming and public health by focusing on their work in livestock health and welfare, which directly impacts food security and public safety. Veterinarians address health challenges through disease prevention, diagnosis, and treatment, and contribute to food security by ensuring safe and productive livestock through herd health programs and biosecurity measures. Their role extends to broader public health issues, including zoonotic disease control and risk management across the entire food chain, from farm to table

1. INTRODUCTION

A veterinarian's role in meeting the challenges of livestock health, welfare, and food security is multi-faceted, encompassing disease prevention and control, ensuring food safety from farm to table, and developing guidelines and standards. This includes crucial tasks like epidemiological surveillance, ante- and post-mortem inspections and managing public health issues like zoonotic diseases. The profession is increasingly embracing a 'One Health' approach, recognizing the interconnectedness of animal health, human health and the environment.

As veterinarians play a crucial and complex role in tackling the problems of livestock health, welfare, and food security. Here, in this chapter some important salient features of their duties are mentioned in brief.

2. KEY ASPECTS OF VETERINARIAN'S RESPONSIBILITIES

A. Disease Prevention and Control - Veterinarians develop and implement vaccination

Absolutely! Disease prevention and control are critical components of livestock management and public health. Veterinarians play a vital role by designing and executing strategies such as:

- i) Vaccination Programs:** Developing schedules and administering vaccines to protect animals from infectious diseases, thereby reducing the risk of outbreaks.
- ii) Biosecurity Measures:** Implementing protocols like controlled access to farms, sanitation procedures, and quarantine practices to prevent the introduction and spread of pathogens.

iii) Disease Surveillance: Monitoring animal health through regular testing, reporting, and data analysis to detect early signs of disease emergence and respond promptly



These efforts help safeguard livestock populations, ensure food safety, and protect public health by minimizing the impact of infectious diseases on the food supply chain. Veterinarians are crucial for disease prevention and control by monitoring animal populations for early outbreak detection, implementing control strategies, and safeguarding public health through a One Health approach.

B. Animal Health and Welfare:

Animal health and welfare are ensured by providing humane handling, proper nutrition, and appropriate living conditions, which improves productivity and product quality. This includes protecting animals from pain, injury, and disease, allowing them to express normal behaviors, and ensuring they are free from fear and distress. A commitment to animal welfare fosters ethical practices and can lead to better growth rates and reproductive performance

Key aspects of animal health and welfare -

- **Humane handling:** Practices like gentle handling and avoiding painful procedures are crucial. For example, routine management practices that can be painful, such as certain mutilations, are banned in organic farming .
- **Proper nutrition:** Ensuring animals receive a balanced diet and sufficient water is vital for their physical and mental well-being. Nutritional welfare can be improved by creating diets tailored to species-specific needs.
- **Appropriate living conditions:** Animals need adequate space, healthy environments, and access to fresh water. This can include a proper living environment for the species, appropriate housing, and protection from physical hazards.
- **Health and disease prevention:** This involves the prevention and treatment of diseases, along with regular hygiene and vaccination programs.

- **Mental and emotional well-being:** This includes ensuring animals are free from fear and distress and can express normal behaviors, which can be supported by appropriate housing and social groupings for social species.

Benefits of good animal welfare

- **Enhanced productivity:** Animals with good welfare often have better growth rates and reproductive performance.
- **Improved product quality:** A focus on animal health and proper nutrition contributes to higher quality products.
- **Economic viability:** Better health and productivity can improve the economic viability of farming operations.
- **Sustainability:** Promoting good animal welfare aligns with creating more sustainable and socially responsible agricultural practices.

C. Food Safety and Public Health:

Food safety and public health are critically supported by veterinarians through their role in monitoring and regulating animal-derived food products. They ensure that meat, dairy, and other animal products are safe for human consumption by inspecting farms, processing facilities, and markets. Veterinarians also work to prevent zoonotic diseases—those transmitted from animals to humans—by controlling infections in animal populations and implementing biosecurity measures. Additionally, they ensure compliance with food safety standards and regulations, reducing the risk of food borne illnesses and safeguarding public health.

How veterinarians ensure food safety -

- **Farm-level surveillance:** Veterinarians monitor animal health and welfare on farms, helping to prevent diseases that could be transmitted to humans
- **Disease prevention:** They play a key role in preventing zoonotic diseases (diseases that can spread from animals to humans) from entering the food supply.
- **Slaughterhouse inspections:** A traditional role has been inspecting carcasses and meat products after slaughter to ensure they meet hygiene and safety standards.
- **Risk management:** They advise farmers and producers on how to control hazards like drug residues and mycotoxins in animal feed and primary production.

- **Regulatory compliance:** They help ensure that food producers comply with national and international food safety regulations and standards.
- **"Stable to table" approach:** Veterinarians are trained to audit and ensure the safety and public health standards throughout the entire food chain, from the animal's living conditions to the final food product.

D. Research and Innovation:

Research and innovation contribute to sustainable agriculture by focusing on emerging diseases, improving sustainable farming practices like crop rotation, and developing new technologies such as AI and sensor-based systems to boost livestock health and food production efficiency. This includes creating tools for better animal welfare, more efficient resource management, and developing healthier, more resilient food systems to meet future challenges.

i) Research on emerging diseases and animal health

- **One Health approach:** A "One Health" approach emphasizes the interconnectedness of human, animal, and environmental health, focusing on disease prevention and managing antimicrobial resistance.
- **Monitoring and early detection:** Health monitoring systems and technologies are being developed to detect illnesses in livestock early, improving welfare and reducing the economic and environmental impact of diseases.
- **Reproductive technologies:** Innovations in artificial insemination (AI) and other breeding technologies have significantly increased dairy and meat production by improving genetic traits..

ii) Innovations in sustainable farming practices

- **Crop rotation and soil health:** Sustainable practices like crop rotation are being updated to improve soil health, control pests, reduce disease, and increase resilience to climate variations and market changes.
- **Water-efficient cropping:** Research focuses on more water-efficient cropping methods to reduce water usage in agriculture.
- **Organic farming:** This approach relies on natural principles like composting and biodiversity to produce food while avoiding synthetic chemicals and reducing carbon emissions.

iii) New technologies to improve efficiency and productivity

- **Precision agriculture:** This involves using technologies like GPS, sensors, and drones to monitor and manage crops and soil with high precision, leading to increased yields and efficiency.

- **Sensors and data analytics:** A wide range of sensors (acoustic, electromagnetic, optical, etc.) are used to collect real-time data on soil conditions, plant health, and animal behavior, which is then analyzed to inform decision-making.
- **AI and machine learning:** Artificial intelligence is being integrated into crop management for tasks such as predictive analytics and resource optimization to increase yields and resilience.
- **Automation:** The use of robots and automation in tasks like planting, monitoring, and harvesting can improve efficiency and reduce labor demands.
- **Digital marketplaces:** The expansion of digital platforms connects farmers to markets, information, and resources more efficiently.

E. Roles in comparative medicine –

Veterinarians are crucial in comparative medicine due to their expertise in the diseases of multiple species.

Roles in comparative medicine

- **Research and discovery:** Veterinarians apply their knowledge of different animal species to study diseases that affect both animals and humans. They have been instrumental in research on diseases like scrapie and kuru, understanding viruses, and developing treatments for conditions like HIV.
- **Bridging human and animal health:** Their expertise helps advance the "One Health" concept by working on issues that impact human, animal, and environmental health. This includes disease surveillance, food safety, and controlling emerging infectious diseases that can spread from animals to humans.
- **Biomedical research support:** Veterinarians specialized in laboratory animal medicine ensure the highest quality of animal care and husbandry in research settings. They are vital for creating and enforcing policies and procedures that protect animal welfare and the integrity of research outcomes.
- **Public health and safety:** They play a key role in preventing and controlling zoonotic diseases (those that jump from animals to humans), like rabies and foodborne illnesses. This includes public health surveillance and biosecurity measures to protect against bioterrorism and other threats.
- **Education:** Veterinarians in academia contribute to comparative medicine by teaching students in human medical schools and encouraging research among their peers. They also help design new research facilities and create institutional policies to further research and maintain animal welfare, bridging human and animal health through research, public health, and clinical practice.

F. Policy and Advocacy:

Veterinarians act as policy advisors on regulations for animal health, welfare, and food security by providing scientific expertise and advocating for sustainable and ethical practices. This involves collaborating with policymakers to create legislation for humane treatment, participating in public

discussions, and promoting awareness through education and partnerships. Their role extends to national and international levels, ensuring policies are built on a foundation of animal health, food safety, and biosecurity.

Key areas of policy and advocacy

Animal health and welfare: Veterinarians advise on legislation to protect animals from abuse and neglect, ensuring humane handling and preventing the spread of disease. They help set standards and provide expertise on everything from proper nutrition to behavioral health issues.

- **Food security and safety:** They ensure food production practices are safe by helping to set regulations for veterinary drugs and pesticides. They also advise on policies that support sustainable livestock farming and protect the food chain.
- **Sustainable and ethical practices:** Veterinarians advocate for practices that consider environmental, economic, and social factors. This includes promoting sustainable methods in animal production, improving animal welfare as a component of sustainability, and aligning animal health with public and planetary health goals, notes this article from the National Institutes of Health .
- **Public health and biosecurity:** Their expertise is crucial for national security, as they are a key defense against the accidental or deliberate introduction of animal diseases that could also impact human health (zoonoses). They also play a role in public health by providing education on issues like zoonotic disease prevention.
- **Collaboration and education:** They work with governments, veterinary associations, non-governmental organizations, and the public to inform policy and promote better animal care practices. They also educate owners and producers on best practices.

G. Emergency Response and Crisis Management:

During emergencies, veterinarians coordinate response efforts to control disease outbreaks and natural disasters by assessing risks, implementing control measures, and providing logistical support to protect animal health and the food supply chain..They work with local, state, and national authorities to manage the immediate crisis and ensure public safety, including managing carcasses and preventing the spread of zoonotic diseases.

Key roles in emergency response:

- **Coordination:** Veterinarians coordinate with various authorities, such as state disaster management and animal health services, to create and implement response plans.
- **Disease control:** They implement local disease control measures like vaccination programs and quarantine procedures.
- **Risk assessment:** Veterinarians are crucial in the preparedness phase for risk assessment, identifying potential weaknesses, and developing mitigation strategies to protect both animal and human health.

- **Public health:** They play a critical role in preventing the spread of zoonotic diseases from animals to humans, especially after events like a natural disaster..
- **Food safety:** They help maintain food safety standards by managing issues like carcass disposal and ensuring the integrity of the food supply chain during a crisis.
- **Logistical support:** They provide logistical and technical support, which is vital for effective management during large-scale emergencies.
- **Animal welfare:** Their work extends beyond medical interventions to include supporting animal recovery efforts, which is essential for community recovery.

H. Education and Extension Services:

Education and extension services are crucial for livestock management by providing farmers, stakeholders, and the public with the knowledge and skills for best practices in animal health, disease prevention, and food safety. These services bridge the gap between scientific research and farm-level application through methods like workshops, demonstrations, and one-on-one consultations.

Their ultimate goal is to improve the quality of life for those involved, increase productivity, and ensure the sustainability and food security of the livestock sector.



Key roles and functions

- **Livestock management:** Extension services offer guidance on a wide range of topics, from optimal feeding and nutrition to herd management and biosecurity practices like cleaning and disinfection.
- **Disease prevention:** They help implement health protocols to prevent and address diseases, contributing to overall herd health and productivity. This includes training on biosecurity measures, such as cleaning and disinfection techniques.

- **Food safety:** Services educate workers on essential hygiene practices and the proper handling of raw materials to ensure the safety of the final food products .
- **Skill development:** They are essential for upskilling and reskilling the workforce to meet current job requirements and prepare for future roles, supporting the overall sustainability of the livestock sector.
- **Knowledge dissemination:** They act as a critical link to spread evidence-based knowledge from research institutions to end-users, although this can be challenged by inefficient communication pathways.
- **Problem-solving:** Extension services help farmers identify problems, find solutions, and evaluate the results within their specific farming system.

Methods and delivery

- **Workshops and demonstrations:** These are practical ways to teach new techniques and procedures.
- **One-on-one consultations:** Personalized advice helps stakeholders make informed decisions tailored to their specific situation.
- **Training materials:** Creating visual aids and using in-person teaching methods can lead to better learning outcomes, especially for employees who may not have English as a first language.

I. Sustainable Livestock Production:

Veterinarians promote sustainable livestock production by using practices that enhance animal welfare, reduce environmental impact, and maintain productivity. These practices include optimizing nutrition, improving animal health through early disease detection and responsible medication use, and integrating technologies like precision farming to monitor and improve resource efficiency. This balanced approach helps ensure long-term food security by making the livestock sector more resilient, ethical, and environmentally sound.



Practices veterinarians promote

- **Animal welfare:** Ensuring ethical treatment through humane slaughter, pain management, and protection from harm.
- **Resource efficiency:** Optimizing feed use, reducing water and land degradation, and managing manure effectively to reduce waste and greenhouse gas emissions.
- **Disease management:** Implementing comprehensive health programs, including early disease detection and using only approved veterinary products to prevent environmental contamination.
- **Environmental stewardship :** Incorporating practices like rotational grazing and agroforestry (integrating trees into livestock systems) to improve soil health, biodiversity, and carbon sequestration.
- **Technological integration:** Utilizing precision livestock farming (PLF) and other data-driven tools for real-time monitoring to improve decision-making and resource management.
- **Responsible medication:** Using medications and vaccines registered and approved by local authorities and avoiding the use of non-therapeutic antibiotics or other harmful substances, as outlined in guidelines like those from www.sustainableagriculture.eco .

How this contributes to long-term food security

- **Economic viability:** Sustainable practices can improve productivity and efficiency, ensuring the long-term financial success of producers, notes [the Journal of STM Journals](#) .
- **Environmental resilience:** By minimizing environmental impact, these practices help protect natural resources needed for future food production.
- **Food availability:** Optimizing production systems ensures a consistent supply of animal-sourced foods, which are vital for nutrition for many people, explains [HealthforAnimals](#) .
- **Ethical and cultural acceptance:** Meeting growing consumer demand for ethically produced food helps maintain public support for the livestock sector, notes [the National Institutes of Health](#) .

In summary, veterinarians play a crucial role in ensuring the health and welfare of livestock, which directly impacts food security, public health and sustainable agricultural development.

3. CHALLENGES AND FUTURE PROSPECTS

Veterinarians face challenges like financial burdens, high emotional and physical stress, understaffing, and a poor work-life balance, compounded by administrative demands. The future holds opportunities in areas like telemedicine , public health roles related to climate change, animal welfare, and food safety, requiring new skills in environmental stewardship, and adapting to corporate veterinary models and consumer demands.

4. CONCLUSION

The conclusion on the role of a veterinarian is that their work extends far beyond individual animal care to encompass a vital, multidisciplinary role in public health, environmental protection, and

society as a whole. They are essential in the "One Health" approach, connecting animal, human, and environmental health, which includes preventing zoonotic diseases, ensuring food safety, and contributing to research and welfare. Their expertise is crucial for both animal well-being and protecting the broader community.

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Research Trends in Environmental Sciences

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Environmental science is undergoing rapid transformation driven by technological advances, growing awareness of planetary limits and an urgent need to translate science into equitable policy and practice. This chapter surveys major research trends shaping the field in the early-to-mid 2020s: applications of artificial intelligence (AI) and remote sensing for environmental monitoring; nature-based solutions (NbS) and ecosystem restoration; microplastics and emerging contaminants; environmental informatics and big-data synthesis; climate-adaptation science and resilience; and innovations in remediation and circular economy approaches. For each theme I summarize drivers, representative methods, recent advances, persistent gaps and suggested directions for future research. The chapter closes with a short synthesis highlighting interdisciplinarity, equity and translational research as cross-cutting priorities. Key claims are supported by recent reviews and scientometric studies.

Introduction

Environmental science has shifted from largely descriptive studies toward highly integrated, data-driven and solution-oriented research. Several forces drive this shift: the proliferation of environmental sensors and satellite assets advances in computational capacity and machine learning, escalating evidence of persistent pollutants and biodiversity loss, and stronger demands from policymakers and communities for actionable science. Together, these forces favor approaches that are interdisciplinary, scalable and geared toward implementation and justice.

Recent bibliometric and review studies corroborate rising research activity in AI-enabled monitoring, nature-based adaptation and microplastics, among other areas.

1. Artificial Intelligence, Machine Learning and Remote Sensing

1.1 Why it matters

The combination of AI and the explosion of remote-sensing data (satellite, airborne, UAV, hyperspectral, radar) has created unprecedented capacity for high-resolution, near-real-time monitoring of land use, water quality, vegetation health, air pollution plumes and disaster dynamics. This enables faster detection of environmental change and improved attribution of drivers. Recent reviews describe substantial uptake of machine-learning techniques in classification, change detection, data fusion and predictive modeling across environmental domains.

1.2 Methods and applications

Common methods include convolutional neural networks for image classification, ensemble learners for land-cover mapping, deep learning for hyperspectral analysis and hybrid physics-ML models (e.g., physics-informed neural networks) for system understanding. Applications span crop yield estimation, inland and coastal water monitoring, wildfire detection, urban heat mapping, and biodiversity surveys using automated image/audio classification. Explainable AI (XAI) is emerging as a priority to improve transparency in regulatory and stakeholder contexts.

1.3 Challenges and research gaps

Key limitations include biases in training data, transferability across regions/sensors, uncertainty quantification, and the need for standards for model validation and interpretability. Future research should emphasize open, reproducible benchmarks, hybrid models that blend domain knowledge with ML flexibility and investment in capacity building for low-resource regions.

2. Nature-Based Solutions (NbS) and Ecosystem Restoration

2.1 The rise of NbS

Nature-based Solutions-interventions that protect, sustainably manage and restore ecosystems to address societal challenges-have gained traction as multi-benefit strategies for climate mitigation/adaptation, biodiversity conservation and human well-being. Bibliometric analyses reveal rapid growth in NbS literature and a broadening of topics from urban green infrastructure to coastal restoration and agroecosystem interventions.

2.2 Research directions

Active research areas include (i) rigorous performance evaluation of NbS under diverse climatic and socio-ecological contexts; (ii) co-design and governance frameworks to ensure equity and local stewardship; (iii) carbon accounting and trade-offs between biodiversity and climate objectives; and (iv) scaling from pilot projects to landscape-level implementation. A consistent finding is an

implementation gap-evidence and pilot successes often do not translate to large-scale action without supportive institutions and finance.

3. Microplastics and Emerging Contaminants

3.1 Why focus on micro/nanoplastics

Microplastics and nanoplastics are now ubiquitous across ecosystems and have been detected in drinking water, food, air and even human tissues. Research attention has surged because of potential ecological and human-health implications and difficulties in detection, quantification and remediation. Scientometric studies indicate sharp increases in publications and new methodological advances for detection and treatment.

3.2 Advances in detection and remediation

Technological advances include improved microscopy and spectroscopy for nano-particle detection, mass-spectrometric approaches and novel sorbents/filters (including bio-based materials) for water treatment. Research into photocatalytic degradation and innovative materials (e.g., composite sponges) shows promise for removing microplastics, but scalability, lifecycle impacts and by-product risks require careful evaluation.

3.3 Knowledge gaps

Uncertainties remain around exposure pathways, dose-response relationships for human health and standardized sampling/analysis protocols. Priorities include harmonizing analytical standards, long-term monitoring and interdisciplinary toxicology studies.

4. Environmental Informatics, Big Data and Synthesis Science

4.1 Integration at scale

Environmental informatics-the fusion of data science, geospatial science and domain knowledge-allows large, heterogeneous datasets (sensors, citizen science, remote sensing, omics, socio-economic datasets) to be integrated for systems-level insights. Recent reviews emphasize the role of informatics in multi-dimensional global change studies and in enabling reproducible, transparent synthesis.

4.2 Tools and methods

Key methods include metadata standards, ontologies, federated data systems, causal inference techniques, and model-intercomparison frameworks. There's also growing interest in digital twins for coupled human-environment systems and in using federated learning to respect data governance constraints while enabling model training across jurisdictions.

5. Climate Adaptation, Resilience and Human Dimensions

5.1 From vulnerability assessment to anticipatory systems

Climate-adaptation research is maturing from vulnerability mapping toward anticipatory and transformational approaches-early warning systems, anticipatory governance and socioecological resilience that explicitly integrates livelihoods, equity and governance. Scientometric studies show increasing attention to adaptation pathways and to human dimensions (health, migration, food security).

5.2 Participatory and transdisciplinary methods

Co-production of knowledge with communities, scenario planning and decision-support tools that incorporate stakeholder values are becoming standard facets of high-impact adaptation research. Embedding monitoring and evaluation into adaptation projects is critical to closing the learning loop.

6. Pollution Remediation, Circular Economy and Green Chemistry

Research is shifting from end-of-pipe solutions toward prevention and circular strategies: designing out toxic substances, material reuse and recovery and green chemistry solutions for safer alternatives. Photocatalysis, bio-remediation, and engineered materials (e.g., carbon-based sorbents, functionalized membranes) are active research fronts that aim to make remediation more efficient and sustainable. Lifecycle assessment and techno-economic analysis are important complements to lab-scale innovation.

7. Equity, Environmental Justice and Policy Translation

Increasingly, environmental research recognizes justice and governance as central scientific questions: who benefits from green interventions, how risks are distributed and how science informs fair policy. Recent literature emphasizes co-design, inclusion of Indigenous and local knowledge systems and the need for research metrics that reward translational impact and community engagement. Translational pathways-clear connections from evidence to policy, finance and practice-remain a bottleneck for many promising solutions.

8. Future Directions and Research Agenda (select priorities)

1. Responsible AI and explainability: develop transparent, uncertainty-aware AI tools with benchmarking datasets and regionally representative training data.
2. Standardization for contaminants: harmonize micro/nanoplastic sampling and analysis methods and invest in longitudinal exposure studies.
3. Scaling nature-based solutions: focus on governance, finance mechanisms and trade-off assessments to move NbS from pilots to landscape scales.
4. Interoperable data ecosystems: build federated, FAIR (Findable, Accessible, Interoperable, Reusable) infrastructures that enable cross-disciplinary synthesis while respecting data sovereignty.
5. Equitable translational science: prioritize co-production with vulnerable communities, embed social science into environmental interventions, and measure social outcomes alongside ecological metrics.

Conclusion

Environmental science in the 2020s is characterized by rapid technological uptake, a stronger orientation toward solutions, and an increasing emphasis on equity and governance. The most impactful research will be transdisciplinary, methodologically rigorous, and tied to mechanisms for implementation. Addressing standardization, transparency and inclusivity will be essential to translate scientific advances into resilient, just environmental outcomes.

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Organic Farming: Principles and Practices

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Since independence, India's primary agricultural challenge has been feeding its booming population. While conventional farming remains necessary to meet immediate demand, the current environmental and economic problems facing the sector demand a shift toward sustainability. Organic farming offers a vital solution. It is recognized as an ecologically, socially, and economically viable agricultural system that directly addresses the objective of sustainable agriculture. Organic practices prioritize biological inputs and natural processes (like composting and crop rotation) over the heavy, often indiscriminate, use of chemical fertilizers and pesticides.. Therefore, organic farming must be certified and supported where it already exists, and systematically promoted into new areas as a key solution to ensure the long-term health and security of India's food production system

Organic Farming

Organic farming is a holistic production system that rejects the use of synthetic fertilizers, pesticides, and genetically modified organisms (GMOs). Its fundamental philosophy is rooted in the belief that the health of the soil, plant, animals, and humans are intrinsically linked. By operating within natural ecological cycles, organic agriculture aims to enhance biodiversity, promote biological activity, and foster a sustainable, long-term food supply.

Concept of organic farming

Organic farming includes the concept that the soil, plant, animals, and humans are linked. Concept of organic farming is based on some below mentioned principles:

1. Nature is considered as a best role model for farming, since it does not use any inputs nor demand unreasonable quantities of water.
2. Organic farming system does not believe in mining of the soil of its nutrients and protect the long-term fertility of soil.
3. The soil in this system is considered as living entity.
4. On sustained basis living population of microbes and other organisms in soil's are significant contributors to its fertility.
5. Development of biological diversity and the maintenance and replenishment of soil productivity is the major concern of organic farming systems.
6. Importance given on crop rotation, natural predators, biological pest, disease and weed management, resistance varieties instead of chemical control.

The main pillars of organic farming

The main pillars of organic farming (Roychowdhury *et al.*, 2013)

1. Organic threshold standards.
2. Reliable mechanisms regarding certification and regulatory affairs.
3. Technology packages.
4. Efficient and feasible market network

The principle aims of organic production and processing

(From IFOAM, 2002)

- To produce food of high quality in sufficient quantities, Operation within natural cycles and closed systems as far as possible, drawing upon local resources,
- To maintain long term fertility and sustainability of soils
- The creation of a harmonious balance between crop production and animal husbandry,
- The securing of high levels of animal welfare,
- The fostering of local and regional production and supply chains, and
- To utilize biodegradable, recyclable and recycled packaging materials;
- To support the establishment of an entire production, processing and distribution chain which is both socially and ecologically responsible;
- To recognize the importance of, and protect and learn from, indigenous knowledge and traditional farming systems.
- To consider the wider social and ecological impact of the organic production and processing system.

The four IFOAM main principles of organic production (IFOAM, 2005)

The Principle of Health – Organic agriculture should sustain and enhance the health of soil, plant, animal, human and planet as one and indivisible.

The Principle of Ecology – Organic agriculture should be based on living ecological systems and cycles, work with them, emulate them and help sustain them.

The Principle of Fairness – Organic agriculture should build on relationships that ensure fairness with regard to the common environment and life opportunities.

The Principle of Care – Organic agriculture should be managed in a precautionary and responsible manner to protect the health and well-being of current and future generations and the environment.

The Important Goals of Organic Farming are:

1. A sufficiently high level of productivity
2. Compatibility of cultivation with the natural cycles of the production system as a whole
3. Maintaining and increasing the long-term fertility and biological activity of the soil.
4. Maintaining and increasing natural diversity and agrobiodiversity
5. Maximum possible use of renewable resources
6. To create an integrated, environmentally sound, safe and economically sustainable agriculture production system.
7. Protection of, and learning from, indigenous knowledge and traditional management systems.

Components of Organic Farming

Different components of organic farming are as follows.

1. **Crop and soil management**
2. **On-farm Waste Recycling**
3. **Non-chemical Weed Management**
4. **Domestic and Industrial Waste Recycling**

Organic Farming in India**Historical background**

The concept of organic agriculture is not alien to India. In fact, the first scientific approach to organic farming dates back to the Vedas of the later Vedic period, the essence of which is to live in harmony with, rather than exploit, Mother Nature. There is brief mention of several organic inputs in our ancient literatures like Rigveda, Ramayana, Mahabharata, Kautilya Arthasashthra etc. In fact, organic agriculture has its roots in traditional agricultural practices that evolved in countless villages and farming communities over the millennium.

More recently, Mahatma Gandhi pioneered organic farming through his constructive programmes in several locations in India. It was the aggressive promotion of the green revolution, which led to a

reverse in the spread of the Gandhian movement's organic farming programme. There is evidence to suggest that small farmers, especially in the Third World, are more likely to apply agricultural practices, such as crop rotations and mixed cropping, which are the essential building blocks of organic farming. They also usually combine agriculture with livestock rearing and utilize the manure to replenish the soil fertility. Empirical evidence further suggests that while conventional agriculture goes better with large holdings, organic farming functions better in small farms. A study by Gupta and Verma (1997) comparing grain production in organic vis-à-vis conventional methods observed that as farm size increases, the advantages of organic rotation become less visible. Further, the study reported that on a smaller scale, organic farming was more profitable and productive than conventional farming.

Government initiative

The lucrative market of the developed world has so far acted as the primary driving force behind the development of the 'certified organic' sector, which is still in a nascent stage in India. According to one estimate, in 1999 merely 0.001 percent of the total agricultural land in India was under certified organic cultivation. It is predominantly the NGOs and people's organizations that have been spearheading organic agriculture movement in different parts of the country during the last two decades. The Government of India has set up a special cell under the Agricultural and Processed Food Export Development Authority (APEDA) of the Ministry of Commerce and Industries (MOCI). The MOCI has come out with the 'National Programme of Organic Products' (NPOP) in 2000, and the 'India Organic' logo in 2002. While these initiatives have been undertaken to promote exports of Indian organic products, the Department of Agriculture and Cooperation has formulated a 'National Project on Organic Farming' to promote organic agriculture as part of an exercise to curb the use of chemical pesticides and make agricultural activity more eco-friendly. Shifting to organic farming, even if it promises higher returns in terms of better prices and international acceptability in the long run, may not be preferred by the majority of farmers as they are dependent on the farm for livelihood and any departure would affect them immediately. To make majority of small farmers shift to organic farming, several subsidies have to be given on organic inputs. Such initiatives are a step in the right direction, but they should be weighed against the huge subsidies that the Central Government has been providing for the production and import of chemical fertilizers and pesticides. There is a great potential for organic farming to flourish in this country and given an appropriate institutional and policy framework, it will not be very difficult to promote the existing 'de-facto organic' farms to the category of certified organic farms. This would enable the small farmers to take advantage of the lucrative market for certified organic products in the developed world, which could directly contribute towards the improvement of their economic well-being.

Accreditation boards in India

Country has recently started to support organic agriculture on a large scale and on various levels. A national regulatory framework (standards, accreditation regulations) has already been passed in 2000. There are various schemes and events to support and facilitate exports of organic products (e.g., the large conference ‘Indian Organic Products – Global Markets’ held in Delhi in December 2002, mainly sponsored by the Indian Government). The Ministry of Agriculture announced that various forms of support for organic producers, processors and traders were to be included within the latest five-year plan. The Ministry of Commerce has identified six organisations as accreditation agencies of organic products, they are (1) Agricultural and Processed Food Products Export Development Authority (APEDA), (2) Tea Board, (3) Spices Board, (4) Coconut Development Board, (5) Directorate of Cashew and Cocoa, and (6) Coffee Board. These accreditation boards give permission to certifying agencies for certifying organic products, following the prescribed norms. Certification through these boards and agencies has been made compulsory, particularly for export market, as ‘the Government of India has issued a public notice according to which no organic products may be exported unless they are certified by an inspection and certifying agency duly accredited by one 22 of the accreditation agencies designated by the Government of India’. Several certifying agencies are functioning in India.

Current situation of organic farming

According to current situation there are two different kinds of organic farm

1. Certified organic farms
2. Non-certified organic farms

In 2015, the export and domestic market of Indian organic industry grew by 30 per cent and 40 per cent respectively. According to IFAD, India has more than 15,000 certified organic farms. These farms provide many ecological benefits and deliver nutritious food. India has 30 per cent of total organic cultivable land in Asia. In India, Sikkim state with 75,000 hectares of land under organic cultivation is an organic state. By 2030, Meghalaya another north-east state of India also eyes to transfer 200, 000 hectares of land into organic cultivation. (NPOF, 2015-16). Due to climate change at global level, organic farming practice has made a significant position. The Indian Government is promoting organic farming through diverse scheme under National Mission Sustainable Agriculture (NMSA). The government has introduced Paramparagat Krishi Vikash Yojna (PKVY) and Organic Value-Added Development schemes under the NMSA to promote organic farming in India. Under this scheme, the state and central government, based on cluster for every 20-hectare land, will support farmers by offering financial assistance for maximum two-hectare land. It primarily aims to

increase soil fertility and thereby helps in production of healthy food through organic practices without the use of agro-chemicals.

Practices guided by core principles of organic farming

Organic farming combines various practices that align with its core principles: health, ecology, fairness, and care. These practices promote sustainable agriculture and safeguard the well-being of the environment, communities, and food systems.

1. Soil management

Composting: Utilizing organic waste to create nutrient-rich compost, enhancing soil fertility and structure.

Crop rotation: Alternating crops on same land in a specific sequence to improve soil health and reduce pest cycles.

Cover cropping: Cover crops are the plants which are grown to improve soil fertility, prevent soil erosion, enrichment and protection of soil, and enhance nutrient and water availability, and quality of soil (Sharma et al., 2018). Planting cover crops during off-seasons to prevent erosion, improve soil structure, and fix nitrogen.

2. Biodiversity enhancement

Polyculture: Growing multiple crops in the same space to enhance biodiversity and reduce pest occurrences.

Native planting: Incorporating inherent species into the farming system to support local ecosystems and pollinators.

3. Pest and disease management

Integrated Pest Management (IPM): Combining biological, cultural, and mechanical control methods to manage pests while minimizing chemical use.

Natural predators: Encouraging beneficial insects and organisms that control pest populations naturally.

4. Water conservation

Drip irrigation: Implementing efficient irrigation systems that deliver water directly to plant roots, reducing waste.

Rainwater harvesting: Collecting and storing rainwater for irrigation to improve water sustainability.

5. Animal husbandry

Free-range systems: Allowing livestock access to outdoor spaces, promoting natural behaviors and animal welfare.

Organic feed: Providing animals with organic, non-GMO feed that meets their nutritional needs without harmful additives.

6. Fair trade practices

Fair pricing: Ensuring that farmers receive fair compensation for their products, supporting economic feasibility and community development.

Community Supported Agriculture (CSA): Establishing direct relationships between consumers and farmers, nurturing community support and local food systems.

7. Sustainable resource use

Minimal tillage: Reducing soil disturbance through reduced-till or no-till practices, preserving soil structure and health. In particular, the widespread use of reduced or no-till farming practices in intensive agriculture has led to great improvements in soil quality, including reduced erosion and retention of soil organic matter (Llewellyn & Ouzman, 2019).

Organic inputs: Using natural fertilizers, such as manure and compost, instead of synthetic chemicals.

8. Education and community engagement

Workshops and training: Providing education on organic practices for farmers and consumers to promote sustainable agriculture.

Collaborative initiatives: Engaging with local communities and organizations to support food security and sustainable practices.

9. Waste management

Recycling and reuse: Executing practices to minimize waste, such as reusing packaging and composting organic materials.

Sustainable packaging: Choosing eco-friendly packaging materials to reduce environmental impact. These practices, rooted in the core principles of organic farming, not only enhance agricultural productivity but also promote environmental stewardship, social equity, and community well-being. By adopting these practices, organic farmers contribute to a more sustainable and reasonable food system.

Challenges of organic farming

Soil fertility management: Maintaining soil health without synthetic fertilizers can be tough. Organic farmers rely on compost, cover crops, and crop rotations, which require careful planning and knowledge of soil dynamics.

Pest and disease control: Organic methods for managing pests and diseases, such as biological controls and natural pesticides, can be less effective and more labour-intensive than conventional methods. This may lead to higher crop losses.

Weed management: Organic farming often struggles with weed pressure since synthetic herbicides are not allowed. Farmers must employ manual weeding, mulching, and crop rotation strategies, which can be time consuming and costly.

Market access and pricing: Organic products typically draw higher prices, but farmers may face challenges accessing markets or competing with cheaper conventional goods. Establishing a reliable customer base can take time.

Certification process: Achieving organic certification can be a lengthy and complex process. Farmers must follow to strict guidelines and maintain careful records, which can be devastating, especially for small-scale operations.

Knowledge and training: Successful organic farming requires a deep understanding of agroecology and sustainable practices. Farmers may need additional training and support to transition from conventional to organic methods.

Climate vulnerability: Organic systems may be more vulnerable to climate variability due to their dependence on diverse crop rotations and organic inputs, which can be impacted by changing weather patterns.

Input availability: Sourcing organic seeds, fertilizers, and pest control materials can be more challenging and expensive compared to conventional inputs, limiting options for organic farmers.

Labor intensity: Organic farming often requires more labour for practices such as hand weeding, planting, and harvesting, which can lead to higher labour costs and difficulties in finding qualified workers.

Yield variability: Organic farms may experience lower yields compared to conventional farms, especially in the early years of conversion. This can impact profitability and economic viability.

Consumer awareness and education: Although demand for organic products is growing, some consumers may lack understanding of the benefits of organic farming, which can hamper market growth.

Policy and regulation: Inconsistent policies and regulations regarding organic farming can create uncertainty and complicate the transition for farmers, especially in regions with less supportive frameworks. By addressing these challenges through education, innovation, and supportive policies, the principles and practices of organic farming can continue to evolve and contribute to sustainable agriculture.

Conclusion

The principles and practices of organic farming represent a general approach to agriculture that prioritizes ecological balance, soil health, and biodiversity. By following to core beliefs such as the avoidance of synthetic inputs, the promotion of sustainable land use, and the emphasis on local

ecosystems, organic farming not only produces healthy food but also fosters environmental stewardship and community resilience. Despite the challenges faced by organic farmers ranging from pest management to market access the potential benefits for both the environment and human health are significant. As awareness of these advantages grows, so too does the demand for organic products, highlighting the importance of continuous education, research, and innovation in organic practices. Moving forward, it is essential for stakeholders' farmers, policymakers, and consumers to work together in overcoming barriers and supporting organic agriculture. By embracing these core principles, we can cultivate a sustainable food system that benefits current and future generations, ensuring the health of our planet and its inhabitants. Ultimately, the evolution of organic farming can serve as a model for a more sustainable and resilient agricultural model.

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Agroforestry: A Multifaceted Solution for Climate Change Mitigation and Adaptation

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Climate change is one of the most serious global threats to human welfare and ecosystem stability in the twenty-first century. Rising temperatures, unpredictable rainfall, and more frequent extreme events like droughts and floods are significantly impacting agriculture, damaging ecosystems, and jeopardizing the livelihoods of millions—especially in developing regions where farming is a primary source of income and food security, as highlighted by the Intergovernmental Panel on Climate Change (IPCC, 2021). The agriculture, forestry, and other land-use (AFOLU) sectors account for about one-quarter of global greenhouse gas (GHG) emissions, driven mainly by deforestation, land degradation, and unsustainable production systems (Smith et al., 2019). Hence, there is an urgent need for land management practices that simultaneously boost productivity, conserve resources, and build resilience to climatic variability (Jose, 2009; Nair et al., 2010). Climate change poses significant challenges to global ecosystems, food security, and rural livelihoods, demanding innovative strategies that enhance both environmental and socio-economic resilience. Agroforestry, the intentional integration of trees with crops and livestock, has emerged as a vital nature-based solution in this context. By combining agricultural productivity with ecological stability, agroforestry systems contribute to carbon sequestration, soil and water conservation, and biodiversity enhancement while offering livelihood diversification for farming communities. As nations strive to meet their climate goals and adapt to changing conditions, understanding the role of agroforestry in mitigating greenhouse gas emissions and building adaptive capacity becomes increasingly crucial. This approach not only bridges the gap between traditional farming and environmental conservation but also paves the way toward a sustainable and climate-resilient future. By combining the ecological benefits of forests with agricultural productivity, agroforestry enhances soil fertility, biodiversity, carbon sequestration, and livelihood diversification (Leakey, 2014; Mbow et al., 2014). Systems such as alley cropping, silvopasture, homegardens, and windbreaks optimize interactions between trees, crops, and livestock, offering both adaptation and mitigation benefits (Nair, 2011; Verchot et al., 2007). Trees moderate microclimates, improve water regulation, and protect soil from erosion while storing carbon in biomass and soil (Nair et al., 2009).

Agroforestry's multifunctionality makes it particularly beneficial for smallholder farmers in tropical and subtropical regions, where land degradation and climate risks are severe (Rahman et al., 2020). Unlike monocropping, which often depletes nutrients and reduces biodiversity, tree-based systems improve soil health, enhance nutrient cycling, and provide valuable products such as timber, fodder, fruits, and non-timber forest products (Mbow et al., 2014; Mercer, 2004). Ecologically, agroforestry maintains habitat connectivity, restores degraded land, and supports vital ecosystem services like pollination and pest regulation (Schroth & Harvey, 2007; Torralba et al., 2016). Globally, agroforestry contributes to major sustainability agendas such as the United Nations Sustainable Development Goals (SDGs), the Paris Agreement, and the UN Decade on Ecosystem Restoration (2021–2030). It directly supports SDGs 2 (Zero Hunger), 13 (Climate Action), and 15 (Life on Land) (World Bank, 2021). Several countries—including India, Kenya, and Brazil—have incorporated agroforestry into their Nationally Determined Contributions (NDCs) to achieve climate and rural development goals (FAO, 2020). India's National Agroforestry Policy (2014) represents a key step toward integrating tree-based farming for productivity, livelihood, and ecosystem resilience (Dhyani et al., 2013). However, large-scale adoption faces barriers such as insecure land tenure, lack of technical knowledge, weak markets, and delayed returns (Mercer, 2004; Rahman et al., 2020). Success depends on locally adapted designs, policy support, and participatory implementation (Nair, 2011; Mbow et al., 2014).

1. Agroforestry as a Strategy for Climate Adaptation

1.1 Microclimate Regulation

Trees provide shade and act as windbreaks, which lowers ambient temperature, reduces wind speed, and increases relative humidity in the immediate area. This buffering protects sensitive crops and livestock from heat stress, wind damage, and cold snaps. Trees within agricultural landscapes create microclimatic buffering by moderating temperature, increasing relative humidity, and reducing evapotranspiration (Lin, 2007). The canopy intercepts solar radiation, lowering soil surface temperatures and protecting crops from heat stress (Jose et al., 2006). Studies in tropical agroforestry systems have shown temperature reductions of 2–4 °C compared with open fields, improving crop performance during droughts (Mutuo et al., 2005).

1.2 Soil Fertility and Nutrient Cycling

Agroforestry improves soil fertility and nutrient cycling by increasing organic matter, enriching the soil with nitrogen, and enhancing nutrient retention. Trees in these systems recycle deep-rooted

nutrients, fix atmospheric nitrogen, and add organic matter through leaf litter and root decomposition, leading to better soil structure and microbial activity. This leads to more efficient nutrient use, reduced losses through erosion, and a more closed and resilient nutrient cycle. Agroforestry systems maintain or enhance soil fertility by improving organic matter inputs through litterfall, root turnover, and nitrogen fixation (Schroth et al., 2001). Nitrogen-fixing species such as *Gliricidia sepium* or *Leucaena leucocephala* improve soil nutrient balance and reduce reliance on chemical fertilizers (Rao et al., 1998). Tree-based systems enhance soil microbial biomass and enzyme activity, improving nutrient cycling efficiency (Cardinael et al., 2017).

1.3 Water Conservation and Hydrological Regulation

Agroforestry conserves water by improving soil's ability to absorb and retain moisture, which is achieved through increased organic matter and root systems that enhance infiltration and reduce runoff. It regulates the hydrological cycle by slowing surface water flow, which prevents erosion and allows more water to recharge groundwater, while also filtering pollutants from runoff to improve water quality. Tree cover and root systems help control soil erosion, prevent runoff, and protect water bodies from agricultural pollutants. Tree roots enhance infiltration and water-holding capacity, reducing surface runoff and erosion (Hairiah et al., 2006). In semi-arid regions, agroforestry increases infiltration rates by 25–40 % compared to annual cropping systems (Bayala et al., 2014). Deep-rooted trees access groundwater, redistributing moisture to the topsoil via hydraulic lift, thereby benefiting adjacent crops (Burgess et al., 1999).

1.4 Diversified Livelihoods and Food Security:

Agroforestry strengthens diversified livelihoods and food security by integrating trees into farming systems, providing a wider range of products for both sustenance and income, and improving soil health and resilience. This diversification reduces the risk of crop failure, provides diverse and nutritious food sources, and creates multiple income streams from products like timber, fruits, nuts, and fodder. Farmers can generate multiple income streams from various tree products (fruits, nuts, timber, fodder) alongside traditional crops and livestock. This diversification acts as "insurance" during crop failures caused by unpredictable climate conditions, enhancing economic resilience.

1.5 Pest, Disease, and Wind Protection

Combining different plant species can help reduce the need for pesticides, as it can disrupt pest cycles. Tree canopies reduce wind velocity, minimizing soil erosion and physical damage to crops (Garrity, 2012). Greater habitat diversity supports natural enemies such as predatory insects and birds, lowering pest incidence (Bender et al., 2016). This ecological regulation enhances system stability under climatic fluctuations.

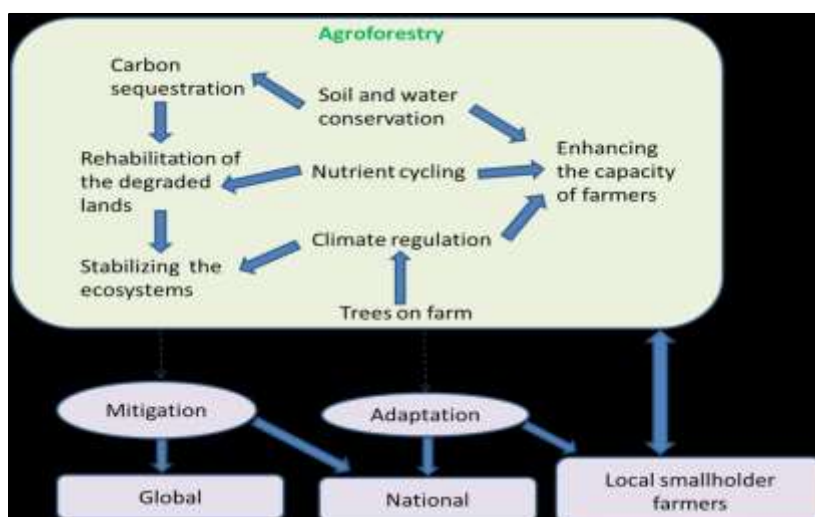


Fig 1: Potential of agroforestry systems as adaptation to and mitigation of climate change (Lasco et al., 2014)

2. Agroforestry and Climate Mitigation

2.1 Carbon Sequestration Potential

The woody perennial plants (trees and shrubs) in agroforestry systems absorb carbon dioxide (CO₂) from the atmosphere through photosynthesis and store the carbon in their biomass (trunks, branches, roots, and leaves). These systems can sequester significantly more carbon than conventional monoculture agricultural systems. Agroforestry sequesters carbon in both above- and below-ground biomass, contributing to mitigation of greenhouse gas emissions (Nair et al., 2010). Global estimates suggest carbon storage in agroforestry systems ranges from 12 to 228 Mg C ha⁻¹ depending on system type and climate (Lorenz & Lal, 2014). Soil carbon accumulation is particularly significant: (Cardinael et al. 2017) found average soil organic carbon (SOC) gains of 19 % over 10–20 years compared to adjacent croplands.

2.2 Greenhouse Gas Reduction

Agroforestry practices can lead to reduced reliance on synthetic fertilizers (especially when using nitrogen-fixing trees), which are a major source of nitrous oxide (N₂O), a potent GHG. The use of trees for fuelwood and timber also reduces pressure on natural forests, preventing deforestation-related emissions. By integrating trees with crops and livestock, these systems act as a carbon sink, improving soil health and fertility, which further enhances carbon storage. In addition to carbon storage, agroforestry reduces nitrous oxide and methane emissions by improving soil aeration and nutrient management (Kim et al., 2016). Integration of leguminous trees can reduce nitrogen fertilizer use by up to 50 %, mitigating emissions associated with fertilizer manufacture and application (Rao et al., 1998).

2.3 Enhanced Soil Carbon Storage:

Agroforestry enhances soil carbon storage by increasing the amount of organic matter returned to the soil from diverse plant sources. This happens through processes like leaf litter and root decomposition, which convert atmospheric carbon dioxide into soil organic carbon (SOC). By integrating trees with crops and livestock, agroforestry systems provide a larger and more diverse carbon supply to the soil compared to traditional monocropping, leading to greater accumulation and stability of soil carbon. Tree roots, leaf litter, and organic matter inputs from the trees enrich the soil, increasing soil organic carbon (SOC) content. Healthy, carbon-rich soils act as long-term carbon sinks.

2.4 Policy and Accounting Implications

Recognizing its carbon benefits, agroforestry has been included in many Nationally Determined Contributions (NDCs) under the Paris Agreement (FAO, 2022). However, quantifying its mitigation potential requires robust monitoring, reporting, and verification (MRV) frameworks (Minang et al., 2015). Standardized methodologies are essential to ensure creditability in carbon markets and payment for ecosystem service schemes.

Table 1. Carbon Sequestration and Storage in Major Agroforestry Systems

Agroforestry System	Typical Carbon Sequestration Potential (Mg C ha ⁻¹)	References
Homegardens / Multistrata systems	50–200	Montagnini & Nair (2004); Kumar & Nair (2011)
Silvopastoral systems	12–60	Nair et al. (2009); Murgueitio et al. (2011)
Alley cropping systems	10–30	Udawatta & Jose (2012); Lorenz & Lal (2014)
Windbreaks and boundary plantations	5–20	Nair (2011); Schoeneberger et al. (2012)
Parkland systems	15–40	Bayala et al. (2014); Mbow et al. (2014)
Average soil carbon increase under agroforestry	15–30% over 10–20 years	Lorenz & Lal (2014); Nair et al. (2009)

Table 2. Carbon Sequestration Potential of Agroforestry Systems in India

(Source: Dhyani, Ram, & Dev, 2020)

Agroforestry System Type	Carbon Sequestration Rate (Mg C ha ⁻¹ yr ⁻¹)	Major Components
Agrisilviculture	0.29 – 0.49	Trees + annual crops
Silvipasture	0.36 – 0.72	Trees + grasses/pasture
Agrihorticulture	0.25 – 0.63	Fruit trees + crops
Boundary plantation	0.05 – 0.15	Trees on field boundaries
Homegarden	0.58 – 2.78	Mixed trees, crops, livestock
Block plantation	0.41 – 0.58	Dense tree stands

3. Challenges and Barriers to Adoption

3.1 Institutional and Policy Constraints

Despite proven benefits, adoption remains limited by unclear tree-tenure rights, restrictive forestry regulations, and insufficient extension services (Mercer, 2004). Many farmers lack incentives due to delayed financial returns (Ajayi et al., 2009).

3.2 Knowledge and Technical Gaps

Limited technical capacity to design site-specific systems constrains scalability (Nair, 2011). Research–extension linkages must be strengthened to translate scientific knowledge into farmer practice (Garrity, 2012).

3.3 Economic and Market Challenges

Agroforestry products often face weak market linkages and low price competitiveness. Developing value chains for timber and non-timber products can enhance profitability and adoption (Rahman et al., 2020).

4. Scaling Up Agroforestry for Climate-Smart Landscapes

Mainstreaming agroforestry into agricultural and forestry policies is critical for scaling (FAO, 2022). Successful implementation requires multi-stakeholder collaboration—farmers, researchers, NGOs, and policymakers (Minang et al., 2015). Financial incentives such as Payments for Ecosystem Services (PES) and carbon credits can accelerate adoption (Kim et al., 2016). Integrating agroforestry in watershed and landscape planning ensures spatial optimization of ecological and social benefits (Jose, 2009).

Conclusion

Agroforestry stands out as a sustainable and resilient land-use strategy that bridges the gap between agriculture and forestry, offering powerful tools for both climate change mitigation and adaptation. By integrating trees with crops and livestock systems, it enhances carbon sequestration, improves soil health, conserves biodiversity, and strengthens ecosystem services. Moreover, it supports rural livelihoods, stabilizes local microclimates, and reduces the vulnerability of farming communities to extreme weather events. As the global community seeks climate-smart solutions, adopting and scaling up agroforestry practices can play a decisive role in achieving environmental sustainability, food security, and long-term climate resilience. Agroforestry offers a holistic and sustainable approach that simultaneously addresses both the causes and consequences of climate change, while providing numerous co-benefits for livelihoods and the environment. Thus Agroforestry offers an integrative approach that reconciles production with conservation. Its ability to sequester carbon, enhance biodiversity, and sustain livelihoods makes it indispensable for climate adaptation strategies.

However, scaling requires enabling policies, institutional support, and capacity building. When integrated into national climate action plans and land-use strategies, agroforestry can serve as a cornerstone for resilient and sustainable landscapes.

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Harnessing Apomixis: A Revolutionary Path to Fix Hybrid Vigor in Crops

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Introduction

Hybrid vigor, also known as heterosis, plays a crucial role in enhancing crop performance as F1 hybrids typically show superior yield and perform better compared to their parental lines. However, one of the major limitations in breeding programmes is the inability to maintain this Vigor in subsequent generations. This is because of genetic recombination in F2 offspring leading to variability and loss of the desirable traits, forcing farmers to buy new hybrid seeds each season. Apomixis is a form of asexual reproduction that occurs via seeds in which embryo develop without fertilization. It allows plants to produce offspring genetically identical to the parent. This means the hybrid's traits and vigor can be maintained across generations. Unlike sexual reproduction, which relies on meiosis and fertilization and often leads to genetic variation among progeny, apomixis skips these steps. Because of this, it holds tremendous potential for creating stable, high-yielding varieties that does not lose their desired traits over time.

Apomixis is known to occur in some 300 species belonging to about 40 families, but it is absent in major cereals and pulses. Recent advances in molecular biology and genome editing have enabled researchers to identify key genetic regulators

Apomixes and engineer synthetic systems that mimic this process. These breakthroughs open new possibilities for stabilizing hybrid traits, reducing seed costs, and improving food security. If successfully integrated into breeding programs, apomixis could fundamentally reshape crop improvement strategies by combining the benefits of hybrid vigor with the reliability of clonal propagation.

1. Types of Apomixis

- Apomixis can be classified based on its origin and how the embryo develops:
- **Sporophytic Apomixis (Adventitious Embryony):** In this type, embryos arise directly from diploid somatic cells of the ovule, such as the nucellus or integuments, without involving the embryo sac. These embryos often develop alongside those formed through sexual reproduction.
- **Gametophytic Apomixis:** This involves the formation of an embryo sac without normal meiosis (a process called apomeiosis), resulting in an unreduced female gametophyte.

- **Diplospory:** In this type, the megaspore mother cell either skips or modifies meiosis, leading to the development of an unreduced embryo sac from the same cell lineage.
- **Apospory:** In this case, a somatic cell from the nucellus changes its developmental path and forms an embryo sac directly, bypassing the meiotic process.

In both diplospory and apospory, the egg cell develops into an embryo through parthenogenesis—without fertilization—thus maintaining the genetic identity of the mother plant.

- **Facultative vs. Obligate Apomixis:** Obligate apomicts reproduce only through apomixis, while facultative apomicts can produce seeds both sexually and asexually within the same generation.

Genetic basis of apomixis

The genetic and molecular basis of apomixis in crops revolves around the coordinated action of three key processes: apomeiosis, where meiosis is bypassed; parthenogenesis, in which an embryo forms without fertilization; and endosperm development to nourish the growing embryo. Advances in comparative genomics, functional genetics, and modern biotechnology have shed light on how these mechanisms work together. Understanding and harnessing these processes could transform crop breeding by allowing the stable transmission of hybrid vigor and valuable traits across generations. The engineering of apomixis relies predominantly on mutations or transgenic incorporation of apomixis-linked genes.

Apomeiosis (Meiosis Bypass): Mutations in three primary genes—*SPO11-1*, *REC8*, and *OSD1*—implement the “MiMe” (Mitosis instead of Meiosis) system. This approach suppresses chromosomal recombination, changes chromatid behaviour, and eliminates the second meiotic division, transforming meiosis into a mitotic-like process and producing unreduced, clonal gametes. In rice, additional genes such as *PAIR1* are included in similar strategies.

- **Parthenogenes is (Embryo Formation):** The most notable gene is *BBM1* (BABY BOOM 1), which when expressed in egg cells triggers the development of the embryo without fertilization. Naturally, the *ASGR- BBML* locus in *Pennisetum squamulatum*, and similar BBM-like genes in other species are central to the switch from sexual to asexual seed formation.
- **Endosperm Development:** Independent or coordinated mutations affecting genes such as *CENH3* (centromere-specific histone H3) can induce maternal haploidy or facilitate proper endosperm formation after apomeiosis and parthenogenesis. Genomic regions like *AutE* in some wild species further modulate endosperm autonomy.

Synthetic Apomixis: Technological Advances

Synthetic apomixis is a new and advanced method in plant breeding that tries to copy the natural process of apomixis. The goal is to keep the good qualities of hybrid plants, such as high yield and strength, for many generations. In normal sexual reproduction, segregation occurs and variation is created but synthetic apomixis avoids this by producing seeds that are exact copies of the parents. In nature, apomixis relies on three main processes: apomeiosis, parthenogenesis, and the development of a functional endosperm to support seed growth. Synthetic approaches attempt to reproduce these steps using precise genetic modifications, offering enormous potential for creating uniformly high-performing crop varieties.

The synthetic approach mainly used to create apomixis is called the MiMe system, which stands for “Mitosis instead of Meiosis.” Normally, meiosis produces haploid gametes (with half the number of chromosomes) and introduces genetic variation. In the MiMe system, this process is changed so that meiosis behaves like mitosis, producing diploid gametes that are exact genetic copies of the parent. This is done by disrupting three important genes—SPO11-1, REC8, and OSD1, which control the key steps of recombination, chromosome separation, and meiotic progression. As a result, the produced gametes keep the full set of chromosomes, making them identical to the parent plant.

Another important step is inducing parthenogenesis, where embryos develop from egg cells without fertilization. The BABY BOOM1 (BBM1) gene, a transcription factor first identified in monocots like rice and Pennisetum, has been utilized to induce parthenogenesis when ectopically expressed in the egg cell, promoting embryo formation autonomously without fertilization.

Detection and Validation of Apomixis Embryo Sac Analysis (Cytological Methods)

Embryo sac analysis involves microscopic examination of developing ovules to determine whether sexual or apomictic reproduction is occurring. Through cytological clearing techniques and differential staining, researchers can directly visualize the formation of unreduced ($2n$) embryo sacs—signature indicators of apomixis such as diplospory or apospory. Observing whether the embryo sac arises from a megaspore mother cell (diplospory) or from somatic nucellar cells (apospory) is critical for distinguishing reproductive modes. Such analyses remain foundational for initial detection and fine characterization of apomictic development in plant material.

Flow Cytometric Seed Screening (FCSS)

FCSS is a high-throughput, quantitative method to detect apomixis at the seed level. This technique involves simultaneous estimation of DNA content in both embryo and endosperms nuclei, allowing deduction of fertilization patterns. In apomictic seeds, characteristic deviations in the usual embryo: endosperm DNA ratios (e.g., 2C:2C rather than the normal 2C:3C in

sexual plants) provide molecular confirmation of asexual reproduction. FCSS is known for its scalability and ability to analyze large seed lots quickly and reliably.

Molecular Markers Linked to Apomixis

Molecular markers offer robust, early detection and allow marker-assisted selection (MAS) of apomictic individuals. Approaches such as bulked segregant analysis, RAPD, and especially sequence-characterized amplified region (SCAR) markers have been highly effective. Markers tightly linked to specific apomixis loci (such as the ASGR—Apospory-Specific Genomic Region) enable the efficient screening of breeding populations. For example, in *Cenchrus ciliaris* and *Brachiaria humidicola*, several SCAR and RAPD markers have shown absolute linkage to apomictic reproduction, drastically improving selection accuracy in breeding pipelines.

Genetic Segregation Studies

Genetic segregation studies use progeny testing and marker analysis to validate the inheritance pattern of apomixis. After controlled crosses or selfing, large segregating populations are evaluated for uniformity of progeny (clonal apomixis yields identical offspring, while sexual reproduction creates variation). Mendelian analysis of marker presence/absence, often using chi-square tests, allows researchers to confirm whether apomixis is under simple genetic control or exhibits more complex inheritance, including facultative cases. Studies have documented single-gene dominant inheritance in some species with expected ratios in F1 and F2 generations and have mapped recombination events with high precision.

Microscopic and Histological Examination

Complementing embryo sac analysis, advanced microscopy (including confocal and histological sectioning) allows precise visualization of ovule tissue organization, cell fate, and abnormal developments associated with apomixis. Histological analysis differentiates apomictic from sexual development by examining the sequence and timing of key events—such as the absence of meiotic divisions or the proliferation of nucellar tissue. Such detailed cell and tissue imaging aids in distinguishing facultative from obligate apomicts, confirming molecular marker results, and solving unexpected segregation patterns in breeding populations.

List of genes involved in Apomixis

Gene Name	Function / Role	Species	Reference(s)
BABY BOOM1 (BBM1)	Induces parthenogenesis; triggers embryo development without fertilization	Rice, Arabidopsis, synthetic apomixis systems	Khanday <i>et al.</i> , 2019
SPO11-1	Meiotic recombination initiation; mutated in MiMe to replace meiosis with mitosis	Arabidopsis, rice (MiMe system)	Herbert <i>et al.</i> , 2025

Gene Name	Function / Role	Species	Reference(s)
REC8	Meiotic cohesion involved in chromosome segregation; mutated in MiMe	Arabidopsis, rice (MiMe system)	Wang <i>et al.</i> , 2017 ; Cromer <i>et al.</i> , 2013
OSD1 (Omission of second division 1)	Controls meiotic division II; mutated in MiMe	Arabidopsis, rice (MiMe system)	Mercier <i>et al.</i> , 2015; Wang <i>et al.</i> , 2017
PAIR1	Meiotic chromosome pairing; mutated in MiMe	Rice	Nonomura <i>et al.</i> , 2004; Marimuthu <i>et al.</i> , 2011
CitRWP	RWP-RK transcription factor controlling sporophytic apomixis via somatic embryogenesis	Citrus (sporophytic apomixis)	Wang <i>et al.</i> , 2019
PARTHENOGENESIS (PAR)	Triggers fertilization-independent embryo development	Dandelion (Taraxacum);	Khanday <i>et al.</i> , 2019; Conner <i>et al.</i> , 2015
Gene Name	Function / Role	Species	Reference(s)
		synthetic apomixis	
ZmBBM2	BBM homolog in maize, involved in embryogenesis initiation	Maize (synthetic apomixis studies)	Lin <i>et al.</i> , 2021

Recent Case Study of Synthetic Apomixes in Plant Breeding

A recent notable case study demonstrated synthetic apomixis successfully induced in hybrid rice by combining mutations in key meiotic genes (PAIR1, REC8, OSD1) with expression of the BABY BOOM1 (BBM1) gene in egg cells. In this study (Vernet et al., 2022), the all-in-one genetic construct enabled the rice plants to bypass meiosis and initiate embryo development without fertilization, producing clonal seeds that retained hybrid vigor with up to 95% efficiency across multiple generations. Whole-genome sequencing confirmed that the heterozygous hybrid genomes were faithfully transmitted without genetic reshuffling. The synthetic apomictic plants showed normal growth and grain quality similar to original hybrids, although fertility was somewhat reduced. This represents a major breakthrough, proving stable synthetic apomixis for fixing hybrid vigor is achievable in an important crop species.

Limitations and Challenges in Synthetic Apomixis

The development of synthetic apomixis for fixing hybrid Vigor holds great potential but also faces significant challenges and limitations. One major limitation is that natural apomixis is very rare, and simply transferring it between species is not possible. Synthetic apomixis requires complex genetic engineering involving multiple genes and pathways, making it technically difficult to develop stable systems in many crops, especially dicots and some major food crops.

Another important concern is the potential loss of genetic diversity. Since apomixis produces clones, accumulation of harmful mutations over generations could occur, and the ability of crops to adapt to new pests, diseases, or environmental stresses may be reduced. Maintaining genetic diversity within breeding populations alongside apomictic lines is therefore a critical challenge.

Technically, achieving highly efficient apomixis with consistent seed fertility and viability remains a hurdle. For example, current systems like MiMe combined with parthenogenesis genes (e.g., BBM1) still suffer from relatively low fertility rates and the need for pollination to trigger endosperm development, sometimes resulting in a mixture of clonal and sexual seeds. Autonomous endosperm formation, essential for fully asexual seed development, is an active area of research but not yet fully solved.

The adoption of apomixis could also disrupt traditional seed industries and intellectual property frameworks, raising regulatory, economic, and social challenges. Issues related to GMO crop acceptance, seed sovereignty, and biodiversity conservation must also be carefully considered.

Conclusion and Future Prospects

Widespread adoption of apomixis promises to transform agriculture by stabilizing food supplies and making superior seeds more accessible to farmers. Apomixis allows plants to produce seeds that are exact genetic copies of the parent. This means that hybrid vigor, which gives hybrids their superior traits like high yield and stress tolerance, can be fixed and passed down unchanged without having to make cross between parents every season. It has the potential to drastically reduce the cost and complexity of hybrid seed production, particularly for important crops like maize, rice, pulses etc.

Moreover, synthetic apomixis can speed up agricultural innovation by enabling breeders to transfer desirable traits such as disease resistance, drought tolerance etc., directly into clonal seed lines. This leads to crop improvement and genetic stability since valuable traits are preserved over multiple generations, avoiding the genetic segregation that occurs as a result of sexual reproduction. Recent progress in molecular biology and gene editing tools like CRISPR has greatly increased the potential of synthetic apomixis. Alongside growing regulatory support, these advances are paving the way for apomixis to revolutionize crop production globally.

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8. Changes chromatid behaviour, and eliminates the second meiotic division, transforming meiosis into a mitotic-like process and producing unreduced, clonal gametes. In rice, additional genes such as *PAIR1* are included in similar strategies.

Biotechnology in Therapeutics: From Natural Products to Biopharmaceuticals

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Biotechnology has influenced modern medicine by using molecular biology, genetic engineering and bioinformatics to produce drugs that are safe, effective and accurate. Biotechnology makes it possible to make drugs on a big scale and in a way that is good for the environment. This includes natural products like penicillin and paclitaxel, as well as biopharmaceuticals made from recombinant DNA, such insulin and monoclonal antibodies. Improvements in mRNA vaccines, gene therapy and nanocarrier drug delivery addressed the prevention and to cure diseases. High throughput screening and omics technologies speed up the process of finding therapeutic targets and new drugs. CRISPR based gene editing, stem cell therapy and customized medicine are the three promising areas for the future.

Keywords: Biotechnology, Biopharmaceuticals, Recombinant DNA, Gene Therapy, Personalized Medicine

IntroductionBiotechnology has transformed the face of modern medicine in ways that have enabled the precise manipulation of biological systems to develop safer, more effective therapeutics. Conventionally, treatments were generated from natural sources such as plants, microbes, and animal extracts, which are precursors of pharmacological compounds (Santos-Beneit, 2024). However, therapeutic biotechnology, by its definition based on recent advances in the field of molecular biology, genetic engineering, and bioinformatics, emerges as a highly sophisticated discipline integrating biological knowledge with technological innovation.

Molecular therapies have replaced empirical natural medicine, and this is one of the biggest paradigms shift in healthcare history. Initial instances comprised penicillin derived from *Penicillium* fungi, insulin extracted from animal pancreas, and then recombinant human insulin produced in *Escherichia coli* (Gaynes, 2017). These findings resulted in a novel category of medications known as biopharmaceuticals, which are therapeutic compounds generated from living cells via recombinant DNA and associated biotechnological methods.

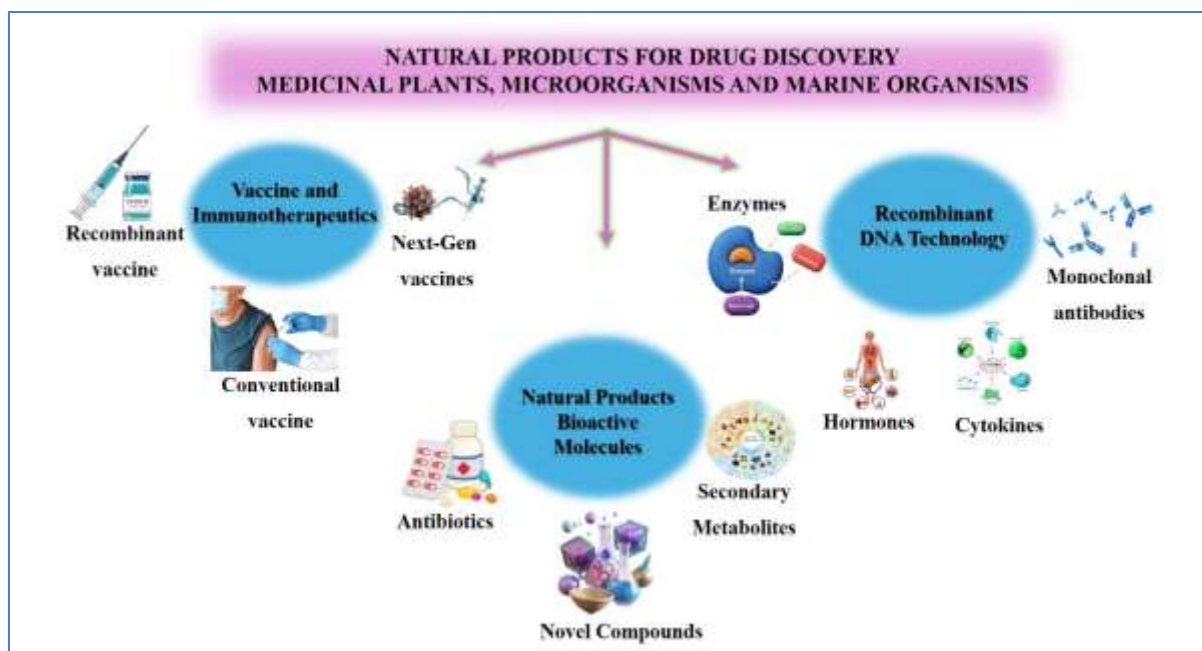


Figure1. From Natural Products to Biopharmaceuticals

Therapeutic biotechnology addresses unmet medical needs, such as rare genetic disorders, autoimmune diseases, cancers, and other emerging infectious diseases. Its importance to global health is underlined by the development of monoclonal antibodies, mRNA vaccines and gene therapies that have changed disease prevention and molecular therapies has replaced empirical natural medicine and this is one of the biggest paradigm shifts in healthcare history (Schürmann et al., 2025). Initial instances comprised penicillin derived from *Penicillium* fungi, insulin extracted from animal pancreas, and then recombinant human insulin produced in *Escherichia coli*. These findings resulted in a novel category of medications known as biopharmaceuticals, which are therapeutic compounds generated from living cells via recombinant DNA and associated biotechnological methods.

2. Natural Products and Bioactive Molecules

Natural products have traditionally been the basic raw materials for drug discovery. They represent a diverse chemical entity that continues to inspire modern drug development. Sources include microorganisms, plants and marine organisms, each one offers unique bioactive metabolites with therapeutic potential.

Microbial Sources

Microorganisms have produced a greater number of significant antibiotics and immunosuppressive agents. *Streptomyces* sp. synthesizes streptomycin, erythromycin and tacrolimus, *Aspergillus terreus* yields lovastatin, while *Penicillium chrysogenum* is renowned for penicillin. Progress in metabolic engineering and synthetic biology has permitted the overexpression of biosynthetic gene clusters,

enhanced yield and enabling the creation of novel analogs via pathway modification (Alam et al., 2022).

Plant Derived Therapeutics

Plants continue to be an important supplier of alkaloids, terpenoids, phenolics, and glycosides. Notable examples include morphine, quinine, paclitaxel (Taxol) and artemisinin. Biotechnology improves production via tissue culture, hairy root systems, and metabolic pathway modification. For example, in the cell culture of *Artemisia annua*, gene is edited by the tool named CRISPR/Cas-mediated have increased artemisinin production for antimalarial treatment (Heinrich et al., 2021).

Marine Derived Compounds

Marine biotechnology has revealed powerful compounds such bryostatin, trabectedin and ziconotide, applicable in oncology and analgesia. Genetic techniques are utilized to clone and express biosynthetic gene clusters from unculturable marine microbes, facilitating sustained production (Karthikeyan et al., 2022).

Screening and Characterization

High throughput screening (HTS) systems, when combined with metabolomics and chemoinformatics, facilitate the rapid discovery of bioactive compounds. Bioassay guided fractionation, NMR and mass spectrometry facilitate structural elucidation, whilst recombinant expression in heterologous systems supports the characterisation and modification of complex natural products (Ayon, 2023).

Case Studies

The semi-synthetic production of artemisinin using modified *Saccharomyces cerevisiae* that expresses *A. annua* genes has revolutionized malaria treatment. Biotechnology assisted large scale synthesis of Paclitaxel uses plant cell fermentation instead of taking it from the endangered *Taxus brevifolia*. Recombinant Erythropoietin improved patients with anemia by replacing purification from human sources. Biotechnology not only helps with the creation of new medicines, but it also helps with the long-term and large-scale manufacture of natural remedies (Elfahmi et al., 2021).

3. Recombinant DNA Technology and Protein Therapeutics

Recombinant DNA (rDNA) technology helps modern biopharmaceuticals and it facilitates the introduction of human genes into microbial or mammalian systems for the synthesis of therapeutic proteins such as hormones, enzymes, cytokines and monoclonal antibodies in order to ensure uniform quality and safety. Recombinant Hormones: Human insulin (Humulin®) was the first recombinant medicine approved in 1982, followed by human growth hormone, interferons and erythropoietin. Recombinant enzymes, including tissue plasminogen activator (tPA) and glucocerebrosidase, are utilized in thrombolytic therapy for the treatment of Gaucher's disease. Monoclonal Antibodies (mAbs), including trastuzumab (Herceptin®) and adalimumab (Humira®), serve as the best examples

of targeted medicines characterized by improved specificity (Khan et al., 2016). The choice of expression host influences yield, post translational modifications such as Bacteria (e.g., *E. coli*) – High yield, low cost; lacks glycosylation capability. Yeast such as *Pichia pastoris* is capable of simple glycosylation, Insect cells such as Baculovirus system is suitable for complex eukaryotic proteins, Mammalian cells such as CHO, HEK293 will provide post translational modifications which are important for therapeutic efficacy (Schütz et al., 2023). Downstream processing involves cell lysis, chromatographic methods such as affinity, ion exchange, size exclusion and formulation with stabilizers all of which ensures biophysical stability and maintaining bioactivity are essential for therapeutic effectiveness (De Luca et al., 2021).

Biosimilars and Regulation

Biosimilars are biological medicines that are highly similar to an already approved "reference" biologic drug play a vital role in reducing costs and expanding the availability to all population. Regulatory frameworks (EMA, US FDA, CDSCO) demand stringent comparability studies which includes the analytical, preclinical and clinical evaluations to ensure the efficacy and safety (Rahalkar et al. 2021).

4. Vaccines and Immunotherapeutics

Vaccines are of the greatest achievement in the field of biotechnology, providing prophylactic and therapeutic approaches against infectious and non-infectious diseases.

Conventional vs. Recombinant Vaccines

Traditional vaccines, such as inactivated or attenuated pathogens, are being replaced by recombinant and subunit vaccines that offer better safety profiles. Recombinant hepatitis B vaccine, produced in *Saccharomyces cerevisiae*, was a milestone in this transition (Zhao et al., 2020).

Next-Generation Vaccine Technologies

DNA Vaccines: Plasmid DNA encoding antigenic proteins triggers host immune responses. Examples are Zika and influenza vaccines. **mRNA Vaccines** are introducing a piece of mRNA that corresponds to a viral protein, usually a small piece of a protein found on the virus's outer membrane. The success of COVID-19 mRNA vaccines named Pfizer (BioNTech), Moderna showed quick scalability and high efficacy. **Viral Vector Vaccines** is a vaccine that uses a viral vector to deliver genetic material (DNA) that can be transcribed by the recipient's host cells as mRNA coding for a desired protein, or antigen, to elicit an immune response. Adenovirus and lentivirus based vectors enable strong cellular immunity, the best example is Oxford-AstraZeneca COVID-19 vaccine (Kozak & Hu, 2024).

Monoclonal Antibodies and Immunotherapies

Therapeutic mAbs, such as nivolumab and pembrolizumab, block immune checkpoints (PD-1, CTLA-4) enhancing antitumor immunity. Bispecific antibodies and antibody-drug conjugates (ADCs) represent next-generation modalities with dual-targeting capability. Engineered T cells (CAR-T

therapy) and immune-modulatory biologics such as TNF- α inhibitors have redefined cancer and autoimmune disease management. For example, CAR-T therapy (Kymriah®, Yescarta®) shows remarkable remission rates in leukemia (Gravbrot et al., 2019).

5. Drug Discovery and Development through Biotechnology

Biotechnology has transformed drug discovery into a data-driven, high-throughput process integrating molecular biology, omics sciences, and computational tools.

HTS platforms evaluate thousands of compounds against biological targets using automated robotics and microarray technologies. With the help of bioinformatic tools such as virtual screening, docking and predictive modelling help the scientist to identify and optimize the lead compounds in a rapid manner. Genomics identifies disease associated genes and single nucleotide polymorphisms (SNPs), facilitating target based drug design. Proteomic profiles provide the differential protein expression and post translational modifications in diseased condition. Insights into biochemical pathways and metabolic indicators are supported by metabolomics to facilitate the elucidation of pharmacological mechanisms (Han et al., 2023).

Identification and Validation of targets

CRISPR-Cas9 and RNA interference (RNAi) tools enable functional genomics-based validation of therapeutic targets. Systems biology integrates multiomics data to map disease networks in order improve precision in target selection. Nanocarriers mainly include liposomes, polymeric nanoparticle, dendrimers were used to enhance bioavailability, targeted delivery and controlled release of drugs. To quote a best example for this is liposomal doxorubicin reduces cardiotoxicity while maintaining anticancer efficacy. Recent advances include stimuli responsive nanoparticles and exosome-based delivery systems for RNA therapeutics (Khan et al., 2025).

6. Future Prospects and Ethical Considerations

CRISPR-Cas9, AAV-mediated delivery, and other gene therapies modulate genetic mutations in the living body. FDA-approved Luxturna assists in treating retinal dystrophy, while Zolgensma is for spinal muscular atrophy, proving its clinical viability. Stem cell products and engineered immune cells are the best cell based therapies to induce cell regeneration along with specific treatment. Moreover, personalized medicine, which includes genomic analysis, facilitates individualized treatment plans (Maguire et al., 2021). Pharmacogenomics offers enhanced drug efficacy and reduced adverse effects. Biopharmaceuticals remain unreachable in low-income areas due to high production expenses, stringent regulations, and cold chain logistics. The development of biosimilars, modular bioprocessing, and decentralized production can mitigate the challenges caused by high costs. Ethical issues related to biotechnological interventions include genetic alterations and data confidentiality. Gene editing in germline cells requires strict oversight because of the effects it can have on future generations.

International regulatory harmonization is essential for ensuring biosafety, efficacy, and ethical adherence (Qahwaji et al., 2024).

7. Conclusion

Therapeutic biotechnology represents the integration of biology and technology, providing novel solutions to worldwide health issues. The evolution of this field, from utilizing natural bioactive substances to developing precise biopharmaceuticals, shows continuous scientific progress. The ongoing progress in genomes, synthetic biology and nanotechnology sets the future of pharmaceuticals for personalized, precise, and sustainable healthcare solutions. Ethical governance and equal participation will decide the level to which society gets the benefits of this biotechnological revolution.

Competing Interests

The authors declare no competing interests.

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Biotechnological Innovations in Human Nutrition: From Genetic Engineering to Personalized Health

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Biotechnology uses biological systems in food science is an important shift in nutrition and health to make nutrients safer, better and more beneficial. Genetic engineering, fermentation and omics technologies are the recent advancements led to the development of nutrient enriched food products, probiotics and nutraceuticals. These products help to mitigate disease occurrence and metabolic imbalance. Nutrigenomics particularly personalized nutrition provide nourishment that are specific to each person's genetics and metabolism, whereas the bioengineering utilised to produce bioactive substances in an environment friendly way. The new areas of innovation are synthetic biology, postbiotics and smart foods for health. Despite ethical and regulatory challenges, biotechnology serves as the foundation for sustainable, individualized nutrition that meets global health and sustainability demands.

Keywords: Biotechnology, Nutrition, Probiotics, Nutrigenomics, Functional Food

INTRODUCTION

Biotechnology, the use of biological systems, organisms, or processes for the development of technology, has emerged as a pillar in contemporary nutrition and human health (Kishore et al., 1999). In the past decades, it has revolutionized food production, nutrient enrichment and disease prevention measures through precision technologies like genetic engineering, fermentation technology and omics-based nutritional mapping (Siddiqui et al., 2023). The application of biotechnology in nutrition is mainly focused on enhancing the quality, safety and functionality of food such that it not only satisfies caloric requirements but also delivers requisite bioactive compounds ensuring well-being (Ranjha et al., 2022). Genetically engineered crops enhanced with more vitamins and minerals to probiotic foods designed to maintain gut health, the advance of biotechnology is reshaping the boundary line between food and medicine (Abdul Aziz et al., 2022) (Figure 1).



Figure1. Biotechnology for Human wellness and Nutrition

The contribution of biotechnology toward human well-being is not restricted to supplement nutrients alone, but by changing the metabolic pathways in plants, microbes, and animals, it enables the synthesis of compounds which can inhibit chronic diseases such as obesity, diabetes, and cardiovascular disease (Alamgir, 2018). Nutritional genomics, for instance, helps in identifying gene–nutrient interactions which influence individual health outcomes, leading the way to personalized diets. Food crop bioengineering lowers allergenicity and increases digestibility, hence improving the nutritional well-being of the vulnerable groups (German et al., 2011). Biotechnology in nutrition has been applied throughout the world to help with hunger, food insecurity and non-communicable diseases. Golden Rice with β -carotene and biofortified cassava with more iron and zinc are two examples of biotechnology-based fortification projects that are beneficial to the public. Microbial fermentation, synthetic biology, and functional foods are sustainable biotechnological methods that collectively influence global health resilience (Avnee et al., 2023). Hence, biotechnology is an important technology linking food technology, medicine and public health in the 21st century.

2. Probiotics and Gut Microbiome Modulation

Probiotics are nothing but live microorganisms supplement with adequate amounts that confer health benefits for the host by improving or restoring microbial balance. They are typically bacteria, including *Lactobacillus*, *Bifidobacterium*, and *Saccharomyces boulardii*, chosen for their safety, resilience during gastrointestinal transit, and capacity to cling to intestinal mucosa (Latif et al., 2023). Biotechnology particularly Genetic engineering has made it possible to create better probiotics that are more resistant to stress, better at colonizing and better at making metabolites. Methods such as CRISPR-Cas9 and transcriptome analysis facilitate the identification of genes associated with probiotic traits and the development of better strains exhibiting higher bioactivity and stability (Liu et al., 2023).

The probiotic mechanisms are complex. Probiotics inhibit adhesion of pathogenic microorganisms and compete for nutrients, restoring gut microbiological balance. They regulate the immune system by inducing mucosal immunity and the production of SCFAs such as butyrate, which increases intestinal barrier function. Probiotics modulate systemic immunity through influencing cytokines, hence affecting inflammation and allergic reactions (Paul et al., 2025). Another emerging interest is the gut-brain axis, which identifies probiotics' potential influence on mood and cognition through the modulation of neurotransmitters, that signal via the vagus nerve, hence its probable application in neuro-nutrition (Kumar et al., 2024). Bioreactor with better fermentation methodology will make the probiotics on a large scale with controlled pH, temperature, and oxygen levels. All these are downstream activities: microencapsulation, freeze drying, and spray drying-all of these enhance the chances of survival through digestion and storage. To protect the microorganisms from degradation by stomach acid, encapsulation with alginate, chitosan, and lipid nanoparticles has been enhanced. By this way, it ensures their optimal functionality in the intestine.

Food and pharma industries use microencapsulation, freeze drying and spray drying for all downstream activities that make things more likely to survive digestion and storage. To protect microorganisms from being broken down by stomach acid, encapsulation materials including alginate, chitosan, and lipid nanoparticles have been improved. This ensures that they work best in the intestine. Clinical applications consist of the prevention and treatment of diarrhoea, irritable bowel syndrome, inflammatory bowel disease and lactose intolerance. They are also associated with enhanced metabolic health through actions on lipid metabolism and insulin sensitivity. Regulatory norms for usage of probiotics are not the same for all the countries and have been developed by organizations such as FDA (USA) and EFSA (Europe) for safety assessment, strain identification, and substantiation of health claims. Despite these challenges to standardization, probiotics remain one of the most promising biotechnological applications in nutritional science.

3. Functional Foods and Nutraceuticals

In addition to the regular nutritional diets, functional foods contribute to health benefits either by supporting optimal physiological processes or lowering the risk of disease occurrences. Fortified foods, fermented foods and natural foods supplemented with bioactive compounds are some examples. Nutraceuticals are more specific and describe concentrated bioactive food compounds extracted from natural products and formulated as supplements (Fekete et al., 2025). Biotechnology has dramatically changed the development of these types of nutraceuticals by allowing for the precise fortification and bio enhancement. The cereals like rice, maize and soya are enriched with key micronutrients by altering genetic ally and by improving metabolic pathways. For example, fermentation using *Lactobacillus* has been utilized to enhance mineral bioavailability through phytate degradation.

Recombinant DNA makes possible the manufacture of vitamins, antioxidants, and amino acids on industrial levels, for e.g., microbial vitamin B12 and coenzyme Q10 production (Sawant et al., 2025). In addition, transgenic expression of bioactive peptides in plants provides new nutraceutical products. Fermented foods like yogurt, kefir, kimchi and tempeh are natural precursors of probiotics and other bioactive substances. In contemporary biotechnology advancements, the nutritional quality of fermented foods is enhanced by the selection of starting cultures possessing beneficial metabolic traits. Genomic and proteomic research help find ways for flavor and health-promoting metabolites that can be used to make functional fermentation processes (Dhiman et al., 2025). Scientists can, for instance, alter lactic acid bacteria such that they make γ -aminobutyric acid (GABA), which has been found to protect the brain. The market for functional foods and nutraceuticals is growing swiftly around the world. This is because people are learning more about how to avoid health problems and manage chronic diseases. Biotechnological verification of functional features, safety analysis, and efficacy certification all help to build consumer trust (Cui et al., 2020). Regulatory harmonization, clear labeling, and long-term clinical verification are still big problems. As biotechnology progresses increasingly into nutrition science, functional foods play a vital role in the dietary approaches to wellness in the future.

4. Bioactive Compounds from Natural and Engineered Sources

Bioactive compounds are a category of chemical substances that elicit desirable biological activities beyond their nutritional properties. They are derived from polyphenols, flavonoids, carotenoids, peptides, and polysaccharides in plants, microorganisms, marine sources and also fermentation systems. Biotechnological techniques are essential in the discovery, extraction and production of bioactive compounds with streamlined processes into efficiency and sustainability. Microbial biotechnology has aided in the large scale biosynthesis of secondary metabolites from medicinal plants with similar product properties. *Escherichia coli* and *Saccharomyces cerevisiae*, for instance, have been used to produce resveratrol, curcumin, and other antioxidant compounds. Fermentation based bioprocesses also facilitate the transformation of agricultural by products into high value nutraceuticals in agreement with circular bioeconomy practices (Khan et al., 2025). Microalgae including *Nannochloropsis* and *Spirulina* have, in marine biotechnology, been utilized for their high concentration of omega-3 fatty acids, carotenoids, and phycobiliproteins with shown antioxidant and anti-inflammatory activities (Papadaki et al., 2024).

Extraction and purification processes have been improved through biotechnological advances like enzyme-assisted extraction, supercritical fluid extraction, and membrane bioprocessing. These techniques enhance yield and maintain structural integrity of sensitive molecules. In addition, metabolic engineering enables overexpression of important biosynthetic enzymes to maximize

accumulation of target compounds. Genome editing in plants and microorganisms by the CRISPR/Cas9 technology has been employed for modulating the metabolic pathway towards enhanced production of nutraceutical compounds (Fierascu et al., 2020; Trombino et al., 2025).

The biological activities of these bioactive are vast, they scavenge free radicals, regulate inflammatory signaling pathways and maintain metabolic homeostasis. For example, green tea and berry polyphenols minimize oxidative stress and decrease cardiovascular disease risk. Angiotensin converting enzyme (ACE) inhibiting peptides from fermented milk display antihypertensive activity (Goszcz et al., 2017). Biotechnological production provides consistency, safety and scalability thereby these compounds have pharmaceutical, cosmetic and food industry value.

5. Personalized Nutrition and Nutrigenomics

Personalized nutrition is a new arena which has personalized diet prescription in light of a person's genetic, metabolic and microbiome population. Nutrigenomics investigates the impact of nutrients on gene expression, while nutrigenetics examines the influence of genetic differences on nutrient metabolism. These two branches combine to utilize biotechnological techniques to get individual variability in response to diet and to create precision based dietary approaches. The genetic underpinning of nutrient metabolism is in polymorphisms in genes coding metabolic enzymes, transporters, and receptors. For example, mutations within the MTHFR gene modulate folate metabolism and its action on homocysteine concentration and cardiovascular risk. Biotechnological methodologies such as NGS and SNP genotyping help delineate such variations at the population and individual levels (Singar et al., 2024). This allows for the fashioning of diets based on genetic predisposition and therefore contributes to healthier outcomes.

Omics technologies—genomics, proteomics, metabolomics, and microbiomics—are central to personalized nutrition studies. Genomics unveils the genetic template; proteomics detects protein biomarkers of nutrient interaction; metabolomics characterizes the metabolites produced by dietary interventions. Such datasets are amalgamated by use of bioinformatics and systems biology strategies to develop predictive nutritional models. For example, metabolomic profiling can be used to predict glucose response to particular foods to inform individualized meal planning for diabetes (Chaudhary et al., 2021).

Biotechnological platforms also help in the personalization of food products. Precision fermentation and synthetic biology allow the tailoring of nutrient profiles to meet genetic and metabolic requirements. Wearable biosensors and digital health platforms also converge with biotechnology for tracking real-time physiological reactions, facilitating dynamic dietary modifications. Personalized nutrition, however, poses ethical and privacy issues surrounding the handling of genetic data and

necessitates rigid data management and informed consent measures. The intersection of biotechnology and digital health therefore marks the dawn of a new age of personalized wellness management.

6. Future Directions and Global Challenges

The ultimate goal of biotechnology in human health and nutrition is defined by breakthroughs in synthetic biology, postbiotics and personalised food diets. Synthetic biology allows the construction of new metabolic routes for the biosynthesis of functional foods, flavor compounds, and bioactives that are not found naturally in traditional foods. Microbes can be engineered to transform simple substrates into sophisticated nutraceuticals, replacing traditional agriculture. Postbiotics is a non-viable microbial derivatives like metabolites, peptides and cell wall constituent that are a next-generation class of functional ingredients that demonstrate stability and targeted physiological effects. Personalised foods link the biosensors and responsive delivery systems, are yet another area of biotechnology. Smart foods can track biomarkers such as glucose or pH and release drugs or nutrients based on that. The synergy between nanotechnology and biotechnology provides controlled release and enhanced bioavailability of nutrients. In addition, plant molecular farming is progressing to allow vaccines and therapeutic proteins to be produced in edible plants as cost-effective tools for preventive medicine.

Yet, a number of global issues follow in the wake of these developments. Safety evaluation of genetically modified organisms (GMOs) and constructed probiotics is a major regulatory issue. Providing ecological sustainability, ethical and fair access to biotechnological nutrition technologies is required. Developing nations with malnutrition and emerging non-communicable diseases are as double burdens that need accommodative policies and infrastructure to implement biotech based nutritional interventions. In addition, mutualization of international legislation and consumer confidence through clear information and education are essential for the ethical use of biotechnology in diet.

In coordination with international health objectives like the United Nations Sustainable Development Goals (SDGs), biotechnology is a pivotal integrative function. It facilitates SDG 2 ("Zero Hunger") by nutritional enhancement of crops, SDG 3 ("Good Health and Well-being") by disease prevention via functional foods, and SDG 12 ("Responsible Consumption and Production") through sustainable bioprocessing. Interdisciplinary convergence of science, industry and policy will define the extent to which biotechnology will advance equitable human health (Vărzaru et al., 2024).

7. Conclusion

Biotechnology has made a big revolution in the field of human nutrition and health by linking food science with innovative biological techniques. From cultivating genetically improved crops and probiotic products to producing high value bioactive ingredients and customized nutrition systems,

biotechnology plays a unavoidable role for enhancing human health. It will fill the gap between traditional and precision medicine, providing direct interventions for disease prevention as well as health optimization.

Without balanced nutrition, the world is grappling with, aging populations and lifestyle diseases the convergence of biotechnology in food systems provides scientifically informed and sustainable solutions. The future development will depend on interdisciplinary research, responsible governance and global cooperation to make these technologies that are accessed to all populations. Finally, biotechnology is the foundation of a new nutrition paradigm where innovation meets wellness, sustainability and global resilience to health.

Conflict of interests

The authors declare that they have no conflict of interest, financial or otherwise.

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Advanced Pest and Disease Management Strategies in Protected Cultivation

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Protected cultivation has emerged as a transformative solution for modern horticulture, offering year-round production of high-value crops through controlled environmental conditions. However, the stable microclimate that enhances plant growth also favors the rapid proliferation of insect pests, mites, nematodes, and pathogens. This chapter presents a comprehensive overview of **advanced pest and disease management strategies** designed for protected horticultural systems. It explores the biology, damage symptoms, and host range of major pest groups such as aphids, whiteflies, thrips, leaf miners, mites, and caterpillars, as well as common fungal, bacterial, viral, and nematode diseases.

The chapter emphasizes an **integrated pest management (IPM)** framework combining physical exclusion, cultural practices, biological control, and selective chemical interventions. Detailed attention is given to preventive measures like insect-proof netting, UV-blocking films, reflective mulches, and sanitation protocols, which collectively reduce pest entry and survival. The role of **beneficial organisms**—predators, parasitoids, and microbial biocontrol agents—is discussed as the foundation of eco-friendly pest regulation. Safe, residue-minimizing chemical molecules are presented as the final step in a layered control approach.

Emerging innovations such as **AI-driven pest surveillance**, **IoT-based climate control**, **drone-assisted monitoring**, **nanotechnology formulations**, and **RNA-based biopesticides** represent a new frontier in smart IPM. The chapter also highlights government initiatives, certification schemes for residue-free produce, and digital extension programs supporting farmers. Altogether, it provides a holistic vision of how protected cultivation can evolve into a **climate-smart, technology-integrated, and environmentally sustainable production system**, balancing high productivity with ecological integrity.

Keywords:

Protected cultivation; Integrated pest management (IPM); Biological control; Smart greenhouse technology; Sustainable horticulture; Climate-smart agriculture; Nanobiopesticides; Artificial intelligence in agriculture; Residue-free produce.

1. Introduction

Protected cultivation has revolutionized the global horticultural landscape, enabling farmers to produce high-value crops throughout the year irrespective of external climatic conditions. This modern farming approach employs controlled structures—such as polyhouses, greenhouses, shade nets, and tunnels—to regulate temperature, humidity, light, carbon dioxide levels, and nutrient supply. By creating a favorable microclimate, protected cultivation enhances productivity, optimizes resource use, and allows cultivation of exotic vegetables, flowers, and nursery plants in regions where they were previously unsuitable.

The technology gained rapid importance during the last two decades due to increasing pressure on land, erratic climatic conditions, and the need for sustainable intensification of agriculture. Globally, China leads with more than **2.8 million hectares** under protected cultivation, followed by countries like Spain, Japan, the Netherlands, and India. In India, the protected cultivation area has expanded significantly under government schemes such as the **Mission for Integrated Development of Horticulture (MIDH)** and **National Horticulture Mission**, reaching nearly **30,000 hectares** by 2024. However, the very conditions that favor plant growth—high humidity, constant temperature, and limited air circulation—also create ideal environments for pest and disease development. The enclosed system limits natural predators and environmental variability, allowing insects, mites, nematodes, and pathogens to thrive rapidly. Hence, **integrated pest and disease management (IPDM)** becomes indispensable in such systems to maintain both yield and produce quality.

2. Evolution of Protected Cultivation in Horticulture

Protected cultivation emerged as a response to the vulnerabilities of open-field agriculture, particularly in horticultural crops that demand stable growing conditions. Initially, simple structures like thatched sheds or low tunnels were used to protect seedlings. Over time, advancements in material science led to the development of UV-stabilized polyethylene films, polycarbonate sheets, and automated climate control systems.

In India, research institutions such as **ICAR-Indian Institute of Horticultural Research (IIHR)**, **TNAU**, and **PAU** have demonstrated successful protected cultivation of tomato, capsicum, cucumber, rose, chrysanthemum, gerbera, and strawberry. Farmers in Telangana, Maharashtra, and Himachal Pradesh now cultivate these crops throughout the year with yields 3–5 times higher than open-field conditions.

Despite these advantages, pest and disease outbreaks often offset the benefits if not managed scientifically. For instance, studies conducted at IIHR Bengaluru (2023) reported that unmonitored pest populations could reduce greenhouse tomato yields by **up to 60%**, primarily due to whiteflies, thrips, and spider mites.

3. Importance of Pest and Disease Management under Protected Conditions

The microclimate inside protected structures provides continuous availability of host plants and stable conditions that accelerate pest multiplication. Pests complete their life cycles faster—aphids and whiteflies can develop from egg to adult within 7–10 days at 25–30°C, leading to overlapping generations and persistent infestations.

Moreover, the restricted movement of air and absence of rainfall hinder the natural dispersal of spores and pests, causing local population explosions. Chemical residues tend to persist longer because sunlight and UV degradation are limited, thus posing risks of phytotoxicity and human exposure.

Hence, the emphasis in protected farming is not merely on control but on **prevention, prediction, and eco-compatible management**. The modern objective is to create a resilient production ecosystem through:

- Early pest exclusion
- Regular surveillance
- Use of biological control agents
- Environmentally safe crop protection chemicals
- Smart technology integration (sensors, automation, AI prediction tools)

4. Common Pests in Protected Cultivation

Several insect pests have adapted successfully to the stable greenhouse environment. These species are typically polyphagous, highly reproductive, and difficult to manage using conventional insecticides due to resistance development.

4.1 Aphids (Aphididae)

Aphids such as *Aphis gossypii* (cotton aphid) and *Myzus persicae* (green peach aphid) are soft-bodied sap suckers commonly seen in capsicum, tomato, and ornamental crops. They cause leaf curling, yellowing, stunted growth, and transmit viral diseases like cucumber mosaic virus (CMV) and lettuce mosaic virus (LMV).

Under protected conditions, aphids reproduce parthenogenetically, completing a generation in less than a week during warm months. Their excreted honeydew encourages the growth of sooty mold, reducing photosynthetic activity and market value.

4.2 Whiteflies (Aleyrodidae)

The greenhouse whitefly (*Trialeurodes vaporariorum*) and cotton whitefly (*Bemisia tabaci*) are among the most destructive pests of protected crops. They feed on the undersides of leaves, causing chlorosis

and leaf drop. More importantly, they act as vectors for viral diseases such as tomato yellow leaf curl virus (TYLCV).

A single female can lay 150–200 eggs, and up to 15 generations may occur annually in controlled environments. In addition, their waxy secretion and honeydew deposition facilitate secondary fungal growth.

4.3 Thrips (Thysanoptera)

Thrips such as *Frankliniella occidentalis* and *Thrips tabaci* are tiny, slender insects with fringed wings. They scrape leaf tissue and suck sap, leading to silverying, curling, and deformation of leaves and flowers. Thrips are also the main vectors of tospoviruses like Tomato Spotted Wilt Virus (TSWV). Their cryptic behavior makes them difficult to detect, and populations often develop resistance to several insecticides.

4.4 Mites (Tetranychidae and Tarsonemidae)

The two-spotted spider mite (*Tetranychus urticae*) and yellow mite (*Polyphagotarsonemus latus*) are frequent in warm, dry greenhouse conditions. They feed on plant cells, causing stippling, bronzing, and leaf drop. Mite outbreaks are particularly severe in cucurbits and ornamentals. Under optimal conditions, spider mites complete a generation in just 6–8 days, leading to exponential growth if unchecked.

4.5 Leaf Miners (Agromyzidae)

Liriomyza trifolii and *L. sativae* are small flies whose larvae mine leaf tissues, forming serpentine tunnels that reduce photosynthesis. Severe infestation leads to leaf necrosis and premature Because larvae reside inside leaves, they are inaccessible to contact insecticides, requiring integrated approaches involving parasitoid release and sticky traps

4.6 Caterpillars and Lepidopteran Pests

Polyphagous species such as *Spodoptera litura* (tobacco caterpillar) and *Helicoverpa armigera* (tomato fruit borer) occasionally invade greenhouses through openings or contaminated planting material.

Larvae feed voraciously on foliage and fruits, rendering them unmarketable. Pheromone-based monitoring and timely biocontrol applications are essential to prevent outbreaks.

4.7 Other Emerging Pests

Recent surveys (2024) by ICAR–NBAIR identified emerging pests such as fungus gnats (*Bradysia spp.*), root aphids, and mealybugs in soilless and hydroponic units. These pests thrive in moist media and often spread through contaminated substrates or irrigation water.

Major Diseases under Protected Cultivation

Fungal Diseases

1. **Powdery Mildew (*Leveillula taurica*, *Oidium* spp.):** A common disease in cucurbits and tomato, favored by moderate humidity and low ventilation.
2. **Botrytis Blight (*Botrytis cinerea*):** A gray mold affecting flowers and fruits, particularly in rose and strawberry.
3. **Fusarium Wilt (*Fusarium oxysporum*):** Persists in soil and infects through roots; often severe in tomato and chrysanthemum.
4. **Downy Mildew (*Pseudoperonospora cubensis*):** Affects cucumber and other cucurbits under high humidity.

Bacterial Diseases

5. **Bacterial Leaf Spot (*Xanthomonas campestris*):** Causes necrotic spots on foliage and fruits in capsicum and tomato.
6. **Bacterial Wilt (*Ralstonia solanacearum*):** Devastates solanaceous crops; survives in soil and irrigation systems.

7. 5.3 Viral Diseases

8. **Tomato Yellow Leaf Curl Virus (TYLCV) and Tomato Mosaic Virus (ToMV)** are the most widespread. They cause severe yield reductions in tomato and pepper crops. Virus transmission is mainly through whiteflies, aphids, and mechanical means such as handling or pruning.

5.4 Nematode Problems

Root-knot nematodes (*Meloidogyne incognita*) frequently infest greenhouse soils, forming galls on roots that hinder nutrient uptake. They often interact with fungi like *Fusarium*, causing complex disease syndromes.

5.5 Recent Trends in Pest-Disease Dynamics

Over the last five years, climate fluctuations and increased cropping intensity have altered pest behavior. Reports from multiple Indian KVKs reveal:

1. Faster pest cycles due to warmer nights in greenhouses.
2. Shifts from single dominant pests to mixed populations (e.g., whitefly + thrips).
3. Increased incidence of virus-vector complexes.
4. Secondary outbreaks of soil-borne fungi following high humidity spells.

Hence, pest and disease management now demands a dynamic, **systems-based approach** integrating physical, biological, and digital tools.

6. Major Arthropod Pests: Biology & Symptoms

Understanding pest life cycles, survival strategies, and crop-wise impact helps in designing stronger management plans. Below are detailed profiles rewritten in new style

6.1 Aphids (Aphididae Family)

Key species:

Aphis gossypii — Cotton aphid

Myzus persicae — Green peach aphid

Appearance:

Aphids are tiny, pear-shaped, soft-bodied insects often seen clustered on tender plant parts, especially shoot tips and undersides of leaves. They reproduce rapidly through **parthenogenesis**, producing live nymphs. Under warm greenhouse conditions, a complete generation may finish within **7–9 days**, leading to explosive population growth.

Symptoms:

- Cup-shaped curling & twisting of young leaves
- Sticky honeydew deposits
- Development of sooty mold
- Wilting during heavy attack
- Transmission of viruses like CMV, LMV, PVY

Favorable Conditions:

Warm temperature (20–28°C)

Soft lush growth from excess nitrogen

Dense plant canopy

Economic

Heavy aphid infestation in capsicum, cucumber, chrysanthemum, and rose can reduce marketable produce by **30–50%**.

Management Summary:

Introduce early traps & scouting

Release *Aphidius colemani*, *Aphidoletes aphidimyza*

Avoid excess nitrogen; prune dense foliage

Neem oil 0.5–1% spray if early infestation

6.2 Whiteflies (Aleyrodidae Family)

Species of concern:

Bemisia tabaci — Cotton whitefly

Trialeurodes vaporariorum — Greenhouse whitefly

Biological

Whiteflies resemble tiny moths; they feed by sucking plant sap, weakening plants and spreading viruses such as **TYLCV**, **ToCV**. In warm protected houses, they complete **12–15 generations/year**.

Symptoms:

Yellowing or chlorotic patches

Leaf wilting and premature leaf fall

Honeydew → sooty mold

Viral infection symptoms

High-Risk

Tomato, capsicum, cucumber, gerbera

Management:

UV-absorbing greenhouse plastic

Yellow sticky traps @ 1 trap/100 m²

Encarsia formosa release early

Use insect-proof nets (40–50 mesh)

Botanical extracts or safer molecules (spiromesifen/buprofezin)

6.3 Thrips (Thysanoptera Order)

Main species:

- *Frankliniella occidentalis* (Western flower thrips)
- *Thrips tabaci* (Onion thrips)

Damage:

Thrips scrape leaf tissue and suck sap, causing:

- Silver streaks on leaves
- Distorted flowers
- Brown patches & fruit scarring
- Transmission of **TSWV**, **GBNV**

Perfect Conditions:

- Hot & dry greenhouse climates
- Flowering stages of crops

Integrated Actions:

- Install **blue sticky traps** (proved best for thrips)
- Introduce *Amblyseius swirskii*, *Orius insidiosus* early
- Maintain humidity >60% to slow reproduction
- Neem soap wash + bio-fungal sprays (*Beauveria bassiana*)

6.4 Mites (Spider & Tarsonemid)

Key pests:

- *Tetranychus urticae* — Two-spotted spider mite
- *Polyphagotarsonemus latus* — Yellow mite

Lifecycle Mites thrive under dry heat & remain hidden on leaf undersides. They weave fine webs and suck cell sap causing **bronzing, speckling, leaf drop**.

Crop Hosts: Tomato, cucumber, capsicum, rose, gerbera

Best Control:

Phytoseiulus persimilis predators

Maintain optimum humidity

Sulfur dusting in early stage (don't use above 28°C)

Avoid pyrethroids — they flare mites

6.5 Leaf Miners (Agromyzidae)

Species:

Liriomyza trifolii

Liriomyza sativae

Symptoms:

Tunneling (mines) inside leaves

Blistered patches and necrosis

Leaf drying under high infestation

Control Package:

Yellow traps near seedling stage

Remove damaged leaves early

Diglyphus isaea parasitoid release

Systemic insect growth regulators

6.6 Caterpillar Complex

Notable species:

Helicoverpa armigera

Spodoptera litura

Though less common than sucking pests, they enter through nets, vents or nursery plants and quickly destroy foliage & fruits.

IPM Approach:

Pheromone traps (2–4/1000 m²)

Light trap (night monitoring)

Trichogramma chilonis parasitoids

Bacillus thuringiensis weekly spray

7. Major Diseases in Protected Crops

Disease	Pathogen	Symptoms	Favorable conditions
Powdery mildew	<i>Oidium, Leveillula</i>	White powder patches, leaf curl	Low air circulation, cool nights
Downy mildew	<i>Pseudoperonospora cubensis</i>	Yellow spots, purple underside mold	High humidity
Botrytis blight	<i>Botrytis cinerea</i>	Gray mold, petal burn	Condensation, dead plant parts
Fusarium wilt	<i>Fusarium oxysporum</i>	Sudden wilting, vascular browning	Warm moist soil

7.1 Fungal Diseases

Updated Control:

Solarization + biofumigation

Trichoderma enriched media

Fogging & humidity control

Hydrogen-peroxide + silver sanitizers

7.2 Bacterial Diseases

Disease	Causal organism	Key symptoms
Bacterial wilt	<i>Ralstonia solanacearum</i>	Sudden vascular wilt
Bacterial spot	<i>Xanthomonas spp.</i>	Angular spots, cracking on fruits
Soft rot	<i>Erwinia spp.</i>	Watery rots in humid conditions

Solutions:

Drip fertigation sanitation

Copper-based sprays (limited & careful use)

UV-treated irrigation water

Resistant hybrid selection

7.3 Viral Diseases

Disease	Major Virus	Vector
Tomato yellow leaf curl	TYLCV	Whitefly
Tomato spotted wilt	TSWV	Thrips
Cucumber mosaic	CMV	Aphids

Management:

Virus-free seedlings

Remove infected plants immediately

Reflective mulches

Biological vector suppression

7.4 Nematodes

Root-knot nematode — *Meloidogyne spp.*

Symptoms:

Root galls

Stunting, poor uptake

Sudden wilting even with moisture

Control:

Crop rotation + resistant rootstocks

Neem cake + consortium bio-nematicides

Soil solarization + formal biological rest period

8. Diagnostic Guide

Problem Type	Typical Signs
Sap-sucking insects	Leaf curling, sticky surface, sooty mold
Borers & miners	Holes, tunnels, frass
Fungal diseases	Mold, mildew, fruit rot
Bacterial diseases	Oozing, wet patches
Viruses	Mosaic, leaf distortion, stunting

Integrated Management Approaches in Protected Cultivation

The success of protected cultivation depends not only on environmental control but also on a strong, layered pest management strategy. Integrated Pest and Disease Management (IPDM) combines physical, cultural, biological, and chemical tools to maintain pest populations below economic injury levels while ensuring environmental safety and consumer health.

9. Preventive and Exclusionary Measures

9.1 The Principle of Prevention

Unlike open-field farming, pest outbreaks inside greenhouses can expand extremely fast due to closed, favorable conditions. Thus, prevention—*keeping pests out before they enter*—is the first and most cost-effective line of defense.

9.2 Structural Exclusion

Insect-Proof Netting:

Install 40–50 mesh nylon or polyethylene nets over ventilation openings. For thrips and leafminers, use 76 mesh ($\approx 192\ \mu\text{m}$) screens. The latest Indian designs use **UV-stabilized, anti-dust netting**, improving light penetration by 15%.

Double-Entry Doors (Airlocks):

A buffer zone between the outside and growing area minimizes pest entry when people or materials move in and out.

Floor and Border Management:

Concrete or plastic-lined floors reduce pest pupation and nematode survival. Clean perimeters prevent external pest migration.

Positive Air Pressure Systems:

In large commercial greenhouses, slight positive pressure (created by filtered air blowers) prevents insects from flying inside through small gaps.

UV-Blocking Films:

Modern polyethylene films embedded with UV absorbers ($<380\ \text{nm}$ cutoff) disrupt insect navigation and virus transmission. Field studies in 2024 at IIHR-Bengaluru found **65% fewer whiteflies** and **90% lower virus incidence** in such houses.

9.3 Quarantine and Hygiene

Nursery Quarantine: Only introduce pest-free seedlings.

Worker Hygiene: Mandatory hand-washing, footwear change, and no movement from infested to clean areas.

Tool Disinfection: Clean pruning shears, trays, and benches with 1% sodium hypochlorite or hydrogen peroxide.

Debris Management: Remove crop residues promptly and dispose outside the structure to eliminate breeding grounds.

9.4 Weed and Alternate Host Control

Weeds harbor whiteflies, aphids, and viruses. Remove them regularly from both inside and around the greenhouse. Grow a narrow strip of *marigold* or *sorghum* around the greenhouse as a trap or barrier crop.

10. Cultural Practices and Crop Management

10.1 Crop Rotation and Fallowing

Continuous monocropping increases pest persistence. Introduce short fallow breaks of 2–4 weeks between crops and rotate with non-host crops such as leafy greens or lettuce.

10.2 Nutrient and Water Management

Overuse of nitrogen results in lush, soft tissue ideal for sucking insects. Maintain balanced N:P:K levels. Potassium strengthens plant cell walls and enhances tolerance to piercing pests.

Drip irrigation should be preferred to avoid excessive humidity that encourages fungal diseases.

10.3 Plant Spacing and Pruning

Adequate plant spacing allows air circulation, reducing humidity-related diseases. Remove lower old leaves regularly to prevent whitefly and mite build-up.

10.4 Mulching

Reflective mulches (silver/white) deter insects by altering light reflection patterns and suppressing soil-borne pests. They also reduce weed growth and maintain soil moisture.

10.5 Resistant and Tolerant Varieties

Breeding programs have produced resistant cultivars such as:

Tomato hybrids tolerant to TYLCV (*Arka Rakshak*, *US 440*)

Capsicum hybrids tolerant to bacterial spot (*Indra*, *Sweta*)

Cucumbers with powdery mildew resistance (*Sheetal*, *Hercules*)

11. Monitoring, Scouting, and Early Warning Systems

11.1 Regular Scouting

Walk through inspections at least twice weekly, covering random spots in a zigzag pattern. Examine both upper and lower leaf surfaces. Use hand lenses to detect early pest presence.

11.2 Visual Indicators

- Sticky cards: yellow for whiteflies/aphids; blue for thrips.

- Trap density: 1–2 traps per 100 m².
- Replace when 70% covered with insects.

11.3 Data Recording

Maintain a pest logbook—date, pest species, intensity, location, and crop stage. This data helps in forecasting and decision-making.

11.4 Digital Monitoring (2024–25 Trend)

Smart cameras and IoT sensors integrated with AI apps (e.g., **Plantix Pro**, **AgriSense**, **TNAU Smart Scout**) now automatically identify insect species and alert growers via smartphone. These systems detect pest population surges **2–3 days earlier** than visual scouting.

12. Biological Control in Protected Cultivation

Biological control is the backbone of sustainable pest management in enclosed systems. Favorable microclimates support the survival of beneficial organisms released into greenhouses.

12.1 Types of Biocontrol Agents

1. **Predators** – free-living insects that consume multiple prey.
 - a. *Chrysoperla carnea* (green lacewing) for aphids.
 - b. *Phytoseiulus persimilis* for spider mites.
 - c. *Orius insidiosus* (minute pirate bug) for thrips.
2. **Parasitoids** – insects that lay eggs on or inside pests.
 - a. *Encarsia formosa* for whitefly.
 - b. *Aphidius colemani* for aphids.
 - c. *Trichogramma chilonis* for lepidopteran eggs.
3. **Pathogens** – microbes causing insect diseases.
 - a. *Beauveria bassiana*, *Metarhizium anisopliae* (fungi).
 - b. *Bacillus thuringiensis* (bacterium).
 - c. Entomopathogenic nematodes (*Steinernema spp.*).

12.2 Mode of Application

- **Inoculative release:** Small numbers introduced early to establish populations.
- **Inundative release:** Large numbers for immediate control during outbreaks.
- **Conservation:** Maintain favorable microclimate and avoid broad-spectrum chemicals.

12.3 Bio-Control Release Schedules (Typical Examples)

Pest	Bioagent	Recommended Release
Whiteflies	<i>Encarsia formosa</i>	2–5 adults/m ² weekly during early crop stage
Thrips	<i>Amblyseius swirskii</i>	100–200 mites/m ² biweekly
Aphids	<i>Aphidius colemani</i>	0.5–1 wasp/plant at early detection
Spider mites	<i>Phytoseiulus persimilis</i>	5–10 mites/m ² in hot season
Caterpillars	<i>Trichogramma chilonis</i>	1 card/50 m ² every 7 days

12.4 Modern Bioformulations

New nano-formulations of entomopathogenic fungi like *Metarhizium anisopliae* and *Beauveria bassiana* show better spore survival in humid greenhouses. Combination bioproducts containing *Trichoderma* + *Pseudomonas fluorescens* + *Bacillus subtilis* improve both root health and pest resistance.

13. Mechanical and Physical Control

- **Hand Collection:** Remove infested leaves or fruits.
- **Water Jet Sprays:** Knock off soft-bodied pests.
- **Sticky Bands:** Trap crawling insects on stems.
- **Soil Solarization:** 30–40 days of clear polyethylene cover kills nematodes and soil fungi.
- **Light Traps:** Effective against nocturnal moths.

Automation is now being introduced—**robotic pest vacuums** and **UV-C light emitters** can suppress fungal spore load without chemicals.

14. Chemical Control in Eco-Intensive Systems

14.1 Principles of Chemical Use

Chemical pesticides are the last line of defense in an IPM program. Their use should be:

- **Need-based** – guided by pest thresholds.
- **Selective** – targeting specific pests only.
- **Safe** – minimal residue and worker protection.

14.2 Modern Safer Molecules

Pest	Preferred Chemicals	Remarks
Aphids/Whiteflies	Spiromesifen, Flonicamid, Pyriproxyfen	Non-systemic; minimal residue
Thrips	Spinosad, Eamectin benzoate	Compatible with biocontrol agents
Mites	Hexythiazox, Fenpyroximate	Rotate to prevent resistance
Caterpillars	Chlorantraniliprole, Bt formulations	Highly specific and eco-safe

14.3 Application Guidelines

- Apply only when pest populations exceed **Economic Threshold Levels (ETL)**.
- Use fine-mist sprayers for uniform coverage.
- Maintain 7–10 day intervals between sprays.
- Ensure ventilation after spraying to avoid phytotoxicity.
- Avoid repeated use of the same molecule.

14.4 Resistance Management

Rotation of insecticides with different IRAC codes prevents pest resistance. Combine chemical and biological approaches rather than using chemicals continuously.

15. Role of Climate and Environment Management

Environmental parameters directly influence pest behavior. Correct microclimate manipulation is a natural way to suppress outbreaks:

Factor	Optimum Range	Impact
Temperature	20–28°C	Above 32°C accelerates mite & whitefly activity
Relative Humidity	60–70%	>85% favors fungal growth
Light Intensity	40–60 klux	Adequate light reduces leaf wetness
Air Circulation	Uniform	Prevents localized condensation

Modern greenhouses now employ **automated sensors**, **foggers**, and **fans** linked to climate software that maintains the above thresholds, keeping both plant growth and pest risk in balance.

16. Integration of All Methods: IPM Model

Phase	Key Action
Pre-planting	Soil sterilization, structure sanitation, trap setup
Early crop stage	Preventive bio-agent release, sticky trap monitoring
Mid-growth	Nutrient balance, pruning, selective chemical rotation
Fruiting	Maintain humidity control, remove infested leaves
Post-harvest	Sanitize greenhouse, solarize soil, data logging

17. Farmer Education and Capacity Building

Effective pest management is incomplete without knowledge dissemination. KVKs, state agriculture universities, and private agritech platforms are now offering:

- Training on **biocontrol handling**
- **Digital pest identification** through mobile applications
- **Safe pesticide usage** and residue testing workshops

Emerging Innovations and Future Pathways in Protected Crop Protection

18. The Rise of Smart IPM in Protected Cultivation

The concept of Integrated Pest Management (IPM) has evolved from a field-based decision system to a **data-driven, technology-integrated discipline**. Protected structures are particularly suited to automation because their climate and environment are already controlled, making digital pest management a natural next step.

18.1 Digital and Sensor-Based Surveillance

Modern greenhouses now deploy sensors and computer vision systems that continuously monitor temperature, humidity, and pest presence.

Key components include:

- **Smart Camera Traps:** These AI-powered traps automatically identify insects (aphids, whiteflies, thrips) through image recognition and send alerts to the grower's smartphone.
- **IoT-linked Micro-Climate Sensors:** Continuously record environmental conditions; when temperature and humidity cross pest-favorable thresholds, the system issues preventive warnings.
- **Thermal Imaging:** Detects early stress or fungal infection before visible symptoms appear.
- **Acoustic Pest Detectors:** Capture the vibrations produced by larvae feeding inside plant tissue.

Recent trials under the **ICAR–Digital Pest Surveillance Project (2024)** reported a **35–40 % reduction** in pesticide use when AI-driven alerts were incorporated into IPM routines.

18.2 Drone and Robotics Applications

Although small greenhouses limit drone use, medium-to-large polyhouse clusters are experimenting with mini-drones for:

- Aerial release of biocontrol agents.
- Precision spot-spraying using micro-nozzles.
- Thermal imaging to map pest hotspots.

Robotic pest vacuums and UV-C light robots are now being adopted for fungal spore sterilization in floriculture units.

18.3 Smart Decision-Support Systems (DSS)

Software platforms such as **e-Greenhouse Manager**, **Krishi-AI**, and **Plant Health Digital Twin (PDT-India)** combine real-time climate, pest, and nutrient data. These DSS tools suggest the **exact timing** for biological releases, watering, and spray schedules—helping farmers make preventive moves rather than reactive ones.

19. Nanotechnology and Biochemical Innovations

19.1 Nano-Formulated Biopesticides

Research at IIHR and NIPHM (2024–25) introduced nano-carriers for *Beauveria bassiana* and *Metarhizium anisopliae*.

Benefits include:

- Longer shelf life (8–10 months).
- Slow-release delivery improving persistence on leaves.
- Compatibility with beneficial insects.

19.2 RNA-Based Biocontrol

Emerging RNA interference (RNAi) technologies can silence specific pest genes, such as those responsible for reproduction in *Helicoverpa armigera*. These are biodegradable and non-toxic to beneficial fauna.

19.3 Botanicals and Semiochemicals

Plant-based volatiles (citronella, neem, pongamia, and tulsi oils) are being standardized for use in vapor diffusers inside greenhouses to repel insects naturally.

Pheromone confusion dispensers reduce mating success of moth pests by more than 80 %.

20. Soil and Substrate Health Innovations

In protected cultivation, soil and substrate are the foundation of plant health. To maintain pathogen-free growth media:

1. **Bio-fumigation:** Incorporation of mustard or cabbage residues releases glucosinolates that suppress soil pathogens.
2. **Beneficial Microbiome Management:** Regular inoculation with *Trichoderma harzianum*, *Pseudomonas fluorescens*, and *Bacillus subtilis* supports plant defense enzymes.
3. **Hydroponic Sanitation:** In NFT and cocopeat systems, peroxide-silver disinfectants prevent algal and bacterial film formation.
4. **Probiotic Compost Teas:** Provide microbial balance and induce systemic resistance in crops.

21. Policy, Certification, and Market Incentives

21.1 Residue-Free Certification

As export markets increasingly demand pesticide-free produce, many Indian states (Telangana, Karnataka, Gujarat) are developing “**Residue-Free Protected Produce**” certification systems.

Greenhouse IPM farmers using biocontrol and smart surveillance gain access to premium pricing, especially in urban retail chains.

21.2 Government Support

The **Mission for Integrated Development of Horticulture (MIDH)** now includes financial assistance for:

- Installation of insect-proof net houses.
- Subsidies for biocontrol laboratories.
- Training on climate-smart pest management.

21.3 Traceability through Blockchain

Pilot programs in Maharashtra link greenhouse batches to QR-coded digital records showing pesticide use, bioagent releases, and harvest dates. Such traceability boosts consumer trust and export eligibility.

22. Socio-Economic and Environmental Impacts

Smart IPM brings triple-bottom-line benefits:

- **Economic:** Reduced input costs and fewer crop losses.
- **Social:** Safer working conditions and improved farmer awareness.
- **Environmental:** Protection of beneficial insects and pollinators; reduced chemical residues in soil and water.

Studies from KVK-Nagarkurnool (2025) show that farmers adopting full IPM schedules in protected tomato achieved **20–25 % higher profit margins** compared to conventional pesticide users.

23. Challenges and Constraints

Despite remarkable progress, several challenges remain:

1. **Lack of Awareness:** Many smallholders still rely solely on chemical solutions.
2. **High Initial Cost:** Automated traps and sensors require capital investment.
3. **Limited Supply of Quality Bioagents:** Commercial mass-rearing units are still insufficient in rural regions.
4. **Skill Gap:** IPM needs consistent observation and technical understanding.
5. **Climate Variability:** Extreme weather outside can still influence inside microclimates and disease patterns.

Addressing these gaps through training, cooperatives, and digital extension will be vital.

24. Future Prospects and Way Forward

Protected cultivation is moving toward an **eco-digital era**—where biological wisdom meets artificial intelligence.

The near future will witness:

- **Predictive Greenhouses:** Self-learning systems that analyze pest-climate correlations and adjust conditions automatically.
- **AI-Integrated Pollination & Pest Drones:** Combining beneficial pollination with pest detection.
- **Climate-Adaptive Plastic Films:** That regulate UV, IR, and pest behavior simultaneously.
- **Genome-Edited Resistant Varieties:** Developed using CRISPR for durable pest resistance.

Collaborations among universities, startups, and farmers will define the next decade of sustainable horticulture.

25. Conclusion

Protected cultivation has become an indispensable pillar of modern agriculture, ensuring food security, high productivity, and resource efficiency. Yet its success relies on one crucial factor—**effective pest and disease management**.

An integrated approach that emphasizes prevention, early detection, biological dominance, and selective chemical use is the key to sustainability. The inclusion of digital tools, nanotechnology, and eco-engineering further enhances precision and resilience.

As India and the world move toward carbon-neutral, residue-free horticulture, smart IPM systems will lead the way. Empowering farmers with knowledge, access to bio-solutions, and real-time data will transform greenhouses into **intelligent bio-ecosystems**—productive, sustainable, and environmentally sound.

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Biological and Ecological strategies for Pest and Disease control in Tuber vegetable crops

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Tuber vegetable crops, including potato (*Solanum tuberosum*), sweet potato (*Ipomoea batatas*), yam (*Dioscorea* spp.), cassava (*Manihot esculenta*), and taro (*Colocasia esculenta*), play a vital role in ensuring global food and nutritional security. However, their productivity is significantly limited by a wide range of pests and diseases such as late blight, bacterial wilt, sweet potato weevil, and tuber moths, which cause substantial yield and quality losses. Conventional pest control methods relying heavily on chemical pesticides have led to resistance development, environmental pollution, and health hazards. In response, biological and ecological strategies are gaining prominence as sustainable alternatives for pest and disease management in tuber crops. Biological control utilizes beneficial microorganisms such as *Trichoderma*, *Bacillus*, *Pseudomonas*, and *Beauveria* to suppress pests and pathogens through mechanisms like competition, antibiosis, and induced systemic resistance. Ecological approaches including crop rotation, intercropping, mulching, use of resistant varieties, and habitat manipulation promote natural enemy activity and improve soil health. Integrating these methods into an Integrated Pest and Disease Management (IPDM) framework minimizes the dependence on synthetic inputs, enhances biodiversity, and maintains ecological balance. Emerging technologies such as endophytic biocontrols, RNA interference (RNAi), CRISPR-based gene editing, and nano-biopesticides further strengthen sustainable pest management. This chapter highlights the role of biological and ecological strategies in achieving resilient and eco-friendly tuber crop production systems. By combining scientific innovation with traditional ecological knowledge, these approaches ensure long-term productivity, food safety, and environmental sustainability.

Keywords: Tuber crops, Biological control, Ecological management, Sustainable Agriculture, Soil health and Biopesticides.

Introduction: Tuber crops such as potato (*Solanum tuberosum*), sweet potato (*Ipomoea batatas*), yam (*Dioscorea* spp.), cassava (*Manihot esculenta*), and taro (*Colocasia esculenta*) are among the most important food crops which serve as both staple and supplementary foods across the world (FAO, 2022).

They are cultivated extensively across tropical and subtropical regions and form a major component of smallholder farming systems. Tuber crops are a crucial source of carbohydrates, dietary fibre, vitamins (notably vitamin C and B6), minerals (especially potassium and iron), and bioactive compounds such as phenolics and carotenoids that contribute to human health and nutrition (Shekhar *et al.*, 2015). Globally, potatoes and cassava rank among the top five food crops in terms of production and caloric contribution (FAO, 2022).

Despite their significance, the productivity and quality of tuber crops are severely constrained by biotic stresses, primarily insect pests, fungal and bacterial pathogens, viruses, and nematodes (Manjunath and Prabhakar, 2019). Yield losses due to pests and diseases can reach up to 60–80% in the absence of effective management (Sharma and Thakur, 2022). The conventional reliance on synthetic pesticides and fungicides has contributed to pest resistance, pesticide residues, and ecological imbalance, necessitating sustainable alternatives (Ploetz and Pegg, 2021).

Traditionally, pest and disease management in tuber crops has relied heavily on chemical control measures such as synthetic pesticides and fungicides (Sharma and Thakur, 2022). Although these methods provide short-term effectiveness, their long-term consequences which includes pest resistance, pesticide residues in food, non-target organism toxicity, and environmental degradation that raise serious ecological and health concerns (Ploetz and Pegg, 2021). Moreover, the extensive use of agrochemicals disrupts soil microbial diversity and beneficial insect populations, ultimately reducing the resilience of farming systems (Gurret *et al.*, 2017).

In response to these challenges, there is a global shift toward biological and ecological approaches that harness natural processes for pest and disease management. Biological control utilizes beneficial microorganisms (e.g., *Trichoderma*, *Bacillus*, *Pseudomonas*, and *Beauveria*) to suppress pathogens and pests through mechanisms such as competition, antibiosis, parasitism, and induction of host resistance (Harman, 2006; Karthikeyan *et al.*, 2018). Ecological strategies, on the other hand, involve manipulating the agroecosystem through crop diversification, habitat management, and organic amendments which enhance natural enemy populations and reduce pest outbreaks (Gurr *et al.*, 2017; Singh and Devi, 2020).

These biological and ecological strategies form the cornerstone of Integrated Pest and Disease Management (IPDM) in tuber crops, aligning with the broader goals of sustainable and climate-resilient agriculture (Pretty *et al.*, 2020). By minimizing the use of synthetic inputs, these approaches help conserve biodiversity, improve soil health, and ensure food safety. In addition, the use of bioagents and ecological practices can significantly lower production costs while maintaining environmental integrity and long-term productivity (Sharma and Thakur, 2022).

The present chapter aims to review and synthesize current knowledge on biological and ecological strategies for pest and disease control in tuber vegetable crops, highlighting the potential of integrating these approaches into holistic IPDM frameworks. It discusses key biological control agents, ecological principles, and field-based practices that support sustainable tuber crop protection.

Major Pests and Diseases of Tuber Crops

1. Major Insect Pests

- a) **Potato tuber moth (*Phthorimaea operculella*):** This pest attacks both foliage and tubers, causing tunnels and secondary infections during storage. Losses may exceed 40% in unprotected conditions (Rondon, 2010).
- b) **Sweet potato weevil (*Cylas formicarius*):** Considered the most destructive pest of sweet potato, larvae bore into tubers, producing bitter compounds such as ipomeamarone that render tubers unmarketable (Nottingham and Kays, 2002).
- c) **Taro leaf beetle (*Papuacum bilineatus*):** A major defoliator of *Colocasia* and *alocasia*, it causes extensive leaf damage and yield reduction (Ram *et al.*, 2017).
- d) **Yam beetles (*Heteroligus spp.*):** These soil-inhabiting pests damage yam tubers by boring holes, leading to tuber rot and reduced market value (Emehute *et al.*, 2020).

2. Major Diseases

1. **Late blight (*Phytophthora infestans*):** The most devastating disease of potato, capable of destroying entire crops under favourable humid conditions (Fry, 2016).
2. **Bacterial wilt (*Ralstonia solanacearum*):** A soil-borne disease affecting potato, sweet potato, and cassava, causing severe wilting and tuber browning (Genin, 2010).
3. **Viral infections:** Diseases such as Potato virus Y (PVY) and sweet potato feathery mottle virus (SPFMV) are transmitted by aphids and reduce yield and quality (Karyeija *et al.*, 2000).
4. **Tuber and storage rots:** Pathogens including *Fusarium oxysporum*, *Pythium spp.*, and *Rhizoctonia solani* cause pre- and post-harvest losses (Singh and Devi, 2020).

3. Biological Control Strategies

3.1 Use of Microbial Bioagents

Biological control involves the use of beneficial microorganisms or their metabolites to suppress pests and pathogens.

3.1.1 Bacterial Biocontrol Agents

- *Bacillus subtilis* and *Pseudomonas fluorescens* suppress *Ralstonia solanacearum* and *Fusarium oxysporum* through the production of antibiotics (iturin, surfactin), siderophores, and enzymes such as chitinases and glucanases (Karthikeyan *et al.*, 2018).

- *Bacillus thuringiensis* (Bt) produces delta-endotoxins effective against *Phthorimaea operculella* larvae in potato fields and storage (Rondon, 2010).

3.1.2 Fungal Biocontrol Agents

- **Trichoderma spp.** (e.g., *T. harzianum*, *T. viride*, *T. asperellum*) are well-known antagonists of soil-borne pathogens, operating through mycoparasitism, competition, and induction of systemic resistance in host plants (Harman, 2006).
- *Trichoderma*-based formulations are extensively used for managing *Rhizoctonia solani* and *Sclerotium rolfsii* in potato and taro (Sharma and Thakur, 2022).

3.1.3 Entomopathogenic Fungi

- *Beauveria bassiana* and *Metarhizium anisopliae* infect larvae and adults of *Cylas formicarius* and yam beetles, reducing pest populations in the field (Ekesi *et al.*, 2011).

3.1.4 Nematode Antagonists

- *Paecilomyces lilacinus* and *Pochonia chlamydosporia* parasitize eggs and juveniles of *Meloidogyne incognita*, effectively reducing root-knot nematode infestation in sweet potato and yam (Kerry, 2000).

4. Ecological and Cultural Management Strategies

4.1 Crop Rotation and Intercropping

Crop rotation with cereals, legumes, or non-host plants disrupts pest life cycles and improves soil health. For instance, rotating potato with maize or cowpea reduces bacterial wilt incidence (Pradhan *et al.*, 2019). Intercropping sweet potato with maize, beans, or groundnut has been shown to suppress sweet potato weevil infestation through spatial interference and altered pest behavior (Smit *et al.*, 2020).

4.2 Soil and Water Management

Maintaining optimal soil moisture and drainage is crucial to prevent tuber rots and bacterial wilt. Incorporation of organic matter enhances soil microbial activity and suppresses pathogens through competitive exclusion (Karthikeyan *et al.*, 2018).

4.3 Resistant Varieties

The use of resistant or tolerant varieties offers the most cost-effective and sustainable approach.

- Potato varieties like ‘Kufri Jyoti’, ‘Kufri Bahar’, and ‘Kufri Himalini’ exhibit partial resistance to late blight and bacterial wilt (CIP, 2020).
- Sweet potato varieties such as ‘Sree Kanaka’ and ‘Sree Nandini’ are resistant to *Cylas formicarius* (CTCRI, 2021).

4.4 Mulching and Trap Crops

Organic mulches (straw, crop residues) conserve soil moisture, suppress weeds, and reduce tuber exposure to insects like the potato tuber moth. Trap crops such as mustard, marigold, or cowpea act as pest sinks, attracting pests away from the main crop (Singh and Devi, 2020).

4.5 Habitat Manipulation

Ecological engineering enhances natural enemy populations through habitat diversification. Planting flowering strips with marigold, coriander, or sunflower provides nectar and refuge for beneficial insects like ladybird beetles, lacewings, and parasitoid wasps (Gurr *et al.*, 2017).

5. Integrated Pest and Disease Management (IPDM)

An IPDM framework integrates biological, cultural, and ecological methods for sustainable crop protection (Ploetz and Pegg, 2021).

Key IPDM Components in Tuber Crops:

- a) **Seed and Planting Material Treatment** – Treating seed tubers or vines with *Trichoderma harzianum* or *Pseudomonas fluorescens* before planting reduces initial inoculum levels (Karthikeyan *et al.*, 2018).
- b) **Field Sanitation** – Removing infected debris and volunteer plants minimizes disease carry-over.
- c) **Monitoring and Trapping** – Using pheromone traps for *Cylas formicarius* helps monitor and mass-trap weevils (Nottingham and Kays, 2002).
- d) **Judicious Irrigation** – Avoiding waterlogging reduces bacterial and fungal infection risks.
- e) **Bioagent Integration** – Combining *Trichoderma* and *Bacillus* inoculants enhances synergistic effects and tuber health (Sharma and Thakur, 2022).

Field studies by CIP (2020) demonstrated that integrating resistant potato varieties, *Trichoderma*-based biopesticides, and proper rotation reduced late blight severity by 60% and increased yield by 25%.

6. Role of Organic Amendments and Biofertilizers

Organic amendments improve soil health, suppress pathogens, and enhance nutrient cycling (Maderet *et al.*, 2002).

- **Vermicompost enriched with *Trichoderma harzianum*** enhances suppressiveness against *Rhizoctonia solani* (Sharma and Thakur, 2022).
- **Biofertilizers** such as *Azospirillum*, *Azotobacter*, and phosphate solubilizing bacteria (PSB) promote plant growth and enhance rhizospheric microbial diversity (Bhattacharyya and Jha, 2012).

Such amendments indirectly suppress pathogens by stimulating beneficial microbial communities and improving soil enzyme activity.

7. Future Prospects

The integration of biotechnology with ecological pest management opens new opportunities for sustainable tuber crop protection.

- **Endophytic microorganisms** offer dual functions—promoting growth and suppressing pathogens (Santoyo *et al.*, 2016).
- **RNA interference (RNAi)** and **CRISPR-Cas** gene-editing tools can be used to develop pest- and disease-resistant cultivars (Koch and Kogel, 2014).
- **Nano biopesticides** and microbial consortia may improve formulation stability and field efficacy (Kumar *et al.*, 2023).
- Farmer field schools and participatory extension programs can accelerate adoption of biological control practices (Pretty *et al.*, 2020).

Conclusion: Biological and ecological strategies are the cornerstone of sustainable pest and disease control in tuber vegetable crops. They combine nature-based solutions such as bioagents, resistant cultivars, and habitat management to maintain ecological balance and reduce reliance on synthetic pesticides. Adoption of these integrated, ecosystem-based approaches ensures enhanced productivity, resilience, and environmental safety and supporting global goals for sustainable agriculture and food security.

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Natural Resource Management through Agroecological Practices for Climate Resilience

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Agriculture is fundamental to global food security and supports the livelihoods of millions of people worldwide. However, the sector faces growing challenges due to climate change, shrinking natural resources, and the rising demand for food. Increasing occurrences of floods, droughts, and other climate extremes are further threatening agricultural productivity and stability. Without timely and effective adaptation strategies, these pressures could severely impact the ability of agriculture to meet future food needs. In this context, agroecology has emerged as a promising approach to build sustainable and resilient agricultural systems capable of addressing these challenges.

Agroecological practices have demonstrated strong potential in mitigating the adverse effects of climate change. Techniques such as contour hedgerows, shade trees, bench terraces, contour bunds, bamboo fencing, hedgerow intercropping, integrated agroforestry systems, residue recycling, improved tillage practices, natural farming, crop diversification, mulching, and crop–livestock integration play a crucial role in reducing soil erosion, enhancing carbon sequestration, conserving water, lowering on-farm pollution, maintaining biodiversity, improving soil health, and ensuring economic profitability for farmers. To fully harness these benefits, it is important to move beyond simplified production systems toward more complex, integrated approaches that consider local contexts and foster collaboration across scales and sectors. Such integration creates an enabling environment for agroecological practices to drive climate-resilient and sustainable agricultural development.

Key words: Agroecology, climate resilience, natural resource management

Introduction

Nature encompasses all biotic, abiotic, and social elements that sustain life, with natural resources—land, water, air, forests, minerals, wildlife, and humans—supporting human well-being. Sustainable development requires conserving these resources for current and future generations, as unchecked exploitation has caused severe ecological degradation. Agriculture, once in harmony with nature, has become increasingly exploitative through mechanization and chemical use.

Natural Resource Management (NRM) is essential for climate mitigation and adaptation, promoting sustainable use of soil, water, and biodiversity, enhancing ecosystem resilience, and reducing carbon emissions. Integrating NRM with climate strategies ensures long-term sustainability, strengthens community adaptation, and supports resilient agriculture and food security.

Agriculture and Climate Change

Agriculture is crucial for global food security and livelihoods but faces increasing challenges from climate change, resource scarcity, and population growth. Rising temperatures, erratic rainfall, prolonged droughts, intense storms, and sea-level rise threaten water and land resources, crop and livestock productivity, and farmer livelihoods (Gemeda et al., 2023). Impacts include reduced water availability, soil degradation, lower yields, increased pest and disease incidence, and socio-economic stresses such as food insecurity and debt. Agroecology offers a sustainable and resilient approach to mitigate these risks and adapt agricultural systems to changing climatic conditions (Martini et al., 2020).

Agroecology: A Solution

Agriculture is both a major driver of climate change and one of its primary victims, requiring adaptive and sustainable solutions. Agroecology integrates ecological, economic, and social principles to enhance the resilience of smallholder farming systems (FAO, 2016). It promotes context-specific, co-created solutions through collaboration between scientists, farmers, and local stakeholders rather than relying on externally imposed methods. Core principles such as diversification, recycling, and stronger producer–consumer connections guide these transitions. Agroecological practices-like contour hedgerows, shade trees, and landscape-level tree cover-help reduce soil erosion, regulate temperature, improve nutrition, and support groundwater recharge (Sinclair et al., 2019). Beyond environmental benefits, agroecology strengthens livelihoods by improving resource management, income stability, and food security.

What is Agroecology?

The concept of agroecology emerged in the early 20th century as the application of ecological methods to agriculture. Odum (1969) introduced the idea of the “agroecosystem,” while Altieri (1987) expanded agroecology to encompass social and cultural knowledge systems. Altieri later defined it as the “application of ecological concepts and principles to the design and management of sustainable agroecosystems.” Similarly, the FAO (2016) emphasizes that agroecological innovations apply principles such as recycling, resource efficiency, diversification, and synergy to strengthen interactions among plants, animals, humans, and the environment for improved food security and nutrition.

Principles of Agroecology

1. **Resource Efficiency:** Use local renewable resources, recycle nutrients, and minimize external inputs.

2. **Soil and Animal Health:** Enhance soil fertility and structure, and ensure livestock well-being.
3. **Biodiversity and Synergy:** Maintain diverse species and promote beneficial ecological interactions.
4. **Economic and Social Sustainability:** Diversify farm income, support fair trade, and ensure nutritious, culturally relevant food systems.
5. **Knowledge and Participation:** Foster collaborative learning, farmer-to-farmer exchange, and inclusive decision-making.
6. **Governance and Connectivity:** Empower farmers as stewards of land and resources while strengthening producer–consumer links.

How agroecology enhances climate resilience:

Meta-analyses show that agroecology enhances climate resilience by improving soil health, promoting biodiversity, and diversifying species and genetic resources. It also aids climate mitigation through

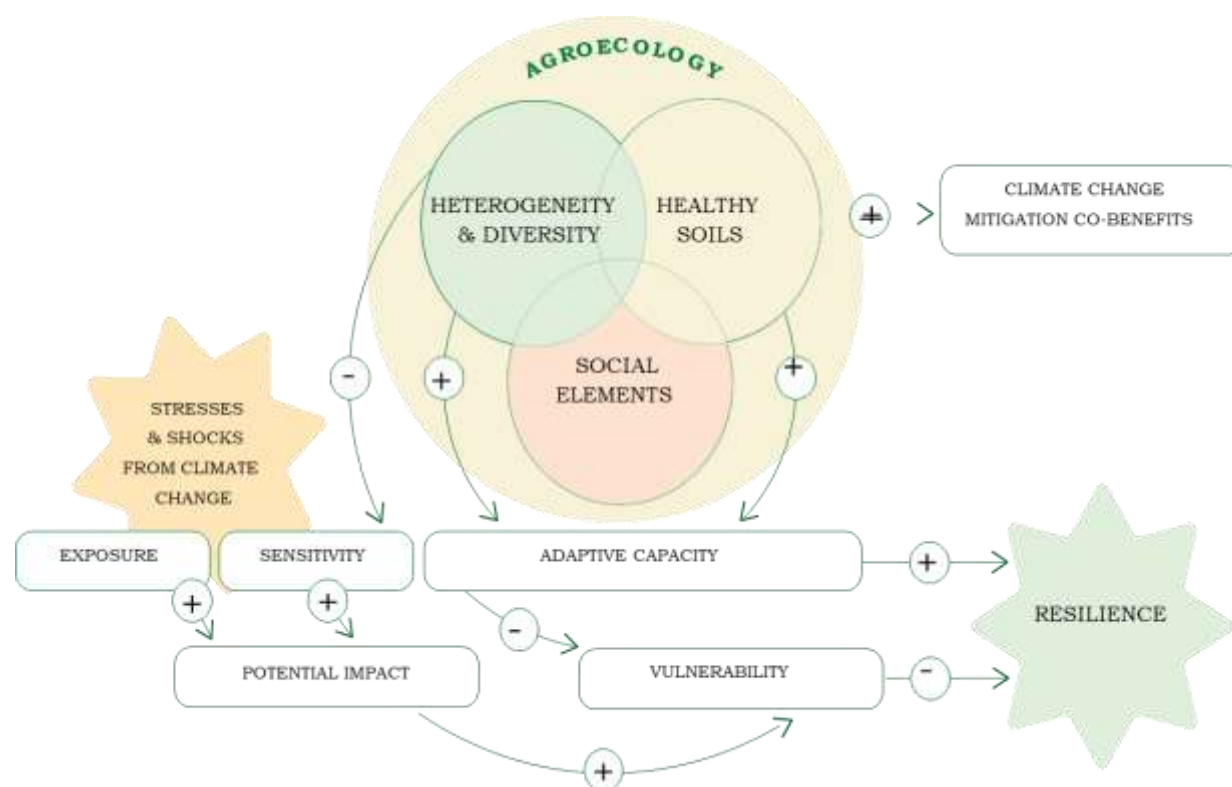


Fig 1. Agroecology increases climate resilience Source: FAO, 2020

carbon sequestration and reduced synthetic fertilizer use. Institutional support, participatory learning, and farmer-to-farmer knowledge sharing are key to advancing agroecological practices and strengthening locally relevant innovation systems.

Key On-farm Features of Agroecology

Agroecological farms aim to enhance productivity, sustainability, and resilience while conserving soil, water, and biodiversity, strengthening ecological interactions, and reducing dependence on external inputs. Key practices include diverse crop rotations, cover cropping,

multicropping, agroforestry, crop-livestock integration, and use of green manures to improve soil fertility, nutrient cycling, and ecosystem synergy. Techniques such as natural irrigation, reduced tillage, and integrated pest and pollinator management support resource conservation and ecological balance. Emphasis is placed on local food production, prioritizing nutrition, food security, and the multifunctional roles of agriculture.

Categories of agroecological practices contributing to climate adaptation

Some of the categories of agroecological practices contributing to climate adaptation are listed below (Sinclair et al. 2019)

Category I: Prevention of land degradation

Agroecological Practice	Climate Adaptation Benefits
▪ Rehabilitation and conservation of vegetation • Hedges • Agroforestry • Conservation of local tree species	• Soil fertility maintenance • Increased vegetation cover • Forage production • Improved water retention • Local biodiversity conservation • Crop protection against extreme climate events • Additional production
▪ Erosion control • Zai holes • Half moons • Trenches • Stone bunds • Bench terracing • Contour bunding • Agricultural and silvo-pastoral benches • Grass strips between crop lines	• Soil fertility improvement • Rehabilitation of degraded (crop) land • Increased vegetation cover • Improved water retention and infiltration • Decreased risk of crop loss during temporary drought in cropping season • Possible off-season production

Category II: Improving soil health

▪ Use of organic fertilizers and material • Crop residues compost • Biogas slurry • Cover crops • Green manure • Mulching	• Soil fertility improvement • Improved water retention • Renewable energy production • GHG emissions reduction (only bio-digester) • Weed control • Erosion control

Introduction of soil-improving woody plants as windbreak and/or hedgerow	• Soil fertility improvement • Legumes for nitrogen fixation • Improved water retention • Wind and water erosion control • Increased permanent vegetation cover • Additional productions

Category III: Better water management

▪ Water collection devices • Permeable stone dam • Sand dam • Mini dam • Weir • Pond Water reservoir • Collective water management	• Improved water retention • Water storage • Increased vegetation cover • Possible off-season production • Groundwater tables filling • Water supply improvement
▪ Drip irrigation	• Improved water use efficiency

Category IV: Diversifying production

▪ Introduction of new crops • Vegetables • Pulses • Castor oil plant • Moringa	• Risk reduction for overall agricultural production • Local diets improvement • Additional income • Resistance to drought (castor oil)
▪ Combination of crops • Wet and dry seasons crop rotation • Diversified crop rotation • Mix of cultivars • Cereal/cowpea intercropping	• Soil fertility improvement • Weed, pest and disease control • Increased crop diversity • Biodiversity enhancement • Risk reduction for overall agricultural production
▪ Introduction of animals • Small ruminants • Poultry • Beekeeping	• Diversification with animals • Risk and reduction for overall agricultural production

Category V: Adapting crop management

▪ Species and cultivar choice • Drought-tolerant crops • Shorter-term crop cultivars • Farmer local seed selection and dissemination	• Crop failure risk reduction • Higher resistance to pests and diseases • Additional income for local seed producers • Agrobiodiversity conservation
▪ Soil working adaptation • Minimum tillage • Zero tillage • Stubble mulch tillage	• Soil fertility improvement • Improved water retention • Early crop maturity

Category vi: Pest and disease control

▪ Compost enriched with <i>Trichoderma</i> ▪ Neem oil-based insecticide ▪ Biopesticides ▪ Natural predators	• Environmentally friendly pest and disease control • Pest attacks reduction • Cryptogamic diseases control

Category VII: Managing Livestock

▪ Feed improvement ▪ Crop residues as forage • Fodder trees (Sesbania, Acacia, Moringa) • Urea treatment of hay mineral supplements for dairy animals • Millet bran supplement ▪ Hay production	• Feed ration and feed quality improvement • Supplementary forage sources, especially during dry periods • Additional wood production
▪ Rotational grazing ▪ Regulation of animal movement • Reduced herd size • Collective grazing on village territory • Selection of drought-adapted cattle breeds	• Overgrazing limitation • Soil fertility improvement • Increased vegetation cover • Biodiversity enhancement • Improved management of forage resources, animal drought resistance

Prominent systems and practices involved in Agroecology

Agroecology integrates diverse farming systems and practices to balance productivity with ecological sustainability. **Organic farming** maintains soil fertility using manures, crop residues, and biofertilizers while avoiding synthetic inputs. **Natural farming**, including Zero-Budget Natural Farming (ZBNF) in India, relies on local resources and minimal inputs, enhancing soil microbial activity and reducing

costs. **Biodynamic farming** treats the farm as a self-sustaining organism, using composts, crop rotations, and natural pest control. **Conservation agriculture** applies minimum tillage, permanent soil cover, and crop diversification to improve soil health, water retention, and climate resilience. **Agroforestry** integrates trees, crops, and livestock to conserve moisture, enhance nutrient cycling, and diversify income. **Integrated farming systems (IFS)** combine crops, livestock, fisheries, and horticulture for resource recycling and risk reduction. **Crop diversification** through rotations, intercropping, and mixed cropping improves resilience, pest management, soil health, and income stability. Collectively, these systems promote ecological integrity, optimize resources, and build climate-resilient, low-cost, and sustainable agriculture.

Impact of agroecological practices across diverse agro-ecological regions

Agroecological approaches integrate ecological principles into farming systems to enhance productivity, resilience, and environmental sustainability. The case studies compiled across diverse agro-ecological regions of India and abroad provide strong empirical evidence supporting the role of agroecological interventions in improving soil health, crop yield, biodiversity, and climate resilience. In the North-Eastern Himalaya, indigenous and mechanical soil conservation measures—such as contour bunding and bench terracing—significantly reduced soil erosion (by 80–98%) and enhanced soybean yield, with contour bunds proving the most effective for both soil conservation and economic return (Ray et al., 2021). In the Eastern Ghats, hedgerow intercropping with *Gliricidia* and *Leucaena* reduced runoff (by 40%) and soil loss (by 35%) while improving SOC sequestration (up to 1.7 Mg C ha⁻¹ yr⁻¹) and millet yield (Hombegowda et al., 2019).

Agroforestry systems based on *Phalsa*, *Karonda*, and *Moringa* increased system productivity, water-use efficiency, and carbon sequestration under semi-arid conditions, demonstrating superior sustainability yield indices (Rathore et al., 2022). Conservation tillage and residue retention in the Indo-Gangetic Plains enhanced soil temperature moderation, earthworm populations, and nutrient recycling efficiency (Mishra et al., 2021).

Incorporation of crop residues in jute-based systems markedly improved soil quality indices (up to 0.63) and system productivity (Kumar et al., 2021). Similarly, natural farming practices involving jeevamrit, ghanjeevamrit, and mulching improved yields and microbial activity in Himachal Pradesh. Studies from the UK confirmed that minimum and no-tillage enhanced soil moisture and organic matter (Alskaf et al., 2021). Mulching practices in NEH India increased soil organic carbon and water-holding capacity (Ngangom et al., 2020). Integrated crop–livestock systems (Shanmugam et al., 2024) and rainwater conservation on degraded ravines (Ali et al., 2020) further demonstrated increased productivity, nutrient cycling, and GHG mitigation. Overall, these studies reinforce that agroecological

interventions significantly improve soil–plant–water relations, enhance ecosystem services, and promote sustainable agricultural intensification under varying climatic and edaphic conditions.

Future Thrust

The future thrust of agroecology focuses on mainstreaming its principles into policies and extension systems, strengthening participatory research across diverse agro-climatic zones, and building the capacity of farmers and rural youth through training programs. Emphasis is placed on ecosystem restoration through soil carbon enrichment, biodiversity conservation, and pollution-free farming, while integrating modern technologies like remote sensing, precision irrigation, and decision-support tools for efficient management. Supportive measures include developing value chains, certification systems, and market access for sustainable products, along with policy incentives, credit, and insurance mechanisms. Collaboration at local and global levels among research institutions, farmer organizations, and policymakers is key to advancing climate-resilient and sustainable agroecological systems.

Conclusion: Agroecology offers a holistic approach to sustainable agriculture by integrating ecological principles with traditional and innovative practices to optimize productivity, strengthen ecosystem resilience, and conserve natural resources. It transforms farms into self-regulating systems through efficient management of crops, soil, water, livestock, and biodiversity. Across organic, biodynamic, conservation, and diversified systems, agroecology emphasizes nutrient recycling, minimal external inputs, and ecosystem service enhancement. Practices like intercropping, cover cropping, green manuring, agroforestry, and livestock integration promote soil fertility, water-use efficiency, climate adaptation, and reduced greenhouse gas emissions. Beyond environmental benefits, agroecology supports smallholder livelihoods, diversifies income, improves food security, and fosters local knowledge and community-based innovation, making it a pathway toward ecologically balanced, economically viable, and socially equitable agriculture.

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Optimising Drip Irrigation and Biodegradable Mulches for Enhanced Resource Use Efficiency and Sustainable Sweet Corn Production in Lateritic Soils

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The research focused on a sustainable cultivation strategy for sweet corn in the lateritic soils of the Konkan region, characterized by low fertility, high acidity, and poor moisture retention. Conducted during the *rabi* season using a split plot design, the study evaluated effects of varying irrigation levels through drip systems and different mulching materials, including traditional and biodegradable options. The findings revealed that full irrigation through drip proved most effective in maintaining optimal soil moisture, enhancing plant growth, and improving nutrient uptake and yield. Among the mulching treatments, wool mulch enriched with micronutrients emerged as the most beneficial, promoting superior crop performance while improving soil microbial activity, organic carbon content, and residual nutrient status, thereby contributing to long-term soil health. Integrated use of full irrigation with nutrient-enriched wool mulch demonstrated the most synergistic effect, resulting in the highest yield, quality, and profitability, while ensuring sustainability in resource use. Overall, the study established that precision irrigation combined with biodegradable nutrient-enriched mulch provides a viable, eco-friendly approach for enhancing productivity and sustainability of sweet corn cultivation in lateritic soils.

Keywords: Sweet corn, lateritic soils, drip irrigation, biodegradable mulch, sustainable cultivation

Introduction

The cultivation of sweet corn offers significant potential for agricultural transformation, particularly in regions like the Konkan, where it provides both economic and nutritional benefits and meets the rising urban demand for the crop (Game et al., 2017). However, lateritic soils characterized by low fertility, high acidity, and poor water retention—pose challenges to sustainable cultivation. Addressing these challenges requires innovative practices rooted in the principles of precision agriculture and soil and water conservation (Kader et al., 2017).

Efficient water management through optimized irrigation levels is crucial, especially in water-limited regions, as it minimizes wastage and maintains optimal water availability for plant development (Fernández and Alcon, 2014). Mulching techniques, on the other hand, offer a cost-effective approach to conserving soil moisture and modifying the microclimate, thereby enhancing crop growth and yield (Sharma and Kumar, 2020).

This research focuses on evaluating the combined effects of varying irrigation levels through drip irrigation and the use of different mulch types, including biodegradable options, on sweet corn production in lateritic soils. The objectives align with promoting eco-friendly, resource-efficient farming strategies by determining optimal irrigation scheduling and assessing the feasibility of sustainable mulching materials that conserve resources and improve soil health.

Methodology

The field experiment was conducted during the *rabi* season (2024–25) at the Agronomy Farm, College of Agriculture, Dapoli, Maharashtra, India, on lateritic soil of sandy clay loam texture. The soil was acidic in nature, with moderate organic carbon and low available nitrogen and phosphorus content.

A Split Plot Design (SPD) with three replications was used, comprising eighteen treatment combinations. The main plots consisted of three irrigation levels (100%, 80%, and 60% ET_c through drip irrigation), while the subplots included six mulching treatments, various biodegradable and conventional options such as wool mulch, wool mulch enriched with micronutrients, silver-black polythene mulch, paddy straw mulch, wool–coir–jute blend, and an unmulched control.

Irrigation scheduling was based on crop evapotranspiration (ET_c) using daily weather data (Allen et al., 1998). Water soluble fertilizers were uniformly applied through fertigation. The sweet corn variety used was ‘Sugar-75’.

Innovations in Water and Soil Conservation

Irrigation Levels and Soil Moisture Availability

Precision irrigation aims to deliver optimal water to crops while minimizing losses. In this study, full irrigation through drip (100% ET_c) maintained the best soil moisture conditions throughout the crop period, ensuring superior plant water status. Intermediate irrigation (80% ET_c) maintained moderate soil moisture, while deficit irrigation (60% ET_c) recorded the lowest values and reflected limited water availability.

Plants under full irrigation consistently showed better relative water content and turgidity compared to reduced irrigation levels, confirming that adequate irrigation via drip ensures optimal physiological functioning and sustained growth in lateritic soils.

Role of Biodegradable Mulches in Soil Health and Water Retention

Mulching significantly influenced soil characteristics and biological activity. Polythene mulch maintained the highest soil moisture, followed closely by wool mulch enriched with micronutrients and pure wool mulch (Zhang et al., 2019). In contrast, the unmulched control plots exhibited the lowest moisture retention.

Biodegradable mulches offered distinct long-term benefits by improving microbial activity and enhancing soil fertility (Bandopadhyay et al., 2018). The wool mulch with micronutrients supported

the highest bacterial and fungal populations, indicating a more favourable soil microenvironment. This enhanced biological activity facilitated nutrient cycling and soil structure improvement, unlike synthetic mulch, which showed limited microbial proliferation.

The wool mulch enriched with micronutrients also resulted in superior organic carbon content after harvest, indicating its contribution to soil organic matter buildup (Abdallah et al., 2019). The gradual decomposition of wool mulch served as a slow-release organic fertilizer, improving soil health and nutrient availability.

Impact on Sweet Corn Growth and Productivity

Vegetative Growth and Dry Matter Accumulation

Plant height, leaf area index (LAI), and dry matter production were all positively influenced by higher irrigation levels. The best vegetative growth was recorded under full irrigation, followed by moderate irrigation, while the lowest values were under deficit irrigation.

Among the mulches, wool mulch enriched with micronutrients exhibited superior performance in terms of plant height, number of functional leaves, leaf area, and total dry matter accumulation, followed by pure wool mulch. The unmulched plots consistently showed inferior growth performance. The combination of full irrigation with wool mulch enriched with micronutrients recorded the best growth parameters, indicating strong synergistic effects between optimal moisture availability and nutrient-enriched biodegradable mulch.

Yield Attributes and Final Yield

Sweet corn yield attributes such as cob length, girth, weight, and grain yield were significantly affected by both irrigation and mulching practices.

Full irrigation produced the highest green cob, stover, and total biomass yields, outperforming the reduced irrigation levels. Among the mulches, wool mulch enriched with micronutrients recorded the best performance for all yield components, followed by pure wool mulch and silver-black polythene mulch. The unmulched control recorded the lowest yields.

The interaction between full irrigation and wool mulch enriched with micronutrients resulted in the overall best yield performance, demonstrating a clear synergistic enhancement in growth and productivity due to efficient water supply and improved soil environment.

Weed Management

Mulching effectively reduced weed density and dry matter accumulation (Mahajan et al., 2007). Silver-black polythene mulch recorded the lowest weed population, closely followed by wool mulch enriched with micronutrients. The combination of deficit irrigation with polythene mulch further minimized weed emergence, suggesting that reduced soil moisture combined with a physical barrier effectively

suppresses weeds. Nonetheless, the combination of full irrigation and wool mulch with micronutrients also maintained effective weed control compared to the unmulched control plots.

Nutrient Dynamics and Soil Fertility Balance

Nutrient Uptake

Full irrigation enhanced the uptake of nitrogen, phosphorus, and potassium by sweet corn plants (Ibrahim et al., 2016). Similarly, wool mulch enriched with micronutrients facilitated the best nutrient uptake among the mulch treatments, followed by pure wool mulch. This improvement was attributed to better nutrient mineralization and microbial activity associated with the biodegradable mulch materials.

The combination of full irrigation and wool mulch enriched with micronutrients achieved the highest total nutrient uptake, confirming its efficiency in promoting nutrient availability and absorption.

Available Nutrient Status and Balance Sheet

Post-harvest soil analysis revealed higher available nitrogen and potassium under full irrigation, whereas moderate irrigation maintained balanced phosphorus levels. Among the mulch treatments, wool mulch with micronutrients maintained the best residual nutrient status, contributing to long-term soil fertility enhancement (Sarkar et al., 2021).

The combination of full irrigation with wool mulch enriched with micronutrients showed a positive nutrient balance, reflecting its sustainability and soil-conserving potential. In contrast, the control treatment under deficit irrigation exhibited nutrient depletion, indicating unsustainable nutrient removal from the soil system.

Quality Parameters and Economic Viability

Quality Studies

Irrigation and mulching treatments significantly influenced sweet corn quality attributes. Full irrigation produced grains with superior protein content, while slightly deficit irrigation promoted higher sugar accumulation (Ertek et al., 2013). Among the mulches, wool mulch with micronutrients yielded grains with the best sugar, protein, and chlorophyll content, followed by pure wool mulch. The combination of full irrigation and micronutrient-enriched wool mulch produced the best overall quality parameters.

Economic Analysis and Water Productivity

Economic analysis demonstrated that full irrigation combined with wool mulch enriched with micronutrients achieved the highest gross and net returns and the most favourable benefit-cost ratio. Although deficit irrigation improved economic water productivity due to reduced water input, the full irrigation and wool mulch combination provided the best balance between yield, profit, and sustainability.

Thus, while deficit irrigation can be suitable in water-scarce conditions, the full irrigation with biodegradable wool mulch enriched with micronutrients remains the most profitable and sustainable system overall (Kuscu et al., 2013).

Conclusion: A Blueprint for Sustainable Lateritic Agriculture

The findings of this study strongly support integrating optimized drip irrigation with biodegradable mulches as a sustainable strategy for sweet corn cultivation in lateritic soils.

Optimal Strategy

The combination of full irrigation through drip (100% ETc) with wool mulch enriched with micronutrients proved to be the most effective approach. This integrated method enhanced crop growth, yield, nutrient uptake, soil microbial activity, and quality parameters while ensuring maximum economic returns and profitability. The wool mulch, particularly when enriched with micronutrients, acted as a soil conditioner—improving organic carbon, microbial biomass, and moisture conservation.

Resource Efficiency

While full irrigation produced the best yield, moderate deficit irrigation improved economic water productivity, suggesting flexibility in irrigation management based on water availability. Nevertheless, the full irrigation with nutrient-enriched wool mulch combination achieved the best overall balance between productivity, profitability, and sustainability.

Final Insight

The integration of precision drip irrigation with advanced biodegradable mulching creates a self-regulating, resource-efficient ecosystem. Drip irrigation ensures precise water delivery to the root zone, while biodegradable mulches act as intelligent covers—reducing evaporation, stabilizing soil temperature, and gradually enriching the soil as they decompose. Together, these innovations ensure that every drop of water and nutrient contributes maximally to yield and profitability. This synergy transforms farming into a model of economic viability and environmental responsibility, paving the way for sustainable agricultural intensification in lateritic regions.

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Rearing of Silkworms from Chawki rearing to formation of cocoons

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Silkworm rearing (sericulture) is important for **production of silk**, a valuable natural fiber for the textile industry. Beyond this primary product, it offers significant **socio-economic and environmental benefits**, particularly in rural areas of developing countries. Silkworm rearing needs proper understanding of techniques involved in chawki and late age rearing. Chawki rearing is the specialized process of raising young silkworms during their first two instars, or stages, of life and late age rearing is raising of young silkworms from their III instars, or stages, of life to formation of cocoons.

A. Chawki Rearing

This initial period, lasting about 12 to 14 days, is crucial for producing healthy larvae that will yield a better silk. Due to the worms' delicate state and vulnerability to disease, this phase requires controlled environmental conditions and care.

Key requirements for chawki rearing

- ✓ Dedicated chawki garden: Silkworms in this stage require very tender, succulent, and high-protein mulberry leaves. A separate, well-maintained mulberry garden is managed to produce this specific quality of leaf.
- ✓ Specialized rearing house: A separate, well-ventilated rearing house is essential to maintain a stable microclimate and practice strict hygiene. It should be isolated from late-age rearing rooms.
- ✓ Optimal temperature and humidity: The environment must be carefully regulated to prevent moisture loss and inhibit the growth of pathogens.

First instar: A temperature of 27–28°C and relative humidity of 85–90%.

Second instar A temperature of 27°C and relative humidity of 85%.

- ✓ Sanitation: Impeccable hygiene is non-negotiable. This includes disinfecting the rearing room and all equipment and promptly disposing of diseased or dead worms to prevent contamination.

Specific Requirements for 100 DFL'S

Instar	Temperature	Humidity	R. Space Sq. ft	Vijetha gm	Leaves /Kg
First	23 – 24	80 - 85	9 – 36	50	5.9
Second	24 - 25	80 – 85	36 – 98	150	16

The chawki rearing process

1. Incubation and hatching:
 - ✓ Silkworm eggs are incubated under controlled temperature (25–26°C) and humidity (80%).
 - ✓ "Black boxing," a process of covering the eggs in a dark box for 1–2 days, synchronizes hatching when the eggs turn bluish.
2. Brushing: This is the process of transferring the newly hatched worms to rearing trays.
 - ✓ Once 80–90% of the eggs have hatched, the larvae are gently transferred with a soft brush or feather onto paraffin paper in the rearing trays.
 - ✓ Wrapping the trays with paraffin paper helps maintain the required humidity and prevents the leaves from drying out.
3. Feeding: Young silkworms are fed finely chopped, tender mulberry leaves.
 - ✓ First instar: Leaves are chopped into small, 0.5–1 cm pieces.
 - ✓ Second instar: The leaf size is increased to 2–4 cm pieces.
 - ✓ Feedings are provided frequently, 3–4 times per day

Leaves Requirement for 100 DFL'S Per DFL

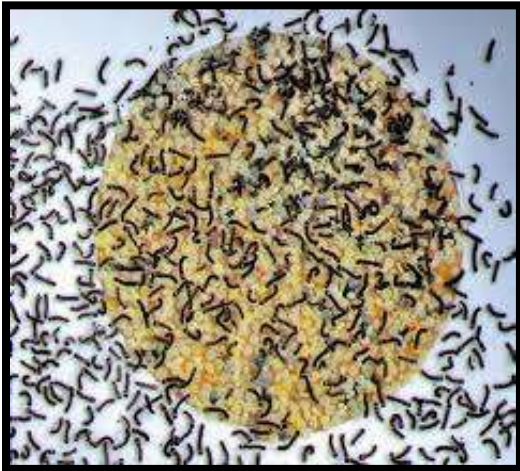
Instar	1 Day	2 Day	3 Day	4 Day	Total
First	1	1.8	2.5	0.6	5.9
Second	5	8	3	0	16

4. Bed cleaning: As leaf waste accumulates, it can become a breeding ground for pathogens.
 - ✓ The rearing bed is thinned and cleaned once during the first instar and once after the second moult.
 - ✓ A bed cleaning net is placed over the worms, and fresh leaves are added. The worms crawl up through the net to the fresh leaves, allowing the old litter to be discarded.
5. Moulting care: When the worms prepare to moult (shed their skin), they become inactive and stop feeding.
 - ✓ Feeding is stopped, and the rearing bed is thinned and dried by sprinkling slaked lime powder.
 - ✓ After the moult, feeding is resumed and a bed disinfectant, such as Vijetha, is applied to the larvae.

Commercial chawki rearing

Commercial Chawki Rearing Centres (CRCs) are specialized facilities that raise chawki worms under optimal conditions and supply them to farmers.

- Benefits for farmers: CRCs save farmers the labor and risk associated with the most vulnerable stage of the silkworm.
- Improved yield: Purchasing high-quality chawki worms from a CRC leads to a higher cocoon yield and better returns.
- Reduced losses: CRCs maintain strict hygiene and environmental controls, which significantly reduce the risk of crop failure due to disease.



Hatching



Feeding



B. (Late age Rearing III- V Instar)

Rearing a silkworm from the third instar involves a period of rapid growth and feeding, called "late-age rearing," which continues until the silkworms are ready to spin their cocoons. This stage is critical for producing high-quality silk

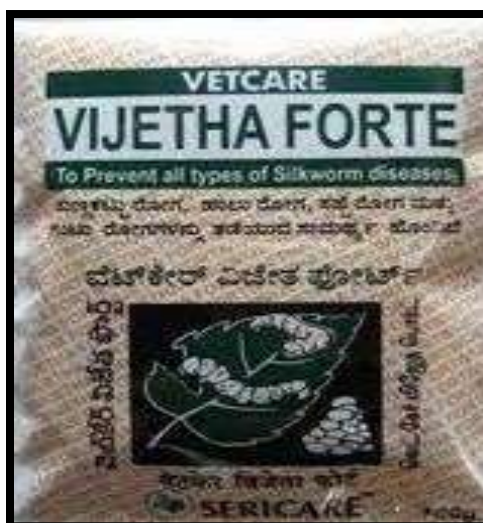
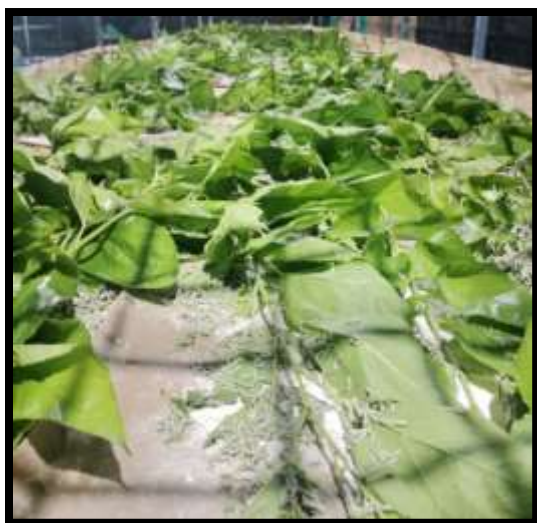
THIRD INSTAR

- ✓ This stage arises after completion of second moult and at these stage farmers receive silkworms from chawki centers
- ✓ Place the larvae on respective stands and provide adequate spacing
- ✓ Dust larvae with Vijetha powder on every alternate day
- ✓ Start Feeding leaves with branches to larvae

- ✓ Provide feeding twice a day
- ✓ Avoid leaves containing dust, yellow, hot and black leaves
- ✓ This stage last for **3 – 3.5 Days**

THIRD MOLT

- ✓ At this stage stop feeding
- ✓ Spray lime powder useful for drying of leaves on stands
- ✓ This molt occurs for 24 – 30 hours
- ✓ After completion of molt dust vijetha powder on larvae
- ✓ Maintain Temperature and humidity in shed



Third Instar larva feeding



FOURTH INSTAR

- ✓ After completion of third molt larva enters into 4th instar
- ✓ Provide spacing on stands
- ✓ Dust Vijetha powder on alternate day basis

- ✓ This stage last for 4 – 4.5 days
- ✓ Maintain temperature and humidity



FOURTH MOLT

- ✓ Stop feeding to Larvae
- ✓ Dust lime on larvae
- ✓ This molt last for 36 – 40 hours
- ✓ Maintain temperature and humidity

FIFTH INSTAR

- ✓ After completion of fourth molt larva enters 5th instar
- ✓ Provide spacing on stands
- ✓ Dust Vijetha powder on alternate day basis
- ✓ This stage last for 7 – 8 days
- ✓ After completion of feeding larvae starts cocoons formation.



Fifth Instar larva

100 DFL Specific Requirements

Stage	Temperature	Humidity	R. Space/ Sq. Ft	Vijetha/ Gm	Leaves/ Kg
Third	25 – 26	75 – 80	98 – 285	900	140
Fourth	26 – 27	70 – 75	325 – 525	1900	460
Fifth	27 - 28	65 - 70	1050	3000	2880

100 DFL FEEDING REQUIREMENTS/ KG

Stage	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Total
Third	25	50	50	15	00	00	00	140
Fourth	60	120	150	130	00	00	00	460
Fifth	220	310	460	560	700	410	220	2880

Molting

For rearing of silkworms knowledge and skill regarding identification and management practices during moulting plays an important role which makes study of moulting essential during rearing of silkworms. Silkworms undergo a process called moulting (ecdysis) in which they shed their old skin to allow for growth. Most commercial silkworm strains are tetra moulters, meaning they moult four times during their larval stage, resulting in five larval instars (developmental stages between moults). To identify a silkworm on molt one has to observe key behavioral and physical changes, including periods of inactivity, a swollen or raised head, and a tightening and darkening of its skin. A typical silkworm undergoes four molts during its larval stage, growing into a new, larger skin each time.

Physical signs of a silkworm preparing to molt

- ✓ Appearance of a new head: A new, larger head capsule can be seen forming underneath and behind the old, smaller one, making the head appear swollen or raised
- ✓ Tight, dark, or wrinkled skin: Before the molt, the silkworm's skin will become tight, dull, and dark. As the new skin forms underneath, the old skin may appear loose and wrinkled.
- ✓ Translucent body: For some strains, the body may appear translucent, particularly in the thorax region.
- ✓ Appearance of a spiracle index: Specialized sericulturists can identify the exact molting stage by observing changes in the appearance of the silkworm's spiracles.

Behavioral signs of a silkworm preparing to molt

- Stops feeding: A silkworm will completely stop eating mulberry leaves as it prepares to molt. This "rest period" can last for 18 to 24 hours.
- Becomes immobile and raises its head: The worm will become still and motionless, often raising its head in an upward position.
- Excretion of soft feces: Just before entering the final molt, a mature silkworm may excrete soft, moist feces before it stops feeding entirely.

Signs of a silkworm completing a molt

- Shedding of the old skin: You will see the discarded old skin, or *exuviae*, on the rearing bed. It is often a tiny, crumpled piece of skin with a head capsule attached.
- Becomes active again: Once the molt is complete, the worm becomes active and resumes eating.
- Body appears shiny: The body will look stout, shiny, and amber-colored after it has molted into its new skin.
- Broader mouthpart: After shedding its old head capsule, the worm's new mouthpart will be noticeably wider.

How to care for silkworms during molting

- Avoid disturbance: Do not disturb the silkworms when they are molting.
- Stop feeding: Stop providing leaves during the molting period. Resume feeding only when all worms have completed the molt.
- Keep the bed dry: Maintain a dry environment during the molt by dusting lime on the bed to reduce humidity.
- Ensure good ventilation: Proper airflow is important to keep the rearing bed from becoming damp





Silkworm on molt

Biotechnological Innovations in Plant Secondary Metabolite Production: In Vitro Strategies and Metabolic Engineering

1. Introduction

Plants possess an extraordinary biochemical capacity, synthesizing a vast and diverse array of organic compounds known as secondary metabolites (SMs). These molecules extend beyond the basic requirements of growth and reproduction (primary metabolism) to fulfill critical roles in the plant's long-term survival, including ecological interactions, defense mechanisms, and adaptation to environmental stresses. The immense commercial and therapeutic value of SMs is underscored by their diverse applications such as pharmaceuticals, agrochemicals, nutraceuticals, and industrial additives. The totality of biochemical processes within an organism is known as its metabolism, and metabolites are the small molecule intermediates and products. While primary metabolites are essential for immediate survival, the absence of SMs typically results in long-term impairment of an organism's survivability or fecundity. In modern industry, SMs serve as invaluable sources for a wide range of products, including pharmaceuticals, flavors, fragrances, and pesticides.

In recent years, the escalating global demand for Plant-Derived Medicinal Compounds (PDMCs) has prompted a critical shift in production methods. Conventional sourcing relies on extraction from native plant tissues, a process plagued by several disadvantages: low yields in native tissues, tissue-specific localization, seasonal and environmental dependency, and significant sustainability concerns leading to the overexploitation of wild populations. In response, in vitro culture technologies have emerged as controlled, reproducible, scalable, and sustainable platforms for the efficient biosynthesis of desired secondary metabolites.

2. Classification

Chemical Diversity of Secondary Metabolites

The classification of SMs can be ambiguous, as many intermediates overlap with primary metabolism, such as amino acids and sterols. However, SMs are broadly categorized into five major classes: alkaloids, terpenoids and steroids, fatty acid-derived compounds and polyketides, non-ribosomal polypeptides, and enzyme cofactors.

They are systematically classified based on criteria such as:

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- **Molecular Composition:** Containing components that include nitrogenous, sulfur-containing, aromatic, or glycosidic groups, which often dictate their pharmacological activity.

3. Commercial relevance of secondary metabolite production

The commercial significance of Plant-Derived Medicinal Compounds (PDMCs) is soaring, making the efficient production of secondary metabolites (SMs) crucial for the pharmaceutical sector. Conventional methods, which rely on extraction from native tissues, face critical limitations such as low yields, seasonal dependence, overexploitation issues, and the requirement for complex purification. The adoption of the *in vitro* system (cell and tissue culture) provides a superior alternative, offering a controlled, scalable, and sustainable platform for the reliable, year-round biosynthesis of these valuable compounds for medicinal and other commercial products.

4. *IN VITRO* CULTURE TECHNOLOGY FOR SECONDARY METABOLITE PRODUCTION

The core commercial relevance of secondary metabolite production lies in the pharmaceutical industry. The disadvantages inherent in conventional extraction methods—such as the necessity for complex purification and the vulnerability to seasonal variation—make them less efficient for large-scale production. *In vitro* systems overcome these limitations by allowing for constant, year-round production in a controlled environment, thus offering a reliable platform for the modern medicinal market.

The term "*in vitro*" (Latin for "in glass") denotes biological material cultured outside its natural environment. Plant cell and tissue cultures are powerful tools for the controlled production of SMs, provided the culture parameters are rigorously optimized.

Table – Baseline

Conditions for Plant Tissue Culture System

4.1. Callus Culture

Callus is an unorganized, pluripotent cell mass typically induced from explants (e.g., leaf segments, root tips) on a solid growth medium supplemented with auxins and cytokinins. This tissue is formed when cell differentiation is reversed, causing the explant to produce new, unspecialized meristematic cells. The most common media include Murashige and Skoog (MS) medium, with Gamborg's B5 and Linsmaier and Skoog (LS) media also frequently employed. Sterilization is a crucial practice, often involving a sequence of treatments like 70% ethanol and 0.1% sodium hypochlorite (NaOCl).

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A cell suspension culture is initiated by placing friable callus fragments into a sterile liquid medium under continuous agitation. This system promotes uniform cell growth and enhanced metabolite production, often exhibiting higher rates of cell division than callus culture.

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Organ cultures involve culturing a whole or part of a plant organ (e.g., shoot or root) to preserve its specific architecture and function, which is often crucial since SMs accumulate in specific organs (e.g., alkaloids in roots, flavonoids in leaves). *In vitro* organ cultures, particularly root and hairy root systems, replicate the natural biosynthetic environment more closely than undifferentiated callus or cell suspensions, often resulting in higher metabolite yields.

Hairy Root (HR) Culture is a specialized and highly effective organ culture technique. It is induced by infecting a plant explant with the soil-borne bacterium *Agrobacterium rhizogenes*, which causes hairy root disease. The bacterium transfers a segment of its Root-Inducing (Ri) plasmid's T-DNA into the plant genome. This T-DNA carries genes (*iaaM*, *iaaH*, *ipt*) that code for enzymes controlling auxin and cytokinin biosynthesis, which in turn induces the formation of rapidly growing, lateral, and *Agrobacterium*-free HRs. HR cultures are favored because of their:

- **Genetic and Biochemical Stability:** They maintain a consistent metabolite profile over long-term subcultures.

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5. Advanced Strategies to Enhance Metabolite Yield

To maximize the output of valuable secondary compounds, several advanced strategies are integrated with *in vitro* culture protocols.

5.1. Elicitation: Stress-Induced Biosynthesis

Elicitation is a strategy where small quantities of stress factors, known as elicitors, are applied to a living system to induce or improve the biosynthesis of specific compounds. This process taps into the plant's natural defense mechanisms to redirect metabolic flux.

Elicitors are classified based on their origin:

- Abiotic Elicitors: Non-biological factors, including physical stresses (light, salinity, temperature), chemical agents (heavy metals like Ni, Ag, Co), and hormones, which modulate key metabolic pathways.
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The success of elicitation hinges on optimizing parameters like elicitor concentration, duration of exposure, and culture age. For instance, a study on ajmalicine production in *Catharanthus roseus* showed that while higher concentrations (5.0%) of an elicitor extract enhanced accumulation, a further increase (10.0%) adversely affected the yield, demonstrating the need for precise dose-response optimization.

A notable example of biotic elicitation involves the co-cultivation of *Centella asiatica* with the endophytic fungus *Colletotrichum gloeosporioides*. This symbiotic co-cultivation resulted in a marked enhancement in root growth and biomass, including a 1.65-fold increase in root number. Crucially, the content of the key triterpenoid, asiaticoside, reached 4.08 mg/g in co-cultivated plants, substantially surpassing the 2.63 mg/g found in control plants. This enhancement is attributed to the fungal elicitor activating the Methylerythritol Phosphate (MEP) pathway, boosting the levels of the terpenoid precursor, squalene, and subsequently stimulating secondary metabolism.

5.2. Precursor Feeding

Precursor feeding involves the exogenous addition of specific biosynthetic intermediates or metabolic precursors to the culture medium. The underlying principle is that supplying these key substrates can bypass rate-limiting steps in the plant's endogenous biosynthetic pathways, thereby increasing the final metabolite yield.

This strategic approach is widely favored for industrial production due to its capacity for providing reproducible results in controlled *in vitro* environments. It also plays a role in conserving wild plant populations by shifting metabolite sourcing to the laboratory.

Despite its efficacy, precursor feeding presents challenges:

- **Cytotoxicity:** High concentrations of precursors can be toxic to plant cells, compromising viability and productivity.
- **Uptake Limitations:** Inefficient uptake and transport across the cell membrane can restrict the precursor's bioavailability.
- **Metabolic Diversion:** The fed precursors may be rerouted into unintended endogenous pathways, reducing the yield of the target compound.

Table -Successful examples for precursor feeding

Medicinal Plant	Precursor Supplemented	Increased Compound(s)	Reference
<i>Coptis japonica</i>	L-tyrosine	Berberine	Ikuta <i>et al.</i> , 1999
<i>Plumbago zeylanica</i>	Acetate	Plumbagin	Mali <i>et al.</i> , 2012
<i>Artemisia annua</i>	Mevalonic acid lactone	Artemisinin	Zhang <i>et al.</i> , 2011
<i>Catharanthus roseus</i>	Tryptophan	Ajmalicine, Catharanthine	Jaleel <i>et al.</i> , 2007
<i>Panax ginseng</i>	Squalene	Ginsenosides	Kim <i>et al.</i> , 2004
<i>Withania somnifera</i>	Sodium acetate	Withanolide A	Sharada <i>et al.</i> , 2007
<i>Taxus wallichiana</i>	Phenylalanine	Taxol	Wang <i>et al.</i> , 2007

A case study on *Sphagneticola calendulacea* demonstrated the power of this technique in transgenic hairy roots. Supplementing the culture with 0.5 mM phenylalanine led to a significant elevation in the coumestan compound, wedelolactone. The precursor-fed system yielded 440.33 µg/g DW of wedelolactone, compared to 422.01 µg/g DW in non-fed transgenic roots and only 294.44 µg/g DW in the wild-type plant. Furthermore, this strategy dramatically increased other phenolic compounds, with ferulic acid and rutin increasing up to 8.11-fold and 33.77-fold, respectively, over controls.

5.3. Genetic Transformation and Metabolic Engineering

Genetic transformation is the process of introducing foreign DNA into an organism's genome to achieve the stable integration and expression of desired genes. *Agrobacterium* -mediated transformation, as used in hairy root culture, is highly efficient and is one of the best techniques for enhancing SM production *in vitro*.

Metabolic engineering is a sophisticated extension of genetic transformation used to redesign cellular pathways for improved biosynthesis. Researchers can manipulate the flow of metabolites by:

- Introducing heterologous genes.
 - Blocking competing pathways.
 - Altering genes that encode rate-limiting enzymes.
- Table - functional analysis of metabolic engineering tools for secondary metabolite enhancement in plants

A study on hairy root cultures of *Momordica dioica*, induced using *A. rhizogenes*, successfully demonstrated enhanced phenolic production. Analysis revealed that transgenic roots produced significantly higher levels of flavonols, hydroxycinnamic acids, and hydroxybenzoic acids compared to non-transgenic roots. antibacterial, and antiviral activities. This work validates HR culture as a stable and reliable platform for the overproduction of valuable phenolic compounds.

Tool	What Does	How It Works	Role in Metabolite Enhancement	Example
CRISPR/Cas9 Genome Editing	Modifies plant genes very precisely	Cuts DNA at targeted sites and edits via repair mechanisms	Silences undesired genes, introduces new biosynthetic pathways	Boosted artemisinin in <i>A. annua</i> (Li <i>et al.</i> , 2020)
Agrobacterium-Mediated Transformation	Transfers new genes into plant cells	Uses <i>A. tumefaciens</i> or <i>A. rhizogenes</i> to deliver DNA into plant tissue	Creates hairy roots with stable expression; boosts production platforms	Enhanced phenolics in <i>Momordica dioica</i> (Thiruvengadam <i>et al.</i> , 2016)
Transcription Factor Modulation	Controls entire biosynthetic pathways	Overexpresses or silences key TFs like MYB, WRKY, bHLH	Turns on multiple metabolite-related genes simultaneously	Flavonols boosted in tobacco by <i>AtMYB12</i> (Grotewold <i>et al.</i> , 2000)
RNAi Silencing	Switches off unwanted or competing genes	Uses siRNA to target and degrade specific mRNA transcripts	Prevents metabolic cross-talk; redirects precursor flow toward desired products	Increased vinblastine in <i>C. roseus</i> (Chen <i>et al.</i> , 2018)

6. BIOTRANSFORMATION

Biotransformation is a chemical modification of some compounds by biological systems, typically involving enzymatic reactions that convert substrates into bioactive and extractable forms. It is widely used to enhance or diversify metabolite profiles. More precisely and it refers to the enzymatic conversion of exogenously supplied precursors into structurally modified or functionally enhanced metabolites by plant cells, tissues, or enzymes. (Yashavardhan *et al.*, 2020)

Table CORE CHEMICAL REACTIONS IN SECONDARY METABOLITE BIOTRANSFORMATION

Reaction	What It Does	Relevance in SM Enhancement
Hydroxylation	Introduces hydroxyl (-OH) groups into a molecule	Increases solubility and bioactivity of compounds (Khirwadkar <i>et al.</i> , 2013)
Glycosylation	Attaches sugar moieties to a molecule	Enhances stability, solubility, and pharmacokinetics (Yashavardhan <i>et al.</i> , 2020)
Oxidation	Adds oxygen or removes hydrogen, increasing polarity	Converts precursors into more reactive forms (Bajaj <i>et al.</i> , 2020)
Reduction	Removes oxygen or adds hydrogen, lowering oxidation state	Balances oxidative reactions, especially in polyphenol transformations
Methylation	Adds methyl (-CH ₃) groups, often affecting activity and mobility	Modifies antimicrobial or anticancer properties of metabolites

This is a multi-stepped process involving a living production platform. It typically initiates with substrate selection, followed by biocatalyst preparation, which involves plant cell cultures, microbial strains, or isolated enzymes. Following enzymatic activation, the transformation reaction is initiated, usually involving hydroxylation, glycosylation, or methylation and the products undergo **isolation**, **purification**, and **characterization** to conform structural integrity and bioactivity. After that optimization and scale up strategies are employed to enhance yield and productivity

7. Challenges and Future Prospects

Despite considerable progress, several challenges must be addressed to ensure the widespread industrial adoption of *in vitro* metabolite production systems.

7.1. Overcoming Key Challenges

- **Low Yield and Metabolic Bottlenecks:** Low yields remain a primary limitation, often due to rate-limiting enzymes and feedback inhibition in biosynthetic pathways. Overcoming this requires a deeper understanding of pathway regulation and the application of robust metabolic engineering.
- **Cytotoxicity and Uptake:** The cytotoxicity caused by high concentrations of elicitors or precursors can compromise cell viability. Furthermore, the lack of efficient membrane transporters can restrict the cellular bioavailability of these compounds, posing a hurdle for standardization.
- **Culture Stability and Scale-Up:** Long-term culture stability is a concern, as genetic and epigenetic drift can lead to the loss of biosynthetic capacity over successive subcultures. Finally, scaling up laboratory protocols to industrial production requires cost-effective, reproducible methods and rigorous quality control frameworks.

7.2. Avenues for Future Advancement

The future of *in vitro* SM production is focused on integration and precision.

Scalable Bioreactor Platforms: Hairy root and cell suspension cultures, when integrated with advanced bioreactor technologies, enable large-scale, high-yield production under fully reproducible conditions, meeting the needs of pharmaceutical and nutraceutical industries.

- **Precision Genetic Engineering:** Tools such as CRISPR/Cas9 and transcription factor modulation offer unprecedented precision for manipulating biosynthetic pathways. These tools allow for the targeted enhancement of rate-limiting steps, suppression of competing pathways, and the activation of 'silent' gene clusters, which promises to dramatically improve metabolite yield and diversity.
- **Synergistic Approaches:** The simultaneous application of advanced strategies, such as the synergistic use of elicitation (e.g., microbial endophytes like *C. gloeosporioides* in *C. asiatica*), precursor feeding, and genetic transformation, has been shown to result in substantial improvements in productivity.

The integration of these *in vitro* culture systems with cutting-edge biotechnological strategies represents a transformative shift in the production of medicinal compounds. By enabling year-round, controlled production, these techniques not only satisfy the growing demand for plant-derived bioactives but also offer a vital and sustainable alternative to conventional harvesting, supporting the conservation of endangered medicinal plant species.

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Table -Successful examples for precursor feeding

Medicinal Plant	Precursor Supplemented	Increased Compound(s)	Reference
<i>Coptis japonica</i>	L-tyrosine	Berberine	Ikuta <i>et al.</i> , 1999
<i>Plumbago zeylanica</i>	Acetate	Plumbagin	Mali <i>et al.</i> , 2012
<i>Artemisia annua</i>	Mevalonic acid lactone	Artemisinin	Zhang <i>et al.</i> , 2011
<i>Catharanthus roseus</i>	Tryptophan	Ajmalicine, Catharanthine	Jaleel <i>et al.</i> , 2007
<i>Panax ginseng</i>	Squalene	Ginsenosides	Kim <i>et al.</i> , 2004
<i>Withania somnifera</i>	Sodium acetate	Withanolide A	Sharada <i>et al.</i> , 2007
<i>Taxus wallichiana</i>	Phenylalanine	Taxol	Wang <i>et al.</i> , 2007

A case study on *Sphagneticola calendulacea* demonstrated the power of this technique in transgenic hairy roots. Supplementing the culture with 0.5 mM phenylalanine led to a significant elevation in the coumestan compound, wedelolactone. The precursor-fed system yielded 440.33 µg/g DW of wedelolactone, compared to 422.01 µg/g DW in non-fed transgenic roots and only 294.44 µg/g DW in the wild-type plant. Furthermore, this strategy dramatically increased other phenolic compounds, with ferulic acid and rutin increasing up to 8.11-fold and 33.77-fold, respectively, over controls.

5.3. Genetic Transformation and Metabolic Engineering

Genetic transformation is the process of introducing foreign DNA into an organism's genome to achieve the stable integration and expression of desired genes. *Agrobacterium* -mediated

transformation, as used in hairy root culture, is highly efficient and is one of the best techniques for enhancing SM production *in vitro*.

Metabolic engineering is a sophisticated extension of genetic transformation used to redesign cellular pathways for improved biosynthesis. Researchers can manipulate the flow of metabolites by:

- Introducing heterologous genes.
- Blocking competing pathways.
- Altering genes that encode rate-limiting enzymes.

A study on hairy root cultures of *Momordica dioica*, induced using *A. rhizogenes*, successfully demonstrated enhanced phenolic production. Analysis revealed that transgenic roots produced significantly higher levels of flavonols, hydroxycinnamic acids, and hydroxybenzoic acids compared to non-transgenic roots. Specifically, levels of catechin, rutin, quercetin, and gentisic acid were markedly higher, correlating with stronger antioxidant, antibacterial, and antiviral activities. This work validates HR culture as a stable and reliable platform for the overproduction of valuable phenolic compounds.

6. BIOTRANSFORMATION

Biotransformation is a chemical modification of some compounds by biological systems, typically involving enzymatic reactions that convert substrates into bioactive and extractable forms. It is widely used to enhance or diversify metabolite profiles. More precisely, it refers to the enzymatic conversion of exogenously supplied precursors into structurally modified or functionally enhanced metabolites by plant cells, tissues, or enzymes. (Yashavardhan *et al.*, 2020)

Table CORE CHEMICAL REACTIONS IN SECONDARY METABOLITE BIOTRANSFORMATION

Reaction	What It Does	Relevance in SM Enhancement
Hydroxylation	Introduces hydroxyl (-OH) groups into a molecule	Increases solubility and bioactivity of compounds (Khirwadkar <i>et al.</i> , 2013)
Glycosylation	Attaches sugar moieties to a molecule	Enhances stability, solubility, and pharmacokinetics (Yashavardhan <i>et al.</i> , 2020)
Oxidation	Adds oxygen or removes hydrogen, increasing polarity	Converts precursors into more reactive forms (Bajaj <i>et al.</i> , 2020)
Reduction	Removes oxygen or adds hydrogen, lowering oxidation state	Balances oxidative reactions, especially in polyphenol transformations
Methylation	Adds methyl (-CH ₃) groups, often affecting activity and mobility	Modifies antimicrobial or anticancer properties of metabolites

This is a multi-stepped process involving a living production platform. It typically initiates with substrate selection, followed by biocatalyst preparation, which involves plant cell cultures, microbial strains, or isolated enzymes. Following enzymatic activation, the transformation reaction is initiated,

usually involving hydroxylation, glycosylation, or methylation and the products undergo **isolation**, **purification**, and **characterization** to conform structural integrity and bioactivity. After that optimization and scale up strategies are employed to enhance yield and productivity

7. Challenges and Future Prospects

Despite considerable progress, several challenges must be addressed to ensure the widespread industrial adoption of *in vitro* metabolite production systems.

7.1. Overcoming Key Challenges

- **Low Yield and Metabolic Bottlenecks:** Low yields remain a primary limitation, often due to rate-limiting enzymes and feedback inhibition in biosynthetic pathways. Overcoming this requires a deeper understanding of pathway regulation and the application of robust metabolic engineering.
- **Cytotoxicity and Uptake:** The cytotoxicity caused by high concentrations of elicitors or precursors can compromise cell viability. Furthermore, the lack of efficient membrane transporters can restrict the cellular bioavailability of these compounds, posing a hurdle for standardization.
- **Culture Stability and Scale-Up:** Long-term culture stability is a concern, as genetic and epigenetic drift can lead to the loss of biosynthetic capacity over successive subcultures. Finally, scaling up laboratory protocols to industrial production requires cost-effective, reproducible methods and rigorous quality control frameworks.

7.2. Avenues for Future Advancement

The future of *in vitro* SM production is focused on integration and precision.

- **Scalable Bioreactor Platforms:** Hairy root and cell suspension cultures, when integrated with advanced bioreactor technologies, enable large-scale, high-yield production under fully reproducible conditions, meeting the needs of pharmaceutical and nutraceutical industries.
- **Precision Genetic Engineering:** Tools such as CRISPR/Cas9 and transcription factor modulation offer unprecedented precision for manipulating biosynthetic pathways. These tools allow for the targeted enhancement of rate-limiting steps, suppression of competing pathways, and the activation of 'silent' gene clusters, which promises to dramatically improve metabolite yield and diversity.
- **Synergistic Approaches:** The simultaneous application of advanced strategies, such as the synergistic use of elicitation (e.g., microbial endophytes like *C. gloeosporioides* in *C. asiatica*), precursor feeding, and genetic transformation, has been shown to result in substantial improvements in productivity.

The integration of these *in vitro* culture systems with cutting-edge biotechnological strategies represents a transformative shift in the production of medicinal compounds. By enabling year-round, controlled production, these techniques not only satisfy the growing demand for plant-derived bioactives but also offer a vital and sustainable alternative to conventional harvesting, supporting the conservation of endangered medicinal plant species.

Promoting Women-Led Micro-Enterprises in Rural Communities: Pathways to Empowerment and Sustainable Development in India

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Introduction

Empowering women through micro-enterprises has emerged as a cornerstone strategy for fostering economic independence and catalyzing sustainable rural development in India. With over **70% of rural women in India dependent on agriculture for their livelihood, yet owning only about 13% of landholdings** (NSSO 70th Round), non-farm micro-enterprises offer a critical pathway to economic agency. These enterprises, flourishing in sectors like food processing, handicrafts, textiles, and agriculture, are vital engines of growth. They extend beyond mere income generation to strengthen family welfare, enhance community resilience, and diversify local economies (World Bank, 2020). The successful promotion of these ventures, however, necessitates an integrated support system tailored to the Indian context, encompassing skill development, financial literacy, access to credit, and robust marketing channels. This chapter examines the multifaceted ecosystem required to nurture women-led micro-enterprises in rural India, analyzing persistent challenges, the pivotal role of extension services, effective promotion strategies, and illustrative Indian case studies, while also proposing future directions for scalable and sustainable impact.

Challenges Faced by Women Entrepreneurs in India

Despite their significant potential, women entrepreneurs in rural India confront a complex array of barriers that stifle their entrepreneurial growth and sustainability.

Social and Cultural Barriers: Deeply entrenched patriarchal norms and gender biases significantly constrain women's entrepreneurial activities. A study by the **International Center for Research on Women (ICRW)** found that **nearly 80% of women in rural North India required permission from a male family member to engage in economic activities outside the home**. The disproportionate burden of unpaid care work—Indian women spend **approximately 297 minutes per day on unpaid domestic services compared to 31 minutes by men** (NSSO, 2019)—severely limits their time availability for business pursuits.

Limited Access to Credit, Technology, and Markets: Women face significant hurdles in accessing formal finance.

While the **Jan Dhan Yojana** has increased account ownership, a credit gap of US\$ 158 billion exists for women-owned MSMEs in India (IFC, 2019). They often lack collateral and face perceived creditworthiness issues, forcing reliance on high-interest informal lenders. Furthermore, the digital divide is stark; **only about 31% of rural women in India have ever used the internet** (GSMA, 2023), restricting their access to online markets and information.

Lack of Business Skills, Financial Literacy, and Mentorship: While many women possess exceptional technical or artisanal skills, they often lack formal training in essential business management. A **NABARD survey (2018)** revealed that **only 22% of women members in Self-Help Groups (SHGs) felt confident in maintaining formal business accounts**, highlighting the critical gap in financial literacy and business skills.

Table 1: Key Challenges for Rural Women Entrepreneurs in India: Data Snapshot

Challenge Category	Specific Barrier	Supporting Indian Data / Statistic	Source
Social & Cultural	Time poverty due to unpaid care work	Rural Indian women spend 297 mins/day on unpaid domestic work vs 31 mins for men.	NSSO Time Use Survey, 2019
	Restricted mobility & agency	Nearly 80% of women in rural North India need permission from a male member to work outside home.	ICRW Study
Financial Access	Credit Gap for MSMEs	A credit gap of US\$ 158 billion exists for women-owned MSMEs in India.	IFC, 2019
	Reliance on Informal Sources	XX% of women entrepreneurs in [State, e.g., Bihar] rely on local moneylenders for initial capital.	[Relevant State-Level Study]
Digital Divide	Internet Access	Only 31% of rural women in India have ever used the internet.	GSMA, 2023
	Mobile Phone Ownership	The gender gap in mobile ownership in India is XX%.	GSMA, 2023
Skills & Literacy	Low Business Formality	Only 22% of women SHG members feel confident in maintaining formal business accounts.	NABARD, 2018
	Lack of Formal Training	Only X% of women micro-entrepreneurs have received any formal business skills training.	[MSME Ministry Survey, if available]

Role of Extension Services and Community Support in India

Extension services and community-based institutions form the critical backbone of support for rural women entrepreneurs in India, effectively bridging the gap between potential and success.

Capacity Building: Government initiatives like the **Deendayal Antyodaya Yojana - National Rural Livelihoods Mission (DAY-NRLM)** and NGOs conduct targeted training. For instance, **Kudumbashree in Kerala has trained over 300,000 women in various entrepreneurial skills**, leading to the establishment of thousands of micro-enterprises (Kudumbashree, 2023).

Access to Microfinance and Collective Action: The SHG-Bank Linkage Programme has been revolutionary. As of **March 2023, over 12 million SHGs had savings-linked with banks, with 88% being all-women groups, channeling credit of over ₹1,48,000 crore (approx. US\$18 billion)** (NABARD, 2023). This collective dynamic empowers women financially and fosters social solidarity.

Technology Transfer and Marketing Support: Institutions like the **Khadi and Village Industries Commission (KVIC)** and state rural livelihood missions provide access to improved technology (e.g., efficient looms, bio-fertilizer units) and marketing support through a network of emporiums and online platforms like **GVMs (Gramin Vikas Madals)**.

Strategies for Promoting Women-Led Enterprises in India

A strategic, multi-pronged approach is essential for creating a sustainable entrepreneurial ecosystem for women in rural India.

Participatory Approaches: Actively involving women in the planning and implementation of programs, as demonstrated by the **Mahila Kisan Sashaktikaran Pariyojana (MKSP)**, which empowers women farmers, ensures interventions are contextually relevant and fosters a strong sense of ownership.

Creating Local Value Chains and Market Linkages: Moving beyond supporting isolated enterprises, efforts should focus on integration into value chains. The **"Tripti" project in Odisha successfully linked women-led SHGs producing turmeric and other spices with large food processing companies, increasing their income by over 40%** (World Bank, 2019).

Leveraging Digital Tools: The strategic adoption of digital tools is key. Initiatives like **UPI-based digital payments have empowered women entrepreneurs by providing direct control over finances**. E-commerce platforms like **Amazon Karigar and Flipkart Samarth** have been instrumental in taking products from rural women artisans to national and international markets

Case Studies / Examples from India

Government Initiative: Deendayal Antyodaya Yojana - National Rural Livelihoods Mission (DAY-NRLM): This flagship program has systematically mobilized **over 8.6 crore women into 77 lakh SHGs**. It provides recurrent capitalization and intensive capacity-building. A notable success is the **promotion of over 300,000 women-led enterprises in non-farm sectors** and the facilitation of community resource persons, empowering women to become change agents in their communities (DAY-NRLM, 2023).

Community-Based Intervention: Shri Mahila Griha Udyog Lijjat Papad: Started in 1959 by seven women in Mumbai with a seed capital of ₹80, Lijjat is a stellar example of a women-led cooperative enterprise. It is entirely owned and managed by women, providing sustainable livelihoods to **over 43,000 women members across India**. Its success is built on the pillars of collective ownership, strict quality control, and the empowerment of women from marginalized backgrounds (Lijjat, 2023).

Private Sector Partnership: Amul (Gujarat Cooperative Milk Marketing Federation Ltd.): While not exclusively women-led, Amul's model has profoundly impacted rural women. It is estimated that **over 3.6 million women are part of dairy cooperatives in Gujarat alone**. By ensuring direct payment to women's bank accounts for milk, Amul has significantly enhanced their economic decision-making power and social status within households and communities (NDDB, 2022).

Table 2: Comparative Analysis of Indian Case Studies of Women-Led Enterprise Models

Feature / Model	DAY-NRLM (National, Govt-Led)	Lijjat Papad (Cooperative Model)	Amul (Dairy Cooperative)
Primary Objective	Poverty alleviation & livelihood generation through collective institutions.	Provide honest & dignified employment to women through collective ownership.	Increase milk producers' profitability & market access.
Structure	Federated structure: SHGs → Village Organisations → Cluster Level Federations.	Decentralized production units with a centralized brand and management.	3-tier structure: Village Dairy Cooperative Societies → District Unions → State Federation.
Key Support Provided	Revolving fund, bank linkage, capacity building, community resource persons.	Raw material supply, quality control, strong brand, 100% profit sharing.	Cattle feed, veterinary care, artificial insemination, guaranteed market & price.
Scale of Impact	8.6 crore+ women mobilized into 77 lakh+ SHGs.	43,000+ women member-entrepreneurs across India.	3.6 million+ women members in Gujarat dairy cooperatives alone.

Key Success Factor	Scalable government architecture and political will.	Utmost emphasis on quality and the power of collective brand.	Robust supply chain management and technological integration.

6. Challenges and Future Directions for India

While considerable progress has been made, persistent and emerging challenges necessitate focused future action.

Policy Gaps and Need for Inclusive Entrepreneurship Policies: There remains a critical need for policies that simplify business registration for home-based enterprises, enforce women's inheritance rights more effectively, and provide targeted tax incentives. The **Stand-Up India scheme, which mandates bank loans for women and SC/ST entrepreneurs, is a step in the right direction, but its reach needs to be expanded in rural hinterlands.**

Scaling Up Successful Models through Replication and Adaptation: Proven models like DAY-NRLM and the SHG-Bank Linkage program need to be scaled up with a greater focus on enterprise diversification and sustainability. This requires building strong, self-reliant institutions led by the women themselves.

Integrating Climate Resilience and Digital Inclusion: Future programs must integrate training in climate-resilient practices, such as organic farming and water conservation, crucial for agriculture-based enterprises. Concurrently, a massive push for digital inclusion through initiatives like **Pradhan Mantri Gramin Digital Saksharta Abhiyan (PMGDISHA)** is imperative to ensure rural women are not left behind.

Table 3: Strategic Interventions and Their Potential Impact for Scaling Up

Strategic Area	Proposed Intervention	Expected Outcome / Impact
Policy & Governance	Simplify "Udyam Shakti" registration for home-based enterprises.	Increased formalization, access to government schemes and credit.
	Mandate a percentage of public procurement from women-led collectives.	Assured market, better price realization, and economic stability.
Digital Inclusion	Integrate digital literacy modules (e-commerce, digital payments) into all SHG training.	Enhanced market access, financial autonomy, and reduced information asymmetry.
	Promote "Digital Sakhis" as community tech-trainers.	Sustainable, community-led support system for digital tools.
Climate Resilience	Train women in sustainable practices (e.g., organic farming, water-saving tech).	Reduced vulnerability to climate shocks, access to green markets.
	Promote enterprises in waste management & upcycling.	New livelihood opportunities while addressing local environmental issues.

Conclusion

Promoting women-led micro-enterprises is a powerful catalyst for sustainable and equitable development in rural India. This chapter has demonstrated that transforming entrepreneurial potential into tangible success requires an integrated ecosystem. This system must strategically address persistent barriers- social, financial, and digital through collective action, targeted skill development, and robust market linkages. The proven successes of models like SHGs under DAY-NRLM and cooperative enterprises highlight the transformative impact of such support. Moving forward, the focus must be on scaling these initiatives with greater inclusivity and innovation. This entails strengthening women-led institutions, bridging the digital divide, and integrating climate-resilient practices. By consolidating these efforts and fostering a responsive policy environment, we can unlock the full potential of rural women entrepreneurs. Empowering them is not merely an economic strategy but a fundamental step towards building a more resilient, prosperous, and self-reliant India.

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DIVERSIFIED CROP ROTATION: AN APPROACH FOR SUSTAINABLE AGRICULTURE

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Crop rotation with species diversification plays a crucial role in pest, disease, and weed management, while enhancing soil fertility and conservation. This approach can significantly boost profitability in grain production systems globally. By adopting diversified crop rotations, farmers can increase system efficiency, improve soil properties, and enhance productivity. Such rotations lead to better soil water uptake and retention, support a higher population of beneficial soil microbes, and increase yield resilience against drought and adverse conditions. Moreover, diversified crop rotations contribute to soil and ecological sustainability, lower production risks, and provide opportunities for revenue diversification. This strategy, increasingly favoured for sustainable crop production, helps build long-term soil health by optimizing the plant-soil relationship. This paper explores the benefits of diversified crop rotation, the challenges associated with its implementation, and potential solutions.

Key words: Crop rotation, diverse cropping system, soil health, crop productivity, sustainable agriculture, etc.

INTRODUCTION

Crop rotation is the practice of planting two or more crops sequentially on the same plot of land to improve soil health, optimize nutrients, and combat pest and weed pressure. Simple rotation may involve two or three crops, while a complex rotation may incorporate a dozen or more. Crops can be rotated on a field within the same year, over two years, or more extended periods. Crop rotation requires the succeeding crops to be of a different species or subspecies (variety) than the previous crop, such as legumes grown after wheat and cereals, however, cereal-cereal crop rotations are predominant in the country with farmers growing green manure before sowing rice for building soil health (stakeholders consulted at ICAR-IFSMR).



Crop rotation diversification

Meeting the ever-increasing food demand with insufficient agricultural supplies presents a significant challenge both nationally and globally (Davis *et al.*, 2016; Renard&Tilman, 2019). The agricultural sector plays a crucial role in addressing this productivity crisis, especially given the dual pressures of rising food demand and declining crop yields due to population growth. Soil texture and fertility are vital for maintaining agricultural efficiency and a healthy environment. As food demand continues to rise and agricultural land becomes scarcer, achieving greater crop yield and soil productivity will be essential (Tahat *et al.*, 2020).

In response to these challenges, there is growing interest in efficient and innovative agricultural practices, particularly to combat soil erosion exacerbated by anthropogenic and unsustainable farming practices. Recent concerns have focused on soil depletion caused by intensive farming (Khaledian *et al.*, 2017; Mekonnen *et al.*, 2015). Since the 1950s, nearly 60% of soil depletion has been linked to various soil ecological processes, with farming practices being a major contributor (Lal, 2015; Snakin *et al.*, 1996). Researchers are increasingly emphasizing the need to transform scientific knowledge about soils into practical techniques that enhance farmers' understanding and management of their agricultural activities. One effective approach in sustainable farm management is crop rotation, which aims to increase soil organic matter and reduce soil erosion.

Crop rotation effectively disrupts the reproduction and life cycles of insects and pathogens, contributing to their control. By including specific plant species in a rotation, this technique helps restore plant nutrients and reduce the need for chemical fertilizers. Crop rotation is a key practice in sustainable agriculture, offering advantages over monocultures or simple double-crop rotations. Diversified crop rotations involve rotating three or more different crops, which can help minimize trade-offs between crop viability and environmental impacts. A well-chosen crop rotation scheme not only maintains long-term soil fertility but also disrupts weed and disease cycles through natural nutrient recycling.

Diverse cropping systems present a promising approach for enhancing agricultural productivity, and diversified crop rotation methods offer substantial benefits for soil quality. By strengthening soil conditions and boosting overall system production, these methods contribute significantly to agricultural sustainability worldwide. As Doran and Zeiss noted, “the soil serves as a vital life cycle within the ecosystem, influencing land-use limitations for sustaining plant and animal growth, controlling or improving water quality, and supporting overall environmental health.”



Maize – Pea crop rotation



Maize – Wheat crop rotation



Durum wheat – Winter pea crop rotation



D. Flax – Chickpea crop rotation

Diversifying crop rotations enhances the agro-ecosystem

Soil health, which refers to the land's ability to sustain crop production despite challenges like soil depletion and environmental degradation, is crucial for the quantity and consistency of soil-based organic produce. By maintaining and improving soil health, diversified crop rotations ensure more resilient and productive agricultural systems. Soil health is defined as the ability of soil to function effectively within an ecosystem, supporting crop development while preserving its own quality and minimizing environmental harm. It plays a crucial role in determining the quality and performance of soil-based agricultural products. Diversified crop rotation (DCR) enhances soil health by increasing crop residues and root diversity, which boost microbial activity and improve soil performance. By managing soil erosion and enhancing crop production, DCR significantly improves soil properties and

optimizes the use of water and nutrients in the soil profile, thereby sustaining productivity (Miller et al., 2003). DCR helps disrupt disease cycles and enhance the physical and chemical properties of soil systems, thereby preserving soil quality and laying the foundation for improved agricultural practices. This study also addresses the challenges, obstacles, and factors influencing the adoption and implementation of DCR in agricultural systems. Thematic areas are outlined briefly in the following subheadings.

Factors affecting adoption of diversified crop rotation

Different studies Alemuet *al.* (2017) and Manjunathaet *al.* (2013) showed that land fragmentation is also seen as the major factor that affects the adoption of diversified crop rotation attributed to inadequate allocation of resources, which is correlated with production costs; suboptimal use of factor inputs that minimize total returns to land due to extra travel time losses, wasted space across boundaries, insufficient surveillance, and reluctance to use predefined types of machinery; and hindering agricultural development and restructuring schemes to adjust adverse implications, among others. The incentive to improve the productivity of smallholders' farmland through area expansion is confined, and farmers are hesitant to diversify crops due to modest outputs from diverse specified crops, given the expanding population versus the subsequent fragmentation of land size and depletion of natural resources.

According to farmers with more and smaller plots prefer employing fewer current technologies, mono cropping, and decreasing the average distance between plots, while an increase in farm size improves the farmer's interest in picking diverse crops with rotation, lowering the overall cost of production per unit. The factors influencing the adoption of these technologies and practices (i.e., DCR), are as follows:

1. *Soil and climate:* The suitability of the soil for different crops in a rotation can impact the effectiveness of diversified crop rotation. Some soils may benefit more from specific rotations than others. Regional climate conditions influence the choice of crops and their performance within a rotation. Farmers need to select crops that are well-suited to their local climate.
2. *Cultural or traditional practices:* Long-standing farming traditions and practices can affect the willingness to adopt new methods. Resistance to change may stem from established routines and beliefs. The practices of neighboring farms and peer influence can impact adoption rates. Farmers may be more likely to adopt new practices if they see success in their community.

3. *Farm infrastructure and resources:* The existing infrastructure, such as irrigation systems and machinery, may need to be adapted or upgraded to support diversified crop rotation. Access to resources such as seeds, fertilizers, and equipment suitable for diverse crops is crucial for successful implementation.
4. *Technical consideration:* Successful implementation of diversified crop rotation requires knowledge of which crops to rotate and the benefits of each. Farmers may need additional training or access to expert advice. Farmers with experience in diverse cropping systems are more likely to adopt these practices. Lack of experience or familiarity can hinder adoption.
5. *Economic considerations:* The upfront costs associated with implementing diversified crop rotation, such as purchasing new seeds or modifying equipment, can be a barrier for farmers. The perceived or actual financial returns from diversified crop rotations compared to traditional monoculture systems can influence adoption. Farmers may need time to see the economic benefits of improved soil health and reduced pest management costs.
6. *Crop marketability:* Farmers may be reluctant to adopt diversified crop rotation if they are uncertain about the market demand for the crops included in the rotation. Access to markets, processing facilities, and distribution channels for diverse crops can impact the feasibility of rotating crops.
7. *Government incentives:* Supportive policies and subsidies can encourage the adoption of diversified crop rotation by offsetting costs and risks. Investments in research and development for crop rotation practices and technologies can facilitate adoption by providing evidence-based benefits and solutions.
8. *Risk management:* Farmers may be concerned about the risks associated with transitioning to a new cropping system, such as potential crop failures or reduced yields during the adjustment period. Availability of insurance and risk management tools can influence the decision to adopt diversified crop rotation.

Role and benefits of diversified crop rotations: Diversified crop rotation involves systematically varying the types of crops grown in a particular field over successive seasons. This agricultural practice plays a vital role in enhancing both farm productivity and environmental sustainability. Here's a detailed look at its roles as follows:

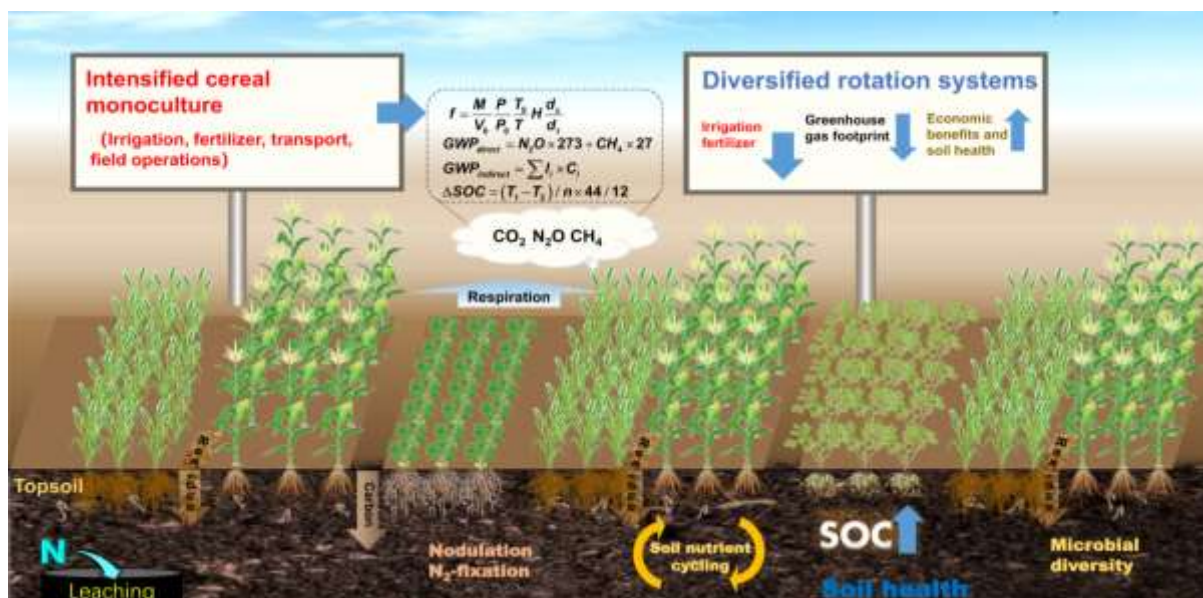
1. *Improving soil health:* Researchers and farmers have been at the forefront of developing and implementing methods to maintain soil health. Both agricultural producers and academics play crucial roles in preventing soil depletion by emphasizing long-term sustainability. Soil health and quality are significantly influenced by agricultural practices such as diverse crop cultivation, crop rotation, and

intercropping, which address soil management from various spatial and temporal perspectives. Diversified crop rotation is particularly important for optimizing crop production and enhancing soil health.

2. *Resistance to pest and diseases:* In diversified crop rotation, breaking the disease cycle in the soil is highly beneficial. Monoculture, where the same crop is grown repeatedly, allows pathogens to build up and spread, leading to more severe plant disease outbreaks. In contrast, rotating crops—such as a seven-year cycle of three years of alfalfa, two years of chili pepper, and two years of cotton before returning to alfalfa—helps disrupt this cycle. Since alfalfa, chili pepper, and cotton come from different plant families, pathogens that affect one type of plant cannot easily infect others, reducing their population in the soil. Diversified crop rotation also aids in pest management by breaking pest cycles, reducing weeds and diseases, improving soil quality, and supporting ecosystem health (Lupwayi et al., 2006). Different plant species contribute unique residues and root exudates that enhance soil microbial diversity and nutrient cycling. This increase in microbial variety and soil heterogeneity promotes better soil quality through interactions with a wider range of soil microorganisms (Berg et al., 2016).
3. *Enhancing soil fertility:* Diversified crop rotation, including the use of cover crops planted between cash crops, helps maintain soil quality by integrating root exudates and biomass. These plant roots and their interactions with soil fungi and microbes contribute to stabilizing soil and creating resilient structures, such as biopores, which are beneficial for soil health. However, non-mycorrhizal plants like lupin and canola, which don't form beneficial relationships with arbuscular mycorrhizal fungi (AMF), make it harder to understand how soil stabilization and glomalin production are affected. Additionally, plants like canola and mustard, which don't form certain beneficial soil relationships, may require more chemical fertilizers and could alter soil structure and fertility over time (Alam et al., 2014; Wright & Anderson, 2000).
4. *Increasing soil quality:* Soil with optimal physical, chemical, and biological properties enhances overall soil quality in farming systems. For instance, a wheat-pulse crop rotation can improve soil conditions and increase productivity. Studies have shown that a four-year rotation of wheat and field pea helps protect soil from degradation by maintaining its physical structure. Diversifying crops with rotation also helps conserve soil water and boost nitrogen levels. For example, cover crops like rye and phacelia effectively absorb nitrogen and water, reduce nitrate leaching in broccoli by 70%, and lower fertilizer needs. Overall, diversified cropping systems offer significant benefits, including better

water use efficiency, increased grain production, improved soil quality, and enhanced management of weeds, insects, and diseases.

5. *Increasing crop productivity*: In parts of the world where diverse cropping is a common element of agro-ecosystem management, productivity is usually far more robust and consistent throughout time. By diversifying crops, farmers can reduce the risk of crop failure due to pests, diseases, or adverse weather conditions. This can stabilize and potentially increase overall yields. Different crops have varying growing seasons and environmental needs, which can optimize land use and increase overall farm productivity.
6. *Enhancing water management*: Diverse root systems and plant residues improve soil's water-holding capacity, reducing runoff and erosion. Different crops have different water requirements, and their rotation can help in better managing soil moisture levels throughout the year.
7. *Mitigating environmental impact*: Diverse root systems and plant residues help prevent soil erosion and degradation. Healthier soils and reduced need for chemical inputs can contribute to lower greenhouse gas emissions.



Benefits of diversified crop rotation

Overall, diversified crop rotation is a cornerstone of sustainable agriculture, offering numerous benefits for soil health, crop productivity, environmental conservation, and economic stability. Its role in promoting resilient and efficient farming systems makes it a valuable practice in modern agriculture.

Challenges in adoption of diversified crop rotation

Diversified crop rotation faces several adoption challenges due to a lack of essential investments in machinery, infrastructure, and expertise, as well as insufficient research evidence. Financial constraints are a major hurdle, as integrating additional crops into existing rotations often requires significant upfront investments in new equipment and incurs short-term costs. Additionally, farmers with short-term rental agreements may lack the long-term stability needed for effective planning and soil or production improvements.

Other obstacles include limited access to financing for investments, insufficient technological support, and a lack of knowledge among stakeholders. Farmers may also face difficulties due to inadequate access to markets, as some crops may not have readily available or regulated markets. Despite the benefits of diversified crop rotation such as enhanced crop yields, improved soil health, and better water-use efficiency.

Challenges like poor communication, outdated knowledge, understaffing, and low morale also impede the adoption of new conservation methods. Furthermore, crop insurance and credit limitations can be significant barriers, as lenders may be hesitant to finance complex rotations due to unfamiliarity with their profitability potential. For broader adoption, farmers need assurance that new methods are feasible, can be effectively implemented locally, and will positively impact their financial outcomes.

CONCLUSION

Diversified crop rotations are gaining popularity as a sustainable approach to crop production due to growing concerns about providing high-quality food while minimizing environmental impact. DCR fosters beneficial soil microbes, disrupts disease cycles, and reduces weed populations. It enhances the physical and chemical properties of soil, boosts land-use efficiency, and increases crop productivity, making it a valuable practice for long-term profitability. Farmers need flexible and cost-effective diversified crop rotations to meet market demands. Supportive policies and organizational backing are crucial for encouraging the adoption of these practices. The scientific community should focus on developing adaptable diversified crop rotation strategies that can respond to changing climatic conditions and improve overall farming sustainability.

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Next-Generation Agriculture: AI Solutions for Plant Disease Detection and Management

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1. Introduction

Agriculture is the backbone of many economies, providing food, raw materials, and livelihood to billions of people. In India, it contributes nearly 18 per cent to the Gross Domestic Product (GDP) and employs over 45 percent of the population. However, the sector faces immense challenges due to climate change, population growth, limited natural resources, and the continuous threat of crop diseases. Plant diseases caused by fungi, bacteria, viruses and nematodes cause annual yield reductions of 15-25% in India and up to 40% globally. Foliar diseases are particularly destructive as they directly impair photosynthesis, reduce crop vigour, and severely limit yield potential.

Traditional approaches to disease monitoring-mainly field scouting and laboratory tests-are labour-intensive, time-consuming, and unsuitable for large-scale surveillance. With the demand for precision agriculture increasing, remote sensing technologies and artificial intelligence (AI) driven solutions have emerged as powerful alternatives. By integrating vegetation indices, multispectral and hyperspectral imaging, drones, and deep learning algorithms, plant health can now be monitored at scale, enabling early interventions and informed decision-making.

This chapter highlights how remote sensing tools and AI-driven models are transforming disease detection and management within the broader framework of smart agriculture.

2. Impact of Plant Diseases on Food, Health, and Environment

2.1 Food Security

Plant diseases directly threaten food availability and stability. Historical events such as the Irish Potato Famine (1845-49) and Bengal Famine (1943) illustrate how crop epidemics can lead to widespread hunger and human migration. In the present era, coffee rust outbreaks in Central America and wheat rust epidemics in Africa demonstrate how plant diseases remain a serious global threat.

2.2 Food Safety Hazards: Mycotoxins such as aflatoxins and ochratoxin A, produced by fungal pathogens, contaminate grains, nuts, and fruits, posing risks of cancer and other health disorders. Disease outbreaks therefore influence not only yields but also food quality and consumer health

2.3 Occupational and Socioeconomic Impacts

Excessive pesticide use to control plant diseases exposes farm workers to health risks and increases environmental contamination. Outbreaks reduce farmers' income, disrupt markets, and contribute to rural poverty

2.4 Environmental Consequences

Plant disease epidemics disturb ecosystems, biodiversity, and nutrient cycles. For example, chestnut blight destroyed American chestnut forests, permanently altering forest ecology. Climate change is expected to increase the frequency and severity of such outbreaks by altering pathogen life cycles and crop susceptibility

3. Remote Sensing in Plant Disease Detection

Remote sensing refers to the acquisition of crop data through sensors mounted on handheld devices, UAVs (drones), satellites, or ground vehicles. It provides non-destructive, rapid, and large-scale measurements of crop health

3.1 Vegetation Indices

NDVI (Normalized Difference Vegetation Index): Sensitive to chlorophyll content and canopy vigour. Declines in NDVI values show a strong correlation with yellow mosaic and other viral diseases in mungbean and soybean

SPAD Chlorophyll Meter: Provides point-based measurements of leaf greenness, correlating with disease severity but limited in scalability

Thermal Indices: Useful for identifying stress-induced changes in transpiration associated with disease

Hyperspectral Imaging: Captures reflectance covering hundreds of narrow bands, enabling detection of slight biochemical alterations preceding visible symptom expression

3.2 UAV and Drone-Based Monitoring

Unmanned aerial vehicles (UAVs) equipped with RGB, multispectral, and hyperspectral sensors facilitate precise, field-level disease surveillance and timely identification of crop health anomalies. High-resolution imagery can identify hotspots, generate disease maps, and guide site-specific management practices.

3.3 Satellite-Based Remote Sensing

Medium- to coarse-resolution satellite imagery (e.g., Sentinel, Landsat) supports regional and national-scale monitoring of epidemics, though limited by cloud cover and resolution constraints

4. AI and Smart Agriculture

AI, especially deep learning (DL), has revolutionized plant disease detection. By automatically extracting features from images, DL models achieve superior performance compared to traditional machine learning methods

4.1 Deep Learning Models

CNNs (Convolutional Neural Networks): Widely applied for disease classification in crops like tomato, potato, and wheat

YOLO (You Only Look Once): Object detection framework that localizes and identifies multiple foliar diseases in real time

ResNet, DenseNet, and EfficientNet: Advanced CNN architectures with superior accuracy and generalization

Vision Transformers (ViTs): Emerging models that capture long-range dependencies in images for improved disease recognition

4.2 Applications in Major Crops

Rice: CNNs detect blast and bacterial blight with >95% accuracy

Wheat: YOLO models identify rusts and field-incited powdery mildew

Soybean and Mungbean: UAV-based YOLO detectors achieve mAP>80% for Yellow Mosaic Virus detection

Horticultural Crops: CNN-ViT hybrids classify grape, apple, and banana leaf diseases with accuracies above 98 per cent

5. Workflow for AI-Based Disease Detection

The typical workflow for deploying AI in plant disease detection includes:

- a. Image Acquisition: Cameras, drones, hyperspectral sensors, and public datasets.
- b. Preprocessing: Resizing, background removal, noise reduction, and normalization.
- c. Image Augmentation: Rotation, flipping, scaling, brightness/contrast adjustments to improve robustness.
- d. Annotation: Labeling of images with bounding boxes, class names, or segmentation masks.
- e. Model Training: Using supervised or transfer learning with CNN, YOLO, or hybrid models.

- f. Evaluation: Accuracy, precision, recall, F1-score, and mAP metrics.
- g. Deployment: Integration into mobile apps, UAV platforms, or IoT-enabled devices for real-time use.

6. Integration of Remote Sensing and AI

The convergence of remote sensing and AI forms the backbone of smart agriculture. UAVs collect multispectral images, AI models analyze disease severity, and IoT devices transmit data for real-time decision support. Precision spraying drones enable targeted pesticide application, thereby reducing chemical inputs, environmental contamination, and associated costs. Decision-support systems (DSS) combine remote sensing data, weather forecasts, and AI-based risk models to predict disease outbreaks and recommend timely interventions.

7. Case Studies and Recent Advances

7.1 Yellow Mosaic Disease Detection in Legumes

Recent work on yard-long bean showed that RGB canopy imaging combined with machine learning can effectively quantify yellow mosaic disease (YMD) severity. From 143 genotypes, vegetation indices like RCC and ExR correlated strongly with disease ($r \approx 0.87$), while GRD was negatively correlated ($r \approx -0.88$). Models such as Random Forest and XGBoost achieved high predictive accuracy ($R^2 \approx 0.88$, d-index ≈ 0.96), demonstrating that low-cost visible imaging can provide reliable, non-destructive disease assessment

7.2 UAV-Based Soybean Disease Monitoring

Studies have shown YOLOv8 object detectors can identify MYMV in soybean with precision up to 98%, enabling real-time diagnosis at field scale.

7.3 Other Global Applications

UAV-hyperspectral imaging detected early blight in potato before visible symptoms appeared. Satellite-based NDVI time-series successfully mapped wheat rust outbreaks across large regions

8. Advantages and Limitations

Advantages

- Non-invasive and scalable disease detection.
- Early detection before visible symptoms.
- Real-time decision-making through mobile apps and drones.
- Reduced pesticide use and input costs.

Limitations

- High cost of sensors and UAV platforms.

- Dependence on large, annotated datasets.
- Environmental variability (lighting, occlusion, leaf overlap) reduces accuracy.
- Farmer's limited technical expertise and slow adoption in rural areas.

9. Future Prospects

Future research should focus on:

- Explainable AI (XAI): Increasing model transparency and farmer trust.
- Drone-Integrated Systems: Real-time disease maps and precision spraying.
- Cross-Crop Models: Generalizable architectures adaptable to multiple crops.
- Integration with IoT and Cloud Platforms: Continuous monitoring and data-driven advisory services.
- Policy and Extension Support: Ensuring accessibility for smallholder farmers.

10. Conclusion

Remote sensing and AI are transforming plant disease management by enabling rapid, precise, and large-scale detection systems. Vegetation indices such as NDVI and SPAD provide early indicators of stress, while AI-based image analytics detect and identify diseases with high precision. Their integration offers a scalable framework for smart agriculture, reducing input costs, improving sustainability, and safeguarding food security. With further advances in UAVs, hyperspectral imaging, and explainable AI, these technologies will play a central role in sustainable crop production and global food security.

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Impacts of Climate Change on Crop Production and Soil Health

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1. Introduction

Climate change has emerged as one of the most pressing global challenges of the 21st century, with far-reaching consequences for natural ecosystems, human societies, and economic systems. Among the sectors most vulnerable to climate variability, agriculture stands at the forefront due to its direct reliance on environmental conditions such as temperature, rainfall, and soil quality [1]. Crop production, the cornerstone of global food security, is highly sensitive to even minor shifts in climate. Small changes in temperature or precipitation patterns can affect plant physiological processes, leading to reductions in growth, yield, and quality. These changes are particularly concerning for staple crops such as rice, wheat, maize, and pulses, which feed billions of people worldwide [2,3]. Soil health, the foundation of agricultural productivity, is also intricately linked to climate. Healthy soils support nutrient cycling, water retention, and microbial biodiversity, all of which are essential for robust crop growth. Climate change can disrupt these soil functions through processes such as erosion, salinization, organic matter depletion, and altered microbial activity. Degraded soils not only reduce agricultural productivity but also contribute to the release of greenhouse gases, creating a feedback loop that exacerbates climate change.

The implications of these changes are not limited to agriculture alone; they extend to socio-economic stability, rural livelihoods, and national economies. Declining crop yields and deteriorating soil health can lead to food insecurity, price volatility, and increased vulnerability for millions of smallholder farmers, particularly in developing countries. The complex interplay between climate, crops, and soils underscores the urgent need to understand, anticipate, and mitigate these impacts.

This chapter aims to provide a comprehensive overview of how climate change affects crop production and soil health. It examines the specific climatic factors driving these changes, explores their consequences on crop growth and soil properties, and highlights socio-economic implications. Furthermore, the chapter discusses adaptive strategies and mitigation measures that can help safeguard agricultural productivity and promote sustainable soil management. By synthesizing current scientific knowledge and real-world examples, this chapter seeks to provide actionable insights for researchers, policymakers, and farmers striving to build resilient agricultural systems capable of withstanding the challenges of a changing climate.

2. Overview of Climate Change Factors Affecting Agriculture

Climate change encompasses a broad range of environmental alterations driven primarily by human activities such as fossil fuel combustion, deforestation, and industrialization. These changes do not occur in isolation; rather, they interact with agricultural ecosystems in complex ways. Understanding the primary climatic factors that influence crop production and soil health is fundamental to assessing agricultural vulnerability and planning adaptation strategies [6].

2.1 Rising Temperatures

Global average temperatures have significantly increased over the past century. Even a 1–2°C rise can lead to heat stress in crops, accelerate evapotranspiration, and shorten the growing period. Heat stress adversely affects flowering and grain-setting stages, leading to reduced yields. High night-time temperatures can increase plant respiration, reducing net photosynthesis and negatively affecting grain quality.

2.2 Changes in Rainfall Patterns

Climate change has caused unpredictable rainfall events, some regions experience prolonged droughts while others face excessive rainfall and flooding. Uneven distribution of rainfall disrupts planting schedules, affects seed germination, and reduces soil moisture availability. Floods lead to waterlogging, root damage, and nutrient leaching, whereas drought conditions limit crop growth and reduce productivity [7].

2.3 Increased Frequency of Extreme Weather Events

Extreme weather phenomena such as cyclones, heatwaves, droughts, hailstorms, and flash floods have become more frequent. These sudden events cause direct crop destruction, loss of topsoil, delayed planting, and long-term damage to soil health. Recurrent climate extremes increase uncertainty in agricultural systems, making farming riskier and less predictable.

2.4 Elevated Carbon Dioxide (CO₂) Levels

While elevated CO₂ can stimulate photosynthesis in some crops (known as the CO₂ fertilization effect), it also leads to higher atmospheric temperatures [8]. Moreover, the benefit is limited by nutrient availability, water supply, and increased pest populations. In many crops, elevated CO₂ can reduce grain protein content and nutrient density.

2.5 Sea-Level Rise and Salinisation

In coastal regions, rising sea levels cause saltwater intrusion into agricultural lands, leading to increased soil salinity. High salinity inhibits plant growth, interferes with water uptake, and degrades soil structure. Many staple crops such as rice and wheat are highly sensitive to saline conditions, resulting in reduced yields [9].

2.6 Increased Pest, Weed, and Disease Pressure

Climate change alters the life cycles and habitats of pests and pathogens. Warmer temperatures may extend breeding seasons, allowing pests to reproduce more rapidly and infest crops more frequently. Changes in humidity and rainfall create favorable environments for fungal diseases and invasive weed species [10].

3. Impacts of Climate Change on Crop Production

Climate change directly affects the biological, physiological, and ecological processes involved in crop growth. Variations in temperature, rainfall, carbon dioxide levels, and the frequency of extreme weather events significantly influence agricultural productivity. These changes affect every stage of crop development—from germination to yield formation—ultimately threatening food security at local, national, and global levels [6,8].

3.1 Temperature Stress and Heatwaves

Rising temperatures are among the most critical factors affecting crop production [11]. Crops have an optimum temperature range for each stage of growth. When temperatures exceed these thresholds:

1. **Germination is delayed** due to reduced water uptake.
2. **Flowering and pollination rates decline**, leading to poor grain or fruit setting.
3. **Accelerated crop maturity** reduces the time available for biomass accumulation.
4. **Heatwaves cause flower sterility**, especially in rice and wheat.

For example, a temperature rise of even 1.5–2°C can reduce wheat yield by 6–10%, while rice experiences spikelet sterility and reduced grain weight.

3.2 Changes in Rainfall and Water Availability

Water is a vital resource for agriculture, and climate change-driven alterations in rainfall patterns severely impact crop productivity:

- **Drought conditions** reduce soil moisture, causing crop wilting, stunted growth, and early plant death.
- **Floods and waterlogging** hinder root respiration, reduce nutrient uptake, and increase susceptibility to fungal diseases.
- Rainfed agriculture, which constitutes nearly 60% of global farming, is more vulnerable to irregular rainfall. Prolonged dry spells force farmers to delay sowing or switch to less productive crops, reducing overall agricultural output.

3.3 CO₂ Fertilization Effect

Elevated atmospheric CO₂ concentrations can enhance photosynthesis, particularly in C3 crops like rice, wheat, and soybeans. This is known as the CO₂ fertilization effect. However, its benefits are limited and often offset by other climate stressors:

- Increased CO₂ can lead to higher carbohydrate content but lower protein and micronutrient levels in crops.
- The positive impact is reduced under conditions of limited nitrogen, phosphorus, or water. Therefore, while the CO₂ effect may temporarily increase biomass, it does not guarantee enhanced food quality or yield sustainability.

3.4 Pest and Disease Proliferation

Climate change alters ecosystems and creates favorable conditions for pests, insects, weeds, and pathogens:

- **Warmer temperatures** extend the breeding season of pests like stem borers, aphids, and locusts.
- **Increased humidity** promotes the spread of fungal diseases such as rust, blight, and mildew.
- **Emergence of new pest species** in regions where they were previously absent. These biological threats lead to increased crop losses and greater dependency on chemical pesticides, raising production costs and affecting environmental sustainability.

4. Impacts of Climate Change on Soil Health

Soil health plays a central role in sustaining agricultural productivity, ecosystem stability, and food security. Climate change affects soil not only physically but also chemically and biologically, disrupting its natural processes and reducing its capacity to support crops. The impact on soil health is long-lasting and can be far more difficult to reverse than the effects on crops. This section highlights the major ways climate change is altering soil properties and functions.

4.1 Soil Erosion and Degradation

Changes in rainfall intensity and frequency have led to increased soil erosion:

1. Heavy rainfall events cause runoff that removes the top fertile layer of soil.
2. Loss of organic matter and nutrients results in decreased soil fertility.
3. In sloping areas, surface erosion leads to land degradation and desertification.

Once lost, topsoil takes hundreds of years to regenerate, making this a critical concern for global agriculture.

4.2 Decline in Soil Organic Matter (SOM)

Soil organic matter is essential for maintaining soil structure, water retention, and nutrient availability.

- High temperatures accelerate decomposition of organic material, leading to a decline in SOM.
- Reduced organic matter decreases soil's ability to hold water, impacting crop resilience during drought.
- Lower carbon content affects soil structure and increases susceptibility to erosion.

4.3 Moisture Stress and Soil Salinization

Climate change contributes to both soil drying and salt accumulation:

- Reduced rainfall and higher evaporation rates lead to soil moisture deficits.
 - In coastal regions, sea-level rise results in saltwater intrusion, increasing soil salinity.
 - Salinity interferes with plant water uptake and reduces germination rates.
- Crops such as rice, wheat, and legumes are highly sensitive to saline conditions, leading to significant yield loss.

4.4 Impact on Soil Microbial Activity and Biodiversity

Soil organisms—such as bacteria, fungi, and earthworms—are critical for nutrient cycling and soil fertility.

- **Elevated temperatures** alter microbial activity, leading to imbalances in nutrient availability.
 - **Flooding and drought** reduce microbial populations and limit beneficial interactions between plants and soil microorganisms.
 - Changes in soil biology impair **nitrogen fixation, decomposition, and carbon sequestration**.
- Overall, this reduces soil resilience and agricultural sustainability.

Integrated Effects on Soil Health

Climate-induced alterations in soil properties have cascading effects:

- **Reduced nutrient availability** leads to lower crop yields.

- **Poor soil structure** affects water infiltration and root development.
- **Loss of biodiversity** disrupts ecological balance.
- **Increased land degradation** threatens long-term food security.

5. Socio-Economic Consequences of Climate Change on Agriculture

Climate change poses severe socio-economic challenges, particularly for agrarian societies where a large population depends directly on farming for livelihood. Declining crop productivity and soil fertility result in reduced agricultural output, diminishing farm incomes and increasing vulnerability among farmers, especially small and marginal landholder. Unpredictable weather events such as droughts, floods, and storms cause crop failures, leading to financial losses, indebtedness, and sometimes forcing farmers to abandon their land. The worsening soil health further escalates production costs as farmers need to invest more in fertilizers, irrigation, and soil restoration efforts, increasing their dependency on external inputs.

Moreover, climate-induced stresses disrupt the stability of food supply chains, causing fluctuations in market prices. When crop yields decline, food scarcity arises, leading to inflation and reduced food affordability, particularly for low-income populations. In regions heavily dependent on agriculture, rising production risks contribute to social instability, rural unemployment, and migration to urban areas in search of alternative livelihoods. As agricultural productivity diminishes, the overall national economy suffers, especially in countries where agriculture forms a significant share of GDP. Climate change also exacerbates inequalities, as vulnerable communities with limited adaptive capacity bear the brunt of the impacts while having fewer resources to cope. In essence, the adverse effects of climate change on crop production and soil health extend far beyond the farm, influencing food security, rural development, and economic stability at both national and global scales.

6. Adaptation and Mitigation Strategies

Adaptation and mitigation strategies are essential to safeguard agriculture from the adverse impacts of climate change on crop production and soil health. Farmers, scientists, and policymakers are increasingly focusing on climate-resilient farming practices to ensure sustainable food production. One of the most effective approaches is the adoption of climate-smart agriculture (CSA), which integrates improved crop varieties, resource-efficient technologies, and ecological management to enhance resilience while reducing greenhouse gas emissions. The development and cultivation of drought-tolerant, heat-resistant, and saline-resistant crop varieties help maintain yields under changing environmental conditions. Additionally, precision farming techniques, such as drip irrigation, sensor-

based nutrient application, and weather forecasting tools, enable efficient resource use and reduce wastage of water and fertilizers.

Maintaining soil health is equally crucial in climate adaptation. Practices such as organic farming, cover cropping, mulching, and conservation tillage improve soil structure, enhance water retention, and reduce erosion. The use of biofertilizers and compost increases soil organic matter, boosting microbial activity and nutrient availability. Mitigation strategies also include agroforestry and crop diversification, which promote ecological balance and act as natural carbon sinks. Government policies and institutional support play a vital role in implementing these strategies by providing subsidies, crop insurance, early warning systems, and access to sustainable agricultural technologies. Collectively, these adaptation and mitigation measures not only protect agricultural productivity in a changing climate but also contribute to long-term sustainability and environmental conservation.

8. Future Outlook and Research Needs

The future of global agriculture in the face of climate change will largely depend on the development and adoption of innovative technologies, sustainable practices, and strong policy support systems. While the negative impacts of climate change on crop production and soil health are expected to intensify, there is also a growing opportunity to transform traditional agriculture into a resilient, adaptive, and climate-smart system. Future agricultural research must focus on breeding crops that can withstand multiple stresses such as heat, drought, salinity, and flooding simultaneously. Advancements in biotechnology, genetic engineering, and CRISPR gene-editing techniques offer significant potential in developing next-generation crops tailored to changing environmental conditions.

Soil health research will play a central role in sustaining agricultural productivity. There is an urgent need to explore soil carbon sequestration methods, enhance microbial diversity, and develop low-cost organic amendments to restore degraded soils. Digital agriculture powered by artificial intelligence, remote sensing, and satellite monitoring will become crucial in predicting climate risks, optimizing resource usage, and providing real-time decision-making support to farmers. Moreover, interdisciplinary research integrating climate science, agronomy, economics, and social studies is essential to design region-specific adaptation strategies. Strong international cooperation, government investment, and farmer-centered innovation will be indispensable in shaping a sustainable agricultural future. Ultimately, the resilience of global food systems will depend on how effectively societies respond to climate challenges through science-driven, inclusive, and environmentally responsible agricultural transformation.

9. Conclusion

Climate change poses a profound and escalating threat to global agriculture by disrupting the delicate balance between crop production and soil health. Rising temperatures, erratic rainfall, and the increased frequency of extreme weather events are already reducing crop yields, altering growing seasons, and compromising the nutritional quality of food. Simultaneously, soil health is deteriorating due to erosion, declining organic matter, moisture stress, and loss of biological activity, all of which undermine the foundation of sustainable agriculture. These impacts are not confined to environmental parameters alone; they extend to economic instability, threats to food security, rural livelihood losses, and increased vulnerability of farming communities, particularly in developing nations.

However, the challenges presented by climate change can be addressed with proactive adaptation and mitigation strategies. Sustainable practices such as climate-smart agriculture, conservation of soil resources, crop diversification, and technological innovation offer practical solutions to build resilience. Continued research, policy support, and global cooperation are crucial to ensure the transformation of agriculture from a climate-affected system into a climate-resilient one. In conclusion, safeguarding crop production and soil health in the changing climate is not just an environmental necessity but a global responsibility essential for food security, economic stability, and ecological sustainability for present and future generations.

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Enhancement of Carbon Sequestration in the Field through Agricultural Activities

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1. Introduction:

Agriculture plays a dual role as both a significant contributor to global greenhouse gas (GHG) emissions and a critical avenue for climate change mitigation through carbon sequestration. Accounting for approximately 25% of anthropogenic GHG emissions primarily methane (CH₄) from livestock, nitrous oxide (N₂O) from fertilizers, and CO₂ from land-use changes and soil disturbance agriculture is a major driver of climate change (IPCC, 2019). However, agricultural systems also offer substantial potential to act as carbon sinks by capturing and storing atmospheric CO₂ in soil organic carbon (SOC) and biomass. Practices such as conservation agriculture, no-till farming, cover cropping, agroforestry, and biochar application can sequester 0.5–5 t C/ha/year, depending on the practice, soil type, and region (Lal, 2020). The "4 per 1000" Initiative suggests that increasing SOC by 0.4% annually could offset 20–35% of global GHG emissions, aligning with the Paris Agreement's climate goals, though achieving this consistently in temperate regions is challenging due to soil and management constraints (Minasny et al., 2017; Poulton et al., 2018). By adopting these practices, agriculture can transform from a net emitter to a key contributor to global climate mitigation, supporting goals like net-zero emissions by 2050 while enhancing soil health and food security.

2. The Role of Agriculture in Carbon Sequestration

Agriculture dual role as a GHG emitter and a potential carbon sink makes it a pivotal sector for climate change mitigation. Practices such as conservation agriculture, no-till farming, cover cropping, agroforestry, and biochar application can sequester 0.5–5 t C/ha/year, depending on the practice and region (Lal, 2020). The 4 per 1000 Initiative suggests that increasing SOC by 0.4% annually could offset 20–35% of global emissions, aligning with the Paris Agreement's goals (Poulton et al. 2018). By adopting these practices, farmers can transform agricultural landscapes into carbon sinks while maintaining productivity and supporting global climate objectives.

3. The Carbon Cycle in Agricultural Systems:

The carbon cycle in agricultural systems governs the balance between carbon inputs and outputs, determining the potential for soil organic carbon (SOC) storage and its role in climate change mitigation. Carbon inputs include carbon fixed through photosynthesis by crops and cover plants, as well as organic matter from crop residues, manure, and compost. These inputs add to the SOC pool, enhancing soil fertility and carbon sequestration. Carbon outputs occur through soil respiration (microbial decomposition releasing CO₂), erosion (loss of carbon-rich topsoil), and tillage (which accelerates organic matter breakdown). For instance, conventional tillage can release 20-50% of SOC as CO₂ within a few years (Lal, 2004). Optimizing this cycle by increasing inputs and minimizing outputs is critical for transforming agricultural lands into carbon sinks, capable of sequestering 1-5 t C/ha/year depending on practices and conditions (Paustian et al., 2016; Oldfield et al., 2022).

4. Practices for Carbon Sequestration through agricultural activity:

India's agricultural sector, supporting over 50% of the rural population and contributing 18% to GDP, emits approximately 400 million t CO₂ e annually, primarily from rice cultivation, livestock, and land management (IPCC, 2019; Sapkota et al., 2020). However, it holds significant potential for carbon sequestration, estimated at 1-10 Gt CO₂/year globally through sustainable practices, offsetting up to 20% of sectoral emissions while enhancing soil health and farmer incomes (Lal, 2020; Minasny et al., 2017; Paustian et al., 2016). With diverse agro-climatic zones from the Indo-Gangetic Plains to the Himalayas practices like agroforestry and conservation agriculture can sequester 0.5-5 t C/ha/year, aligning with India's net-zero by 2070 pledge (Nair et al., 2009; Government of India, 2022). Over 50 carbon credit projects are active, generating credits via voluntary markets like VERRA, though challenges like measurement, reporting, and verification (MRV) persist (Oldfield et al., 2022; Smith et al., 2019; VERRA, 2023). The following practices, drawn from meta-analyses and field studies in India, focus on enhancing soil organic carbon (SOC), biomass storage, and reducing emissions. They are adaptable to India's rainfed (68% of arable land) and irrigated systems.

1. **Agroforestry Systems** Integrating trees with crops or livestock is highly effective in India, sequestering 1-5 t C/ha/year through biomass and SOC gains (Nair et al., 2009). Common systems include:

- **Agrisilviculture:** Trees like poplar or eucalyptus with crops (e.g., wheat in Indo-Gangetic Plains) yield 180 Mg/ha biomass over 6 years, generating INR 0.72 million/ha income while sequestering 5.05 Mg C/ha/year (Dhyani et al., 2016; Chavan et al., 2023).
 - **Agrihorticulture and Silvopastoral:** Fruit trees with vegetables or grazing in Himalayan foothills increase SOC by 25% and labile carbon pools for long-term stability (Dhyani et al., 2016; Nair et al., 2009).
 - **Boundary Planting and Homegardens:** Low-cost integration on field edges sequesters 83.27 Mg C/ha total, covering 95,748 km² of trees outside forests (Dhyani et al., 2016).
2. **Conservation Tillage and No-Till Farming** Reducing soil disturbance preserves SOC, sequestering 0.5–1 t C/ha/year (West & Post, 2002). In rice-wheat systems, no-till with residue retention cuts emissions by 10–20% and boosts yields (Sapkota et al., 2020; Jat et al., 2020). Combined with rice straw application, it achieves 1.39 Mg C/ha sequestration (Bhattacharyya et al., 2022).
 3. **Cover Cropping and Crop Rotation:** Legumes or grasses between main crops add 0.3–0.6 t C/ha/year via root biomass and nitrogen fixation (Poeplau & Don, 2015). In diverse rotations (e.g., cereals-legumes), SOC rises 5–15%, reducing erosion in rainfed areas (Minasny et al., 2017). Bundled with reduced tillage, effects amplify by 25% in Indian agro-ecosystems.
 4. **Organic Amendments and Biochar** Manure, compost, or biochar (from crop residues) increases SOC by 10–20% over 5–10 years (Gattinger et al., 2012). Biochar sequesters 2.5 t CO₂/t applied, improving fertility in acidic soils; scalable via pyrolysis facilities (Lehmann et al., 2011). In rice systems, rice straw combined with fertilizers yields the highest SOC gains (Bhattacharyya et al., 2022).
 5. **Residue Management and Regenerative Grazing** Retaining rice and wheat residues by eliminating stubble burning in Punjab, Haryana, and western Uttar Pradesh using zero-tillage and mulching sequesters 0.5–2.0 t C/ha/year in the rice–wheat system, with field-verified rates of 0.68–1.85 t C/ha/year under full retention and machinery based management (Sapkota et al., 2017, Lohan et al., 2018; Shyamsundar et al., 2019). Adaptive multi-paddock (AMP) grazing in silvopastoral systems of Rajasthan, Gujarat, and Madhya Pradesh enhances root biomass and soil organic carbon (SOC) by 15–20% (ICAR-Central Soil Salinity Research Institute, 2021).

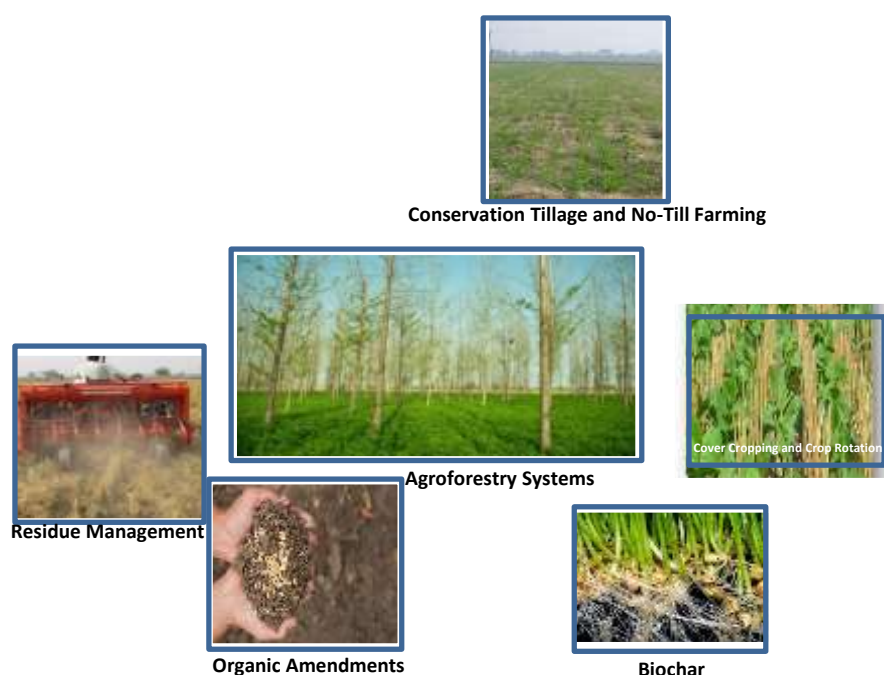


Fig. 1 Options for enhancing SOC sequestration in field through agricultural Activity

Table: Sequestration Potential and Co-Benefits from major agricultural activity

Practice	Sequestration Rate (t C/ha/year)	Regions	Co-Benefits
Agroforestry (e.g., Poplar-based)	1.0-5.0	Indo-Gangetic Plains, Himalayas	Income diversification, erosion control, biodiversity (Nair et al., 2009, Dhyani et al., 2016; Chavan et al., 2023)
Conservation Tillage and No-Till Farming	0.5-10	Rice-Wheat belts (Punjab, UP)	Fuel savings, yield stability, reduced emissions (West & Post, 2002 Sapkota et al., 2020; Jat et al., 2020, Bhattacharyya et al., 2022)
Cover Cropping/Rotation	0.3- 0.60	Rainfed Deccan Plateau	Nutrient cycling, drought resilience (Poeplau & Don, 2015, Minasny et al., 2017)
Organic Amendments/Biochar	1.0-2.5	Tropical/Coastal areas	Soil fertility, water retention (Lehmann et al., 2011 Gattinger et al., 2012)
Residue Management and Regenerative Grazing	0.5-20	Arid/Semi-Arid zones	wheat residues, Pasture quality, livestock health (Sapkota et al., 2017, Lohan et al., 2018; Shyamsundar et al., 2019; ICAR-Central Soil Salinity Research Institute, 2021)

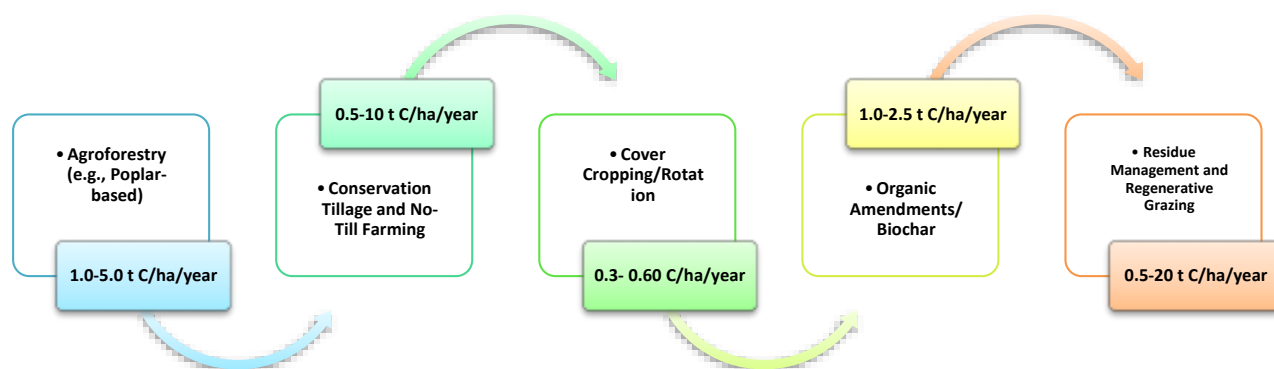


Fig. 1 SOC sequestration potential of various agricultural activity (tons C/ha/year) (Data source: West & Post, 2002, Nair et al., 2009, Lehmann et al., 2011, Poeplau & Don, 2015, Dhyani et al., 2016, Gattinger et al., 2017, Minasny et al., 2017, Shyamsundar et al., 2019, Sapkota et al., 2020; Jat et al., 2020, Bhattacharyya et al., 2022 and Chavan et al., 2023,)

5. Co-Benefits of Carbon Sequestration Practices

Adopting carbon-sequestering practices yields multiple co-benefits, enhancing the sustainability and resilience of agricultural systems:

- 1. Improved Soil Health:** Cover cropping and organic amendments increase SOC, improving soil structure, water retention, and nutrient availability. Cover crops reduce erosion and enhance SOC by 0.3–0.6 t C/ha/year (Poeplau & Don, 2015).
- 2. Increased Crop Resilience:** Higher soil organic carbon (SOC) improves soil's capacity to withstand drought (via enhanced water-holding capacity of 120 m³/ha per 1% SOC increase) and heavy rainfall (via better infiltration and reduced erosion). Agroforestry systems provide shade and windbreaks, reducing crop stress (20–40% lower evapotranspiration) and boosting yields by 10–30% (Minasny et al., 2017; Verchot et al., 2007; Lasco et al., 2014).
- 3. Sustainable Food Production:** Practices like rotational grazing and crop rotation maintain soil fertility, reducing reliance on chemical inputs and supporting long-term productivity (Conant et al., 2017).

6. Challenges and Barriers to Scaling Up

India possesses significant potential to enhance soil organic carbon (SOC) through climate-smart agriculture (CSA). However, the transition from isolated success stories to widespread adoption is hindered by a complex web of interconnected economic, knowledge-based, and policy-related barriers.

1. **Economic and Financial Hurdles:** The economic case for carbon sequestration is often weak or inaccessible for the average Indian farmer, creating a fundamental adoption barrier.

High Upfront Capital Costs: The initial investment required for climate-smart technologies—such as zero-till seeders and happy seeders (costing ₹1.5–2.0 lakh per unit) is prohibitive for the vast majority (86%) of India's small and marginal landholders (Kaur et. al., 2023, Villalba et. al., 2024).

Limited Access to Carbon Finance: Farmer participation in carbon markets remains negligible (<3%) due to high transaction costs for Monitoring, Reporting, and Verification (MRV) and a lack of institutional mechanisms to aggregate and sell credits on their behalf (Cariappa, et. al., 2024, Khurana, et. al., 2024).

Delayed Economic Returns: Practices like agroforestry and organic amendment application deliver SOC and economic benefits over a medium to long-term horizon (3-7 years), which misaligns with the short-term financial needs of resource-constrained households (Pretty and Bharucha, 2014).

2. **Knowledge, Awareness, and Technical Gaps:** A deficit in targeted information and technical support creates uncertainty and impedes effective implementation.

Inadequate Extension Systems: Only about one-third of Indian farmers have reliable access to updated knowledge on CSA, leading to improper implementation of SOC-enhancing practices and sub-optimal results (Barooah, et. al., 2023).

Spatial Variability in Sequestration Potential: SOC gains are highly dependent on local conditions, ranging from 0.2–0.6 t C/ha/yr in conservation agriculture to 1–3 t C/ha/yr in agroforestry systems. This variability creates unpredictability for farmers and policymakers, complicating target-setting and incentive structures (Page, et. al., 2020, Panwar, et. al. 2022,).

Low Awareness of Co-Benefits: Farmers often prioritize immediate yield over long-term soil health. The secondary benefits of SOC sequestration such as improved water retention, reduced fertilizer requirement, and enhanced climate resilience are not widely recognized, diminishing the perceived value of adoption.

3. **Policy, Institutional, and Governance Constraints:** The existing policy landscape is not fully aligned to incentivize and sustain carbon sequestration efforts.

Fragmented and Misaligned Incentives: While schemes like the National Mission for Sustainable Agriculture (NMSA) and PM-KUSUM promote climate-resilient infrastructure, they do not explicitly reward or quantify carbon sequestration outcomes, missing a key motivational lever.

Mismatched Time Horizons: Government programs typically operate on short political and funding cycles (3-5 years), which is insufficient for the long-term process of significant SOC accumulation, leading to discontinued support before benefits are realized.

Underutilized Mitigation Focus in Climate Policy: India's National Determined Contributions (NDCs) rightly emphasize adaptation in agriculture, but this has come at the expense of developing a robust strategy for soil carbon mitigation, leaving a major climate opportunity untapped (IPCC, 2019).

Solutions to Overcome Barriers

Addressing the multifaceted barriers to adopting soil organic carbon (SOC)-enhancing practices in Indian agricultural systems requires a tailored, multifaceted approach that aligns with the country's diverse agro-ecological zones, smallholder-dominated farming (86% of farmers operate on less than 2 ha), and national priorities like food security and the Nationally Determined Contributions (NDCs) under the Paris Agreement.

Financial Incentives: Accelerate the Green Credit Programme (GCP) and Voluntary Carbon Market (VCM) by aggregating smallholders through Farmer Producer Organizations (FPOs), offering ₹2,000–5,000 per t CO_{2e} for verified SOC gains. This offsets initial costs of practices like zero-tillage or agroforestry, directly supporting India's NDC target of a 2.5–3 Gt CO_{2e} carbon sink by 2030 (Khurana et al., 2024; Cariappa et al., 2024).

Education and Extension: Expand the National Innovations in Climate Resilient Agriculture (NICRA) and digital tools like Kisan Suvidha to train over 1 million farmers annually. Use self-help groups (SHGs) and women-led cooperatives to reach underserved rainfed regions and female farmers, increasing CSA adoption by 20–30% and enhancing resilience in drought-prone areas (Barooah et al., 2023).

Research: Prioritize SAUs, ICAR-led, region-specific trials on biochar, conservation tillage, and agroforestry to quantify SOC sequestration (0.4–2.5 t C/ha/yr) across India's 21 agro-ecological zones. This reduces variability-driven risks and yield trade-offs, enabling evidence-based practice selection for diverse soils and climates (Panwar et al., 2022).

Policy Integration: Integrate SOC enhancement into the National Mission for Sustainable Agriculture (NMSA) and the 2023 Carbon Credit Trading Scheme, using Soil Health Cards and remote sensing for robust monitoring, reporting, and verification (MRV). Link carbon payments to PM-KISAN transfers to drive systemic adoption and co-benefits like water retention and food security.

Table 2: Solutions to Overcome Barriers to SOC-Enhancing Practices in Indian Agriculture

Category	Key Actions	Mechanisms	Expected Outcomes	References
Financial Incentives	Accelerate Green Credit Programme (GCP) & Voluntary Carbon Market (VCM)	Aggregate smallholders via Farmer Producer Organizations (FPOs); Offer ₹2,000-5,000 per t CO ₂ e for verified SOC gains	Offset initial costs of zero-tillage, agroforestry; Support India's NDC target of 2.5-3 Gt CO ₂ e sink by 2030	Khurana et al., 2024; Cariappa & Krishna, 2024
Education & Extension	Expand NICRA and digital platforms (e.g., Kisan Suvidha); Train >1 million farmers annually	Leverage SHGs and women-led cooperatives to target rainfed regions and female farmers	Increase CSA adoption by 20-30%; Enhance resilience in drought-prone areas	Barooah et al., 2023
Research	Prioritize SAUs & ICAR-led region-specific trials on biochar, conservation tillage, agroforestry	Quantify SOC sequestration (0.4–2.5 t C/ha/yr) across 21 agro-ecological zones	Reduce yield trade-offs and variability risks; Enable evidence-based practice selection	Panwar et al., 2022
Policy Integration	Integrate SOC enhancement into NMSA and 2023 Carbon Credit Trading Scheme	Use Soil Health Cards + remote sensing for robust MRV; Link carbon payments to PM-KISAN transfers	Drive systemic adoption; Deliver co-benefits (water retention, food security)	-

7. Opportunities and Future Directions

Scaling up carbon sequestration in agriculture offers significant opportunities to align with global climate goals.

Technological Innovations:

8.1 Precision Agriculture: Satellite imagery and AI optimize inputs, increasing sequestration efficiency by 10–20% (Paustian et al., 2016).

Soil Sensors: Real-time SOC monitoring with 0.1% accuracy improves carbon market verification (Nevalainen et. al., 2022).

Carbon Monitoring Tools: Remote sensing and blockchain ensure transparent quantification, with US pilot projects verifying 1–2 t C/ha/year in no-till systems (Oldfield et al., 2022).

Policy Opportunities

National Climate Strategies: Integrating sequestration into NDCs can prioritize funding, as seen in the EU’s €10 billion allocation for climate-friendly practices (European Commission, 2020).

Carbon Markets: Expanding markets could generate \$1–10 billion annually for farmers, as demonstrated by Australia’s Emissions Reduction Fund (Lam et al., 2013).

Global Potential: Widespread adoption of sequestration practices could offset 1–10 Gt CO₂/year, or 10–20% of agricultural emissions (Lal, 2020). Tropical regions show high potential for agroforestry (2–5 tC/ha/year), while temperate regions excel in no-till and cover cropping (0.5–1 tC/ha/year) (Nair et al., 2009).

Global Networks: The 4 per 1000 Initiative unites 40+ countries to share knowledge and resources (Minasny et al., 2017).

Conclusion

Enhancing carbon sequestration through agricultural activities is a critical strategy for mitigating climate change while supporting sustainable food production. Practices like no-till, cover cropping, agroforestry, biochar, and rotational grazing offer significant sequestration potential (0.5–5 t C/ha/year) and co-benefits, including improved soil health, crop resilience, and productivity. However, economic barriers, knowledge gaps, and policy inconsistencies limit adoption. Solutions such as financial incentives, education, research, and policy integration can overcome these challenges. Technological innovations, expanded carbon markets, and global collaboration further enhance the

scalability of these practices. By aligning science, policy, and practice, agriculture can play a transformative role in achieving net-zero emissions by 2070, contributing to a resilient and sustainable future.

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Cutting-Edge Innovations and Sustainable Solutions Shaping the Future of Environmental Science

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Introduction

The future of environmental science is changing due to innovations and sustainable solutions that tackle urgent issues like climate change, biodiversity loss, pollution, and resource depletion. Advances in renewable energy, artificial intelligence, and green chemistry are transforming how we interact with and protect our planet, leading to a new age of environmental responsibility. Recent progress in solar, wind, hydro, and geothermal energy systems is making clean energy more dependable and affordable, significantly cutting our reliance on fossil fuels and greenhouse gas emissions. The rollout of perovskite solar cells, floating solar farms, improved geothermal systems, and smart grids shows the potential for scalable, sustainable power. At the same time, artificial intelligence (AI), Internet of Things (IoT) sensors, and drones are allowing for real-time monitoring of ecosystems, predictive analytics, and better resource management. Green innovation extends beyond energy, including sustainable materials, eco-friendly packaging, bioremediation methods, and circular economy models that reduce waste and enhance resource efficiency. Breakthroughs in bioremediation and green chemistry use natural and synthetic biology to clean pollutants, create safer products, and repair damaged environments. These sustainable methods are also seen in smart agriculture systems, carbon capture technology, sustainable hydrogen production, and improved energy storage. All these elements contribute to a resilient, low-carbon global economy. The use of these technologies is sparking significant change across industries and communities. From climate adaptation and resilience planning to citizen science involvement and policy reforms, environmental science professionals are leading efforts to protect vulnerable populations, conserve resources, and plan for sustainable growth. Strong regulatory frameworks and business tactics further promote environmental innovation, supporting a shift to a healthier and more sustainable planet. This chapter looks at how new innovations and sustainable solutions are reshaping environmental science, not just as an area of study but as a basis for action, policy, design, and restoration. We will examine key areas of innovation: data-driven monitoring, renewable energy and storage, advances in materials and the circular economy, nature-based and biomimetic strategies, and the connection between systems thinking, adaptation, and resilience.

In each area, we will outline the technology or method, its potential, notable examples, upcoming challenges, and the impact on the future of environmental science, including research, practice, policy, and education.

2. Renewable Energy, Energy Storage & Decarbonization

2.1 Advanced Solar, Wind & Novel Renewables

The shift from fossil fuels to renewable energy is crucial for sustainability. However, the next phase involves more than just installing more panels and turbines. It focuses on smarter, more efficient systems, new materials, and incorporating renewables into larger systems. For instance, advancements include tandem solar cells with multiple layers that increase efficiency, bifacial solar panels, and flexible solar membranes. Wind technologies are also progressing. This includes floating offshore wind farms, variable speed turbines, and the integration of storage to stabilize output.

2.2 Energy Storage, Green Hydrogen & Carbon Capture

Energy storage, green hydrogen, and carbon capture are three key areas in modern environmental science. They are essential for moving towards sustainable energy systems and reducing climate impact.

2.2.1 Energy Storage

New energy storage technologies make it easier to use renewable resources by providing electricity on demand and stabilizing the grid (Popescu et al., 2025). Key innovations include:

Lithium-ion batteries: These are the preferred choice for grid storage because they are cost-effective and efficient. New types like sodium-ion and solid-state batteries offer better sustainability and resource security.

Flow batteries: Enhanced designs, such as organic and metal-air flow batteries, provide long life, scalability, and low degradation. They are well-suited for large-scale integration of renewable energy sources.

Compressed Air Energy Storage: This method stores energy as compressed air in underground facilities. It can be released and converted into electricity, helping to manage fluctuations in the grid. These technologies reduce dependence on fossil fuels, address the variability of renewable sources, and improve grid resilience.

Green Hydrogen

Green hydrogen is created by using renewable energy to electrolyze water, resulting in no greenhouse gas emissions. It is valued for its ability to serve as a clean fuel in hard-to-electrify sectors, such as heavy industry and transportation. It helps with industrial decarbonization and utilizes surplus renewable energy (Popescu et al., 2025). However, it faces challenges like high production costs, infrastructure needs for transportation and storage, and ongoing technology improvements. As technology develops and costs decrease, green hydrogen is poised to have a significant impact on future energy systems.

2.2.3 Capture Technologies

Carbon capture means capturing carbon dioxide emissions from industrial processes or directly from the air. It helps address climate change. This technology is vital for reducing emissions in sectors like cement, steel, and chemical production (Valavanidis, 2016; Popescu et al., 2025). These sectors are hard to electrify and contribute a large amount of CO₂ emissions. There are a few main methods in carbon capture, utilization, and storage:

2.2.3.1 Utilization and Storage

Once captured, CO₂ can be transported to storage sites. Here, it is stored in deep geological formations, such as depleted oil and gas reservoirs or salt beds. This method ensures permanent containment. Alternatively, CCUS technologies enable the conversion of CO₂ into useful products like chemicals, fuels, and building materials. Ongoing research also explores biological methods, such as growing algae and mangrove trees, which naturally absorb carbon and provide long-term storage.

2.2.3.2 Market Growth and Impact

The CCUS market is growing quickly because of stricter climate policies, higher carbon prices, and more government support. Investments in large-scale CCUS infrastructure are expected to hit \$80 billion worldwide by 2030, with North America and Europe leading the way. As projects increase in size and costs go down, CCUS will capture and store a large part of the world's CO₂ emissions. It will play a key role in reaching net-zero goals and helping industries that cannot completely decarbonize by switching to electricity alone. In summary, carbon capture is at a critical point. Driven by new technology, supportive policies, and rising investment, it is set to become a key part of global efforts to reduce carbon emissions as we work toward a more sustainable future.

2.3 Implications & Challenges

Implications: Environmental science now merges with energy systems science. Understanding the life cycle impacts, system boundaries, and integration across sectors is essential. Decarbonization strategies involve not just energy but also land use, agriculture, buildings, and transport.

Challenges: Cost and scalability are key issues. Storage remains expensive. Green hydrogen infrastructure is still developing. Carbon capture has unresolved questions about permanence, leakage, cost, and energy use. Policies need to support technology deployment, and supply chains must be sustainable, for example, in mining for battery materials.

3. Materials Innovation, Circular Economy & Waste-Reduction Solutions

3.1 Biodegradable, Bio-Based & Advanced Materials

Biodegradable, bio-based, and natural materials are quickly becoming important in the effort for sustainable development. They offer eco-friendly options to traditional petroleum-based products while tackling environmental pollution and resource depletion (Popescu et al., 2025).

Biodegradable Materials

Biodegradable materials are made to break down into natural substances like water, carbon dioxide, and biomass through biological processes. This helps reduce long-lasting plastic waste. The biodegradable plastics market is growing quickly because of pressure from regulations, rising consumer demand, and tech improvements that boost the durability and performance of these materials in areas such as packaging, agriculture, and textiles. However, challenges still exist in making sure these materials fully degrade in natural settings and do not leave behind microplastic residues.

Bio-Based Materials

Bio-based materials come from renewable biological sources like plants, algae, and organic waste, instead of fossil fuels. These materials include biopolymers such as polylactic acid, polyhydroxyalkanoates, and bio-based polyamides. They perform similarly to traditional plastics but greatly reduce carbon footprints. Production methods now often use microbial fermentation and composite formation to improve material properties and sustainability.

Advanced Materials

New materials in this sector include new features such as better biodegradability, mechanical strength, and environmental adaptability. Innovations involve adding substances for photodegradation, making composites of synthetic and natural polymers, and creating biodegradable electronics to address e-waste. These materials support circular economy principles by allowing reuse, recycling, and safe disposal. Together, these materials play a key role in reducing plastic pollution, conserving resources,

and helping the shift towards a more sustainable and circular material economy that matches global environmental goals.

3.2 Waste-to-Value, Upcycling & Circularity

Beyond material innovation is the broader idea of the circular economy. This approach focuses on eliminating waste, keeping materials in use, and regenerating natural systems. For instance, selective upcycling of plastics using catalytic technologies turns waste into valuable products instead of downcycling or sending it to landfills. Other innovations address ocean plastics, such as floating “Seabins” that capture plastic debris and collect IoT data. Thirumalaiyammal et al. 2024). Smart recycling systems also play a role through AI-driven sorting and chemical recycling.

3.3 Implications & Challenges

- Implications: Environmental scientists must work with ecosystems and industrial ecologies, including life cycle analysis, industrial metabolism, and material flows. Education and curriculum need to include design thinking, materials science, and industrial ecology.
- Challenges: Bio-based materials often struggle with performance, scalability, and cost. The “bio” label doesn't guarantee low impact; it needs careful evaluation. Transitioning to a circular economy requires significant changes in industries, supply chains, consumer behavior, and regulations.

4. Nature-Based and Biomimetic Solutions

Nature-based and biomimetic solutions are new strategies that use the power of natural ecosystems and processes to tackle environmental problems, build resilience, and support sustainable development. These approaches focus on collaborating with nature, imitating biological systems, and restoring ecosystems to deliver ecological, social, and economic benefits amid climate change and resource pressures.

4.1 Nature-Based Solutions

Nature-based solutions involve protecting, restoring, or managing natural ecosystems to lessen environmental impacts and adapt to climate change. They consist of practices like reforestation, wetland restoration, sustainable agriculture, and managing coastal ecosystems, such as planting mangroves to reduce flooding, erosion, and storm impacts. NbS effectively sequester carbon dioxide, improve biodiversity, enhance water and air quality, and boost community resilience. International policy increasingly acknowledges the importance of NbS; over 84% of countries involved in climate commitments include these solutions in their national plans. These strategies not only help cut greenhouse gases but also provide additional benefits like reducing disaster risks, saving money, and

improving ecosystem health. Examples include restoring coastal wetlands to guard against storm surges or using agroforestry and sustainable land management practices to increase carbon storage.

4.2 Biomimetic Solutions

Biomimicry focuses on designing materials, structures, and systems inspired by biological processes and organisms to create efficient, sustainable technologies. For example, researchers develop water filtration systems based on mollusk shells or energy-efficient buildings that take cues from the natural ventilation found in termite mounds (Popescu et al., 2025). These innovations aim to lower energy use, cut down waste, and promote circular economies by imitating nature's effective solutions.

4.3 Applications of Biomimicry

- Water resource management: Structures that mimic natural buffers and wetlands can control flow, enhance water quality, and lower flood risks.
- Material development: Creating biodegradable plastics based on natural decomposition processes or developing sustainable textiles inspired by spider silk.
- Renewable energy: Designing wind turbines that copy the flapping of bird wings or ocean wave energy devices influenced by marine creatures.

Nature-based and biomimetic solutions play a vital role in building sustainable futures. They address climate and environmental issues through eco-friendly practices and innovative designs that reflect nature's efficiency and resilience.

4.4 Implications & Challenges

- Implications: Environmental science is becoming more interdisciplinary. Ecology, engineering, design, and economics intersect. Studies must include a holistic view of natural processes, the value of ecosystem services, and non-linear interactions. Monitoring and evaluation methods need to evolve.
- Challenges: Scaling up nature-based solutions and biomimicry is still tough. How can small-scale successes be replicated in larger infrastructure or landscape settings? Who pays the costs, who gains the benefits, and how are unexpected consequences managed? Also, issues with classification, standardization, metrics, and policy alignment still exist.

5. Climate Adaptation and Resilience Innovations

5.1 From Mitigation to Adaptation

While reducing emissions remains vital, the increasing reality of climate impacts means more emphasis is now given to adaptation and resilience-designing systems to withstand, absorb, and recover from shocks. According to one review, 2025 marks “a shift from mitigation to adaptation” in

many areas: vulnerability assessments, disaster risk reduction, resilient infrastructure, and planning for future scenarios.

5.2 Soft Robotics, Autonomous and Smart Systems for Environmental Remediation

Emerging technologies underpin new approaches to restoration and adaptation. For instance, the field of soft robotics is proposed to address sustainable development and climate action by deploying autonomous robots in land and ocean preservation, disaster remediation, and clean energy support.

5.3 Implications & Challenges

- 1) Implications: Environmental science becomes proactive: not only asking “how bad is it?” but “how do we design for a changed world?”. Researchers will need to embrace scenario planning, risk analysis, and resilient design. Systems thinking and socio-ecological approaches become core.
- 2) Challenges: Many adaptation solutions are context-specific and localized; scaling and financing remain concerns. Equity issues loom large: vulnerable communities may lack the capacity to implement high-tech adaptive systems. Measuring resilience and attributing impact is complex.

While reducing emissions remains important, the growing reality of climate impacts means we now emphasize adaptation and resilience. This involves designing systems that can withstand, absorb, and recover from shocks. One review states that 2025 will be a turning point, shifting focus from mitigation to adaptation in several areas: vulnerability assessments, disaster risk reduction, resilient infrastructure, and planning for future scenarios.

6. Data-Driven and Digital Technologies

6.1 Sensor Networks, IoT & Remote Sensing

One of the biggest changes in environmental science is the surge of data. Sensors, satellites, drones, Internet of Things (IoT) networks, and machine learning systems now allow for real-time, high-resolution monitoring of ecosystems, pollutants, land use, and air and water quality. Recent surveys show that by 2025, technologies like AI, IoT sensors, and drone platforms will allow for ecosystem monitoring at scales we couldn't have imagined before. For instance, drones with imaging spectrometers have been used to check forest health and biodiversity in remote areas. They provide details and speed that far exceed traditional ground surveys. These tools support new decision-making methods, such as early warnings for floods, wildfires, and deforestation, tracking illegal logging or poaching, and mapping biodiversity through environmental DNA and remote sensing.

6.2 Artificial Intelligence and Machine Learning

Collecting data is just the first step. The real power comes from analyzing it, finding patterns, forecasting, and suggesting actions. AI and ML play a key role in environmental science. They help model how pollution spreads, predict how ecosystems respond, optimize resource use, and even design resilient infrastructure (Al-Darkazali et. al. 2025). For example, AI and ML reportedly transform environmental monitoring by offering better predictive abilities and real-time data analysis. Another area of exploration is using quantum computing for climate models. The work "Quantum Computing for Climate Resilience and Sustainability Challenges" suggests that quantum algorithms might handle the scale and complexity of climate modeling better than traditional methods.

6.3 Implications & Challenges

- 1) **Implications:** Environmental scientists need to understand data science, think systemically, and work across different fields. Monitoring should be ongoing instead of occasional. Management and policy can become more flexible and based on evidence.
- 2) **Challenges:** Big data tools can lead to bias and misinterpretation. Sensor networks require infrastructure and upkeep. Equity issues arise, such as who monitors and who owns the data. Concerns about privacy and governance also exist. Data overload can make it hard to find useful insights.

7. Integrative Insights & The Future of the Discipline

7.1 Blurring Disciplinary Boundaries

The innovations mentioned earlier highlight a common theme: the lines between environmental science, data science, engineering, materials science, systems design, policy studies, and business are quickly fading. Future environmental scientists will work in teams that include sensors and AI, ecology, and industrial process design. This requires changes in educational programs, institutional structures, and funding models.

7.2 Metrics, Governance & Ethical Considerations

High-tech solutions bring new governance issues: data ownership, privacy, algorithmic bias, technological lock-in, unintended ecological effects, and access equity. The circular economy and biomaterials raise concerns about biodegradability, sourcing, land use, and trade-offs. Environmental science must increasingly incorporate ethics, governance, and justice into its practice.

7.3 Future Scenarios and Research Directions

- 1) Digital twins of ecosystems: combining sensor networks, high-resolution remote imagery, and AI models to create live "digital twins" of landscapes. This allows for real-time management, simulation, and "what if" planning.

- 2) Material systems as carbon sinks: materials that are not just inert but also actively capture carbon. This includes bio-based composites that store carbon for decades.
- 3) Negative emissions infrastructure at scale: deploying technologies like DAC, CCU, bioengineering, and nature-based systems widely as part of a common environmental strategy.
- 4) Resilient landscapes and cities: integrated design of urban, peri-urban, and rural systems that produce food, energy, materials, water, and ecosystem services, closing loops and building resilience.
- 5) Socio-ecological transformation: moving beyond technological fixes to a systemic redesign of business models, consumption patterns, governance, and behavior change, connecting human systems with planetary boundaries.

Conclusion

The future of environmental science is shaped not only by what we know but by how we act and innovate. As sensors and AI reveal insights into our planet's dynamics, as renewable energies and materials change our industrial systems, as biomimicry and nature-based solutions ground our technologies in living systems, and as adaptation and resilience become essential, the field is undergoing a significant transformation. This moment offers unprecedented opportunity for students, researchers, practitioners, and policymakers. If we embrace interdisciplinary approaches, link innovation with equity and justice, connect knowledge to implementation, and adopt a systems mindset, we can address the planet's challenges. These challenges are vast, but so are the tools we have to meet them. Environmental science must shift from simply diagnosing issues to providing solutions: from understanding ecosystems to designing regenerative ones; from monitoring change to actively shaping it. In the chapters that follow, we will explore several areas of innovation, including material solutions, carbon removal technologies, urban ecosystem design, and more. This overview highlights a key message: sustainability is no longer just an aspiration but increasingly a frontier for technological, ecological, and social innovation. The science of the environment is evolving into the science for the environment.

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Climate Change in the 21st Century: Risks, Resilience, and Policy Frameworks

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Climate change represents a long-term alteration in global and regional climate patterns driven by both natural processes and, predominantly, human activities. Rising concentrations of greenhouse gases due to fossil fuel combustion, deforestation, and land-use change have intensified the greenhouse effect, resulting in higher global temperatures, extreme weather events, and widespread environmental disruptions. The consequences of climate change are evident across ecosystems, oceans, and the atmosphere, affecting human health, infrastructure, and biodiversity. Agriculture, one of the most climate-sensitive sectors, faces declining crop yields, altered rainfall patterns, increased pest and disease incidence, soil degradation, water scarcity, and reduced livestock productivity, ultimately threatening global food and nutritional security. Poor and vulnerable populations are disproportionately affected due to limited adaptive capacity and higher exposure to climate related hazards. To address these challenges, climate change adaptation and disaster risk reduction (DRR) frameworks aim to build resilience through integrated planning, sustainable resource management, and institutional coordination. International mechanisms such as the UNFCCC, Kyoto Protocol, and Paris Agreement guide global climate governance, while national strategies like India's NAPCC help align climate action with sustainable development priorities.

Keywords: climate change, environmental disruptions, greenhouse effect, productivity, strategies.

Introduction to Climate Change

To understand climate change, we first need to understand the difference between weather and climate. Weather refers to the actual atmospheric conditions that are being experienced now. It also includes changes that are forecast over the next few days, for example, in temperature and rainfall. Climate refers to the kind of weather that's typically expected in a region. This includes describing the range of conditions that are possible. Climate change is a long-term shift in the average weather conditions of a region, such as its typical temperature, rainfall, and windiness. Climate change means that the range of conditions expected in many regions will change over the coming decades. This means that there will also be changes in extreme conditions.

The climate varies naturally from year to year and decade to decade. This is caused by natural processes linking the atmosphere, ocean and land, as well as variations in heat output from the sun. In addition to changes in climate that are caused by natural climate variability, climate change can be caused by human activity. The kind of climate change we are experiencing now is being caused primarily by these human factors (IPCC, 2021).

Basics of Climate Change (NASA, 2020)

- Changing temperature and precipitation patterns.
- Increases in ocean temperatures, sea level, and acidity.
- Melting of glaciers and sea ice.
- Changes in the frequency, intensity, and duration of extreme weather events.
- Shifts in ecosystem characteristics, like the length of the growing season, timing of flower blooms, and migration of birds.
- These changes are due to a buildup of greenhouse gases in our atmosphere and the warming of the planet due to the greenhouse effect.

Climate Science

Climate science studies how changing climates affect the natural order on a global level. Rising global temperatures bring with them the potential to raise sea levels, change precipitation and local climate conditions (Dessler, 2021).

Climate Change Science

The earth's climate is changing. Multiple lines of evidence show changes in our weather, oceans, ecosystems, and more. EPA is committed to providing the most up-to-date and accurate climate-science information, including basics, causes, indicators, and impacts of climate change (EPA, 2022).

Causes of Climate Change Current climate change is mainly caused by human activities that release greenhouse gases to the atmosphere. The sun's energy warms the Earth and the warmed Earth releases heat to the atmosphere. Certain gases in the atmosphere trap this heat and act like the glass of a greenhouse. Such gases are called greenhouse gases. The main greenhouse gases are water vapour, carbon dioxide and methane. Greenhouse gases absorb heat and radiate some of it back to the Earth, raising surface temperatures. This process is often called the greenhouse effect. The greenhouse effect is a natural process, but it is being intensified by human activities that increase greenhouse gas levels in the atmosphere, especially carbon dioxide. Increasing greenhouse gas levels in the atmosphere

makes it more effective at trapping heat, resulting in overall warming of the earth. Burning fossil fuels (coal, oil, gas) and some industrial processes are the main sources of carbon dioxide. Climate change caused by human activity is referred to as anthropogenic climate change (IPCC, 2014).

Impacts of Climate Change

Climate change affects people all over the world. Young and old people are particularly at risk from extreme heat or air pollution. Scientists are studying global climate change and studying how it affects our environment and our health. There are some effects like sea-level rising from the melting of glaciers. There are more dramatic impacts like animal extinctions, more heat waves, heavy rain in some areas and drought in others. Weather disasters increase costs for homeowners, insurers and governments. Extreme weather damages important infrastructure like schools, hospitals and roads. High temperatures impacted agriculture, horticulture, and forestry (WMO, 2020).

Anthropogenic Contributors to Climate Change

People who are studying the climate believe that most contemporary climate change is caused by humans. Burning fossil fuels, cutting down forests and converting land for agricultural, industrial, and other human purposes is causing atmospheric CO₂ to increase rapidly, mostly from fossil fuel emissions. Plants and the ocean remove CO₂ from the atmosphere. Fossil fuel emissions and the destruction of natural “sinks” have disrupted the natural carbon cycle that kept greenhouse gases in the atmosphere relatively constant for the thousands of years before industrialisation. As carbon dioxide in the water increases, the water becomes more acidic over time (UNEP, 2019).

Impact on Agriculture Climate change contributes to declining crop productivity, income losses, asset losses, and food insecurity. Climate change has contributed to higher temperatures and altered weather patterns. Scientists predict global crop production will decline in the near future. Agriculture is particularly vulnerable. In 2017, India was home to more than 17 percent of the world’s population. Food and nutritional security remain key challenges. To keep pace with the growing food demand, it is estimated that Indian agriculture needs to grow at least at the rate of 2.0 percent annually (ICAR, 2018). Indian Council of Agricultural Research is working to increase the productivity of agriculture crops such as rice, wheat, ornaments, medicinal and aromatic plants and horticultural crops. Scientists have increased productivity of agriculture, livestock, and fishery sectors through research and development activities (ICAR, 2018).

Climate Change Adaptation

Climate change adaptation includes adjusting to long-term environmental risks. People adapt to the climate in many different ways. Adaptation helps individuals, communities and organizations deal with new climate realities. This includes altering lifestyle choices or infrastructure, and changing environmental or social policies. Climate change and environmental risks pose significant challenges for development strategies and poverty eradication (UNFCCC, 2007).

Disaster Risk Reduction (DRR)

Disaster Risk Reduction (DRR) focuses on dwindling the risks associated with disasters – everything from earthquakes to floods to hurricanes. DRR includes strategies for managing environmental risks. The goals of DRR are managing disaster risks, preventing disasters, minimizing impacts of disasters and building resilience in individuals, communities, industries, and governments. Countries must integrate DRR into development plans, laws and business operations. Increasing urbanization has intensified the impacts of disasters in cities (UNDRR, 2015).

Differences between Climate Change Adaptation (CCA) and Disaster Risk Reduction (DRR)

- CCA addresses climate-related disasters (like hailstorms and avalanches) but does not address hazards that are unrelated to climate, like earthquakes and tsunamis.
- CCA focuses on long-term shifts in climate patterns due to human activity or natural variability and focuses on managing effects of these changes.
- Climate change is a permanent change affecting the environment and permanently altering the life of people.
- DRR addresses all types of hazards, climate-related and non-climate-related.
- DRR focuses on reducing risks associated with specific events and focuses on vulnerability to hazards and reducing those vulnerabilities.
- DRR focuses on specific actions intended to reduce disaster risks.

Both DRR and CCA focus on minimizing environmental risks. Experts say that coordination between DRR and CCA is essential to reduce global disaster losses. Countries need to strengthen action, policies and investments for improved risk assessment, preparedness and enhanced climate outcomes (UNDRR, 2020).

Impact on the Poor

Climate change impacts human health, food systems, water supplies, infrastructure, and energy systems in growing urban areas. Some effects are particularly severe for poor populations. Climate change may drive millions into poverty. Many people live in areas that are becoming more vulnerable

due to sea-level rise, droughts, and floods. Poor communities have limited access to clean water, healthcare, and food, and may lack resources to adapt (World Bank, 2016).

United Nations Framework Convention on Climate Change (UNFCCC)

While the global North has historically caused the most CO₂ pollution, its effects impact the poorest countries and poorest people the most. The UNFCCC (1992) convention showed the relationship between the economy, technology, and high levels of CO₂ emissions. The goal of the UNFCCC is to stabilize greenhouse gas emissions at safe levels. The Convention established frameworks, rules and processes for international cooperation on mitigation, adaptation, finance, and technology (UNFCCC, 1992).

UNFCCC Institutions

The Conference of the Parties, UNFCCC (COP) is the decision-making body. It is held every year and decisions are made by consensus of all Parties. There are 198 Parties to the Convention. The Paris Agreement works with the UNFCCC. The Paris Agreement is a legally binding international treaty on climate change that was adopted by 196 Parties at COP 21 in Paris in 2015. The UNFCCC also includes subsidiary bodies, expert groups, multilateral funds, organizations and agencies which contribute to climate action (UNFCCC, 2022).

Kyoto Protocol

In 1997, the Kyoto Protocol was signed in Kyoto, Japan, and entered into force on 16 February 2005 in Montreal, Canada. Kyoto Protocol commitments also included international cooperation in clean development and sustainable development programmes. Kyoto Protocol (KP) outlines greenhouse gas emissions targets which are legally binding on Annex I countries. These include the United States, the European Union, and Japan (UNFCCC, 2005).

Kyoto Protocol – Flexibility Mechanisms (KP Mechanisms)

- International Emissions Trading (IET)
- Clean Development Mechanism (CDM)
- Joint Implementation (JI)

Countries engage in flexible and cost-effective ways. It helps developing countries to contribute towards sustainable development and build climate resilience. These mechanisms support businesses, organizations and governments to engage in greenhouse gas reduction activities (UNFCCC, 2006).

National Action Plan on Climate Change (NAPCC)

Eight national missions: National Solar Mission, National Mission for Enhanced Energy Efficiency, National Mission on Sustainable Habitat, National Water Mission, National Mission for Sustaining Himalayan Ecosystem, National Mission for a “Green India”, National Mission on Sustainable Agriculture, National Mission on Strategic Knowledge for Climate Change. India’s climate and sustainable development priorities: clean and efficient energy systems / healthy living / sustainable mobility / waste to wealth / water conservation / resource efficiency / air pollution (Government of India, 2008).

Strengthening Institutional Arrangements

Major institutions:

- Ministry of Environment, Forest and Climate Change (MoEFCC)
- Sectoral Ministries
- NITI Aayog

Sponsor consultative groups like the Prime Minister’s Council on Climate Change (PMCCC). India’s climate governance system coordinates national missions, policies, and schemes that focus on long-term climate action and sustainable development. It strengthens coordination between ministries, promotes research, builds institutional capacity, and aligns national and state-level climate strategies (Government of India, 2015).

Climate Change and Agricultural Productivity: Deepening the Impacts Agriculture is one of the most climate-sensitive sectors, and even small variations in temperature, rainfall, or humidity can significantly impact crop growth, soil properties, and overall productivity. Climate change has already begun influencing key physiological processes in plants, including photosynthesis, respiration, and transpiration. While rising CO₂ levels may offer short-term benefits for some crops through enhanced photosynthesis, these benefits are often offset by higher temperatures, moisture stress, pest outbreaks, and declining nutrient content in grains. Studies show that staples such as wheat, rice, and maize experience reductions in protein, iron, and zinc concentrations under elevated CO₂ conditions, directly affecting food quality and nutrition security (Smith and Myers, 2019)

In tropical countries, where temperatures are already near the upper threshold for crop tolerance, a further increase of even 1°C can drastically reduce yields. Heat stress during the flowering stage leads to pollen sterility, grain abortion, and poor grain filling. Crops such as wheat and maize are particularly vulnerable to heatwaves, which shorten growing periods and accelerate water loss. Increasing night-

time temperatures, which many regions are experiencing, also reduce yields by increasing plant respiration rates, leading to lower biomass *accumulation* (Lobell *et al.*, 2011).

Effects of Changing Rainfall Patterns on Farming Systems

Irregular rainfall is among the most visible impacts of climate change on agriculture. Rain-fed agriculture, which supports most smallholder farmers in Asia and Africa, is increasingly exposed to high uncertainty. Regions that once relied on predictable monsoon patterns now face delayed onset, long dry spells during crop-growing stages, and sudden heavy downpours that cause flooding and soil erosion. This variability disrupts sowing calendars, increases the risk of crop failure, and reduces the reliability of traditional farming knowledge (Rao *et al.*, 2014).

Floods wash away topsoil rich in organic matter, reducing soil fertility. Droughts harden the land, degrade soil structure, and reduce groundwater recharge. In many regions, farmers have been forced to switch crop varieties, alter planting schedules, or even reduce the area under cultivation. In India, erratic rainfall patterns have also reduced the success of post-monsoon crops, with rabi (winter) crops increasingly dependent on residual soil moisture that is now less reliable (ICAR, 2020).

Soil Degradation and Climate Stress

Climate change accelerates soil degradation through increased temperature, heavy rainfall, reduced vegetation cover, and frequent extreme events. Soil erosion, salinization, nutrient depletion, and loss of organic carbon are becoming widespread problems. Higher temperatures increase the rate of organic matter decomposition, reducing soil carbon stocks that are essential for soil fertility and water retention. Heavy rains and floods remove fertile topsoil, which takes decades to naturally rebuild (FAO, 2017).

In coastal areas, sea-level rise causes saltwater intrusion into agricultural lands. Salinity reduces crop growth by inhibiting water uptake and damaging root systems. Farmers in these regions are being forced to adopt salt-tolerant varieties, shift to aquaculture, or move away from traditional livelihoods (World Bank, 2016).

Impact on Livestock and Dairy

Livestock systems are extremely vulnerable to climate stress. Rising temperatures reduce feed intake in animals, lower milk production, and increase susceptibility to diseases. Heat stress reduces fertility in cattle and poultry, leading to lower reproduction rates and increased economic losses. Droughts impact pasture availability, forcing farmers to depend on expensive supplementary feed (Thornton *et al.*, 2009).

Water scarcity affects livestock directly and indirectly reduced water availability not only limits drinking water for animals but also lowers fodder production. The increased prevalence of livestock diseases, such as tick-borne infections, is linked to warmer temperatures and shifting humidity levels. Dairy production, which is sensitive to temperature, faces declining yields, especially in tropical regions where cows are often exposed to high heat stress (Nardone *et al.*, 2010).

Pest and Disease Dynamics

Climate change has altered pest and pathogen distribution. Warmer temperatures, higher humidity, and erratic rainfall support rapid multiplication of insects such as locusts, whiteflies, and fall armyworms. Due to warmer winters, many pests now survive year-round instead of being eliminated seasonally, leading to heightened crop damage (CABI, 2018).

Additionally, climate variability has led to the emergence of new agricultural diseases and the spread of existing pathogens to previously unaffected regions. For example, rice blast disease and wheat rust have expanded their geographical range, causing serious yield losses. Increased pest and disease pressure increases farmers' reliance on pesticides, leading to higher production costs and environmental harm (FAO, 2019).

Water Scarcity and Irrigation Challenges

Agriculture consumes nearly 70% of the world's freshwater resources. Climate-induced changes in rainfall, glacier melt, and river flow directly affect irrigation systems. Reduced river flow and falling groundwater levels are becoming major constraints for staple food crops. In South Asia, millions rely on major river systems fed by Himalayan glaciers, which are rapidly melting. This temporarily increases water flow but will ultimately lead to long-term water shortages (WMO, 2018). Groundwater over-extraction due to irregular rainfall is leading to widespread aquifer depletion. Canal irrigation is also affected, as many canals depend on monsoon-fed reservoirs that increasingly fill below capacity (Central Water Commission, 2020).

Climate Change and Agricultural Labour

Climate change influences not only crops but also agricultural labour. Higher temperatures reduce the number of workable hours for farm labourers, risking heatstroke, exhaustion, and productivity loss. Labor shortages during critical agricultural periods, especially harvesting, have been reported in several regions due to rising temperatures (ILO, 2019).

In countries where agriculture is the primary rural livelihood, diminished labour capacity affects household income and leads to greater migration toward cities. Women who form a major portion of agricultural workers face increased workloads as climate stress reduces water availability and increases time spent collecting water or fuelwood (UN Women, 2020).

Climate Change and Food Security

Food security rests on four pillars: availability, access, utilization, and stability all of which are threatened by climate change. Declining yields reduce food availability; increased prices affect economic access; reduced nutrient quality affects utilization; and climate variability undermines stability (IPCC, 2019).

Regions dependent on agriculture for income and food is particularly vulnerable. Climate shocks reduce household food reserves and purchasing power, pushing more families into hunger and malnutrition. Children suffer most, as climate-driven food shortages increase stunting, wasting, and micronutrient deficiencies (UNICEF, 2021).

Conclusion

Climate change is a long-term shift in weather patterns primarily driven by human activity. It has far-reaching effects on agriculture, ecosystems, public health, infrastructure, and vulnerable populations, especially the poor. Adaptation, mitigation, and disaster risk reduction must be integrated into national and global strategies to address the threats posed by climate change. Frameworks such as the UNFCCC, Kyoto Protocol, and India's National Action Plan on Climate Change help countries coordinate efforts toward achieving sustainable development, reducing environmental risks, and building climate-resilient societies.

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Emerging Frontiers in Soil Fertility

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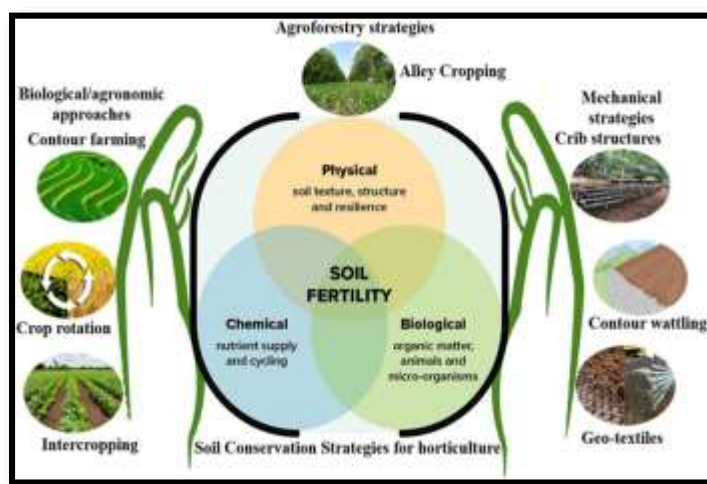
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Introduction

Soil fertility is the foundation of global food security and sustainable agriculture. In 2025, scientific and technological innovations are transforming how soil fertility is studied, monitored and managed.

The integration of machine learning, remote sensing and geoinformatics enables precise mapping of soil nutrients and data-based fertilizer recommendations that enhance productivity while reducing environmental impact. Simultaneously, the use of biological amendments such as biochar, compost and microbial

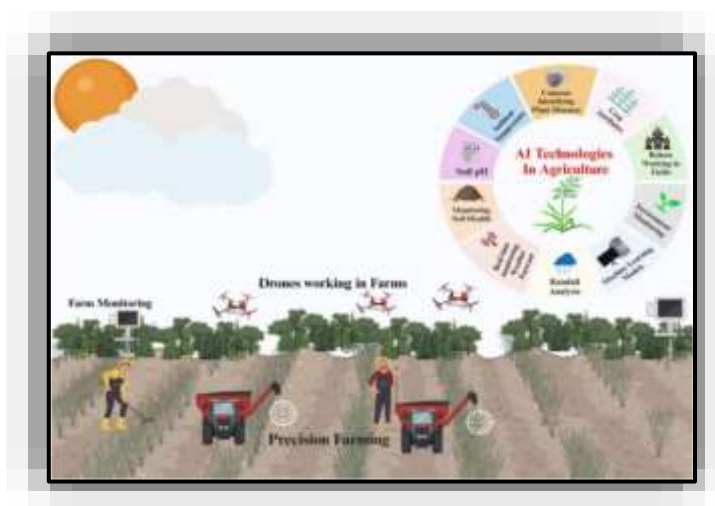


inoculants is shifting fertility management from chemical-based systems toward biologically driven nutrient recycling. These amendments enhance soil organic carbon, stimulate beneficial microbial activity and improve nutrient use efficiency- ensuring long-term soil health and productivity. The preservation and restoration of soil assets are critical to ensuring horticulture's long-term sustainability. Soil properties, irrigation, and nutrient availability must all be managed effectively for horticultural facilities to last and maintain ecological balance (Timmis and Ramos, 2021). Research in climate-smart agronomy underscores the role of soil in carbon storage and resilience against drought and degradation. Advanced methods, including soil micro biome and enzyme profiling, are revealing how microorganisms regulate nutrient transformations and contribute to soil stability under changing climates.

Together, these developments represent a new era of integrated and intelligent soil management, combining digital technology, biological innovation and ecological principles to achieve sustainable productivity and environmental balance.

Recent Advancements in Soil Fertility:

- **AI and Machine Learning-based Soil Fertility Modeling** – predictive analytics for real-time nutrient estimation and soil health diagnostics.
- **Advanced Geoinformatics and Remote Sensing** – precision fertility mapping using satellite, UAV and GIS-integrated data.
- **Biochar Engineering and Biological Amendments** – tailored carbon-based materials improving nutrient retention and soil structure.
- **Soil Microbiome Engineering** – use of metagenomics and microbial consortia for nutrient cycling and stress resilience.
- **Climate-Smart Soil Management** - strategies enhancing carbon sequestration, water retention and drought tolerance.
- **Nuclear and Isotopic Tracing Techniques** – molecular-level tracking of nutrient transformations and fertilizer efficiency.
- **Precision and Decision Support Systems (DSS)** - IoT and cloud-based tools for fertilizer scheduling and soil monitoring.
- **Sustainable Organic and Conservation Practices** – integrated nutrient management using cover crops, composts and green manures.
- **Nanotechnology in Soil Fertility** - nano-fertilizers and sensors for controlled nutrient release and monitoring.
- **Socio-technical Innovations** - participatory digital platforms improving farmer access to soil data and training.



1. Machine Learning and Real-Time Analytics

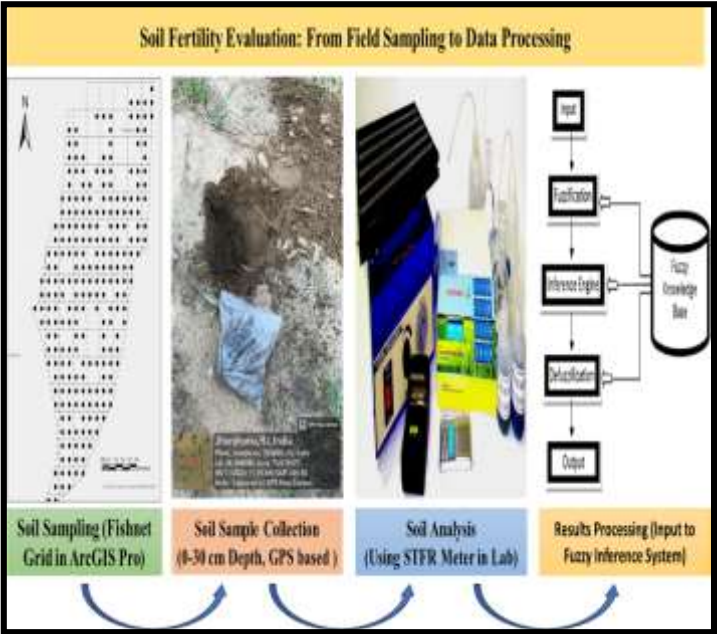
Recent breakthroughs in Machine Learning (ML) and Artificial Intelligence (AI) have transformed soil fertility assessment by making it significantly faster, more precise and data-driven. Advanced algorithms such as Random Forest (RF), Gradient Boosting (GBM) and Long Short-Term Memory (LSTM) neural networks can accurately predict soil fertility based on multiple parameters including pH, organic carbon content, soil texture, cation exchange capacity and weather data. By learning complex relationships among these variables these models can achieve over 90% prediction accuracy thereby supporting highly reliable and site-specific fertilizer recommendations.

The integration of IoT (Internet of Things)-enabled soil sensors with AI platforms has further enhanced real-time monitoring capabilities. These sensors continuously measure soil moisture, nutrient concentration, temperature and electrical conductivity transmitting data to cloud-based analytics systems for instant processing. The combination of AI models and IoT systems enables dynamic soil fertility mapping, precision nutrient management and improved Nutrient Use Efficiency (NUE). Additionally this technology minimizes fertilizer losses, reduces nutrient runoff and promotes environmentally sustainable soil management. Together AI and ML-based systems represent a paradigm shift toward smart, sustainable and resilient agricultural practices ensuring both productivity and ecological balance.

2. Geoinformatics and Spatial Zonation

Geoinformatics and spatial-analysis tools have indeed become central to modern fertility-management practices. High-resolution data from satellites, UAVs and GIS-based surveys help map soil variability across landscapes

Advanced fuzzy-logic models and kriging interpolation are now routinely used to delineate fertility zones for precision input application. This spatial approach enables location-specific fertilizer recommendations, reduces input wastage, supports soil-health monitoring programmes at regional and national scales, and when integrated with machine-learning frameworks, further refines soil-fertility prediction models.



Recent research supports this trend. For example, a study developing a “macro- and micronutrient-based soil fertility zonation using fuzzy logic and geospatial techniques” found that a fuzzy-GIS framework generated high-resolution fertility maps with strong crop-yield correlations (R^2 values up to ~ 0.92) using kriging interpolation. Likewise, a 2025 paper on “Soil Fertility Prediction Using Spatiotemporal Graph Neural Networks (STGNN)” combined spatial and temporal data to improve prediction accuracy by 3.8-6.4 % over traditional ML models, demonstrating how the embedding of spatial context enhances prediction. Another recent investigation used visible-near-infrared spectroscopy (VNIR) integrated with machine-learning (SVM, Random Forest, CNN) to rapidly estimate soil-fertility indices (SFI), achieving an R^2 around 0.80 for multiple soil properties

3. Climate-Smart Soil Management

Climate-smart soil management integrates soil fertility enhancement with climate resilience, aligning agricultural productivity with environmental sustainability. Practices such as conservation tillage, residue retention and diversified crop rotations play a dual role in maintaining soil health while mitigating greenhouse gas (GHG) emissions. Conservation tillage reduces soil disturbance, preserves soil organic matter and minimizes CO_2 release, whereas residue retention enhances soil organic carbon and nutrient cycling. Crop diversification-especially with legumes and cover crops-improves nitrogen fixation, reduces pest pressure and stabilizes yields under variable climatic conditions. Together, these strategies optimize nutrient use efficiency, reduce input dependency and enhance the soil’s biological activity, thereby contributing to both climate change mitigation and adaptation.

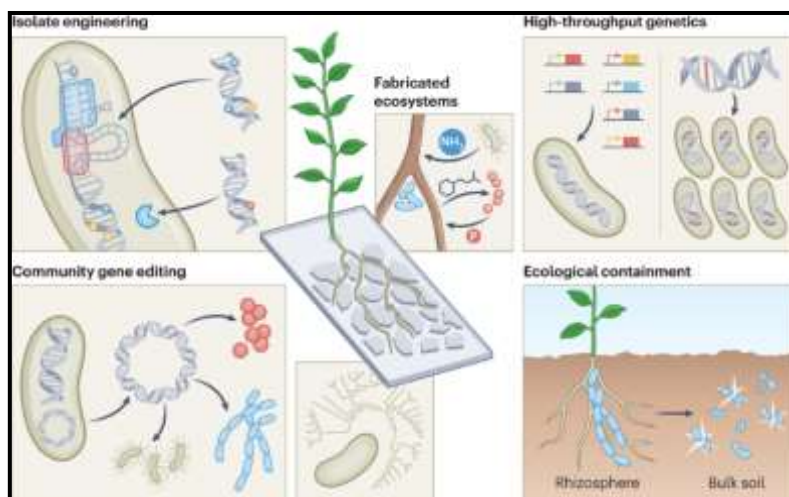
Recent scientific research emphasizes the carbon sequestration potential of deep-rooted crops, biochar application and organic amendments such as compost and vermicompost. These interventions enhance the soil’s capacity to store carbon in stable forms, improving aggregate stability and water-holding capacity. Enhanced soil organic carbon also strengthens soil resilience to drought, salinity and flooding-three key stressors intensified by climate change. Advances in remote sensing, AI-based soil carbon mapping and biogeochemical modeling are now enabling the quantification and monitoring of soil carbon dynamics with high accuracy. Overall, climate-smart soil management represents a holistic



approach that links fertility restoration, carbon storage and ecosystem resilience, ensuring sustainable agricultural productivity under a changing global climate.

4. Soil Microbiome Engineering

Recent advances in metagenomics, transcriptomic and enzyme-based assays have transformed the understanding of soil microbiota and their essential role in maintaining soil fertility. The soil microbiome, comprising bacteria, fungi, actinomycetes and archaea, regulates key biogeochemical processes such as nitrogen fixation, phosphorus solubilization and organic matter decomposition. With the help of next-generation sequencing and molecular profiling, scientists can now identify beneficial microbial species and understand their interactions within the rhizosphere. These insights have led to the development of synthetic



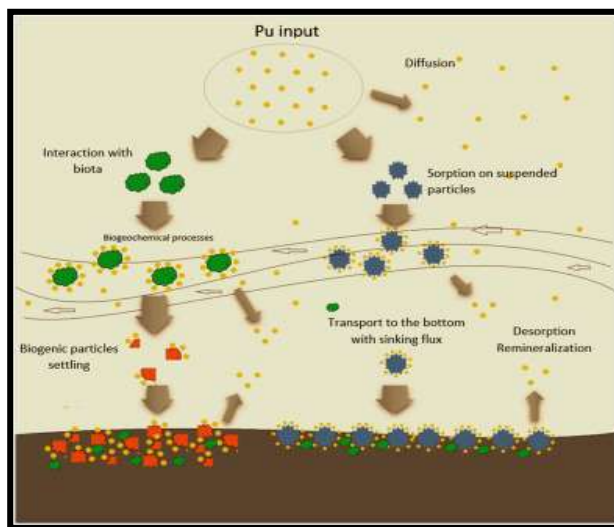
microbial consortia and bio-fertilizers designed to enhance nutrient availability and promote plant growth. Engineered microbial formulations are being tailored to specific crops and soil types, allowing precise management of soil fertility with minimal ecological disturbance.

At the molecular and systems level, research on the soil–microbe–root interface is uncovering new mechanisms of nutrient exchange and signalling. Studies on root exudates, microbial metabolites and enzyme activity are providing critical data for designing microbial-based nutrient delivery systems. Such innovations significantly reduce dependence on chemical fertilizers, improve nutrient use efficiency and strengthen the resilience of agroecosystems against stress factors such as drought and salinity. Overall, soil microbiome engineering represents a frontier in sustainable agriculture, offering a biologically driven approach to maintaining fertility, enhancing productivity and protecting soil health.

5. Nuclear and Isotopic Techniques: Nuclear and isotopic techniques have become powerful tools in modern soil fertility research, providing precise and quantitative insights into nutrient dynamics, water movement and soil–plant interactions. The use of stable and radioactive isotopes such as nitrogen-15 (^{15}N), phosphorus-32 (^{32}P), sulfur-35 (^{35}S) and carbon-13 (^{13}C) allows scientists to trace the movement,

transformation and utilization of nutrients within the soil–plant system. These tracers help in identifying nutrient losses through leaching, volatilization or denitrification, thereby improving fertilizer management strategies. Isotopic dilution and enrichment methods enable accurate estimation of biological nitrogen fixation by legumes and other crops, while ^{13}C studies provide valuable data on soil organic carbon turnover and sequestration rates under different land-use and management practices.

Recent advances in mass spectrometry and nuclear sensors have enhanced the precision of isotope ratio measurements, making isotopic analysis faster and more environmentally safe. Nuclear techniques such as neutron moisture meters and gamma-ray spectrometry are also being used to study soil water content, bulk density and sub-surface nutrient distribution without disturbing the soil structure. The integration of these nuclear and isotopic tools with remote sensing and machine learning further refines predictive models for nutrient use efficiency and soil fertility monitoring. Overall, nuclear and isotopic techniques offer a scientific, non-destructive and highly accurate means to understand and optimize soil fertility, contributing to the development of sustainable and resource-efficient agricultural systems.



6. Precision and Decision Support Systems

Precision agriculture and decision support systems (DSS) have emerged as integral components of modern soil fertility and crop management. Precision agriculture relies on spatial and temporal data to manage variability within agricultural fields, ensuring that inputs such as fertilizers, irrigation and pesticides are applied at the right place, right time and in the right quantity. Tools such as GPS-guided machinery, remote sensing, unmanned aerial vehicles (UAVs) and soil sensors collect real-time data on soil moisture, nutrient status and crop growth. This data is processed through Geographic Information Systems (GIS) and predictive algorithms to create high-resolution fertility



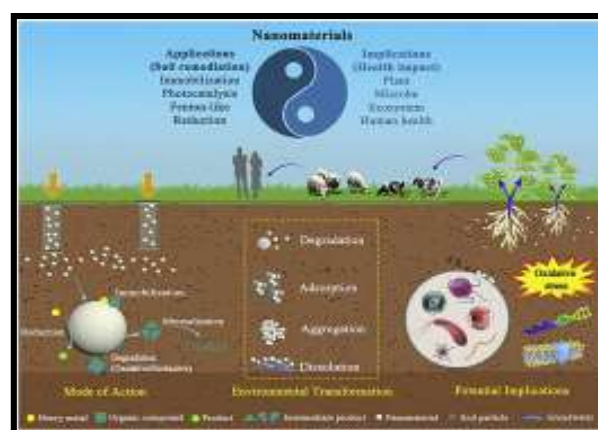
and productivity maps. As a result, farmers can make informed decisions that optimize nutrient use efficiency (NUE), enhance yield quality and minimize environmental impacts such as nutrient leaching and greenhouse gas emissions.

Decision Support Systems (DSS) combine agronomic models, weather forecasts, soil data and machine learning algorithms to provide actionable recommendations for nutrient management and crop planning. These systems simulate various management scenarios, helping users evaluate fertilizer schedules, irrigation regimes and crop rotations based on economic and environmental outcomes. Cloud-based DSS platforms now allow real-time data integration from IoT-enabled devices, enabling adaptive management even under changing climate conditions. Recent advances in AI and big data analytics have further improved DSS accuracy, allowing region-specific and crop-specific nutrient prescriptions. Together, precision agriculture and decision support systems represent a data-driven revolution in sustainable farming—bridging the gap between technology, science and practical field management for enhanced soil fertility and food security.

7. Nanotechnology in Soil Fertility:

Nanotechnology has emerged as a transformative approach in soil fertility management, offering innovative solutions for efficient nutrient delivery, soil remediation and sustainable productivity. Nanoparticles, typically ranging from 1 to 100 nanometers in size, possess unique physicochemical properties such as high surface area, reactivity and controlled release potential. These characteristics make them ideal for formulating nanofertilizers that enhance nutrient availability, reduce losses through leaching or volatilization and improve plant uptake efficiency. For instance, nano-urea, nano-phosphate and nano-zinc formulations provide a slow and targeted release of nutrients directly to the rhizosphere, minimizing environmental pollution and improving crop yield. Studies have shown that nanofertilizers can increase nutrient use efficiency (NUE) by 20–40% compared to conventional fertilizers while reducing input requirements.

In addition to nutrient management, nanotechnology plays a crucial role in soil health monitoring and remediation. Nano sensors are being developed to detect soil parameters such as moisture, pH, and nutrient concentrations in real time, facilitating precision agriculture practices. Furthermore, engineered nanoparticles like nano-clay, nano-zeolite and nano-biochar have shown promise in



immobilizing heavy metals, enhancing soil structure and improving water retention capacity. Recent research also explores the integration of nanomaterials with beneficial microbes to create nano-biofertilizers, which combine the advantages of biological and nano-scale inputs. Despite its vast potential, careful assessment of the environmental fate and toxicity of nanoparticles is essential to ensure safe and sustainable application. Overall, nanotechnology in soil fertility represents a frontier that merges material science, biology and agronomy to achieve efficient, eco-friendly and resilient agricultural systems.

8. Biochar and Soil Amendments

The use of biochar has gained significant scientific attention for its dual role in enhancing soil fertility and mitigating climate change. Biochar is a stable, carbon-rich material produced through the pyrolysis of organic biomass such as crop residues, wood, or manure under limited oxygen conditions. When applied to soil, biochar improves soil structure, porosity and water-holding capacity, creating a more favorable environment for root growth and microbial activity. It also enhances nutrient retention by increasing the soil's cation exchange capacity (CEC), thereby reducing nutrient losses through leaching or volatilization. Biochar acts as a slow-release nutrient carrier, particularly when combined with organic compost or mineral fertilizers, promoting balanced nutrient availability and sustained crop productivity.

Recent research has focused on the development of engineered and functionalized biochars modified with essential nutrients or nanoparticles to further enhance their performance. These advanced biochars exhibit improved CEC, pH buffering capacity and microbial colonization potential, leading to better soil fertility and biological activity. In addition to agronomic benefits, biochar plays a vital role in carbon sequestration, as its stable carbon structure resists microbial decomposition, locking atmospheric CO₂ into the soil for hundreds of years. This contributes to climate change mitigation by reducing greenhouse gas emissions and improving soil resilience under stress conditions such as drought or salinity. Overall, biochar technology represents a sustainable and scientifically proven approach to achieving soil regeneration, nutrient efficiency and environmental stability in modern agriculture.

9. Organic and Ecological Approaches

Organic fertility management promotes soil health through biological nitrogen fixation, composting and green manuring. These approaches enhance organic matter, microbial diversity and natural nutrient cycling.

Emerging innovations include liquid biofertilizers, vermicompost teas and plant-growth-promoting rhizobacteria (PGPR) formulations. Combining these with digital monitoring enables organic systems to maintain fertility while meeting productivity targets.

Challenges and Future Directions

- ✓ Despite technological progress, challenges persist in scaling innovations to smallholder farmers. Issues include limited access to technology, data literacy and infrastructure gaps in developing regions. There is also a need to integrate socio-economic and ecological perspectives into fertility research.
- ✓ Future work will emphasize affordable sensor systems, biodegradable controlled-release fertilizers and open-access digital platforms. Collaborative research linking AI modeling, microbiome engineering and participatory soil management will define the next decade of soil fertility science.

Conclusion

The year 2025 marks a pivotal moment in the evolution of soil fertility research. The convergence of digital analytics, biological amendments and climate-smart management is reshaping how soils are understood and managed. By merging technology with ecological wisdom, agriculture is transitioning toward data-informed, resource-efficient and environmentally balanced production systems. These innovations promise not only higher yields but also resilient and regenerative soils-ensuring global food security for future generations.

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Physiological disorders of Fruit Crops

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Fruit crop physiological disorders are aberrant states that are not brought on by insects or diseases, but rather by non-pathogenic factors such as environmental stress, nutritional imbalances, water problems, and hormonal irregularities. Fruit cracking, blossom-end rot, and browning are just a few of the symptoms that these illnesses induce. They can be controlled by treating their underlying causes, which include temperature swings, mineral deficiencies (such as calcium, copper, and zinc), inadequate watering, and poor soil health. Fruit crop problems are caused by environmental factors such as temperature, light, aeration, and nutritional imbalances, as well as disturbances in plant tissue metabolic activity. Micro-nutrient deficiencies, such as those in Zn, Bo, and Mn, cause far greater problems in fruit crops than macro-nutrient deficiencies.

A. Physiological disorders of Mango:

1. Spongy Tissue in Mango: This physiological abnormality, which is a significant issue in Alphonso, causes fruit pulp to remain unripe due to unhydrolyzed starch as a result of physiological and biochemical disruptions brought on by high temperatures, convective heat, and sun exposure after harvest.

Caused by: The main causes of mango spongy tissue include high temperatures and uneven ripening, which can result in a physiological condition where the pulp does not form normally. Convective heat during development, calcium insufficiency, water stress, and exposure to sunshine after harvest are further significant variables

Primary causes

- **High temperature and uneven ripening:** High temperatures during fruit growth may prevent the pulp from ripening properly, producing a mushy, spongy patch, especially in varieties like Alphonso.
- **Convective heat:** Heat emanating from the ground may result in uneven ripening that impacts the fruit's inside.
- **Post-harvest exposure:** Exposing fruits to excessive heat or sunlight after harvesting can trigger the disorder.

Symptoms: Mangoes' spongy tissue is only noticeable when the fruit is sliced open and does not exhibit any outward symptoms. Internal symptoms include mushy, spongy, pale yellow or off-white pulp that occasionally has air cavities and may taste and smell bad. The impacted flesh frequently lacks the proper firmness and succulence and is mealy.

Management: Mangoes' spongy tissue can be managed with a mix of pre-harvest and post-harvest techniques, such as managing environmental elements including high temperatures and sunlight, applying pre-harvest pesticides, and handling and storing them carefully. Fruits should be shielded from direct sunshine, mulched to regulate soil temperature, harvested at the proper ripeness, handled properly, and cooled after harvest..

- **Environmental control:** Use strategies like fruit bagging, mulching, shading, and windbreaks to protect trees and fruit from high temperatures, direct sunlight, and heat stress.
- **Pre-harvest sprays:** Applying salt solutions, nutrient mixes, or specific formulations like Arka Saka Nivarak to fruits between 50% and 70% maturity can help prevent the onset of spongy tissue.
- **Balanced nutrition:** Implement a balanced nutrition plan tailored to the specific requirements of the mango tree.

2. Biennial Bearing in Mango:

This is one of the most pressing issues since it makes growing mangoes less profitable for the growers. Another name for a biennial bearing is an alternative bearing. It shows yield fluctuations every other year, such as a year with heavy or optimal fruiting followed by a year with minimal or no fruiting. The issue of biennial bearing is a genetically controlled varietal trait that begins to manifest in mango plants as early as the second year of fruiting and gets worse with age.

Caused by:

- **Tree exhaustion:** A heavy crop load in an "on" year depletes the tree's nutritional reserves, making it unable to produce many flowers in the following year.
- **Hormonal imbalance:** High levels of gibberellins, produced by the developing fruit, can inhibit flowering in the next year.
- **Nutrient imbalance:** A high Carbon to Nitrogen (C/N) ratio can contribute to biennial bearing.

Management and control measures:

- **Deblooming:** This technique lowers the crop load in a "on" year by removing some of the flowers, which might assist balance the production in the "off" year.

- **Chemical applications:** To assist control flowering, some chemicals can be used. Spraying potassium nitrate, paclobutrazol, or a combination of potassium nitrate and NAA are a few examples.
- **Pruning:** Regular and appropriate pruning can help regularise bearing and is crucial for maintaining the health of the tree.
- **Cultural practices:** Over time, consistent bearing can be enhanced by maintaining the orchard with efficient pest and disease control, supplying sufficient irrigation, and applying specified rates of fertiliser and manuring.

3. Fruit Drop in Mango:

The percentage of hermaphrodite flowers and immature fruits in mangos is at least 99%. During the first three weeks of April, the commercially grown kinds, Langra and Dasheri, are more prone to drop. The fruit drop can be divided into three stages: (i) pinhead drop, (ii) post-setting drop, and (iii) May-month drop. This process is essentially continuous. In contrast to the third phase, which has a substantial impact on the ultimate output and requires greater attention, the fruit drop in the first two phases is negligible.

Caused by:

- **Natural thinning:** Mango trees naturally shed a large percentage of flowers and young fruits to ensure the remaining ones can develop properly.
- **Environmental factors:** Strong winds, high temperatures, drought, or excessive rainfall can stress the tree and cause fruit drop.
- **Nutritional deficiencies:** Lack of essential nutrients like boron, calcium, and potassium weakens the fruit and its attachment to the stem.
- **Pests and diseases:** Pests like mango hoppers and mealy bugs, as well as diseases like anthracnose and powdery mildew, can weaken fruits and cause them to fall.
- **Poor pollination:** Insufficient pollination and fertilization can lead to early fruit drop.

Management strategies

- **Nutrient management:** Spray Borax (0.2%) before flowering to improve pollination, Apply calcium nitrate (1%) during the fruit development stage to strengthen fruit walls and Spray potassium nitrate (1-1.5%) to help reduce fruit drop.
- **Pest and disease control:** Regularly inspect for pests and diseases, Apply appropriate insecticides and fungicides as needed and Remove any infected leaves or fruits to prevent spread.

- **Plant growth regulators:** Apply NAA (20 ppm) or Alar (100 ppm) at the "pea stage" of fruit development to help control drop.
- **Watering and mulching:** Maintain consistent soil moisture. Avoid both drought and overwatering and Mulch the area around the tree to retain moisture and regulate soil temperature.
- **Pruning:** Prune the tree to improve air circulation and light penetration, and remove weak or diseased branches.

3. Black Tip in Mango:

Black tip is a dangerous condition caused by coal emissions from brick kilns that contain carbon monoxide, sulphur dioxide, and ethylene. Mango orchards up to 200 meters away from brick kilns have been shown to have damage. Fruit damage begins at the marble stage, when the tissues at the distal end begin to turn yellow. The colour gradually becomes more intense, turning brown and then black. At this point, the fruit's growth and development are slowed down, and the black ring at the tip reaches the upper portion of the fruit.

Caused by:

- **Air pollution:** Proximity to brick kilns and industrial areas is a major cause, as their fumes contain toxic gases like sulfur dioxide, carbon monoxide, and ethylene.
- **Boron deficiency:** A lack of boron in the soil can also contribute to the disorder.
- **Environmental factors:** Irrigation, tree condition, and management practices can influence the severity of the problem.

Symptoms

- **Initial signs:** The first signs typically appear at the marble stage, with a yellowing of the fruit's tip.
- **Progression:** The yellow tissue gradually turns brown and then black.
- **Damage:** The affected area spreads upward, slowing the fruit's growth and development.
- **Tissue breakdown:** The inner part of the black tip can become soft and rotten, sometimes secreting a dark liquid.
- **Affected varieties:** Some mango varieties are more susceptible than others. The Dasheri variety is highly susceptible, while the Lucknow Safeda is less susceptible.

Management

- **Distance from pollution sources:** Planting new orchards at a sufficient distance from brick kilns is crucial.

- **Boron application:** Sprays of borax can be applied, especially at the "pea stage" of fruit development, followed by two more sprays at 15-day intervals.
- **Orchard orientation:** Another way to manage the issue is to orient the orchard. Planting mango orchards five to six km from brick kilns and in a north-south direction may significantly lower the occurrence of black tip. Spraying borax (1%) or other alkaline solutions, such as 2% sodium carbonate, might help reduce the occurrence of black tip. At the pea stage, the first borax spray should be applied favourably, followed by two further sprays spaced 15 days apart.

4. Mango Malformation in Mango:

There are two types of mango malformations: vegetative and floral. The vegetative deformity typically affects young plants' seedlings, causing the buds to enlarge tiny shoots to emerge with short inter nodes at the apical section, and a structure like a witch's broom. When there is a floral malformation, the inflorescence appears as a cluster because the panicles are malformed, the axis is short, and the rachis is thick. The majority of malformed panicles are male and have larger flowers than normal flowers.

Symptoms

- **Vegetative malformation:** Short internodes and tiny, scaly leaves, as well as abnormal, stunted, and swollen growth, particularly in early seedlings.
- **Floral malformation:** The growth of a dense, compact cluster of flowers that do not open, with some buds turning into green branches. This kind can result in a 50–80% yield reduction and is the most harmful.
- Other symptoms include stunted growth, brittle and malformed leaves, and branches that resemble a "witches' broom."

Caused by:

- **Primary cause:** The fungus *Fusarium mangiferae* is the primary causal agent. Other strains like *Fusarium moniliforme* var. *subglutinans* have also been identified.
- **Contributing factors:** Other factors, such as certain viruses, mites, and nutritional deficiencies, can also contribute to the disease or increase susceptibility.
- **Environmental factors:** Environmental conditions, particularly low temperatures during the flowering period, can increase the incidence of malformation.

Management

- **Pruning:** Remove and destroy malformed shoots and branches during pruning.

- **Sanitation:** Ensure that pruning equipment is cleaned between uses to prevent the spread of the fungus.
- **Planting material:** Use certified plant material from reliable sources to avoid introducing the disease.
- **Chemical treatments:** Various chemical treatments have shown effectiveness, including fungicides, plant growth regulators like NAA and ethrel, and other agents like copper oxychloride.
- **Biological control:** Use biological control agents such as Trichoderma.
- **Nutritional management:** Applying humic acid and boron has been shown to improve tree health and reduce malformation.

5. Clustering in Mango ('Jhumka') in Mango: The formation of fruitlets in clusters at the tips of panicles is a characteristic of a fruiting condition known locally as "Jhumka." After a month of fruit set, these fruits fall and stop growing past the pea or marble stage. The main cause is that there aren't enough pollinators in the orchards. The use of synthetic pyrethroids, elderly and overcrowded trees, indiscriminate insect and disease treatment, Dashehari monoculture, and unfavourable weather during flowering are additional factors contributing to the illness.

Caused by:

- **Poor pollination and fertilization:** This is the primary cause, often linked to a lack of pollinators.
- **Adverse weather:** Inclement weather, such as low temperatures, during the fruit set period can trigger the disorder.
- **Pesticide use:** Heavy spraying of certain pesticides can kill pollinating insects, leading to a lack of pollination.
- **Improper fruit embryo development:** The fruits abort with shriveled embryos, indicating a role for proper embryo growth in further development.

Management and control

- **NAA (naphthalene acetic acid) spray:** Applying NAA at concentrations of 100-300 ppm during November can help reduce the occurrence of clustered panicles.
- **Borax spray:** Spraying a 0.6% borax solution at 10–14 day intervals starting from fruit set is another effective control method.
- **Pest and weather management:** Managing pests and protecting the trees from adverse weather conditions can also help prevent Jhumka.

B. Physiological disorders of Banana

1. Formation of slitted fingures in Banana

Caused by:**Water inconsistency**

- **Inconsistent watering:** Fruit splitting in many plants, including bananas, is mostly caused by irregular moisture levels. The fruit absorbs a lot of water too quickly when dry spells are followed by heavy rains or overwatering. The peel cracks open due to the abrupt internal pressure.
- **Rapid growth spurts:** The fruit's pulp experiences abrupt development spurts that the outer skin is unable to handle due to the fruit's rapid and irregular water intake. Environmental conditions
- **High relative humidity and temperature:** High relative humidity and temperature can hasten the ripening process, weakening the peel and increasing its susceptibility to splitting. This may occur during post-harvest storage or in the field.
- **Temperature extremes:** Abrupt, sharp changes in temperature put the fruit under stress and may cause it to crack.
- **Ripening on the plant:** Bananas that are allowed to fully mature on the plant are more likely to split because of their exposure to erratic external conditions. Commercial bananas are harvested when still green and grown under strict supervision to prevent this.
- **Nutrient imbalance:** Fruit splitting may result from imbalances caused by excessive nitrogen in the soil, which promotes rapid foliage growth at the expense of fruit development.

Management strategies

- **Optimal harvesting time:** Harvesting bananas at the right ripeness is the best management practice. When the fingers are plump but still green, cut the bunch off the plant. The blooms at the tips of the fingers should be dry, and the individual fruits should feel hard.
- **Balanced nutrition:** Make sure the plant is well fertilised, especially with potassium, which aids in water regulation. Applying borax as a foliar spray or to the soil will help stop fruit cracking in regions where deficits are known.
- **Consistent irrigation:** To avoid stress caused by irregular water availability, stick to a regular watering plan. Don't allow the plants dry out too much before giving them a quick, intense watering.
- **Proper spacing:** Bananas should not be planted too near to one another. This guarantees adequate air circulation, which aids in controlling the humidity and temperature surrounding the growing bunches.

2. Chilling Injury in Banana:

Bananas that are exposed to temperatures below their ideal range—typically below 13°C—develop chilling damage, a physiological disease.

Caused by:

- **Low temperatures:** Bananas are tropical fruits and are susceptible to damage when stored below their critical temperature threshold.
- **Improper storage:** Refrigeration is the most common cause of chilling injury.
- **Exogenous stress:** Other factors like mechanical damage, abrasions, and impacts can also contribute to or exacerbate chilling injury.

Symptoms

- **Surface discoloration:** The peel develops smoky or greyish patches, which can progress to brown or black discoloration.
- **Failure to ripen:** The fruit cannot ripen normally, meaning the starch does not convert to sugar, and the characteristic banana flavour and colour do not develop.
- **Internal browning:** In severe cases, the flesh can turn brown.
- **Reduced flavour:** The taste may become off or bland.

Prevention

- **Proper temperature control:** Store bananas at temperatures above 13°C raised to the composed with power cap C 13°C.
- **Gradual pre-conditioning:** Avoid sudden temperature changes by gradually adjusting the fruit's temperature before exposure to cold environments.
- **Use of protective packaging:** Insulated packaging can help maintain a more consistent temperature during transport.
- **Physical and chemical treatments:** Techniques like low-temperature conditioning, controlled atmosphere storage, and chemical treatments can also help alleviate chilling injury.

3. Choke throat in Banana

Low temperatures have an impact on the plant's ability to develop actively. The tissue dies and the leaves turn yellow. When the period of flowering coincides with a low temperature, the bunch cannot fully emerge from the pseudo stem. In the case of usually flowering plants, the stalk holding bunches elongates freely so that the complete inflorescence emerges out of the pseudo stem and hangs down. The basal portion of the inflorescence becomes lodged at the throat while the distal portion emerges. That's why it's called choke throat. The bunch takes 5–6 months to reach maturity rather than 3.5–4 months to harvest.

Caused by:

- **Low temperature:** This is the most common cause. Cold conditions slow down the plant's growth at a crucial time, so the bunch can't emerge from the pseudostem in time.
- **Nutritional deficiencies:** Lack of essential nutrients like nitrogen, phosphorus, and zinc can contribute to the disorder.
- **Water stress:** Both drought and waterlogging can trigger the condition.

Symptoms

- The fruit bunch gets trapped at the "throat," or the top of the pseudo stem.
- The distal part of the bunch emerges, but the basal part remains stuck.
- Leaves at the top of the pseudo stem may become tightly bunched together.
- The bunch can be misshapen, leading to lower quality fruit.

Prevention and management

- **Shelterbelts:** Planting windbreaks like Casuarina or Eucalyptus around the orchard can protect against cold winds.
- **Variety selection:** Plant low-temperature tolerant varieties.
- **Balanced nutrition:** Ensure the plant receives the proper balance of nutrients, as deficiencies can worsen the problem.
- **Water management:** Avoid both waterlogging and severe drought stress through proper irrigation and drainage.

C. Physiological disorders of Citrus

1. Fruit Splitting in citrus

It is mostly caused by intense irrigation during hot weather or high air humidity after heavy rains. There have been two forms of splitting identified: transverse and radial. Compared to transverse cracking, radial cracking is more frequent. Disease-causing organisms like *Aspergillus*, *Alternaria*, *Fusarium*, and *Penicillium* frequently infect the fruit's fractured surface, causing partial rotting and early fruit dropping off trees.

Caused by:

- **Water stress:** After a drought, an unexpected flood of water may force the fruit to swell too rapidly, shattering the rind.
- **Environmental fluctuations:** The rind may become less elastic during hot, dry spells, increasing its susceptibility to splitting when moisture levels fluctuate.

- **Nutrient deficiencies:** A thinner, structurally weaker rind that is more prone to breaking can result from a lack of calcium and potassium. A lack of boron may also be a factor, particularly in immature fruit.
- **Fruit characteristics:** Larger fruits are more susceptible to cracking.
- **Genetics:** Some varieties and rootstocks are more prone to cracking than others.
- **Sunlight:** Fruit developing in full sun can be warmer, increasing the tendency to split.

Prevention and management

- **Irrigation:** To prevent drastic variations in moisture, water frequently. Water thoroughly and deeply, and think about watering in advance of hot, dry spells.
- **Nutrition:** Use a balanced fertiliser, paying particular attention to calcium and potassium, which can be applied topically or in the soil. Take care of any inadequacies in boron.
- **Crop load management:** Reducing the number of fruits on the tree can reduce stress.
- **Rootstock selection:** Choose crack-resistant varieties or rootstocks when possible.
- **Growth regulators:** Fruit cracking is decreased and fruit weight and size are improved by applying potassic fertilisers correctly, maintaining regular hydration during the summer around bearing trees, and spraying 40 ppm NAA after the monsoon crop's fruit set (the third week of May). Additionally, GA₃ 10–20 ppm can enhance fruit quality and somewhat lessen fruit breaking.

2. Fruit Drop in Citrus

Water stress, high temperatures, nutritional shortages, insect pests, illnesses, and inadequate fertilisation or pollination are some of the main causes.

Caused by:

Physiological factors

- **Nutrient deficiencies:** Fruit drop might result from the tree being weakened by low levels of vital nutrients.
- **Hormonal imbalance:** Growth regulator imbalances can interfere with fruit's proper development.
- **Water stress:** Both drought and overwatering can induce fruit drop.
- **Poor tree health:** An unhealthy tree is more susceptible to dropping its fruit.

Environmental factors:

- **Extreme temperatures:** High heat and frost can stress the tree and cause fruit to drop.
- **High humidity:** Fruit drop can result from excessive humidity, particularly when combined with other stresses.

- **Inadequate sunlight:** Proper flowering and fruiting might be hindered by insufficient direct sunshine.
- **Pathological and entomological factors:**
- **Pests:** Insect infestations, such as fruit flies can cause premature fruit drop.
- **Diseases:** Pathogens that spread to the plant can trigger fruit drop.
- **Poor pollination:** A decrease of flowers and very early fruit may arise from inadequate or poor pollination.

Management and control

- **Integrated Pest and Disease Management:** Monitor and control insect pests and diseases that contribute to fruit drop.
- **Proper Irrigation:** Ensure the tree receives consistent and appropriate watering to avoid water stress.
- **Balanced Nutrition:** A balanced fertiliser program should be implemented, and foliar sprays containing micronutrients like manganese and zinc should be taken into consideration.
- **Pruning and tree care:** Maintain good tree health through proper pruning and other care practices.
- **Growth regulators:** Apply a monthly pre-harvest drop of NAA (10 ppm) between August and October. When applying 2,4-D for pre-harvest drop, pay close attention to the label's recommendations about dilution and application timing.
- **Pollination:** Ensure proper pollination, as it is a critical factor in successful fruit set.
- **Cultural practices:** Young trees are more likely to drop fruit, especially in the first few years of production, so take care of these problems. To enhance air circulation and get rid of dead wood, prune correctly.

3. Citrus Dieback/Decline

Citrus dieback, also known as decline, is the slow degradation of citrus trees brought on by a number of reasons, such as poor soil, diseases like citrus greening, inadequate water management, and mineral shortages like copper.

Symptoms

- **Loss of vigour:** Trees become generally unhealthy and weakened.
- **Twig and branch death:** This is the most prominent symptom, starting at the tips.
- **Leaf yellowing and shedding:** Leaves may turn yellow, and premature leaf fall is common.
- **Reduced fruit quality:** Fruits may be small, have thick skin, be low in juice, and have blemishes.

- **Retarded growth:** Terminal growth is stunted and leaf flushes are delayed.

Caused by:

- **Abiotic factors:** Dieback is associated with inadequate soil, inadequate drainage, poor site selection, copper deficiency, and excessive or insufficient watering.
- **Biotic factors:** Using diseased material to propagate new trees and pests that spread viruses.

Management and control

- **Prevention is key:** Start with certified, healthy plants on the correct rootstock and in the right location.
- **Good nutrition:** Use a balanced fertiliser regimen, paying particular attention to micronutrients such as copper.
- **Proper watering:** Ensure adequate and timely irrigation, and avoid water logging.
- **Pruning:** Regularly prune to remove dead or diseased branches.
- **Pest and disease control:** To control pests and diseases that can spread the issue, adhere to a scientific spray schedule.
- **Rejuvenation:** Improved methods are required to boost productivity and health in diminishing orchards.

4. Granulation in citrus

Granulation is an anomaly that starts at the fruit's stem end and progressively spreads to the stylar end. The impacted fruits grow larger, turn grey, taste flat and insipid, and take on a granular texture; the juice sacs become dry and hard. Because most of the juice transforms into a gelatinous mass, granulated fruits have less extractable juice.

Caused by: Citrus granulation is a physiological condition brought on by a number of causes, including as inadequate irrigation, uneven nutrition (particularly excessive nitrogen and high calcium/low phosphorus), delayed harvesting, and environmental factors like high temperatures and moisture stress. These problems cause the juice sacs to become swollen, dry, and hard rather than juicy.

Management: Applying two or three sprays of NAA (300 ppm) in August, September, and October could lower the granulation incidence to 50%. Granulation is also decreased by spraying GA 15 ppm and NAA 300 ppm in October and November.

D. Physiological disorders of Grape

1. Water Berries in Grape

Water berries typically start to form soon after berry softening and are linked to fruit ripening. When ripe, the impacted berries become soft, flabby, and watery. By harvest time, they are dry and shrivelled. These berries are primarily found on the tips of the main rachis or its branches.

Caused by: Excessive cropping, which results in insufficient nutrition for all berries, is the cause of water berries in grapes. Other contributing variables include high nitrogen levels in the vineyard, excessive nitrogen fertilisation, and cool, overcast weather during ripening. Shade inside the vine canopy and excessive irrigation during berry development are two more possible contributing variables. Berries with this condition become mushy, watery, and shrivel at harvest.

Symptoms: During ripening, water berries in grapes become mushy, flabby, and watery; by harvest, they frequently shrivel and dry out. Additional signs include the berries becoming duller, more opaque, or changing in colour, as well as the emergence of tiny, black patches on the cluster framework (pedicels and peduncles), which may enlarge and result in necrosis. Parts of the cluster, particularly the tips and top shoulders, may turn necrotic, dark brown, or purplish-black in extreme circumstances.

Management: During fruit development, steer clear of excessive nitrogen fertilisation and irrigation to concentrate on balanced nutrition. Reducing shading also requires effective canopy management, which includes branch thinning. Proactive vineyard management is essential to preventing the problem, even though there is no cure. Affected berries are frequently trimmed during harvest.

2. Cluster-Tip Wilting of Grape

The conductivity of the rachis is impacted by light brown lesions on its apical end. The rachis at the tip of the bunch dry out and shrivel as a result. In extreme circumstances, up to 30–40% of the bunch's tips completely dry up, leaving only hard, tiny, light brown berries. This condition is more likely to affect Thompson Seedless.

Symptoms: Cluster-tip wilting in grapes is characterised by abrupt wilting, shrivelling, and discolouration of berries and leaves, which frequently begin at the cluster tips. Shoots may also exhibit wilting or curling, and black patches may appear on the cluster stem and pedicles, which may cause the cluster as a whole to dry out.

Caused by: Factors that interfere with water flow to the cluster induce cluster-tip withering in grapes, which results in the tip drying out and wilting. The main reason is disruption of the rachis (stem of the cluster), which is frequently caused by light brown lesions on the rachis. These lesions can be caused by fungal infections like *Botrytis cinerea* or environmental stress or diseases like Grapevine Leafroll-associated Virus (GLRaV). In certain instances, an increased crop burden may also be the cause.

Management and prevention

- **Cluster thinning** To lessen the total load on the vine, remove a few growing bunches. Additionally, this enhances fruit quality and air circulation.
- **Adequate irrigation** Make sure there is a steady and adequate supply of water, particularly when the berries are developing.
- **Shade protection:** Shield grape bunches from direct, harsh sunlight.
- **Monitor and record:** To foresee and handle potential problems, keep track of the weather and vine health.

3. Shot Berries of Grape

They develop as a result of either insufficient carbohydrate supply into the set berries or a delay in the pollination and fertilisation of a few flowers. The known causes of shot-berry production include girdling, inappropriate phases of GA treatment, and boron deficiency.

Caused by Shot berries are tiny, underdeveloped grapes that result from a variety of circumstances during flowering, such as bad weather (wind, rain, or cold), inadequate pollination, or nutritional deficits like boron. Their creation can also result from certain horticultural methods, such as applying gibberellin (GA) at the wrong time, and inadequate glucose flow to the berries.

Symptom: The presence of tiny, spherical, seedless berries that are sweeter than usual is the primary sign of shot berries. This condition is caused by either insufficient carbohydrate supply to the developing berries or a delay in pollination and fertilisation of some flowers.

Management: Application of GA at proper stage should be ensured. Boron or Zinc deficiencies should be corrected.

4. Pink Berry of Grapes

The primary sign of "pink berry" in grapes is when the berries take on a pink or reddish hue rather than their typical ripened hue, which subsequently becomes bland and unsightly.

Caused by A genetic deficiency in the color-regulatory gene MybA1, which results in a brief insertion in the intron, is the cause of pink berries in grapes

Management: Indiscriminate use of Etherel for berry colouration can also cause this disorder.

5. Bud and Flower Drop in Grapes

Numerous factors, including improper nitrogen application, improper fertilisation, ambient temperature, heavy crop load, uneven ripening, and endogenous auxin deficiency at a specific stage of berry development, have been linked to the disease.

Caused by: Improper fertilisation and nutritional imbalances, particularly excessive nitrogen, can lead to bud and blossom drop in grapes. The issue is additionally exacerbated by extreme temperatures, poor pollination, high humidity, and stress from pests, diseases, heavy crop loads, and water problems.

Control and prevention

Grape bud and flower drop can be controlled by controlling water and nitrogen to prevent excessive vegetative growth, ensuring good sunlight exposure through pruning and canopy management, and applying plant growth regulators such as NAA (Naphthalene Acetic Acid) at 20 ppm during berry formation and GA3 (Gibberellic Acid) combined with cytokinin at 30–40 ppm at the 3–4 mm berry size stage to increase bunch size, which can help improve overall yield by preventing some drop.

6. Poor Cane Maturity in Grape:

This kind of disease causes the bark to stay green until late fall and prevents the shoots from maturing. The low temperatures throughout the winter cause these shoots to turn pink-red. It is more severe in vineyards with dense, strong branch growth, closely spaced plants, and an abundance of nitrogen and irrigation.

Caused by: Unfavourable weather, disease and insect infestations, and excessive vegetative growth all contribute to poor cane maturity in grapes.

Management and prevention: Reducing vegetative growth through pruning and shoot pinching is the main strategy for managing low cane maturity in grapes. Increase sunshine exposure to the canopy and steer clear of excessive nitrogen delivery, particularly during the cane maturation period (60–90 days following pruning) and irrigation. Maturity can also be aided by the prudent application of potassium fertiliser.

E. Physiological disorders of Litchi

1. Poor Fruiting/ non Fruiting / Irregular Fruiting in Litchi

The issues in the country's litchi-growing regions are poor fruiting, non-fruiting, and irregular fruiting. Unpredictable bearing in litchi is also caused by environmental conditions, nutrition, and irrigation. It was also discovered that irregular or alternate bearing was related to the kind of soil and management

techniques. Late harvesting and severe pruning and training operations may lead to non bearing in that particular year of operation.

Management and cultural practices

Focus on appropriate pruning, nitrogen management, and insect control, particularly after harvest, to manage poor or irregular litchi fruiting. Controlling vegetative growth and enhancing flowering can be achieved through timely pruning, proper fertiliser application, and sprays of plant growth regulators such as paclobutrazol. Fruit set and output are also greatly increased by protecting blooms from pests with neem oil or other insecticides and making sure pollination is done well.

2. Flower and Fruit Drop in Litchi

Because litchi trees experience a heavy flower and fruit drop between flowering and fruit maturity, inadequate yield has occasionally been noted even after abundant flowering and fruit set. In many cultivars, just a little percentage of it (2–18%) is carried to maturity. The flower and fruit drop is thought to be due to failure of fertilization, embryo abortion, nutrition and hormonal imbalance and external factors like high temperature, low humidity and strong westerly winds as well as due to fruit borer and heavy mite attack. Compared to elder trees, younger bearing trees have fewer decreases.

Management In litchi, honey bees are the primary pollinators. Increasing the number of bees in orchards promotes improved fertilisation and pollination, which raises fruit retention and set. When the orchard is in bloom, it is recommended not to use any kind of spray. ZnSO₄ at 0.2% sprayed 30 days before to panicle emergence promotes healthy inflorescence and increases fruit set. Fruit retention, cracking, colour, sweetness, and maturity are all improved by foliar spraying of 0.2% boron two to three times during the fruit's growth and development period, increasing the percentage of high-quality production. Planofix @ 4 ml/5 litre water may be applied topically to peanut-sized fruits twice at intervals of 15 days.

3. Fruit cracking in Litchi

It is the most significant litchi disease, appearing in nearly every significant litchi generating tract. It is a significant issue in India's litchi. Fruits that are broken lose their fresh market value and are mainly utilised for processing, particularly for fruit juice. Fruits with cracks have a reduced shelf life and are prone to storage diseases.

Caused by: The main cause of litchi fruit splitting is moisture stress, particularly heavy rainfall or irrigation following a dry spell, which causes the fruit's internal tissues to quickly expand against an

inelastic skin. High temperatures, low relative humidity, and nutritional deficiencies-particularly those of calcium and boron-are other contributing factors. Some cultivars may also be more vulnerable due to certain genetic characteristics, such as a thin skin or fruit shape.

Management and prevention

Litchi fruit cracking can be effectively managed by controlling watering, offering shade, and applying certain foliar sprays. Using shade netting or bagging the fruit, providing minerals like calcium at 2 m/l liquid formulations and Gibberellins at 20 ppm, which lower cellulose activity and hence minimise cracking, and keeping constant soil moisture with sunshine are all important techniques. Cracking is decreased by spraying 2,4-D and NAA at quantities of 20 ppm or 20 mg/litre. Fruit cracking is considerably reduced by applying boron sprays in the form of borax or boric acid @ 2g/l during the early stages of aril growth with enough soil moisture in the root zone.

F. Physiological disorders of Pineapple

1. Multiple Crowns in Pineapple

Fruit typically has a single crown, but occasionally it has many crowns. As a result, the fruit will have a broad, flat top and be unsuitable for canning. These fruits are corky and have a bland flavour. It is thought to be a heritable trait, primarily present in the Cayenne group, which includes the Kew variety.

Caused by: Hormonal imbalances, environmental stress during plant development, or an uncommon genetic mutation may be the reason. Although it is uncommon, this condition usually has no negative effects on the fruit, albeit its shape may be less consistent.

Management

A pineapple with several crowns can be managed by either separating them for propagation or controlling the factors that lead to the disorder if a single giant fruit is the desired outcome. The crown can be divided into portions and planted to produce several new plants. Avoid ratoon cropping and maintain ideal soil fertility by avoiding high calcium or zinc levels, which are associated with strong plant vigour and particular environmental variables, in order to control and prevent numerous crowns on a single plant.

2. Fruit and Crown Fasciation in Pineapple

Fruits with fasciation are completely unusable due to their extreme deformity. Sometimes the multiplication is so severe that the fruit is twisted with countless crowns and severely flattened. Fasciation of fruits and crowns is linked to plants with strong vigour that take longer to blossom.

Caused by: Pineapple fruit and crown fasciation is brought on by a number of environmental factors, including as warm weather and high soil fertility, as well as high plant vigour, which delays flowering. The condition, which is a mutation that causes the fruit to flatten and twist, frequently with multiple crowns, can also be favoured by deficiencies in calcium or zinc.

Management

Avoiding overly fertile soil, controlling soil fertility by making sure calcium and zinc levels are sufficient, and avoiding specific cultivation techniques like ratooning that might raise the risk in order to prevent the high plant vigour that leads to it. Despite being a physiological problem, the condition can be reduced by using excellent agricultural methods.

3. Collar of Slips in Pineapple

The presence of several slips that originate from the stem near the fruit's base or even directly from the fruit itself characterises the collar of slips. The fruit suffers as a result of the excessive slip development, producing little, tapering fruits that frequently have knobs at the base.

Caused by: A number of conditions, such as high nitrogen fertilisation, heavy rainfall, and cold temperatures, contribute to the "collar of slips" in pineapples. A significant number of tiny, developing fruitlets, or slips, are growing at the base of the main fruit in this situation, which hinders the fruit's development and results in a small, tapering fruit.

Management

As advised, provide a balanced quantity of nitrogen to promote appropriate drainage and prevent high nutrient levels that may cause the condition.

4. Dry Fruit and Bottle Neck in Pineapple

It's possible that the bottle neck fruit and dry fruit are descended from the same parent.

Caused by: Inadequate fruitlet growth, which is frequently brought on by climatic elements including hot temperatures, cold snaps, or irregular watering. These problems hinder the fruit's natural development, resulting in symptoms such as undeveloped parts or a stunted look.

Management

Focus on appropriate fertilisation and irrigation to control dry fruit and bottle neck in pineapples, paying particular attention to balanced nitrogen and phosphorus delivery and preventing water stress during dry spells. Both fruitlets fail to develop in dry fruit, whereas the upper fruitlets in bottle neck do not develop normally. However, both can be treated using identical management techniques.

5. Sun-Scald in Pineapple

The physiological condition known as "sun-scald" in pineapples is brought on by prolonged exposure to high temperatures and strong sunlight, which harms the fruit's exposed skin. One side of the fruit is frequently exposed to the sun when the plant tilts or topples over.

Caused by: The main causes of sun-scald in pineapples are high temperatures and intense solar radiation, which harm the fruit's cells. A long stalk, a weak leaf system, or the fruit being exposed due to lodging (leaning or dropping) are other variables that promote vulnerability.

Management strategies

Use reflecting covers, shade, or attaching leaves to provide physical protection. You should also think about utilising reflective sprays like kaolin clay or calcium carbonate. Use high-density planting as well, and maintain plants upright to prevent fruit exposure.

G. Physiological disorders of Pomegranate

1. Fruit Cracking

Pomegranate fruit cracking is a major issue. Fruit cracking can also result from a prolonged delay in harvesting completely matured fruits or from a serious pest or disease infestation.

Caused by: The cause of pomegranate fruit cracking is erratic watering, which causes the fruit to absorb water quickly and expand, particularly after a dry spell followed by intense rain or irrigation. Extreme day-to-night temperature swings and nutrient shortages, especially in boron and calcium, in immature fruits are other concerns.

Control measures

Infected branches should be pruned, dead plant matter should be removed to improve cleanliness, copper-based fungicides like Bordeaux combination or copper oxychloride should be applied, and overhead irrigation should be avoided. Additional techniques include employing systemic fungicides, applying bio-fungicides like Trichoderma or Pseudomonas to the soil, and occasionally planting disease-resistant cultivars. using micronutrient sprays such as GA (40 ppm) and boron (50 ppm).

H. Physiological disorders of Guava

1. Die-back: The fungus anthracnose (*Gloeosporium psidii*), which destroys branches from the tip backward, is the main cause of guava dieback. Additional reasons include algal leaf spot in severe cases, which can result in dieback symptoms, and fungal infections like wilt, which are brought on by pathogens like *Fusarium* and *Botryodiplodia theobromae*. These illnesses are also influenced by environmental elements like high humidity and inadequate air circulation.

Control measures The afflicted branches appear barren as they lose their leaves. By preserving the pH of the soil, applying 1.8 kg of lime or gypsum lowers tree mortality.

2. Bronzing: Guava bronzing is a complicated nutritional condition. When fruiting begins in a P and K-deficient soil, nutrients are transferred from older leaves to the fruits, resulting in bronze-colored leaves and decreased uptake and photosynthetic transfer to the roots. A combination of 20 kg of FYM, 1 kilogramme of SSP, 0.5 kg of MOP, and 100g of ZnSO₄ per tree should be added to the soil for mild symptoms. In severe cases, the dose may be doubled except FYM and foliar spraying of DAP 0.3% and SOP 0.5% is to be given 45 days after the emergence of leaves.

Caused by: Low soil fertility and pH are frequently coupled with nutrient deficits, especially those of zinc, phosphorus, and nitrogen.

Management and treatment

For two months, apply weekly foliar sprays of a 0.5% diammonium phosphate and zinc sulphate solution to address nutrient shortages. Additionally, to lessen tree stress and enhance general health, maintain appropriate cultural practices including sufficient fertilisation, trimming, irrigation, and sanitation. Copper-based fungicide preventive treatments, such Bordeaux combination, can also help stop the spread of related illnesses like canker.

3. Boron deficiency: In guavas, a lack of boron results in stunted growth, brittle, swollen leaves, and deformed forms. Additionally, it can cause red stains on newly emerging leaves and, in extreme situations, the demise of branches and developing points. Fruits may shrivel, split, and develop brown, corky patches.

Management and treatment

The deficit is corrected by spraying 0.3% boric acid 10–15 days prior to flowering. Zinc and boron deficits are generally addressed by foliar treatment of 0.5% zinc sulphate and 0.4% boric acid 10–14 days before to flowering.

I. Physiological disorders of Papaya

1. Boron deficiency: One of the major conditions limiting papaya productivity is boron shortage. It produces fruits that are deformed.

Management and treatment

The deficiency is corrected by spraying 0.1% boric acid every three months starting in the sixth month after planting.

J. Physiological disorders of Sapota

1. Fruit drop: Low fruit setting and shedding is a concern for sapota. Merely 10–12% of all fruits grow, set, and remain until they reach maturity. The majority of the fruit-drop happens right after the fruit sets.

Management and treatment

Adequate diet suitable growth regulators, disease and pest management, and hygienic procedures. Applying plant growth regulators such as GA3 or NAA at 25 to 100 ppm during blooming and at intervals of 15 days is a crucial tactic (Rathod and Amin, 1981). maintaining proper potassium, zinc, and boron nutrient levels and using bait sprays and sanitation to keep pests like fruit flies at bay.

K. Physiological disorders of Custurd apple

Fruit cracking, woodiness, stone fruit production, and tree decline are examples of physiological abnormalities in custard apples. These are brought on by things like erratic water supplies, inadequate pollination, nutritional imbalances (particularly those involving calcium and boron), and water logging. Fruit thinning, balanced fertilisation, and appropriate watering can all help control these problems.

1. Stone fruit formation

As a result, the tree may continue to bear little, dry, brown fruits. High temperatures and competition amongst developing fruits are the causes.

Management: Thin fruits during the initial stage of development.

2. Woodiness

Hard, woody, and gritty seed pockets in the fruit flesh are indicative of this. It is frequently associated with a calcium and boron deficit.

Management: Use fertilisers with calcium and boron. Fruit calcium concentration can gradually increase with repeated calcium soil treatments. Zinc availability can be improved and toxicity can be decreased by keeping the pH of the soil at 6.5 or higher.

3. Fruit cracking

This happens as a result of an abrupt change in the water supply, particularly following a dry spell that is followed by a lot of rain or irrigation.

Management: To prevent abrupt fluctuations in moisture levels, schedule watering properly and consistently.

4. Tree decline

This may occur as a result of problems like waterlogging, which can lead to water stagnation in the soil.

Management: Ensure proper drainage and avoid water logging in the orchard.

5. Sunburn

This can happen when leaves or fruits are exposed to direct sunlight and overheat, sometimes made worse by spray drift or drought stress, and it affects immature tree parts.

Management: Ensure adequate water supply to the plant.

L. Physiological disorders of apple

1. Bitter Pit: This physiological condition lowers the fruit's fresh market quality. The most vulnerable trees are young ones that are just starting to develop fruit. Compared to fruits that are collected at the appropriate maturity stage, immature fruit are more prone to bitter pit. The fruit's flesh develops tiny brown lesions that range in diameter from 2 to 10 mm, depending on the cultivar. The tissue beneath the skin turns corky and black. Depressed areas appear on the skin's surface during harvest or during storage. On the skin next to the calyx, these areas show up as water-soaked patches. After one or two months in storage, the spots usually darken, grow more sunken than the surrounding skin, and fully develop.

Control: Bitter pit is prevented with calcium sprays before harvest and calcium dips before storage. The plants should be sprayed 45 days before harvest, and then again 15 days later. Before storing, the post-harvest dip should be applied for one to two minutes.

2. Brown Heart: Large, overripe fruits are linked to this physiological condition. Additionally, it can happen when storage CO concentrations rise above 1%. Usually starting in or close to the core, the symptoms manifest as brown to discolouration in the skin. Brown regions may contain dry voids caused by desiccation and have distinct borders. A tiny patch of dark flesh or complete browning of the flesh with a margin of healthy white flesh remaining just below the skin are examples of symptoms. Early in storage, symptoms appear, and as storage duration increases, they may become more severe.

Control: Fruits that are too mature should not be harvested. When storing fruits in a controlled atmosphere (CA), they should be picked when they are at their peak maturity. To lower the incidence of brown heart formation, CO concentrations in CA 2 should be less than 1%.

3. Cork Spot: A tiny flushed patch on the fruit's surface above the afflicted brown spot is the first sign of this physiological condition. Generally speaking, the damaged tissue is far harder than the healthy tissue. Cork patch can occasionally be attributed to deficits in calcium and boron.

Control: This condition can be avoided with proper nutrition management, particularly with regard to calcium and boron.

4. Scald: For apple growers, this physiological condition is a major worry. The environment, cultural customs, and apple variety all affect how susceptible a person is to this storage condition. Hot, dry weather prior to harvest, immature fruit during harvest, and high nitrogen and low calcium concentrations in the fruit all contribute to the incidence and severity of scald. This disease is further exacerbated by inadequate ventilation in packaging boxes or storage rooms. After the fruit is removed from cold storage, it warms up and develops irregular brown patches of dead skin within three to seven days. The scald is not brought on by the warm temperatures, but they do create symptoms from an earlier damage that happened during cold storage. When an injury is serious, symptoms could be apparent in cold storage.

Control: Scald incidence can be decreased by harvesting at the right maturity and using air in cold storage. Applying an antioxidant just after harvest is the most popular way to prevent scald. DPA, or diphenylamine, is frequently utilised. While it can harm other apple kinds, ethoxyquin is also beneficial for other varieties. For optimal management, antioxidants should be given within a week of harvest.

5. Water Core: High leaf to fruit ratios, high fruit nitrogen and boron levels, low fruit calcium levels, excessive thinning, and fruit exposure to high temperatures all contribute to this physiological problem. This kind of disease primarily affects large fruits. Water-soaked areas in the meat form as a result of pre-harvest disorder. Only when the illness is extremely serious can these hard, glassy areas be seen from the outside. Fruits that are severely impacted may smell and taste soured. Water-soaked patches can be found all over the apple or close to its core. In storage, mild to moderate symptoms may entirely go away.

Control: Avoiding delayed harvests is the best approach to lower the occurrence. Fruit samples should be checked for water core growth as they get closer to maturity. Fruit should be picked before a large water core forms. Fruit bunches with moderate to severe water core symptoms should be sold as soon as possible rather than being stored in a controlled atmosphere (CA).

6. Sun Burn: The extreme heat of the sun causes this physiological condition. Usually, the fruit on the southwest side of the tree is impacted. Sunburn incidence can also be raised by water stress. White,

tan, or yellow blotches on sun-exposed fruits are the first signs. Fruit with extensive skin injury may become dark brown before being harvested. These regions could get sunken and spongy. After harvest, fruit exposed to the sun may sustain a serious sunburn.

Control: Avoiding abrupt exposure of fruit to extreme heat and sunlight is the greatest control strategy. Pruning and tree training must be done correctly. Care must be taken when pruning in the summer to prevent severe sunburn. To lessen heat stress, pruned orchards should get regular irrigation. Once the damage has been done, the only way to fix it is to carefully sift and remove the damaged fruit before packing.

7. Russetting: Fruit growers are often concerned about the russetting of apples in a humid atmosphere. Shortly after petals fall, russetting takes place. Russetting is more likely to occur in apple cultivars with thin cuticles. Fruits that are exposed are more likely to exhibit it than those that are kept in the shade. Russetting can also result from frost during the flower or early stages of fruit production. Fruit skin ruptures and cracks appear as a result of russetting.

Control: Russetting can be decreased by choosing less vulnerable clones, providing sufficient watering, manuring, and efficient pest control.

8. Fruit Drop: The majority of commercial apple cultivars have three fruit drop cycles: early drop, June drop, and pre-harvest drop. Due to a lack of pollination and fruit competition, the early decline is thought to be natural. The June decline is caused by environmental factors and moisture stress. These two declines are neither adequately controlled by artificial means nor result in significant economic losses. Due to the decrease in auxin levels, fully grown, marketable fruits abscise prior to harvest, resulting in significant economic loss.

Control: By applying NAA (15 ppm) 20 days prior to the anticipated fruit drop or 20–25 days prior to harvest, the pre-harvest drop can be managed.

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Irradiation and Food Safety

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Food spoilage, caused by physical, chemical, and microbial factors, results in significant food waste. To combat this, various preservation techniques are employed, ranging from traditional methods like drying and freezing to advanced technologies like cold plasma, high-pressure processing, and irradiation. Among these, food irradiation, utilizing gamma rays, X-rays, or electron beams, offers a promising solution for pathogen inactivation, shelf-life extension, and improved food safety. While thermal processing is energy-intensive and can degrade nutritional quality, non-thermal methods like irradiation minimize these drawbacks. Irradiation effectively inactivates microorganisms and prevents insect infestation, enhancing food safety and quality. Gamma irradiation, using isotopes like Cobalt-60, provides deep penetration, while X-rays and electron beams offer alternative sources with specific advantages. Electron beams, in particular, offer high efficiency and no radioactive waste. Despite its benefits, public perception remains a challenge due to fears of induced radioactivity, although extensive research and regulatory approvals from organizations like the FDA and WHO confirm its safety. Food irradiation, when conducted according to established guidelines, is a safe and effective method for food preservation, contributing to reduced food waste and enhanced global food safety.

Keywords: Food irradiation, food safety, ionizing radiation, kGy, shelf life extension.

Introduction

Food spoilage is the result of several causes including physical injury, insect damage, chemical changes such as oxidation reactions and microbial activities. The adverse modifications induced by the aforementioned factors renders foodstuff inappropriate for the human consumption. Food preservation encompasses various processes, methods, and technologies aimed at preventing spoilage, maintaining quality, and extending shelf life. Conventional preservation techniques include drying, freezing, the use of chemicals, smoking, pickling, and canning, among others ¹. The need to reduce the food wastage is an alarming problem, to minimize the losses, food processing techniques such as Cold plasma, High pressure processing, Pulse electric field, Irradiation etc. are being used. About 25 % of food loss takes places during the postharvest operation including, storage, processing, handling and distribution.

To avoid the losses several food technologies are used to minimize the losses occurring in the food processing sectors like thermal and non-thermal processing, which are used substantially to maintain the safety and quality of foods. Thermal processing methods of food preservation are long term process, consumes high energy thereby increasing preservation costs. On the other, hand non-thermal methods are continuous, less time consuming making it the more suitable choice for the preservation causing less detrimental nutritional loss in product i.e., vitamins, antioxidant properties, denaturation in protein, sensorial properties etc.²

Food irradiation is a physical treatment involving exposure to a defined dose of ionizing radiation. These short-wavelength radiations possess the capability to inactivate microorganisms, including those responsible for food contamination, spoilage, and deterioration. Irradiation encompasses the treatment of food with ionizing radiation to achieve desired outcomes such as pathogen inactivation, shelf-life extension, sprout inhibition, and replacement of chemical fumigation, all while minimizing significant temperature increases within the food³. Irradiation, whether employed independently or in conjunction with other preservation methods, effectively prevents insect infestation and inhibits microbial growth, thereby enhancing the shelf life of food. The application of irradiation represents a relatively innovative technological approach for enhancing food safety, quality, and international trade of food products.

Sources of radiation

According to the Codex Alimentarius General Standards, ionizing radiation, which exists in two forms i.e. (i) electromagnetic, (ii) particulate⁴ For food irradiation three major source types are as followings:

- (a) Gamma radiation from radionuclides such as ⁶⁰Co or ¹³⁷Cs.
- (b) Electron beams with energies up to 10 MeV.
- (c) X- rays with electron energies up to 5 MeV.^{5, 6}

Gamma radiation

Gamma radiation, a form of electromagnetic energy, originates from the decay of specific radioactive materials. Radioisotopes continuously emit these high-energy photons, known as gamma rays. In food preservation, common gamma ray sources include Cobalt-60 (Co-60) and Cesium-137 (Cs-137). These powerful rays possess the ability to penetrate food to considerable depths (several centimeters). However, their use necessitates substantial safety measures, including heavy shielding, and results in the generation of radioactive waste⁷. Most of the food irradiation is done by Cobalt-60 isotope. It is an artificially produced radioactive isotope created by bombarding stable natural Cobalt-59 (Co-59)

with neutrons. Co-60 undergoes beta decay, transforming into the stable Nickel-60 (Ni-60) isotope and releasing one electron (beta ray) along with two gamma rays. While the emitted beta radiation from Co-60 is relatively weak (approximately 0.3 MeV), the gamma rays, with energies of 1.17 and 1.33 MeV, are the effective radiation utilized for food treatment. Cobalt-60 has a radioactive half-life of 5.27 years.

2.2 X-rays

X-rays for food treatment are typically generated by specialized machines that accelerate a beam of electrons in a vacuum to bombard a metallic target, often made of tungsten or molybdenum. These X-rays share the short-wavelength nature of gamma rays but differ in their origin. Food irradiation using X-rays employs energy levels of 5 MeV or lower, allowing them to penetrate thick food products ⁴. However, similar to gamma rays, the use of X-rays necessitates substantial safety shielding. A notable drawback of X-ray technology for food irradiation is its relatively inefficient production process, leading to higher operational costs compared to both electron beam irradiation and gamma ray methods.

2.3 Electron beam radiation

Electron beam irradiation utilizes a stream of high-energy electrons generated from an electron gun. Characterized by a higher energy level (up to 10 MeV) but limited penetration depth, this method is particularly well-suited for treating the surface and subsurface of food products ⁸. While electrons have less penetrating power than gamma rays, electron beams prove highly effective for irradiating large quantities of small food items. Key advantages of electron beam technology include its switch on/off capability, high energy efficiency, rapid processing rates (high throughput), the absence of radioactive waste generation, and a superior dose rate compared to gamma rays. Furthermore, electron beam sources exhibit greater efficiency in electron utilization as the beam is directly targeted at the product, unlike gamma rays which are emitted omnidirectionally. Economic analyses suggest that electron beam irradiation is a competitive and often more cost-effective preservation method compared to alternatives. Its high efficiency in inactivating microorganisms in diverse food types makes it a safe and viable alternative for food decontamination.

Table 1: Different sources of irradiation and their characteristics

Sources	Characteristics
Cobalt- 60	High penetration power Permanent radioactive source Higher efficiency Source replenishment requirement Low throughout
Electron beams	Lower penetration power Higher efficiency High throughput Power and cooling requirement Technically complex
X-rays	Higher penetration power Lower efficiency High throughput Technically complex Power and cooling needed

2.2 Unit of radiation

The quantity of ionizing radiation taken up by a material is now expressed in **gray (Gy)**, a unit that has replaced the older term **rad**, where **1 Gy is equivalent to 100 rad**. One gray corresponds to the absorption of **1 joule of energy per kilogram** of the material being irradiated. The actual amount of energy absorbed varies based on the **food's mass, density, and thickness**.

1. Mechanism of irradiation technology

Food irradiation is treating food with powerful rays, like gamma rays. This process interacts with the food material, leading to both direct and indirect effects. Primarily, it targets insect pathogens and microbes that can deteriorate the food's quality and safety. Ionizing radiation interacts with these microorganisms in various ways: it can strip electrons from their atoms (ionization), break chemical bonds within their molecules (dissociation), and excite their electrons to higher energy levels. The ionizing radiation can cause either direct or indirect damage on microorganisms. A microorganism's genetic material (DNA or RNA) is directly harmed by ionising radiation. The photons or electrons will randomly strike the genetic material and cause breaks or lesions. Single-strand lesions may not be lethal or may cause a mutation, but multiple lesions would make the microorganism nonviable. The indirect effect of ionizing radiation on microorganisms involves the interaction of the radiation on other molecules or atoms within the organism and they produce positively charged water radicals

(H_2O^+) and negative free solvated electrons(e^-) by lose of electron when radiolysis of cellular water. When food is exposed to ionizing radiation, water molecules and other food components undergo radiolysis, resulting in the formation of reactive chemical species. These include molecular hydrogen (H_2), oxygen (O_2), hydrogen peroxide (H_2O_2), hydroxyl radicals ($\cdot\text{OH}$), hydrogen radicals ($\text{H}\cdot$), and hydroperoxyl radicals ($\text{HO}_2\cdot$)⁴. Of these, hydroxyl radicals and hydrogen peroxide are particularly significant due to their ability to damage nucleic acids by disrupting their molecular bonds. These free radicals are highly reactive and short-lived, with a lifespan of less than 10^{-5} seconds, rendering them lethal to microorganisms. Furthermore, ionizing radiation can disrupt critical cellular components such as membranes, enzymes, plasmids, and proteins, thereby inhibiting the proliferation of targeted microorganisms, insect gametes, and plant meristems.

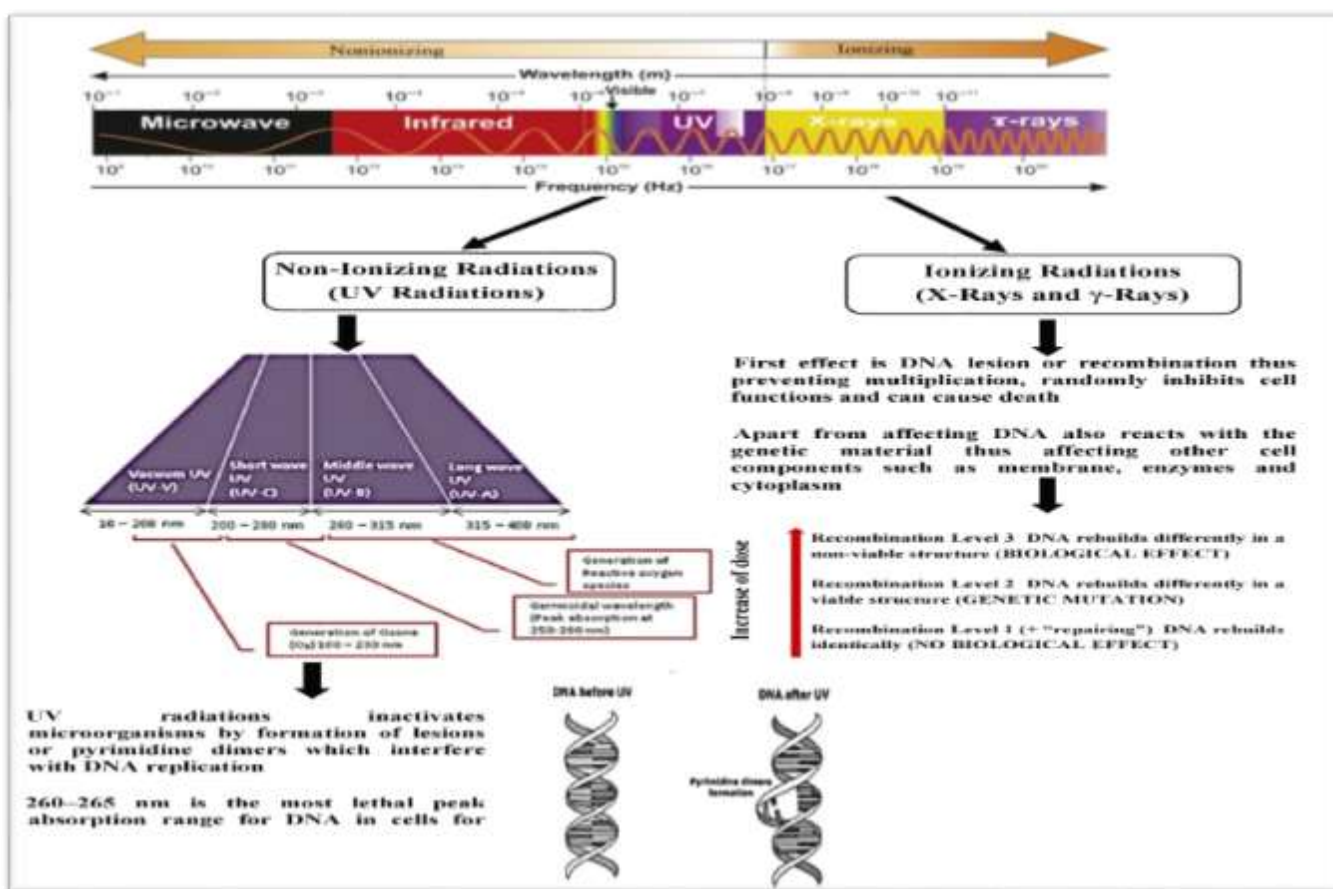


Figure 1: Effect of ionizing and non-ionizing radiations on microbial destruction².

3.1 Factors affecting the food irradiation process

The efficiency of food irradiation is influenced by number of factors such as medium composition enclosing the micro-organism, water activity of the food to be treated, temperature to which product is being exposed, freezing and oxygen ². The fig shows factors affecting irradiation process.

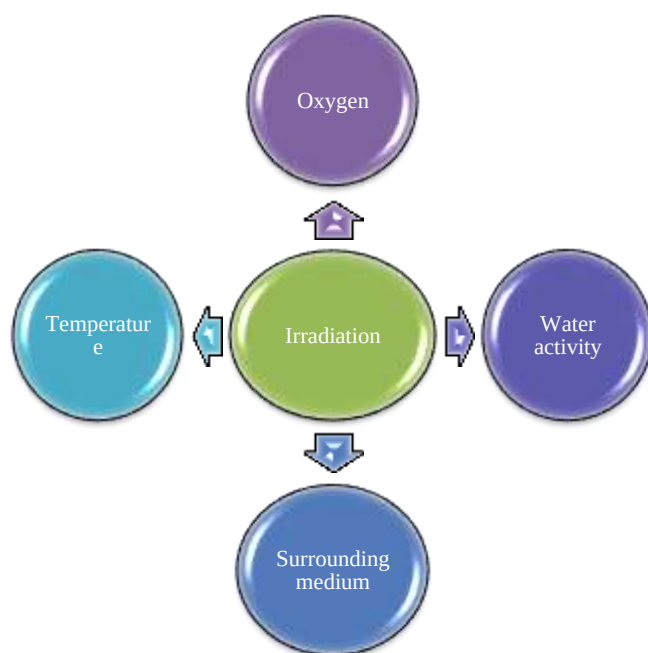


Figure 2: Factors affecting the radiation process in food

2. Food irradiation application

4.1. Low dose levels (0.1 to 1 kGy)

At lower irradiation doses, sprouting of potatoes, onions, garlic, shallots, yams, etc. can be inhibited by irradiation in the dose range. Insect disinfestations by radiation in the dose range 0.2-1 kGy is aimed at preventing losses caused by insect pests in stored grains, pulses, cereals, flour, coffee beans, spices, dried fruits, dried nuts, dried fishery products and other dried food products by sterilizing and interrupting the breeding cycle of insects. The inactivation of some pathogenic parasites of public health significance such as tapeworm and trichina in meat can be achieved at doses in the range 0.3-1 kGy ¹⁰. Low doses of irradiation can also delay the ripening process of certain fruits hence, improving the quality for export of the product at its maturity, but the effect depends upon the type of cultivar and growth conditions of the fruit. Low dose levels cannot eliminate some phytopathogenic fungi which are one of the reasons for the storage losses of products.

4.2 Medium dose levels (1 to 10 kGy)

At a medium irradiation dose levels, fresh meat, seafood, vegetables, and fruits can be subjected to such treatments with doses ranging from approximately 1 to 10 kGy, depending on the product. Irradiation pasteurization of solid foods like meat, poultry, and seafood is an effective method for eliminating pathogenic organisms and microorganisms, except for viruses. A dose range of 2 to 8 kGy is effective in reducing the number of specific viable non-spore-forming pathogenic microorganisms. Reduction in microbial spoilage at dose levels of 1- 3 kGy can extend the shelf- life of perishable products like fish, meat and fruits ¹⁰. Some food- poisoning causing organisms like *Salmonella*, *Listeria* and *Campylobacter* are resistant towards the irradiation causing only few log cycle reductions within this dose range. *Clostridium botulinum* bacteria are more radiation resistive whereas viruses are unaffected till the high dose levels.

4.3 High dose levels (>10 kGy)

At higher dose levels, 10 kGy dose offers an effective alternative to ethylene oxide for microbial decontamination of dried spices, herbs, and other dried vegetable seasonings. Radiation sterilization, with doses of 25 kGy, greatly prolongs the shelf life of precooked or enzyme-inactivated food products packaged in airtight containers, potentially for an indefinite period. One of the main application of high irradiation doses such as meals for immune disorders patients for hospital which can only eat bacteria free food.

3. Food irradiation safety and consumers acceptance

Food irradiation stands as the most thoroughly researched food preservation method. Its safety has been rigorously investigated since the 1950s, when the US Army initiated studies on its effects. Hundreds of subsequent research projects have evaluated the toxicological safety and nutritional impact of irradiated foods. Notably, the FDA reviewed over 400 toxicology studies following the 1981 report by the joint FAO/IAEA/WHO expert committee, which concluded that food irradiation at doses up to 10 kGy poses no significant toxicological, nutritional, or microbiological risks ^{13,14}. Backed by over half a century of research, and with FDA approval for various food categories, food irradiation has been proven to be a safe and effective food safety measure. Consequently, when conducted according to established guidelines and good manufacturing practices, food irradiation presents minimal health risks.

Despite over a century of use and regulatory approval for specific food applications in more than 55 countries, the broader adoption of food irradiation remains limited due to persistent environmental and

health safety concerns. Decades of extensive research have consistently affirmed the general safety of this technology. Although food irradiation has undergone unparalleled scrutiny as a food preservation method over the past 40 years. Recognizing its potential for pathogen reduction and public health, many countries have integrated irradiation into their good manufacturing practices (GMP) and hazard analysis critical control points (HACCP) systems. In the USA, mandatory labeling with terms like "Treated with irradiation" or "Treated by irradiation" must appear on the food package¹⁵.

5.1 Consumer acceptance

Despite the strong scientific evidence supporting its safety, consumer acceptance of irradiated food remains a challenge. This reluctance often stems from:

- **Misconceptions about Radioactivity:** The association of "radiation" with nuclear weapons and accidents creates fear and misunderstanding. Many consumers incorrectly believe that irradiated food becomes radioactive.
- **Lack of Awareness and Education:** A significant portion of the public has limited knowledge about the food irradiation process, its benefits, and its safety. This lack of understanding contributes to skepticism and resistance.
- **"Unnatural" Perception:** Some consumers view irradiation as an "unnatural" or "artificial" food processing method, preferring traditional methods.
- **Concerns about Nutritional Loss:** Despite evidence to the contrary, some consumers worry that irradiation significantly diminishes the nutritional value of food.
- **Labeling Issues:** While mandatory labeling of irradiated foods is intended to inform consumers, it can sometimes be misinterpreted as a warning sign, further fueling negative perceptions. The international Radura symbol, intended to identify irradiated food, is not always widely recognized or understood.
- **Mistrust of Technology:** A general mistrust of technological interventions in the food supply can contribute to resistance towards irradiation.

5.2 Future capabilities of food irradiation

The future of food irradiation holds considerable promise for addressing evolving challenges in food production, distribution, and consumption. Potential future capabilities include:

- **Advanced Irradiation Technologies:** Development of more precise and efficient irradiation sources, such as pulsed electron beams and low-energy X-rays, could optimize treatment for specific food types and minimize energy consumption.

- **Combination Processing:** Integrating irradiation with other preservation techniques (e.g., modified atmosphere packaging, mild heat treatments) could lead to synergistic effects, enhancing food safety and quality while minimizing the intensity of individual treatments. This "hurdle technology" approach can improve overall preservation efficacy.
- **Real-Time Monitoring and Control:** Implementing sophisticated sensors and data analytics to monitor irradiation processes in real-time will ensure consistent and accurate treatment, enhancing safety and efficiency.
- **Tailored Applications for Specific Food Needs:** Research will likely focus on optimizing irradiation protocols for niche food products and specific quality attributes, such as improving the texture of certain fruits or enhancing the flavor profile of irradiated spices.
- **Addressing Food Security and Sustainability:** In a world facing increasing population and climate change, food irradiation can play a vital role in reducing post-harvest losses, enabling wider distribution of food to underserved regions, and potentially decreasing the environmental footprint associated with food production and spoilage.
- **Enhanced Traceability and Authentication:** Integrating irradiation data with food traceability systems could provide consumers and regulators with enhanced information about food processing and quality assurance. This can build trust and transparency in the food supply chain.
- **Development of Portable and On-Site Irradiation Units:** Smaller, more mobile irradiation units could be developed for use in remote agricultural regions or processing facilities, reducing transportation costs and improving access to this technology.
- **Biopreservation Enhancement:** Research could explore the potential of irradiation to enhance the effectiveness of biopreservatives (naturally derived antimicrobial compounds), leading to more sustainable and consumer-friendly preservation strategies.
- **Addressing Antimicrobial Resistance:** As antimicrobial resistance becomes a growing concern, irradiation offers a non-chemical method to eliminate foodborne pathogens, potentially reducing the reliance on antibiotics in the food production chain.

In conclusion, food irradiation stands as a well-established and scientifically validated technology offering significant advantages in enhancing food safety, extending shelf life, and reducing post-harvest losses. Its ability to effectively eliminate harmful pathogens, control pests, and delay spoilage contributes to a more secure and sustainable food supply. While concerns regarding nutritional changes and consumer perception exist, scientific evidence largely supports the safety of the process when

applied correctly within recommended dosage limits. Overcoming misconceptions through education and transparent communication remains crucial for wider acceptance and utilization of this valuable tool.

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Little Leaf and Witches' Broom Disease of Jackal jujube (*Ziziphus oenoplea* L caused by Phytoplasma: Etiology, Impact, and Management

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Ziziphus oenoplea, L. a plant belongs to Rhamnaceae family is a medicinally important for its therapeutic and nutritional benefits. Despite of its medicinal importance, this plant facing severe threat by Little Leaf and Witches' Broom (LLWB) disease caused by phytoplasma. The phytoplasmas are small wall less prokaryotes belongs to the mollicutes. They are phloem limited organisms cause many abnormalities in various plants. In this present chapter we discussed about symptomatology such as severe reduction of leaf size and length, yellowing of leaves due the lack of chlorophyll. A dense mass of shoot appear as cluster which resembles broom and finally defoliation occurs drying death of tree gradually. This disease is primarily spread by the phloem feeding insects such as leaf hoppers. The main diagnostic tools available are visual symptoms observation, Dein's staining technique, scanning electronic microscopic and transmission electron microscopic assay, PCR detection using universal and nested primers, sequencing 16Sr ribosomal gene and blast analysis. The integrated approach is required to manage the disease such as use of healthy planting material, removal and destruction of infected plants and trees, liming of trunk or stem of the tree, insect management using yellow and blue sticky traps, use of parasites and predators of leaf hoppers, spraying of neem oil and insecticides. The current knowledge aids to support the ongoing efforts to develop sustainable management solutions for little leaf and witches broom disease of *Ziziphus oenoplea*

Key words: Phloem limited organisms, *Ziziphus*, detection, vector and management

1. Introduction

The genus *Ziziphus* (family: Rhamnaceae) comprises perennial, thorny shrubs known by common names such as wild jujube and Jackal jujube. Ecologically and economically significant across tropical and subtropical regions, various species are cultivated for their nutritional fruit and hold a esteemed place in traditional medicine (Eswari *et al.*, 2014). In India, their distribution spans the Western Ghats and includes states such as Maharashtra, Karnataka, Telangana, Andhra Pradesh, and Kerala (Ajay *et al.*, 2016).

Among these, *Ziziphus oenoplea* is a key species of profound importance. Despite its resilience, the productivity and health of this plant are critically compromised by various pathogens. Phytoplasma infections, in particular, represent a highly destructive group of diseases that threaten its cultivation. These phytoplasmas are minute, cell wall less bacteria of the class *Mollicutes* that reside obligately within the phloem of plants and the bodies of insect vectors Doi *et al.* (1967). Globally, they incite a spectrum of diseases marked by distinctive syndromes such as phyllody, virescence, yellowing, severe stunting, and the namesake symptoms of little leaf and witches' broom (Marccone *et al.*, 2018; 2021; Lee *et al.*, 1993; 2000). These morphological anomalies arise from the pathogen's interference with plant development pathways, ultimately crippling growth and reproductive potential (Rao, 2021).

In *Ziziphus oenoplea*, the Little Leaf and Witches' Broom (LLWB) disease complex presents a major phytosanitary challenge (Ghosh *et al.*, 1985). Afflicted trees exhibit dramatic foliar reduction and a profuse, abnormal proliferation of shoots forming dense, broom-like structures (Snehi *et al.*, 2020). This symptomatic profile severely impairs photosynthesis and tree architecture, frequently resulting in significant yield loss and eventual plant death, with direct consequences for both economic livelihoods and biodiversity.

Given the significant threat posed by LLWB disease, a cohesive synthesis of existing knowledge is urgently needed. This chapter consolidates contemporary research on the etiology and impact of the disease in *Z. oenoplea*. It comprehensively addresses the identification and molecular classification of the associated phytoplasma, outlines the disease cycle and symptomatology, reviews advanced diagnostic methodologies, and critiques current and prospective management strategies.

2. Etiology

Causal agent : *Phytoplasma*

Nomenclature: *Candidatus phyoplasma balanitae*

Group : 16SrVF

Phytoplasmas are the major group of plant pathogens which has economically significant comes under the class Mollicutes. These pleomorphic organisms were first discovered by Doi *et al.* (1967). Earlier these were called as Mycoplasma like organisms (MLOs) due to their lack of a cell wall and pleomorphic structure and later in 1995 renamed as phytoplasma. These group of bacteria specifically reside in the phloem sieves of plants and the bodies of insect vectors. Since their inability to cultivate artificial culture media, they are taxonomically classified under the "*Candidatus*" genus based on molecular charactrization of their 16S ribosomal gene (16SrRNA) (Lee *et al.*, 1993; 2000).

These phytoplasmas are primarily transmitted by phloem feeding insects like leafhoppers, plant hoppers and psyllids as well as through grafting (Weintraub & Beanland, 2006). They can infect and colonize more than 1000 plant species, including agricultural, horticultural, forestry and weeds (Rao, 2021). The pathosystem associated with phytoplasma infections can cause significant economic losses and threaten food production by altering biodiversity and reducing productivity (Marccone *et al.*, 2018; 2021).

Symptomatology

Main symptoms produced by LLWB include:

- **Yellowing:** Yellowing (chlorosis) of foliage due to reduced chlorophyll production
- **Stunting and little leaf:** Severe reduction in growth, shortening internodes, particularly affecting leaf size and stem length.
- **Witches' broom** - many secondary branches covered with small leaves giving a bushy appearance resembles broom.
- **Proliferation of branches:** Due to excessive axillary branching, appear as abnormal bushy type
- **Defoliation and drying** : Progressive defoliation leading to bare, drying branches
- **Decline** : Overall plant weakness and reduced vigor
- **Flower abnormalities** : including phyllody (leaf-like flowers)

4. Vector and Epidemiology

In natural condition, phytoplasmas are transmitted by phloem feeding insect vectors such as leafhoppers, plant hoppers and psyllids (Bertaccini, 2022). Many researchers has specifically identified several insects which has vector role in spreading of Jujube Witches' Broom (JWB) phytoplasma, including *Hishimonus sellatus* and *Hishimonus phycitis* (La and Woo, 1980; Wang et al., 1981).

Later work by Chen et al. (1984) added *Hishimonoides aurifascialis* and *Hishimonus lamellatus* to this list. Furthermore, PCR-based analysis by Hao et al. (2015) indicated that *Cicadella viridis* and *Kolla paulula* are also likely vectors.

Beyond insect transmission, a significant route of spread is through grafting with infected plant materials, a common propagation method for jujube (Liu et al., 2010). However, studies have confirmed that the pathogen is not transmitted through pollen, seeds, or soil (Chen et al., 1984). While the jujube tree is the primary host, the JWB phytoplasma has been found to infect a range of other fruit trees and herbaceous plants (Chen et al., 2022).

5. Detection

The LLWB can be detected and diagnosed using different methods such as symptomatology, staining techniques, microscopic analysis, PCR based detection and sequencing of 16Sr ribosomal gene.

a. Symptomatology: The LLWB disease in *Ziziphus* presents unique and easily identifiable symptoms. Most notably, plants exhibit the characteristic "little leaf" and dense "witches' broom" growths. Beyond these distinct signs, infected plants also commonly suffer from general yellowing, severely stunted leaf size, premature leaf loss, and eventual drying.

b. Staining techniques : after the symptomatology, staining techniques are quite often used method for the detection of phytoplasma especially staining of phloem tissue because of the colonization of phytoplasma particularly in phloem tissues of the plants. Fluorescence microscope after staining with 4'-6- diamidino-2-phenylindole·2HCl (DAPI) of the phloem tissues can easily distinguish the presence of phytoplasma cells in infected tissue compared to healthy one (Liu, et al., 2010).

c. Microscopic analysis: Following symptom assessment, staining techniques offer a direct method for detecting phytoplasmas. Since these pathogens specifically colonize the plant's phloem tissue, staining this area is key. Using a DAPI (4'-6- diamidino-2-phenylindole·2HCl) stain under a fluorescence microscope, for instance, clearly reveals the presence of phytoplasma cells in infected samples, providing a visible contrast to healthy tissue.

d. PCR based detection: Application of PCR and nested PCR method for the detection of LLWB phytoplasmas in *Ziziphus* diseased plants has significantly facilitated the identification (Gundersen and Lee, 1996, Bertaccini and Lee, 2018). PCR-based detection targeting the 16S ribosomal genes of phytoplasmas, using universal and nested primers, remains one of the most reliable and widely adopted molecular approaches for identifying these pathogens. Techniques such as restriction fragment length polymorphism (RFLP) analysis and sequencing of the 16S rRNA gene are also commonly employed to ensure precise detection of phytoplasmas in both plant and insect samples.

e. Sequencing and phylogenetic tree analysis: The 16S rRNA gene in phytoplasmas stays mostly unchanged across different strains, which makes it a dependable choice for identifying them and studying their genetic relationships. By looking at this gene, researchers can figure out how different phytoplasma isolates are related and how they vary (Bertaccini *et al.*, 2014). It has been found that the particular phytoplasma affecting *Ziziphus* plants belongs to the 16SrV-F group.

f. Other methods: Recent progress in molecular diagnostics has led to the development of advanced tools such as LAMP and CRISPR/Cas12-based assays for identifying plant pathogens (Han et al.,

2015). One significant breakthrough is the “all-in-one” Dual CRISPR assay, which has proven effective in detecting JWB phytoplasma. This method offers high sensitivity, precision, and efficiency. Its results can be seen with the naked eye, and it does not rely on expensive laboratory instruments, making it ideal for quick, on-site detection in the field.

6. Management

- Field sanitation to removal and destruction of the infected plants are the first line of disease management as it reduces the source of inoculum.
- Phytoplasma disease free planting materials can be get through tetracycline treatment, heat therapy (Tian *et al.*, 1993) or cryopreservation (Wang *et al.*, 2015).
- Use of yellow or blue sticky traps to control the leaf hoppers which that can carry and transmit LLWB phytoplasma.
- Application Neem oil for the control of leaf hoppers.
- Use selected ecofriendly insecticides to manage the leaf hoppers and not affecting the other beneficial insects.
- The use of comprehensive control measures may not eliminate phytoplasmas completely, but control the phytoplasma density at a relatively economical level.

7. Conclusion

As conclusion remarks, the medicinal Jackal jujube shrub is highly vulnerable to the devastating Little Leaf and Witches' Broom disease, caused by a phytoplasma. Critical knowledge gaps persist, especially regarding the natural insect vectors that spread the disease. To develop effective control strategies, focused research is urgently needed to understand the pathogen's host range, insect transmission cycle, and plant-pathogen interactions. Future efforts must prioritize disease surveillance, identifying key vectors, and developing protective measures to safeguard this vital plant.

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Nanofertilizers — A Smart and Sustainable Approach toward Modern Agriculture

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1. Introduction

Agriculture in the twenty-first century faces immense challenges due to rapid population growth, shrinking arable land, and the impacts of climate change. Conventional fertilizers, though critical to boosting productivity, have led to issues such as nutrient leaching, soil degradation, and environmental pollution. In this context, nanotechnology has emerged as a promising tool to revolutionize crop nutrition management by enhancing nutrient-use efficiency and minimizing environmental losses (Kah et al., 2018). Nanofertilizers represent a new generation of agro-inputs designed to deliver nutrients in a controlled and efficient manner at the nanoscale, aligning with the goals of precision and sustainable agriculture.

In India, increasing emphasis has been placed on developing eco-friendly, high-efficiency fertilizers through nanotechnology. The Indian Farmers Fertilizer Cooperative (IFFCO) introduced Nano Urea, a liquid formulation that delivers nitrogen at nanoscale size for improved foliar absorption and reduced wastage. This innovation reflects India's shift toward sustainability and aligns with the ICAR's national framework for nano-based agricultural inputs (IFFCO, 2021).

2. Concept and Classification of Nanofertilizers

Nanofertilizers are nutrient carriers engineered with at least one dimension below 100 nanometers. Their small size, large surface area, and enhanced reactivity facilitate better nutrient solubility, uptake, and translocation within plants (Rameshaiah et al., 2015). These fertilizers can be broadly classified into three categories:

1. **Nanoscale fertilizers:** Nutrients directly prepared in nano form, such as nano-ZnO, nano-Fe₂O₃, or nano-TiO₂.
2. **Nutrient-loaded nanomaterials:** Nutrients encapsulated or adsorbed onto nanomaterials such as nano-clays, zeolites, or carbon nanotubes for slow and controlled release.
3. **Nanosensors and smart delivery systems:** Advanced formulations that respond to plant physiological signals or soil conditions to release nutrients precisely when needed.

Compared to conventional fertilizers, nanofertilizers offer prolonged nutrient availability, minimal leaching, and improved crop growth parameters due to enhanced nutrient uptake kinetics (Liu & Lal, 2015).

3. Synthesis of Nanofertilizers

Nanofertilizers can be synthesized through physical, chemical, and biological routes, each offering different advantages regarding cost, scalability, and environmental impact.

- **Physical methods** involve high-energy milling, vapor deposition, and ultrasonication techniques.
- **Chemical methods** use precipitation, sol-gel, or hydrothermal reactions to produce nanoparticles with specific size and morphology.
- **Green synthesis**, however, is gaining popularity because it employs plant extracts, bacteria, or fungi as reducing and stabilizing agents, eliminating toxic chemicals and reducing environmental footprint (Shaikhaldin et al., 2021).

In India, various research institutes such as ICAR-NIASM Baramati and ICAR-IARI New Delhi have successfully synthesized nano-Zn, nano-Fe, and nano-Cu using biological and chemical routes for field evaluation in crops like rice, wheat, and onion (Raliya et al., 2017). The green synthesis approach using neem, aloe vera, and moringa extracts has shown promising results in creating bio-compatible nanofertilizers that enhance both yield and soil health.

4. Mechanisms of Nutrient Uptake and Release

The efficiency of nanofertilizers lies in their targeted delivery and controlled nutrient release mechanisms. Nanoparticles can penetrate plant surfaces either through stomatal openings (foliar application) or root epidermis (soil application). Their small size allows for enhanced mobility in plant tissues and sustained nutrient availability. Controlled release can occur through:

- Diffusion (from porous nanocarriers),
- Biodegradation (of organic coating materials),
- Physicochemical triggers (pH, moisture, or enzymatic activity).

For instance, nano-urea molecules exhibit gradual nitrogen release that synchronizes with the plant's demand, reducing volatilization and nitrate leaching losses (Singh et al., 2023). This efficiency supports India's national mission of reducing chemical fertilizer use by 20–25% without compromising yield.

5. Applications in Crop Growth and Productivity

Numerous studies worldwide and within India demonstrate that nanofertilizers can significantly improve growth rate, chlorophyll content, photosynthetic efficiency, and yield. For example:

- Nano-ZnO improved seed germination and root elongation in wheat and maize (Dimkpa et al., 2020).
- Nano-Fe₂O₃ enhanced chlorophyll biosynthesis and increased biomass accumulation in rice.
- Nano-TiO₂ stimulated enzymatic activity and promoted carbon fixation efficiency.

In India, field trials conducted by IFFCO and ICAR reported that foliar application of Nano Urea (500 mL per acre) could replace up to 45 kg of conventional urea, enhancing nitrogen-use efficiency by 8–10% (IFFCO, 2022). Similarly, nano-Zn fertilizers applied in onion, green gram, and wheat showed higher nutrient uptake and yield under semi-arid conditions (Rameshaiah et al., 2015).

Overall, nanofertilizers act as smart nutrient systems capable of precise delivery, improving both the quantitative and qualitative parameters of crops while minimizing nutrient losses.

6. Role in Nutrient Use Efficiency and Environmental Sustainability

One of the most compelling advantages of nanofertilizers is their ability to enhance nutrient-use efficiency (NUE)—that is, the proportion of applied nutrients actually absorbed and utilized by plants. Conventional fertilizers often exhibit low NUE due to volatilization, leaching, and runoff losses. In contrast, nanofertilizers release nutrients slowly and steadily, ensuring synchronization between nutrient availability and plant demand (Liu & Lal, 2015).

The nano-scale size and surface charge of these particles allow them to adhere more effectively to plant tissues, improving absorption. Research has shown that nano-urea, for example, can improve nitrogen-use efficiency by nearly 20 %, while reducing greenhouse gas emissions associated with excessive urea use (Singh et al., 2023).

In India, the IFFCO Nano Urea initiative represents a breakthrough in sustainable nutrient management. By replacing a 45-kg bag of conventional urea with a 500-mL bottle of nano urea, farmers achieve comparable yields while reducing nitrogen losses and production costs. This approach supports India's commitment to sustainable agriculture and carbon neutrality under national programs such as the *National Mission on Nano Science and Technology* and ICAR's *climate-smart agriculture projects*.

Nanofertilizers also contribute to environmental protection by reducing eutrophication and soil contamination. Controlled-release formulations minimize the frequency of fertilizer application, reducing labor and fuel usage. Additionally, nano-zeolite and nano-clay carriers help retain nutrients in the root zone, limiting their mobility into groundwater (Kah et al., 2018).

7. Comparative Performance with Conventional Fertilizers

Comparative trials between nano- and conventional fertilizers have consistently demonstrated improved performance with nanofertilizers in terms of yield, nutrient uptake, and physiological efficiency. For example, studies by Raliya et al. (2017) revealed that nano-ZnO application in wheat enhanced grain yield by 12 % and zinc content by 18 % compared with traditional zinc sulfate.

Similarly, foliar spraying of nano-Fe₂O₃ improved photosynthetic pigment concentration, and nano-TiO₂ accelerated nitrate reductase activity, resulting in improved nitrogen metabolism (Dimkpa et al., 2020). These findings indicate that nanofertilizers not only enhance productivity but also enrich the nutritional quality of crop—a significant consideration for addressing micronutrient malnutrition.

In onion and green gram experiments under Indian field conditions, nano-Zn and nano-Fe treatments improved chlorophyll content, plant height, and bulb weight significantly over conventional treatments (Shaikhaldain et al., 2021). Therefore, integrating nanofertilizers into existing fertilizer schedules can optimize plant nutrition and reduce dependency on bulk chemical inputs.

8. Challenges and Risk Assessment

Despite their potential, nanofertilizers face several challenges that must be addressed before large-scale adoption. Production cost, regulatory gaps, and potential nanotoxicity remain areas of concern. The behavior of nanoparticles in soil ecosystems—including their interaction with microorganisms, bioaccumulation, and long-term fate—is not yet fully understood (Kah et al., 2018).

Excessive nanoparticle accumulation could disrupt soil microbial communities or enter the food chain through edible plant parts. Therefore, risk assessment frameworks need to be strengthened, and standardization in nanoparticle size, composition, and release behavior must be established (Rameshaiah et al., 2015).

Moreover, public perception and farmer awareness pose social challenges. Many small-scale farmers in India still rely on conventional fertilizers due to affordability and familiarity. Government-supported extension programs and demonstrations, such as those initiated by IFFCO and ICAR, are crucial to build trust and adoption of nanofertilizers among farming communities.

9. Future Prospects

The future of nanofertilizers in modern agriculture lies in smart, integrated nutrient-delivery systems that combine nanotechnology with digital agriculture tools. Developments such as sensor-based nutrient monitoring, bio-degradable nanocarriers, and hybrid nano-bio fertilizers will further revolutionize crop nutrition (Singh et al., 2023).

In India, the government's support for nanotechnology through the Department of Science and Technology (DST) and ICAR-Nanotechnology Centers promises rapid progress. Collaborative research between public institutions and private industry (such as IFFCO, Rashtriya Chemicals and Fertilizers, and Tata Chemicals) is accelerating the translation of laboratory research into commercial products.

Future work should also focus on eco-toxicological studies, field-scale validation, and economic viability to ensure that nanofertilizers are both safe and beneficial for diverse agro-ecological zones. Incorporating nanofertilizers into integrated nutrient management (INM) frameworks can balance productivity with sustainability, a goal central to India's agricultural policy.

10. Conclusion

Nanofertilizers offer a smart, sustainable, and transformative approach to modern agriculture. Their ability to deliver nutrients efficiently, enhance plant metabolism, and minimize environmental losses positions them as key tools for achieving food and environmental security. In the Indian context, innovations such as IFFCO Nano Urea and ICAR's nano-nutrient research have already shown how these technologies can contribute to self-reliant and climate-smart agriculture.

However, widespread adoption requires addressing challenges related to safety, regulation, and cost-effectiveness. With responsible innovation and policy support, nanofertilizers could redefine agricultural input management—leading to a future where high yields coexist with ecological balance.

Relevant Research Findings at Departmental of Agronomy

Cereal crop: In rice crop the application of 50 % N + 100 % P₂O₅ and K₂O + 3 spray of Nano-Urea @ 0.4 % at 30, 45 and 60 DAT recorded significantly higher grain yield (4817.80 kg ha⁻¹), straw yield (5956.26 kg ha⁻¹), biological yield (10774.06 kg ha⁻¹) and harvest index (44.72%) (Parve et al., 2023). In sweet corn experiment the significantly higher green cob, green stover and total biological yield was recorded by 75% RDF (CFB) + foliar application of synthesized nano urea conjugate @ 30 ppm (Patil et al., 2024).

In rice crop the treatment combination of nano urea conjugate at 40 ppm with 100% Wool mulch with micronutrients (200GSM) showed higher values for plant height, dry matter accumulation, and all growth indices (Vinod et al., 2025).

Oil seed crop: The mustard crop grown with application of 75 % of RDN along with 40 ppm foliar spray of nano urea combination produced significantly higher seed yield (Pandav et al., 2022).

In mustard crop the treatment combination of synthesised nano phosphorous conjugate with concentration of 100 ppm showed highest seed yield, straw yield than rest of the treatment (Pisal et al., 2024)

Pulse crop: In green gram the treatment combination of 100% RD of N and P + 100% RD of K and 50% RD of N through nano-urea + 50% RD of P through nano-phosphorous (2 sprays at 45 and 60 DAT)] resulted in higher growth attributes, such as number of functional leaves plant⁻¹ and number of branches plant⁻¹ as well as higher yield attribute viz., weight of pods plant⁻¹ and grain yield (Chavan et al., 2024b).

Another one experiment on green gram finding reveals that the treatment combination of nano phosphorus conjugate @ 200 ppm combined 100% wool mulch with micronutrients (200GSM), resulted in significantly higher growth attributes, such as plant height, number of trifoliolate leaves plant⁻¹, number of branches plant⁻¹, and plant dry matter accumulation and number of root nodule (Surendra et al., 2025).

Horticultural crop: In okra crop the treatment combination of synthesised nano urea conjugate with concentration of 120 ppm showed highest plant height, Number of nodes plant⁻¹, Dry matter accumulation plant⁻¹ (g), Number of fruits plant⁻¹, Fruit yield (q ha⁻¹) (Ghuge et. al., 2024)

In white onion crop the treatment combination of 100% RD of N and P + 100% RD of K and 50% RD of N through nano-urea + 25% RD of P through nano phosphorous (2 sprays at 45 and 60 DAT)] showed higher growth attributes i.e. number of functional leaves plant⁻¹ and bulb neck diameter as well as yield attributes i.e. bulb fresh weight bulb⁻¹ and mean bulb yield. (Chavan et al., 2024a)

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Microclimate Controlled Dairy Housing System - A novel approach

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Introduction

Climate change can securely affect the productivity and economic viability of the Livestock Production Systems. High ambient temperature affects particularly high yielding, low disease resistant and low heat tolerant crossbred population than the indigenous livestock. Heat stress induced by change in climate also affects the reproductive performance of dairy animals compared to meat producing animals. The negative impact of temperature rise on total milk production in India has been estimated to be 1.6 million tonnes by 2020 and more than 15 million tonnes by 2050. (Upadhyay et al, 2007). An increase in the global temperature of only 1.5 to 2.5°C may lead to extinction of approximately 20-30% of livestock and animal species in the world (Agarwal and Upadhyay, 2013). To mitigate the adverse effects of extreme weather events, technologies suitable to local conditions and modifications in housing of animals, management and feeding practices need to be designed for future. Environmental modifications aiming at microclimate of the animal to help alleviate heat stress problems are structural orientation, ventilation, use of cooling system along with shade is the best (Mech et al, 2013). An integrated approach was made in the model project on the “Environmentally Controlled Housing for Dairy cattle” designed at Dairy Experimental Station, College of Veterinary Science, Tirupati under Sri Venkateswara Veterinary University, Tirupati.

Environmentally Controlled Dairy Housing System Model:

The environmentally controlled dairy housing model was designed by giving due consideration to orientation of shed, roof projection and micro-environment of the animal by providing cooling system besides comfort to the animal and reducing the labour costs involve in feeding and management. The micro environment around the animal is modified by to increase the heat dissipation and cooling the air around the animal to reduce the heat load and stress on the animal.

The model (Illustration No: 1) is designed for the first time in India with the funds of Rs.12.0 Lakhs under “Modernization of the Agriculture Farms” sponsored by ICAR, New Delhi. The various components of the designed model installed are

- 1 .Free stall system
2. Rubberized Mattress
3. Cooling Pads
- 4 .Exhaust Fans
- 5 .Automatic Waterers
6. Automatic Concentrate dispenser system
7. Control unit containing temperature sensor
8. Water tank (outside the shed) for re-circulation of water
- 9 .Drinking water tank
10. Feed bin



Illustration: I Model of Environmentally controlled Dairy housing system

1. Free stall system: The standing animal space in the shed for is divided into compartments (Free stalls) and one animal is placed in each stall with adequate space for the animal to lie down and space for the milker to milk each animal. The free stall compartment keeps provision to eat its own share of concentrate and green fodder besides preventing to consume other animals share of feed and water. Within the free stall, above the manger for offering green fodder, provision is made for bowl type Waterers for drinking water and cup type concentrate feeders for consuming concentrate feed.



Free stall or Cow stall compartment for each animal

2.Rubberized Mattress:

Rubberized mattress is provided within each free stall for the animal to lie down to have comfort. The rubberized mattress helps in prevention of the udder injuries to high yielding animals beside slippage of animal resulting in lameness. Provision of rubberized mattress is aimed at increasing cow comfort which improve milk production, herd health and longevity of animal. Cows which lie down for 12 to 14 hr/day are more productive than those with lower lying times (Dressler et al 2005).



3.Cooling Pads.

A total of four cooling pads are arranged on one side of side wall which gets moistened once the control unit is switched on. These cooling pads are arranged exactly opposite to the exhaust fans on other side of wall of the shed. Provision of the cooling pads inside the shed helps in cooling of the air around the animal there by reducing the temperature load on animal and prevents stress on animal to maintain the productivity in high yielding animals.



Cooling Pads for cooling of air in the shed

Exhaust Fans:

Exhaust fans are arranged on other side operate to the cooling pads so as to pump out the hot air from the shed and supply fresh air into the shed with 0.25 HP motor to each exhaust fan installed in the shed. The exhaust fans inside helps in increasing air movement there by enhancing / improving rate of heat loss from the animal. An integrated approach of exhaust fans and cooling pads is more effective mean of reducing the heat load on the animal. The cooling pads and exhaust fans helps in evaporative cooling and forced ventilation of body surface maximizing the cooling effect on the animal. Venugopal (2011) reported that the economic analysis has shown the break even point for fan and mist system is an increase in milk production of 0.81 kg / day.



Exhaust fans for exchange of air inside the shed

5. Control Unit:

The control unit fixed in the middle of the on one side of long-axis of shed controls the number of fans to be operated depending on the temperature outside the shed besides controlling the cooling pads. A temperature sensor is attached to the control unit which helps in reading the internal temperature of shed / controlling the temperature inside the shed. A hygrometer is placed in the shed separately to know the humidity inside the shed. Research shows that in lactating animal's humidity above 70% effect the milk production besides reducing evaporative cooling adding to head load on animal.



Automatic Temperature control unit

Regulation of temp is possible as per need (Picture showing animal shed temp as 27.2 °C)

6. Water tank for re-circulation of water:

A water tank of 500 liter capacity is installed outside the shed with a 0.5 HP motor fixed to the shed for recirculation of the water through the cooling pads for better utilization of the water. The water after circulating through the cooling pads again enters the water tank from where the water was pumped into the cooling pads, they by conserving the water.

7. Automatic Bowl drinkers (Waterers):

A water tank arranged inside the shed is connected to the automatic bowl drinkers placed in each free stall for drinking purpose. The animal can drink the water as and when its likes to drink and once the water bowl fills up, it stops the water automatically preventing the wastage/overflow of water.

8. Automatic Cup Concentrate feeder:

A concentrate feeder is arranged in the free stall where provision is made for dispensing of correct quantity of concentrate mixtures for each animal as per the production requirement of animal coming from the fed bin.



Each cow stall with Automatic waterer and concentrate feeder

9 .Water tank:

A water storage tank is provided at certain height within the shed along the short axis of shed to supply the required quantity of water needed for all animal which is connecting to the automatic bowl drinker system.

**10. Feed Bin/ Feed Bunk:**

Adequate quantity of concentrates required for all the animals in the shed is provided in the feed bin/ feed bunk. The feed in the Feed bin/ Bunk is turn channelizes into the automatic concentrate dispensing system which distributes to the concentrate feeder for consumption by the animal.

**Conclusion:**

A housing system was designed to maintain/sustain the productivity of high yielding animals giving due consideration to orientation, effective cooling systems (Exhaust fans + Cooling Pads), reducing feed and labour requirements (Automatic Waterers and Automatic Concentrate Dispenser system) besides the control of the temperature and humidity inside the shed. Depending on the climatic conditions, suitable building materials, cooling systems can be selected for installation in the dairy farms. The micro-environment around the animal needs to be modified to a greater extent to reduce heat load on the animal besides making the animal more comfortable (Rubberized Mattress) and provision of free space for the animal to lie down.

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Parasitic Helminths and Host Microbiota: Exploring the Connection

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There are the complex biological interplay between parasitic helminths and the host microbiota. Parasitic helminths-multicellular worms that inhabit the host body-have evolved mechanisms to survive and modulate host immunity, often interacting with the gut microbiota in complex ways. The microbiota, comprising bacteria, fungi, and viruses, plays an essential role in regulating host metabolic balance, immune regulation, and overall health. This chapter analyzes the varied associations between these organisms, their co-evolutionary significance, and their potential therapeutic applications in modern biomedical research.

Keywords

Helminth-Microbiota Gut Microbiota; Host Immune Modulation; Co-existence; Helminth Therapy

Introduction

Parasitic helminths represent a diverse group of multicellular eukaryotic organisms that inhabit the tissues or organs of their hosts, extracting nutrients and modulating host physiology to ensure their survival. The host microbiota, by contrast, consists of a vast community of commensal, symbiotic, and sometimes pathogenic microorganisms that inhabit various niches such as the skin, oral cavity, and intestinal tract. The largest and most studies gut microbiota, which inhabits the gastrointestinal tract.(Jarett et al., 2021) There are equilibrium between animal host, pathogen, which is parasitic helminth and microbiome in the gut of host, host get protection, immune system modulation and vitamin supplementation. When this equilibrium breaks pathogen and microbiome causes infection and side by side inflammation. Recent research have a profound bidirectional relationship between helminth parasites and the host microbiota, revealing how these interactions shape immunity, metabolism, and disease outcomes.

Biology of Parasitic Helminths

Helminths are broadly classified into three major groups: nematodes (roundworms), cestodes (tapeworms), and trematodes (flukes). Each group displays unique life cycles, infection routes, and host adaptation mechanisms. Nematodes like *Ascaris lumbricoides* reside in the intestine, absorbing the nutrients directly from the gut lumen. Cestodes, such as *Taenia* spp. attaches to the intestinal wall using scolex structure, while trematodes often infect blood vessels or liver tissues. Their prolonged persistence in the host is facilitated by immune evasion strategies such as secretion of immunomodulatory molecules, antigen mimicry, and induction of regulatory T-cell responses.

Composition and Role of the Host Microbiota

The host microbiota comprises a dynamic consortium of microorganisms, dominated by bacterial phyla such as *Firmicutes*, *Bacteroidetes*, *Actinobacteria*, and *Proteobacteria*. In the gut, these microbes contribute to digestion, synthesis of vitamins, and production of short-chain fatty acids (SCFAs) like acetate, propionate, and butyrate, which maintain intestinal epithelial health. The microbiota also serves as an immunological educator, training the immune system to distinguish between pathogens and commensals. Alterations in microbial composition called dysbiosis - can trigger diseases such as inflammatory bowel disease (IBD), obesity, and type 2 diabetes

Helminth–Microbiota Interactions: Co-evolution and Mutualism

The relationship between helminths and microbiota reflects a co-evolutionary process that enables both entities to coexist within the host ecosystem. Helminths may promote the growth of bacterial species that favor their survival by altering gut pH, mucus production, or nutrient availability. Conversely, certain bacterial metabolites enhance helminth reproduction or survival, exemplifying mutualistic cooperation.

Importance of Helminths - Microbiota Association

The association between parasitic helminths and host microbiota is not merely incidental; it is a co-evolutionary relationship that affects both partners and the host. This association influences immune regulation, nutrient metabolism, and the survival strategies of both helminths and microbial species. Understanding this relationship is essential to reveal how parasites manipulate host biology and how the microbiota can either support or hinder parasitic colonization. This dynamic relationship involves direct and indirect mechanisms and has potential implications for novel therapeutic strategies.

- **Immune System Modulation:** Helminths interact with the host immune system by promoting immune tolerance and modulating inflammatory responses. This helps parasites survive and influences how the host's immune system reacts to infections.
- **Effect on Gut Microbiota:** Helminth infections can alter the diversity and composition of gut microbiota. The induced immune responses, such as Th2 activity, affect bacterial populations, increasing some beneficial species while reducing harmful ones.
- **Nutrient Absorption and Parasite Survival:** Both helminths and microbiota influence nutrient absorption. Microbes aid in nutrient breakdown, while helminths can modify gut function to support their own survival. In turn, the microbiota creates favorable conditions for helminth reproduction.
- **Gut Dysbiosis and Health Impact:** Helminth infections may cause microbial imbalance (dysbiosis), making the host more vulnerable to diseases such as inflammation and metabolic disorders
- **Therapeutic potential:** Recent studies have explored the therapeutic potential of controlled helminth exposure in treating autoimmune and allergic disorders. By modulating immune responses and influencing gut microbiota, helminths could restore immune tolerance and reduce inflammation. This concept, known as “helminth therapy,” is being investigated for conditions like inflammatory bowel disease, multiple sclerosis, and asthma. The underlying mechanism involves rebalancing the microbiota and promoting regulatory immune pathways.

Microbiota-Gut -Brain Axis

The gut microbiota communicates bidirectionally with the central nervous system, forming what is known as the microbiota–gut–brain axis. This connection means that gut health can influence mood, behaviour, and cognitive function. Helminth infections may affect this axis by altering microbial composition and gut-derived neurochemicals. For example, helminth-induced immune changes can modify signaling through the enteric nervous system, leading to alterations in gut motility and neural communication.

Enteric nervous system (ENS) and parasitic infections: The enteric nervous system (ENS) also known as “SECOND BRAIN” is a vast network of neurons embedded in the walls of the GI tract. It regulates digestive processes and communicates directly with the CNS.

- **Parasitic infection and ENS modulation: three major mechanisms** Modification in nerve distribution – parasites may induce structural changes in gut wall, affecting the distribution and density of enteric neurons. This can disrupt motility and gut-brain communication.
- **Alteration of neurochemicals levels**

Parasites can influence the level of neurotransmitters (e.g. serotonin, acetylcholine, dopamine) and neuropeptides in the gut, which in turn affect mood and behaviour.

➤ **Altered neuronal functions**

Chronic infection or inflammation can impair neuronal signaling and cause long-term changes in gut reflexes and sensory processing.

Example of parasite affecting the ENS: *Cryptosporidium parvum*, *Giardia duodenalis*, *Trypanosoma cruzi*, *Schistosoma mansoni*

How do helminths and microbiota interact?

Helminths and microbiota interact through both direct and indirect mechanisms. Direct interactions occur without host mediation, involving physical contact, nutrient competition, and biochemical signaling. Certain helminths can attach to bacterial surfaces, ingest microbes, or harbour bacterial endosymbionts that influence gut ecology. For example, the nematode *Trichuris suis* benefits from bacterial biofilms that promote egg hatching and colonization efficiency. In contrast, some helminths may eliminate pathogenic bacteria through ingestion, indirectly favoring beneficial commensals.

Indirect interactions involve host-mediated processes such as immune regulation, epithelial remodeling, and changes in gut physiology. Helminth infections can alter mucus composition, increase goblet cell numbers, and modulate mucin gene expression (e.g., MUC2, MUC5AC), thereby shaping microbial niches. Additionally, by disrupting epithelial tight junctions, helminths increase epithelial turnover rates, influencing bacterial adherence and colonization. The gut microbiota, in turn, supports epithelial repair and homeostasis by producing short-chain fatty acids (SCFAs), demonstrating mutualistic adaptation.

From an immunological perspective, helminth–microbiota interactions profoundly affect both innate and adaptive immune responses. In innate immunity, Toll-like receptors (TLRs) recognize microbial and helminth-derived ligands, triggering regulated cytokine production. Helminth-secreted antimicrobial peptides can selectively protect commensals, while suppressing pathogenic species. These activities modulate inflammasome signaling and contribute to immune tolerance.

Adaptive immune regulation is a hallmark of helminth–microbiota symbiosis. Helminths such as *Heligmosomoides polygyrus* and *Trichuris muris* induce T-helper 2 (Th2) and regulatory T-cell (Treg) responses, promoting anti-inflammatory states. They stimulate transforming growth factor-beta (TGF- β) and interleukin-10 (IL-10), both essential for maintaining immune balance and preventing excessive

inflammation. Through these effects, helminths can reduce autoimmune and allergic conditions, partly mediated by the microbiota's influence

Functionally, these interactions maintain gut homeostasis, epithelial integrity, and microbial diversity. Helminth infection enhances interleukin-25 (IL-25) signaling, which supports mucosal repair and barrier function. Simultaneously, helminths promote colonization resistance by encouraging the proliferation of beneficial bacteria such as *Lactobacillus* and limiting the overgrowth of pathogenic species. The resulting ecological balance contributes to reduced inflammation and enhanced resilience against enteric diseases.

Overall, helminths and microbiota coexist in a complex ecological and immunological network within the gut. Their interactions involve molecular cross-talk, structural modulation, and immune adaptation. Recognizing these relationships has therapeutic significance - helminth-derived molecules and microbiota-based interventions hold assurance for treating inflammatory bowel disease, allergies, and other immune-mediated disorders.

Impacts of Helminths - Modified Microbiome on Host Immunity

- **Helminths modify the bacterial intestinal microbiota:** The bacterial microbiota provide several functions that support host well-being. Helminth infections can modify and disturb the natural structure of these microbial communities. Research shows that *lactobacilli* can support helminth survival, indicating a mutualistic relationship between certain bacteria and helminths.
- **Metabolites produced by helminths and microbiota:** Both helminths and microbes produce short chain fatty acids (SCFAs) such as acetate, propionate, and butyrate. SCFAs improve gut barrier function and can protect against bacterial infections like *E.coli*.

Helminth infection affects SCFAs levels differently: Increased during *H.polygyrus* and *Haemonchus contortus* infections. Reduced during Ascaris infection in human and *T. suis* in pigs. These metabolic changes directly impact immune balance and infection susceptibility.

- **Helminths modified microbiome and host immunity:** A helminth-altered microbiome shapes host immunity by promoting regulatory immune responses. This can reduce allergic and inflammatory diseases but may also increase vulnerability to microbial coinfections. Experiments show helminth-modified microbiota alone can suppress airway inflammation in asthma models.
- **Immunological effects:** The immune regulation from helminth infection can decrease allergic inflammation, maintain gut immune homeostasis, increase risk of secondary infections.

Emerging Tools to Explore Helminth, Microbial, and Host Interplay

The complex relationships between helminths (parasitic worms), gut microbiota, and their host have gained significant attention in current parasitological research and microbiome studies. Recent advance in multi-omics technologies including genomics, transcriptomics, metabolomics and proteomics along with systems biology now allow scientists to investigate these interactions with exceptional detail. This comprehensive strategy reveals how helminth infections can reshape microbial communities and how these microbes, in turn, affect the host's physiology, immune system and influence the course of disease.

- **Sequencing Technologies:** The advent of next-generation sequencing (NGS) has revolutionized studies of host–parasite–microbiome interactions. Key sequencing technologies include:
 - **16S rRNA Sequencing:** Targets bacterial and archaeal communities, enabling the identification and classification of microorganisms within the host gut.
 - **Internal Transcribed Spacer (ITS) Sequencing:** Used to identify and characterize fungal communities within the microbiota.
 - **Shotgun Metagenomic Sequencing:** Provides comprehensive analysis of microbial genomes, capturing both taxonomic and functional diversity.
- **Flow Cytometry:** Flow cytometry offers a high-throughput technique to study cellular interactions between the host immune system and microbial populations.
 - **Microbial Flow Cytometry:** Enables real-time analysis of bacterial heterogeneity without cultivation.
 - **Host Immune Profiling:** Quantifies immune cell populations and cytokine production, revealing immune modulation during helminth–microbiota interactions.
- **Metabolic Profiling:** Metabolomics, particularly mass spectrometry-based approaches, identifies metabolic changes resulting from helminth infections. These metabolites mediate communication between microbes and host cells, influencing immunity, energy balance, and inflammation.
- **Organoid Culture Models:** Intestinal organoids, or mini-gut cultures, mimic the intestinal epithelium's structure and function. Co-culturing organoids with microbiota and helminths enables controlled examination of host–parasite–microbe interactions. (Loke & Harris, 2023)

Current status in the field of Veterinary Parasitology: Examples from Recent Studies

- Barelli *et al.*, 2021 examined wild tropical primates (red colobus and yellow baboons) across intact and fragmented habitats. Helminth infection types (*Trichuris* spp. and *Strongyloides* spp.) were

associated with altered gut microbiota richness and diversity, demonstrating how environmental and parasitic factors jointly shape microbial communities.

- Mafuna *et al.*, 2021 studied *Haemonchus contortus* infections in Dohne Merino sheep and found distinct bacterial genera such as *Succiniclasicum*, *Rikenellaceae* RC9, and *Candidatus Saccharimonas* dominating the gut microbiome.
- Ramos *et al.*, 2025 suggested microbial composition correlates with infection intensity under natural conditions, highlighting potential biomarkers for haemonchosis control.
- Duarte *et al.*, 2016 studied cats infected with *Toxocara cati* and observed significant shifts in gut bacterial phyla. The infected microbiota showed predominance of Firmicutes, followed by *Proteobacteria*, *Actinobacteria*, and *Bacteroidetes*.

Outcome of Host-Microbiota relationship

The outcome of the helminth–microbiota relationship can be positive or negative. The host-microbiota relationship is a double edged sword that maintains immune and metabolic homeostasis but can also cause disease when imbalanced. Helminths play a unique role in this ecosystem, offering potential therapeutic benefits for autoimmune disorders through immune modulation. Nevertheless, their pathogenic potential and nutritional burden must be carefully managed in therapeutic applications.

- **Positive Outcomes of Host–Microbiota Relationship**

- **Immune System Modulation:** The microbiota significantly influences the development and function of the host immune system.

Helminths and Immune Regulation: Parasitic helminths can modulate the host’s immune responses by promoting a more regulatory and anti-inflammatory environment. This process helps in reducing chronic inflammation and preventing autoimmune reactions. Microbiota contributes by influencing how the immune system reacts to helminth infections, promoting tolerance and reducing excessive immune activity.

Symbiotic Relationship with Microbiota: In several cases, the microbiota assists in immune modulation by releasing signaling molecules that help the host tolerate helminth infections. This symbiotic relationship minimizes inflammatory damage, maintaining a balance that benefits both host and parasite. Such mutual tolerance prevents tissue injury and supports long-term immune homeostasis.

- **Gut Health and Digestion:** The gut microbiota is fundamental for digestion and nutrient absorption.

Helminths and Gut Microbiota Balance: Some helminth infections are associated with improved gut microbiota diversity and composition. They can help restore microbial balance in the intestine, thereby enhancing digestion and gastrointestinal function. The interaction between helminths and beneficial bacteria creates a more resilient gut ecosystem.

Potential for Nutrient Acquisition: Helminths can influence microbial activity to facilitate better nutrient absorption, particularly in malnourished environments. By modifying the gut environment, they may enhance the uptake of essential nutrients, contributing indirectly to the host's nutritional well-being.

- **Protection Against Other Pathogens:** A diverse and healthy microbiota acts as a natural defense mechanism against pathogenic microorganisms.

Microbiota as a Defense: Beneficial microbes compete with pathogens for resources and ecological niches in the gut. Helminths may indirectly assist in this protective mechanism by modulating the microbiota in ways that prevent harmful pathogens from colonizing intestinal surfaces.

- **Helminth Therapy or Immune Therapy:** More than 80 autoimmune diseases have been identified, including inflammatory bowel disease (IBD), multiple sclerosis (MS), rheumatoid arthritis (RA), and type 1 diabetes (T1D), which collectively affect around 10% of the global population. These disorders are characterized by an overactive immune system that mistakenly targets the body's own tissues. Currently, most autoimmune diseases have no definitive cures and are not preventable. Consequently, research into helminth-based immunotherapies has gained attention as an alternative approach to restoring immune regulation. Studies have shown promising results in animal models and early human clinical trials (Smallwood et al., 2017).
- **Mechanism of Helminth Therapy:** Helminth therapy leverages the immunomodulatory properties of parasitic worms to restore immune balance. The mechanism involves shifting the immune response from a pro-inflammatory (Th1/Th17) profile toward an anti-inflammatory or regulatory (Th2/Treg) profile. **Helminth-Derived Products:** Excretory/secretory (ES) products from helminths stimulate immune cells such as macrophages, dendritic cells, and T cells. These secretions enhance the production of cytokines like IL-4, IL-10, and TGF- β , which suppress excessive inflammation and promote tissue repair.

Cellular Pathways: Helminth infection activates M2 macrophages and regulatory T cells (Tregs), which secrete anti-inflammatory mediators. This modulated response counterbalances hyperactive Th1 and Th17 pathways that drive autoimmune pathology. As a result, immune tolerance is restored, reducing the symptoms of autoimmune diseases such as multiple sclerosis, rheumatoid arthritis, and inflammatory bowel disease.

➤ **Examples of Helminths Used in Therapy:**

Helminths Used	Autoimmune Disease Targeted	Reference
<i>Schistosoma mansoni</i> , <i>Schistosoma japonicum</i> , <i>Trichinella spiralis</i>	Inflammatory bowel disease	Smallwood et al., 2017
<i>S. mansoni</i> , <i>S. japonicum</i> , <i>Fasciola hepatica</i> , <i>T. spiralis</i>	Multiple sclerosis	Smallwood et al., 2017
<i>S. mansoni</i> , <i>T. spiralis</i> , <i>Ancylostoma caninum</i>	Type 1 diabetes	Smallwood et al., 2017
<i>S. mansoni</i> , <i>S. japonicum</i>	Rheumatoid arthritis	Smallwood et al., 2017
<i>A. vitatae</i>	Systemic lupus erythematosus	Smallwood et al., 2017

• **Negative Outcomes of Host–Microbiota Relationship:**

- While the host–microbiota interaction can offer multiple benefits, it also poses risks under unfavourable conditions.
- Immune System Dysregulation: Excessive immune tolerance or overactivation can result in either susceptibility to infections or autoimmune reactions, respectively.
- Nutrient Depletion: Certain parasitic helminths compete with the host for essential nutrients, leading to deficiencies, anemia, and growth retardation, especially in children and malnourished individuals.
- Pathogen Transmission: Some helminths act as vectors for other infectious agents or damage intestinal barriers, increasing susceptibility to secondary infections.

Conclusion

The relationship between parasitic helminths and microbiota is bidirectional and context-dependent. The microbiota influences the presentation and evolution of helminth infections and, vice versa,

helminth infections alter the microbiota diversity and composition. The presence of helminths can lead to immune modulation and gut health benefits in certain cases, but it can also cause immune dysregulation, microbiota imbalances, and increased susceptibility to other diseases. The overall outcome depends on factors such as the species of helminth, the health of the host, the composition of the microbiota, and the environmental conditions in which the host lives. Current knowledge of the consequences of worm establishment on the microbiome structure and function, as well as on the pathophysiology of helminth disease is very limited. Helminth–microbiota interactions represent a key determinant of healthy homeostasis.

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Organic Farming: Principles and Practices

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1. Introduction

Agriculture, the backbone of human civilization, has undergone rapid transformation over the last century. The Green Revolution, while significantly increasing food production, also introduced challenges such as soil degradation, reduced biodiversity, contamination from agrochemicals and loss of ecological balance. In this context, organic farming has emerged as a sustainable alternative that harmonizes agricultural productivity with ecological integrity. Organic farming is defined as a production system that sustains the health of soils, ecosystems and people by relying on ecological processes, biodiversity and cycles adapted to local conditions, rather than the use of synthetic inputs such as fertilizers, pesticides and growth regulators. It combines tradition, innovation and science to benefit the shared environment and promote fair relationships and good quality of life for all involved. (IFOAM, 2020) Globally, organic agriculture now covers more than 76 million hectares (FiBL–IFOAM 2024), with India ranking 4th in terms of the area under organic management. The organic movement in India is expanding rapidly through initiatives like the Paramparagat Krishi Vikas Yojana (PKVY), Mission Organic Value Chain Development for North Eastern Region (MOVCDNER) and various state-level policies in Sikkim, Karnataka and Madhya Pradesh. Organic farming not only provides safe and nutritious food but also plays a crucial role in soil health improvement, climate resilience and natural resource conservation- making it a central component of sustainable agriculture.

2. Concept and Evolution of Organic Farming

The concept of organic farming originated in the early 20th century as a response to industrialized agriculture. Sir Albert Howard's classic work *An Agricultural Testament* (1940) laid the foundation by emphasizing the recycling of organic wastes and maintaining soil fertility through composting and



biological activity. Other pioneers include Rudolf Steiner (biodynamic farming, 1924) and J.I. Rodale (promoter of organic movement in the USA).

Evolution in India

India's organic agriculture has deep roots in traditional farming systems where organic matter recycling, mixed cropping and integration of livestock were practiced naturally. After independence, the rise of chemical inputs marginalized these practices, but environmental concerns and consumer demand for chemical-free products revived organic farming from the 1990s onwards.

Milestones include:

- 2000 – Establishment of the *National Programme for Organic Production (NPOP)*.
- 2015 – Launch of *Paramparagat Krishi Vikas Yojana (PKVY)*.
- 2016 – *Sikkim declared the first fully organic state in the world.*
- 2020–2025 – Expansion of organic clusters across India covering >20 lakh hectares.

3. Principles of Organic Farming

Principles of Organic Farming (According to IFOAM)

Organic farming represents a holistic system of agricultural production that aims to maintain ecological balance, enhance soil fertility and ensure the sustainability of natural resources while providing safe and nutritious food. It is not merely the rejection of synthetic inputs but a comprehensive philosophy based on harmony between humans, animals and the environment. The



International Federation of Organic Agriculture Movements (IFOAM), established in 1972, has defined four guiding principles for organic agriculture- Health, Ecology, Fairness and Care. These principles serve as ethical and practical foundations for developing organic systems globally.

3.1 Principle of Health

The Principle of Health emphasizes that organic agriculture should sustain and enhance the health of soil, plants, animals, humans and the planet as one indivisible entity. Health in organic farming is viewed as a dynamic state of well-being rather than merely the absence of disease. Healthy soil forms the basis of this principle. Soils rich in organic matter and microbial life are capable of producing robust and nutritious crops. Practices such as green manuring, compost application, crop rotation, vermicomposting and



biofertilizer use are promoted to maintain soil fertility and microbial diversity. These methods reduce dependency on chemical fertilizers, which may lead to soil acidification, nutrient imbalance and loss of soil structure over time. Healthy soils lead to healthy crops and livestock, which ultimately contribute to the health of humans consuming those products. For instance, organically grown fruits and vegetables are often reported to contain higher levels of antioxidants, vitamins and essential micronutrients. Moreover, avoidance of chemical pesticides ensures minimal residues in food, promoting consumer health and environmental safety.

Example: In India, states like Sikkim, which became the world's first fully organic state in 2016, have demonstrated improved soil organic carbon levels and biodiversity through sustained organic practices, reflecting the successful implementation of this principle.

3.2. Principle of Ecology

The Principle of Ecology asserts that organic agriculture should be based on living ecological systems and natural cycles, working with them, emulating them and helping sustain them. Agriculture is seen as part of the larger ecological system, where the farm operates as a living organism with interdependent components-soil, plants, animals, microorganisms and humans. This principle encourages recycling of organic matter, use of renewable resources, maintenance of biodiversity and protection of natural



habitats. Instead of relying on external chemical inputs, organic farming seeks to utilize natural processes for nutrient cycling and pest regulation. For instance, leguminous crops are integrated into crop rotations to fix atmospheric nitrogen, while natural enemies like ladybird beetles and parasitoid wasps help regulate pest populations. The use of cover crops, agroforestry systems and conservation tillage are also aligned with this principle, as they prevent soil erosion, conserve water and enhance biodiversity.

Example: In southern India, Karnataka's organic farmers under the *Savayava Krishi Mission* follow ecological practices like rainwater harvesting, mulching and mixed cropping systems that mimic natural ecosystems, thereby conserving soil moisture and improving productivity under dryland conditions.

3.3. Principle of Fairness

The Principle of Fairness highlights that organic agriculture should build relationships that ensure equity, respect and justice for all involved—from farmers and workers to traders and consumers. Fairness extends beyond human relationships to include respect for animals, plants and the environment. In practice, this principle promotes fair trade, ethical labor practices and equitable distribution of benefits. It also discourages the exploitation of natural resources and the use of technologies that may harm communities or ecosystems. For farmers, fairness means obtaining a fair price for their produce that reflects the true cost of sustainable production. For consumers, it guarantees access to healthy, safe and honestly labeled organic food. For animals, fairness means providing living conditions that allow them to express their natural behavior, with sufficient space, proper feed and humane treatment. Example: The Fairtrade and Organic Cotton Project in Gujarat and organic spice cooperatives in Kerala ensure that smallholder farmers receive better income and recognition for their eco-friendly production methods. These initiatives embody the spirit of fairness and sustainability by linking rural livelihoods with ethical markets.



3.4. Principle of Care The Principle of Care emphasizes that organic agriculture should be managed in a precautionary and responsible manner to protect the health and well-being of current and future generations as well as the environment. This principle is rooted in the precautionary approach, advocating that agricultural innovations and technologies must be carefully assessed for their long-term impacts before adoption. Organic farming rejects practices and technologies that pose potential harm to human health or the environment, such as genetically modified organisms (GMOs), irradiation and synthetic growth promoters. Instead, it encourages research and development of safe, ecologically sound technologies like microbial bioinoculants, natural pest repellents and renewable energy-based farm operations. The principle also emphasizes the ethical responsibility of scientists, policymakers and farmers to make decisions that do not compromise the ability of future generations to meet their needs. Care in organic farming therefore extends beyond immediate productivity to long-term ecological resilience and sustainability.



Example: In Andhra Pradesh Community Managed Natural Farming (APCNF), the adoption of low-cost, non-chemical inputs like *Jeevamrutha*, *Beejamrutha* and *mulching* illustrates precautionary and caring practices that safeguard soil biota, reduce input costs and improve farm sustainability.

Integration of the Four Principles

These four principles-Health, Ecology, Fairness and Care-are interrelated and reinforce one another. Healthy soils and ecosystems (Health and Ecology) are achieved through fair relationships and responsible stewardship (Fairness and Care). Together, they form the ethical and scientific basis for a sustainable agricultural paradigm that nurtures both people and the planet.

In practice, implementing these principles requires a systems-based approach involving community participation, policy support, market linkages and farmer education. India's National Programme for Organic Production (NPOP) and Paramparagat Krishi Vikas Yojana (PKVY) promote these principles through certification, capacity building and incentives for farmers adopting organic methods.

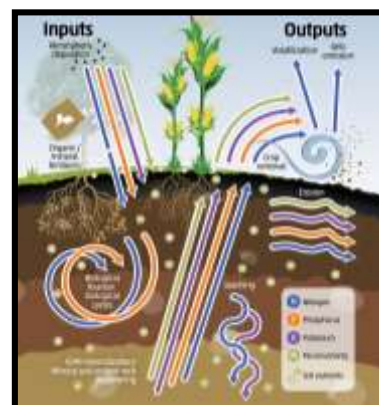
4. Core Practices of Organic Farming

4.1. Soil Fertility Management

Soil fertility in organic systems is maintained and enhanced through organic matter recycling and biological processes rather than synthetic fertilizers.

Key practices:

- Farmyard manure (FYM): Rich in nutrients and beneficial microbes.
- Composting: Converts organic wastes into nutrient-rich humus.
- Green manuring: Incorporation of legumes (*Sesbania*, *Crotalaria*) to add nitrogen.
- Vermicomposting: Earthworms convert organic residues into vermicast.
- Biofertilizers: *Rhizobium*, *Azospirillum*, *Azotobacter*, PSB and Mycorrhizae inoculants enhance nutrient cycling. These practices improve soil structure, water-holding capacity, C:N ratio and biological activity.



4.2. Crop Rotation and Diversification

Crop rotation is the sequential cultivation of different crops to balance nutrient demands and break pest and disease cycles.

Example Rotations:

- Cereals → Legumes → Oilseeds
- Rice–Green gram–Sesame
- Maize–Cowpea–Vegetable

Diversification with intercropping and multi-tier systems (e.g., coconut + banana + pineapple) enhances resource use efficiency and biodiversity.

**4.3. Pest and Disease Management**

Organic pest management emphasizes prevention and ecological balance over chemical eradication.

Methods:

- Cultural: Crop rotation, resistant varieties, sanitation trap crops.
 - Mechanical: Hand-picking, traps, light traps, pheromone traps.
 - Biological control: Predators (ladybird beetles), parasitoids (Trichogramma) and microbial bio-pesticides (Bacillus thuringiensis, Beauveria bassiana).
 - Botanical pesticides: Neem oil, pongamia extract, garlic–chili spray.
- Integrated approaches such as Integrated Pest and Disease Management (IPDM) are widely used.

**4.4. Weed Management**

Weeds are managed through non-chemical means:

- Mulching with crop residues or organic waste.
- Crop rotation and cover cropping.
- Manual weeding and mechanical weeder.
- Use of smother crops and stale seedbed technique.

**4.5. Water and Irrigation Management**

Efficient irrigation practices are vital in organic systems. Drip irrigation, mulching and rainwater harvesting minimize water wastage. Organic matter improves soil moisture retention, reducing irrigation frequency.

**4.6. Livestock Integration**

Livestock forms an integral part of organic farms, providing manure, draught power and diversified income. Animal welfare and balanced feeding (no growth hormones or antibiotics) are essential in certified organic systems.

4.7. Post-Harvest and Processing

Organic produce must be handled separately from conventional products to prevent contamination. Certification standards mandate traceability and approved organic processing inputs.

Storage: Hermetic storage, solar drying and botanical protectants prevent losses.

4.8. Certification and Standards

In India, organic certification is governed by:

- National Programme for Organic Production (NPOP) under APEDA.
- Participatory Guarantee System (PGS-India) under the Ministry of Agriculture for smallholder groups.
- Third-party certification for export markets.

Labels: India Organic, Jaivik Bharat, PGS-Green and PGS-Organic.

Certification ensures consumer confidence and market access, although high cost and documentation remain barriers for smallholders.

Steps for Obtaining Organic Certification (OCF Certificate)

5. Benefits of Organic Farming

5.1. Environmental Benefits

- Restores soil organic carbon and microbial diversity.
- Reduces nitrate leaching and water pollution.
- Enhances biodiversity and pollinator abundance.
- Sequesters carbon and mitigates climate change.

5.2. Economic Benefits

- Premium prices for organic produce (10–40% higher).
- Lower input costs over time.
- Diversified farm income through mixed farming.

5.3. Social and Health Benefits

- Reduced exposure to pesticides for farmers and consumers.
- Improved nutrition due to higher antioxidant content in organic crops.



- Empowerment of smallholders and women through community certification.

6. Limitations and Challenges

Despite its advantages, organic farming faces constraints such as:

- Yield gap: 10–25% lower yields during transition period.
- Nutrient deficiency: Especially nitrogen, in intensive systems.
- Pest and weed pressure: Require skill and labour.
- Market access: Limited local markets and certification costs.
- Knowledge gap: Need for scientific validation and extension services.

However, long-term studies show that after a few years of transition, yields stabilize and profitability improves due to higher prices and reduced input costs.

7. Organic Farming in India – Case Studies

7.1. Sikkim – The Organic State

Sikkim became the first fully organic state in 2016, converting 75,000 ha to organic management. The government banned chemical inputs and promoted bio-inputs, composting and certification. Outcomes include higher soil organic matter, better ecological balance and tourism benefits.

7.2. Karnataka – Organic Villages

Under the *Savayava Bhagya Yojane* scheme, organic clusters were developed, especially in Belagavi and Dharwad districts. Farmers adopted crop diversification, biopesticides and composting. Karnataka also established organic markets under *Raitha Santhe*.

7.3. Madhya Pradesh - Organic Cotton and Soybean

MP has India's largest certified organic area, focusing on cotton, soybean and pulses under contract farming with export companies. It exemplifies organic value chain development.

8. Innovations and Future Trends

- Bio-input technology: Use of microbial consortia and bioenzymes for nutrient release.
- Digital organic farming: GIS, IoT and blockchain for traceability.
- Climate-smart organic systems: Combining organic farming with agroforestry and conservation agriculture.
- Urban organic farming: Rooftop and hydroponic organic systems.
- Market innovations: E-commerce platforms (Jaivik Kheti portal) and organic cooperatives.

9. Conclusion

Organic farming embodies a holistic and ecologically balanced approach that integrates the health of soil, plants, animals, humans, and the entire ecosystem. It goes beyond the mere avoidance of synthetic chemicals, focusing instead on rebuilding natural cycles, enhancing biodiversity, and restoring soil vitality. By harmonizing traditional wisdom with modern scientific innovations, organic agriculture promotes long-term soil fertility, water conservation, and ecological balance.

The future of sustainable agriculture lies in strengthening organic farming systems through supportive policies, farmer education, and market incentives that encourage environmentally responsible practices. Integration of digital tools such as precision agriculture, remote sensing, and AI-based monitoring can further optimize organic management and traceability. Moreover, fostering fair trade practices ensures economic justice for smallholders, empowering rural communities and promoting inclusive growth.

In essence, by embracing the four guiding principles of **Health, Ecology, Fairness, and Care**, organic farming offers a transformative pathway toward climate-resilient and socially just food systems. It ensures not only safe and nutritious food for present generations but also safeguards the natural resource base for future ones- making it a cornerstone of global sustainability and food security in the 21st century.

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COMMUNICATION THROUGH SOCIAL MEDIA IN AGRICULTURE AND ALLIED SECTORS

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Effective communication is a central pillar of successful agricultural extension programs, especially as the field navigates a period of rapid technological advancement and global challenges such as food security, climate change, and sustainable farming. Modern communication techniques are instrumental in bridging the gap between farmers, researchers, policymakers, and other stakeholders, fostering collaboration and knowledge transfer for the adoption of best practices in agriculture.

Transforming Agricultural Extension through Advanced Communication

With the advancement of Information and Communication Technologies (ICTs), agricultural extension has witnessed a paradigm shift in communication practices. The proliferation of computers, mobile phones, and internet connectivity has opened up new avenues for knowledge sharing, interactive learning, and collaboration. Mobile applications, online platforms, and digital resources have revolutionized the way agricultural information is accessed, shared, and utilized.

In agricultural extension, multimedia and audio-visual techniques have also become effective communication tools. Extension agents can improve the impact and clarity of their messaging by utilising presentations, films, and visual aids to make difficult agricultural subjects more approachable and interesting. These resources offer an engaging and participatory experience that promotes learning and aids in memory retention. Furthermore, social media sites and internet forums have revolutionised agricultural extension by facilitating discussions and spreading information. Social media platforms have developed into useful tools for increasing the reach and impact of agricultural extension initiatives, from networking with similar people to sharing success stories and best practices to crowdsourcing solutions.

Social media has become a key tool for communication in agriculture, allowing farmers and businesses to connect with each other and share information.

- **Information sharing:** Farmers can use social media to access information about agriculture, such as from extension experts, officials, and other farmers. Farmers can use social media to access information about agricultural extension, technological developments, irrigation methods, and soil testing.
- **Marketing:** Businesses can use social media to promote their products to customers.
- **Problem solving:** Businesses can connect with scientists on social media to get real-time solutions to problems.
- **Building relationships:** Social media can help farmers build relationships with people from different places.
- **Community building:** Social media can help farmers develop a community of like-minded people.
- **Transparency:** Farmers can use social media to be transparent with consumers.

Social media can also help the farmers to connect with the Agri- dealers, weather advisories etc. and new agricultural methodologies can be exchanged by the farmers. Agricultural Information on social media is scattered and complex to understand for the farmers. Further, it develops into different types of advanced communication media used in the agricultural field, such as mobile applications, online platforms, social media, video, and multimedia resources, webinars, and interactive decision support tools

Social Media as a Catalyst

Social media is a transformative force in agricultural communication, providing platforms for farmers, agribusinesses, and extension agents to interact directly, share experiences, and solve problems in real time. Its core benefits include:

- Social media can be used to communicate with farmers in a number of ways, including
- Creating and sharing content: Farmers can create, share, and exchange information on social media.
- Selling products: Farmers can sell their products directly to consumers or retailers using social media.
- Engaging with extension agents: Farmers can use social media to engage with extension agents and specialists in real time to get solutions to their challenges.
- Participating in virtual communities: Farmers can participate in virtual communities and networks on social
- Farmers can use social media to engage with extension agents and specialists in real time to get solutions to their challenges.

- Participating in virtual communities: Farmers can participate in virtual communities and networks on social media.
- Marketing: Social media can be used as a tool for agricultural marketing.
- Reducing social isolation: Social media can help reduce farmers' social isolation.
- Networking: Social media can increase networking platforms for farmers. Some popular social media platforms for agricultural extension include: Facebook, Twitter, YouTube, WhatsApp, and Instagram

Advanced Communication Media in Agriculture

1. Mobile Applications: Mobile applications provide farmers and extension professionals with instant access to agricultural information, weather updates, market prices, pest and disease management tools, and best agricultural practices. Examples include apps for crop identification, nutrient management, and market analysis.

2. Online Platforms: Online platforms and portals serve as centralized hubs for agricultural information and resources. They offer access to research papers, case studies, training modules, and interactive forums where farmers and extension professionals can connect, share experiences, and seek advice. Examples include e-learning platforms, knowledge-sharing portals, and agricultural forums.

e--Agriculture: An online platform that offers a wide range of resources, including articles, case studies, and discussion forums, facilitating knowledge sharing and networking among agricultural stakeholders.

Farm Logs: A web-based platform that combines farm management tools, satellite imagery, and weather data to help farmers make informed decisions about crop planning, irrigation, and yield forecasting

3. Social Media Platforms: Social media platforms like Facebook, Twitter, and Instagram serve as effective channels for agricultural extension. Extension professionals can create dedicated pages or groups to share updates, disseminate information, answer queries, and engage with farmers and stakeholders. Social media platforms also facilitate networking and knowledge exchange among farmers themselves.

□ **Facebook Groups:** Farmers can join agriculture-related Facebook groups to exchange information, seek advice, and engage in discussions with fellow farmers and experts.

□ **Twitter:** Hashtags like AgChat and AgTech provide platforms for farmers to share experiences, learn from each other, and stay updated on industry trends and innovations.

4. Video and Multimedia Resources

□ Multimedia resources, such as videos, infographics, and animations, are powerful tools for effective communication in agricultural extension. They can simplify complex concepts, demonstrate practical techniques, and engage farmers visually. These resources can be shared through various channels, including websites, social media, and mobile applications.

□ **YouTube:** A vast repository of agricultural tutorial videos, documentaries, and expert interviews, covering topics ranging from livestock management to precision agriculture.

□ **TED Talks:** The TED platform hosts talk by agricultural thought leaders and innovators, sharing ideas and insights on sustainable farming practices, food security, and rural development.

5. Webinars and Online Courses: Webinars and online training sessions enable extension professionals to conduct virtual workshops, seminars, and capacity-building programs. These platforms allow participants to interact, ask questions, and access training materials remotely. Webinar platforms like Zoom, Microsoft Teams, and Google Meet are commonly used for this purpose.

□ **Extension:** Offers online webinars and courses on various agricultural topics, allowing farmers to learn from experts and access specialized knowledge from the comfort of their homes.

□ **Coursera:** Provides online courses from renowned universities and institutions, covering subjects like agribusiness, organic farming, and agricultural economics.

□ **E-Agriculture:** An online platform that offers a wide range of resources, including articles, case studies, and discussion forums, facilitating knowledge sharing and networking among agricultural stakeholders.

6. Interactive Decision Support Tools

Climate Field View: Offers digital tools that collect and analyse field data, providing farmers with insights for precise planting, fertilization.

Crop Nutrient Recommendation Tools: Online tools like Nutrient Expert and Nutrient Management Advisor assist farmers in optimizing fertilizer applications based on soil analysis and crop requirements.

7. Remote Sensing and Satellite Imagery: NASA's Earth Observing System Data and Information System (EOSDIS): Provides satellite imagery and remote sensing data, allowing farmers to monitor crop health, detect anomalies, and assess vegetation indices.

8. Interactive Voice Response (IVR) Systems: IVR systems use automated phone calls to deliver pre-recorded messages to farmers. These systems can provide information on weather alerts, market prices, government schemes, and agricultural practices. IVR systems are particularly useful in areas with limited internet connectivity or low smart phone penetration.

9. Radio and Podcasts: Radio programs and podcasts remain popular communication media in rural areas. Agricultural extension programs can be broadcasted on local radio stations, covering topics such as crop cultivation, livestock management, pest control, and market information. Podcasts, on the other hand, offer the flexibility for farmers to access and listen to pre-recorded agricultural content at their convenience.

10. SMS and Voice Messages: SMS and voice messages are widely used to disseminate timely and targeted information to farmers. Extension professionals can send alerts, reminders, and advisory messages directly to farmers' mobile phones, keeping them informed about important updates, seasonal practices, disease outbreaks, and market trends.

11. Augmented Reality (AR) and Virtual Reality (VR): AR and VR technologies provide immersive experiences that can enhance agricultural learning and extension. These technologies can simulate farm environments, showcase interactive virtual tours, and offer hands-on training in a virtual setting. They are particularly useful for demonstrating complex machinery operations or advanced farming techniques.

The modernization of communication strategies in agricultural extension driven by digital technologies, multimedia tools, and interactive platforms is transforming the sector. Advanced communication media not only enhance the accessibility and clarity of agricultural information, but also support the development of communities and responsive networks. Agricultural extension programs can foster innovation, accelerate adoption of best practices, and better address the challenges of sustainability, climate resilience, and food security in the twenty-first century.

Ultrasonographic Evaluation of Forestomach and Abomasal Disorders in Bovines: A Diagnostic Insight

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Ultrasonography has emerged as an essential, non-invasive diagnostic imaging modality in bovine medicine for the evaluation of abdominal organs and detection of gastrointestinal disorders. It utilizes high-frequency sound waves to produce real-time images of internal structures, allowing assessment of both physiological and pathological changes within the bovine abdomen. This review focuses on the ultrasonographic anatomy of the forestomach and abomasum, as well as its diagnostic applications in common disorders like traumatic reticuloperitonitis, Left and right abomasal displacements, abomasal ulcers. The examination was performed using a real-time B-mode ultrasound scanner equipped with 3.5–5 MHz convex and 5–7.5 MHz linear transducers. Scanning was conducted from the left and right lateral abdominal walls and the ventral abdomen, following both longitudinal and transverse orientations. Normal ultrasonographic findings revealed the typical wall thickness, echogenicity, and motility patterns of the rumen, reticulum, omasum, and abomasum, which are crucial for accurate interpretation of abnormal conditions. Overall, ultrasonography offers significant diagnostic value due to its ability to provide detailed, real-time visualization of abdominal structures, enabling early detection and differentiation of gastric diseases in cattle. Its accuracy, safety, and portability make it an indispensable tool for clinicians and field veterinarians involved in large animal practice.

Keywords: Bovine ultrasonography, Forestomach, Abomasum, Traumatic reticuloperitonitis, Abomasal displacement, Diagnostic imaging

Introduction

Diagnostic imaging has become an essential component of modern veterinary practice. Among the available modalities, ultrasound, also known as ultrasonography (USG), is a diagnostic imaging

technique that utilizes high-frequency sound waves (typically 2–15 MHz) to produce images of internal body structures such as soft tissues, organs, and vessels. It is a non-invasive, real-time, and radiation-free imaging modality widely used in both human and veterinary medicine (Fossum, 2019; Mattoon & Nyland, 2021).

In bovines, abdominal ultrasonography is increasingly used to evaluate conditions affecting the gastrointestinal tract, liver, spleen, urinary bladder, and reproductive organs (Braun, 2009; Kumar, 2024). Conventional diagnostic methods such as clinical examination, percussion, and auscultation are often limited in sensitivity, especially in detecting internal lesions or fluid accumulation. Ultrasonography bridges this gap by providing detailed insight into organ structure and function.

Principle of Ultrasonography

Ultrasonography is based on the piezoelectric effect, where electrical energy is converted into high-frequency sound waves by piezoelectric crystals within the transducer. These sound waves are transmitted into tissues and reflected back from interfaces of differing acoustic impedance. The returning echoes are processed to form a two-dimensional grayscale image (Lumb and Jones, 2020). Different tissues produce varying echogenic patterns: fluids appear anechoic (black), soft tissues hypoechoic to isoechoic (gray), and mineral or gas interfaces hyperechoic (bright white) with distal acoustic shadowing (Thrall, 2018).

Instrument

Equipment:

A real-time B-mode ultrasound scanner equipped with 3.5–5 MHz curvilinear transducers (probes) is most suitable for deep abdominal imaging in adult cattle. For superficial structures such as the umbilicus or urinary bladder in calves, 5–7.5 MHz linear probes are preferred (Braun, 2009 and Kumar, 2024).

Probe: Ultrasound is produced by piezoelectric crystals within the probe, which vibrate when stimulated by mechanical or electrical impulses, emitting ultrasound waves. These waves reflect off tissues and return as echoes, causing the crystals to vibrate again and generate electrical signals. The computer then amplifies and converts these signals into an image. Most probes are real-time sector scanners, producing a triangular or sector-shaped image (usually around 90°), while linear probes create a rectangular image. Older ultrasound scanners used mechanical systems with moving crystals

that rotated or oscillated to generate sound waves. Modern probes, called arrays, contain multiple stationary crystals activated electronically.

These arrays can be linear, curvilinear, or annular (fig. 1). Curvilinear probes provide a wider field of view than linear ones, while phased arrays electronically steer the beam, allowing wide imaging through narrow spaces, ideal for cardiac or intrathoracic studies. Although sector scanners have a limited near-field view, they are useful for imaging deep or restricted structures. In bovine practice, linear or curvilinear probes with frequencies of 3.5–8 MHz are most common, with multifrequency transducers extending up to 8.5 MHz (Blond and Buczinski, 2009).

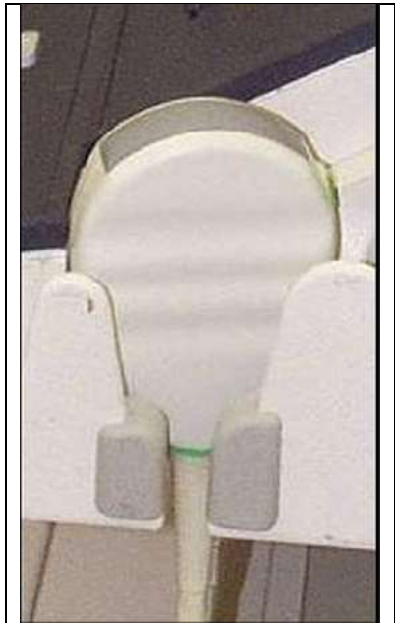


Fig. 1: Curvilinear probe

Scanning

Technique:

A systematic approach is followed:

- **Left side:** Rumen and spleen (fig. 2)
- **Right side:** Liver, gall bladder, omasum, abomasum, small intestine (fig. 3)
- **Ventral abdomen:** Urinary bladder, uterus, umbilical remnants (in calves) (Munday, 2016)

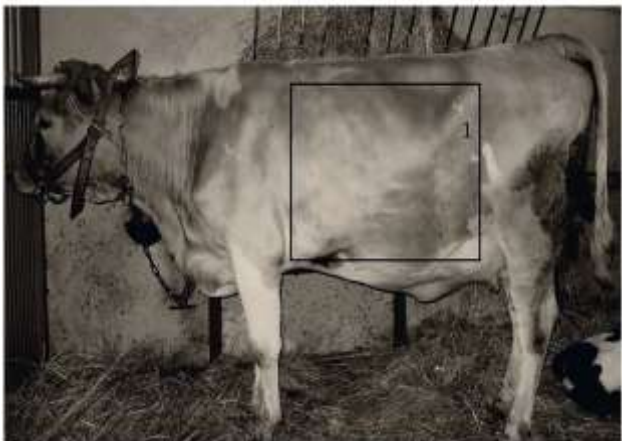


Fig. 2: Left abdomen:1-Rumen

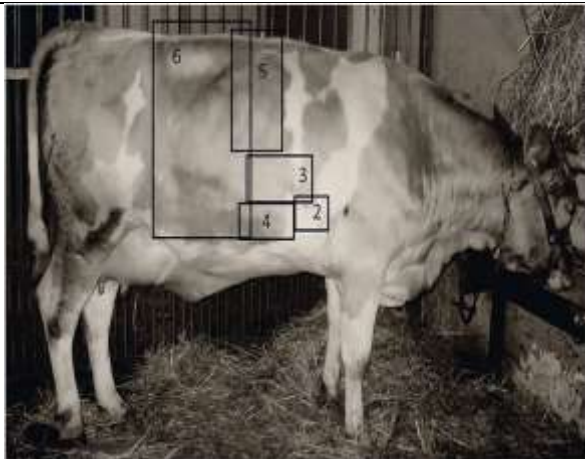


Fig. 3: Right abdomen: 2-reticulum, 3-omasum, 4-abomasum, 5-duodenum, 6-jejunum, and 7-colon

Both longitudinal and transverse planes are evaluated for each organ. The examination is complemented by transrectal ultrasonography when deeper pelvic or abdominal structures are of interest.

Normal Ultrasonographic Findings of Bovine Abdomen

Comprehensive knowledge of the **normal ultrasonographic appearance** of bovine abdominal organs is essential for accurate interpretation of pathological findings.

Rumen

On ultrasonography, the rumen wall appears as a thick hypoechoic layer (3–5 mm) with an echogenic interface at the mucosal surface (fig. 4), adjacent to the left abdominal wall from left flank to the 8th Intercoastal space (ICS) (Imran *et al.*, 2011). The gas cap produces a bright, hyperechoic line with distal acoustic shadowing, while the fibre mat appears as mixed echogenicity, and the fluid zone is anechoic with fine echogenic particles (Braun *et al.*, 2013). Rumen contractions (1–3 per 2 minutes) can be observed as rhythmic inward and outward movements of the rumen wall (Braun, 2009) (fig. 5).

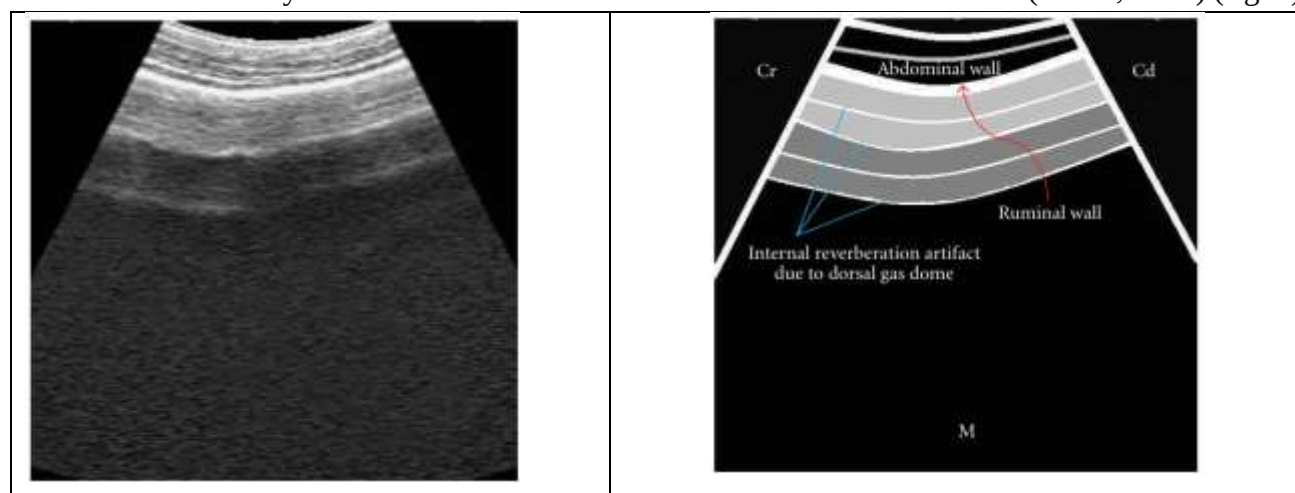


Fig. 4: Ultrasonogram of the rumen obtained from dorsal left paralumbar fossa, by placing the transducer parallel to the longitudinal axis of the cow. Cr: cranial, Cd: caudal, and M: medial.

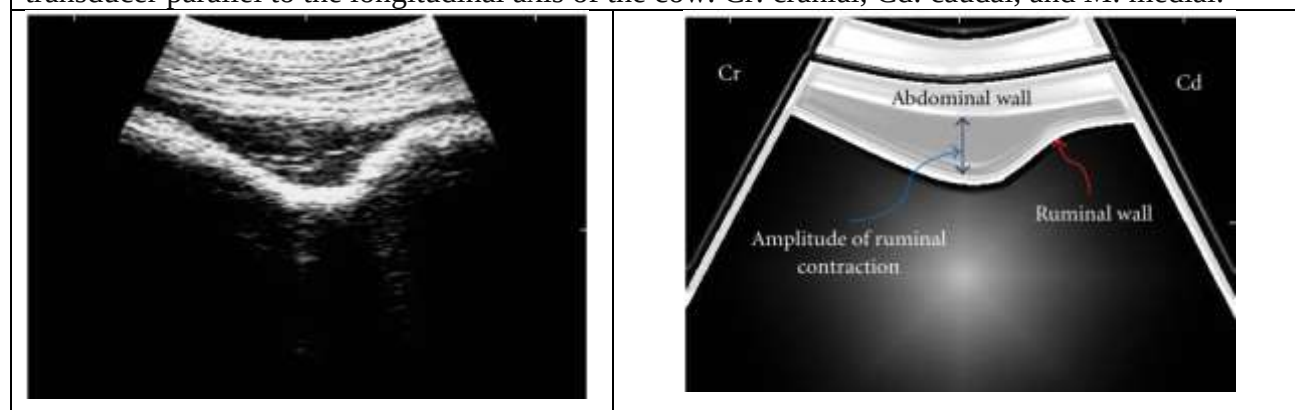


Fig. 5: Ultrasonogram of the rumen obtained from left mid-paralumbar fossa at the peak of its contraction, by placing the transducer parallel to the longitudinal axis of the cow.

Reticulum

In the ultrasonographic assessment of the reticulum, the transducer is positioned on the ventral aspect of the thorax, both to the left and right of the sternum, as well as on the left and right lateral thorax up to the level of the elbow. The examination of the reticulum commences from the left side, followed by the right (fig. 6). The normal reticulum is characterized by a half-moon shape with a smooth contour (Braun, 2009). It contracts at regular intervals and, when relaxed, is located immediately adjacent to the diaphragm and the ventral portion of the abdominal wall. Typically, the distinct layers of the reticular wall are not visible, and the honeycomb-like structure of the mucosa is seldom clearly observed. The contents of the reticulum are generally not imaged due to their partially gaseous composition. Foreign bodies and magnets are also usually not visible within the reticulum because of its gas content (Braun, 2003). The characteristic reticular contractions occur at regular intervals approximately once per minute in a biphasic manner and can be recorded ultrasonographically (Braun, 2009). During contraction, the reticular wall moves cranially and dorsally and then returns to its resting position.

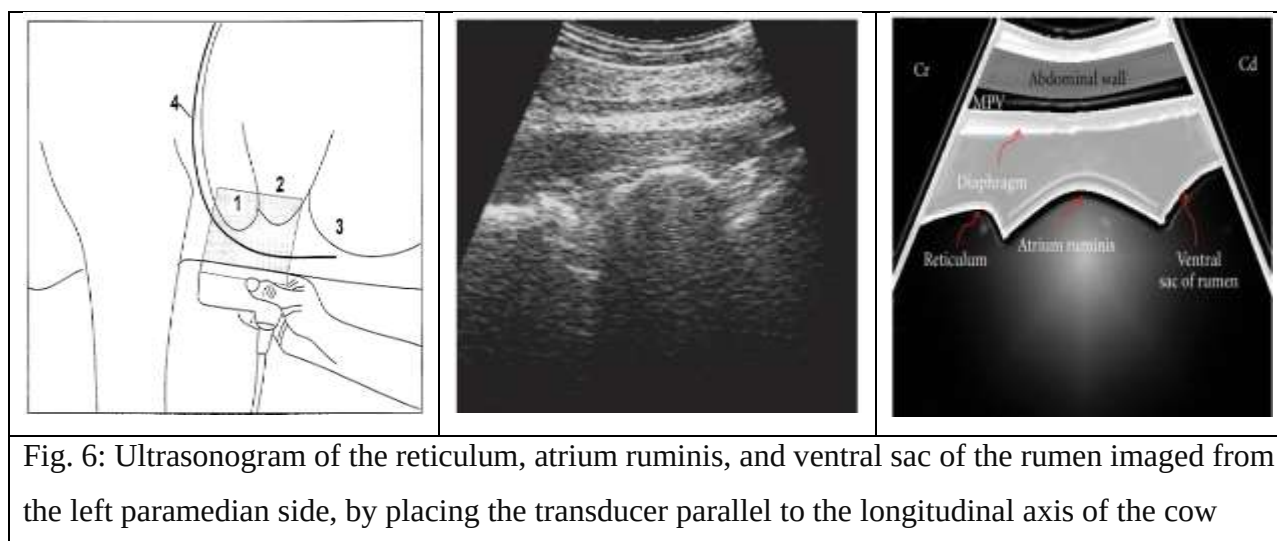


Fig. 6: Ultrasonogram of the reticulum, atrium ruminis, and ventral sac of the rumen imaged from the left paramedian side, by placing the transducer parallel to the longitudinal axis of the cow

Omasum

The omasum appeared as rounded, hypoechoic structure or curved line with a thickness of 3.8 ± 0.9 mm, located medial to the liver on the right side between the 7th and 11th intercostal spaces in all calves (fig. 7). Only the omasal wall closest to the transducer was visible on ultrasonography (Braun, 2009). The omasal leaves, contents, and the far wall could not be visualized. Occasionally the omasal laminae can be seen in ill cattle (fig. 8). The ultrasonographic appearance remained unchanged before,

during, and after feeding hay and grass silage (Braun *et al.*, 2012). The omasum lacks discernible active motility of its own and its movements are believed to be largely passive, resulting from the coordinated activity of the reticulum and rumen (Dirksen, 2005).

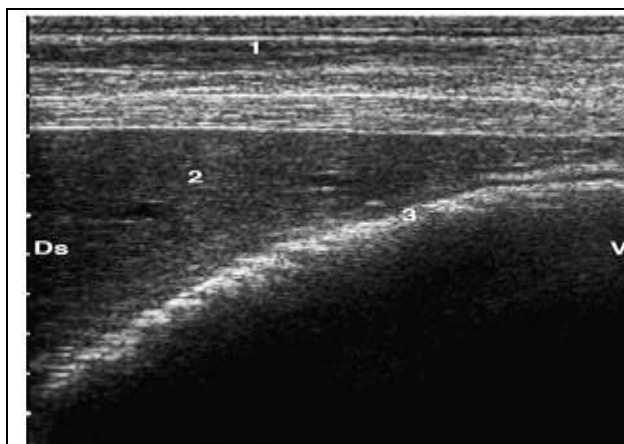


Fig. 7: Ultrasonogram of the normal omasum imaged from the 8th ICS of the right side. Costal part of the abdominal wall (1), liver (2), omasal wall (3). Ds, dorsal; Vt, ventral

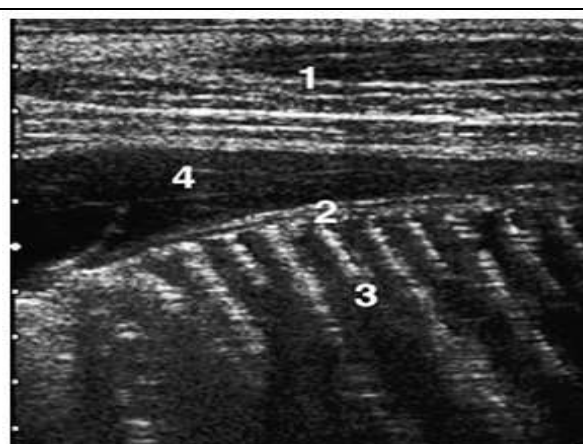


Fig. 8: Ultrasonogram of the normal omasum imaged from 10th ICS. The omasal laminae appears as thin echogenic lines. The abdominal wall (1), omasal wall (2), omasal laminae (3), greater omentum (4).

Abomasum

The abomasum can typically be visualized approximately 10 cm caudal to the xiphoid process through the left and right paramedian regions, as well as from the ventral midline. The abomasal wall appeared as an echogenic line with a thickness of 2.5 ± 0.6 (Braun *et al.*, 2012). Most of the abomasum occupies the right ventral abdomen, typically located immediately caudal to the reticulum and positioned between the craniodorsal blind sac of the rumen or the rumen itself and the ventral abdominal wall. Sonographically, the abomasal wall is identified as a distinct, thin echogenic line. The abomasum is easily distinguishable from surrounding organs due to the unique echotexture of its luminal contents, which



Fig. 9: Normal abomasum, depicting a vermiform hypoechoic rugal fold

manifest as a moderately echogenic, heterogeneous mass with fine, stippled echoes (fig. 9). Occasionally, one can observe echogenic folds of the abomasum within this material. The gentle, passive movement of the abomasal contents is frequently noted and is regarded as a normal physiological occurrence (Braun, 2009).

Abnormal Ultrasonographic Findings of Bovine Abdomen

Traumatic reticuloperitonitis

Ultrasonography is a key diagnostic tool for identifying morphological changes in cattle with traumatic reticuloperitonitis (TRP). The findings can vary depending on the severity of the inflammation and can be categorized as follows as per suggested by Barun 2003:

Morphological Changes

- a) **Fibrinous Deposits:** Deposits of fibrinous tissue are frequently seen on the reticular serosa, often interspersed with fluid pockets. Ultrasonographically, these appear as echogenic (bright) areas that are cavitated by hypoechogenic (dark) fluid pockets (fig. 10). Deposits consisting only of fibrinous tissue appear as homogeneously echogenic areas.
- b) **Abscesses:** Reticular abscesses typically have an echogenic capsule of varying thickness surrounding a centre that can be hypoechogenic to moderately echogenic. The contents may be partitioned by echogenic septa (fig. 11). These abscesses, which can range from a few centimetres to over 15 cm in diameter, are usually located caudoventral to the reticulum but can also be found in other adjacent areas.
- c) **Peritoneal Effusion:** This is seen as an accumulation of anechoic or hypoechogenic fluid in the reticular area without an echogenic margin. Bands of fibrin may be visible within the fluid, which can sometimes extend to the caudal abdomen (fig. 12).

Splenic Involvement: The spleen is often affected, showing fibrinous changes as echogenic deposits, sometimes surrounded by fluid. The spleen may be entirely covered with these deposits, and splenic abscesses or dilated vasculature (indicating splenitis) can also be seen.

Reticular Motility Changes: Reticular activity is almost always affected in cattle with TRP. The contraction frequency can be reduced from the normal three biphasic contractions per 3 minutes to two, one, or even zero. The amplitude of contractions may be reduced. In cases with extensive adhesions, reticular contractions can appear indistinct, with the reticulum contracting by only 1–3 cm.

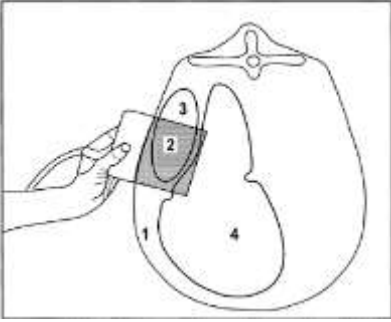
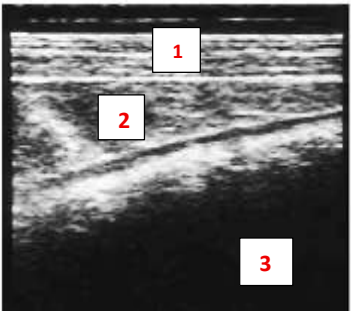
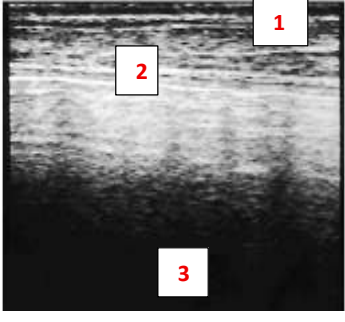
		
Fig. 10: Ultrasonogram TRP viewed from the left ventral thorax. There are echogenic deposits cavitated by hypo echogenic fluid ventral to the reticulum and craniodorsal blind sac of the rumen.	Fig. 11: Ultrasonogram of an abscess caudoventral to the reticulum and ventral to the craniodorsal blind sac of the rumen of a cow with TRP viewed from the left ventral thorax.	Fig. 12: Ultrasonogram showing echogenic deposits on the reticulum and craniodorsal blind sac of the rumen and accumulation of fluid in a cow with TRP viewed from the left ventral thorax.

Ultrasonography is not effective for visualizing metallic foreign bodies or magnets within the reticulum due to the gas content. Radiography is the preferred method for this purpose. While ultrasonography excels at detecting fibrinous deposits and abscesses, which radiography cannot, neither technique alone provides a complete diagnosis. The two methods are complementary.

Left displacement of the abomasum

Normally, the rumen is positioned directly against the left abdominal wall. However, in cases of left displacement of the abomasum (LDA), last three intercostal spaces on the left side are examined ventrally to dorsally with the transducer held parallel to the ribs (fig. 13). The abomasum is found positioned between the left abdominal wall and the rumen, which has been pushed medially. As the ultrasound transducer is moved dorsally, the rumen, initially visible ventrally, is no longer imaged and is replaced by the abomasum (Barun *et al.*, 1997). Further dorsally, the abomasum disappears and the rumen reappears. The contents are not uniform. The ventral portion contains fluid ingesta, while the dorsal portion has a gas cap (fig. 14 and 15). The fluid ingesta in the ventral part of the abomasum

appears hypoechogenic (dark). The rumen wall can often be seen medial to this fluid. Occasionally, the abomasal folds are visible within the ingesta as elongated, echogenic, sickle-shaped structures. The gas cap in the dorsal part is identified by reverberation artifacts. These artifacts appear as parallel lines of varying echogenicity that run parallel to the abomasal surface and prevent visualization of the rumen wall medial to the gas cap (Barun 2003).

		
Fig. 13: Ultrasonographic examination of LDA from the 12th intercostal space. (1) Abdominal wall, (2) ventral part of abomasum containing ingesta, (3) dorsal portion of abomasum with gas cap, (4) rumen.	Fig: 14: Ultrasonogram of LDA from the ventral region of the 12th intercostal space. (1) Abdominal wall, (2) abomasum with hypoechogenic ingesta, (3) rumen displaced medially.	Fig. 15: Ultrasonogram of LDA from the dorsal region of the 12th intercostal space. (1) Abdominal wall, (2) abomasal wall, (3) reverberation artifacts.

Right displacement of the abomasum

Ultrasonography is a useful diagnostic tool for confirming right displacement of the abomasum (RDA), particularly in doubtful cases. The examination involves scanning the area immediately caudal to the last rib and the last two to three intercostal spaces on the right side, moving from ventral to dorsal. In a healthy cow, the ventral abdomen on the right side shows loops of the small intestine, and further dorsally, the liver is seen immediately adjacent to the right abdominal wall (Barun 2003). In animals with right displacement of the abomasum, the liver is displaced medially away from the abdominal wall and cannot be imaged in its normal position. Instead, the abomasum is found immediately adjacent to the right abdominal wall where the liver would normally be. However, it is important to note that ultrasonography cannot reliably differentiate between RDA and Abomasal Volvulus (AV) (Barun *et al.*, 2022).

Defects in Abomasal Emptying

Functional or mechanical narrowing of the pylorus can lead to impaired abomasal emptying. Furthermore, abomasal motility may be reduced secondarily due to intestinal ileus. In such situations, the abomasum becomes enlarged without showing dorsal displacement or torsion. The degree of abomasal distension influences its ultrasonographic detectability; a markedly dilated abomasum can typically be visualized on the right side within the ventral regions of the 8th to 12th intercostal spaces, and occasionally from the ventral abdominal wall caudal to the costal arch. Unlike left or right abomasal displacement, gas accumulation is not a feature of non-displaced abomasal dilatation. The luminal contents generally appear homogenous and hypoechoic, reflecting the fluid nature of the retained hypochloric acid (fig. 16). This anechoic environment often provides distinct visualization of the abomasal folds, which appear as thin, echogenic, undulating structures. In cases where the dilatation results from intestinal ileus, the intestinal loops also appear distended and are easily identifiable on ultrasonography. Conversely, in pyloric stenosis, the small intestinal loops are typically empty (Barun, 2009).

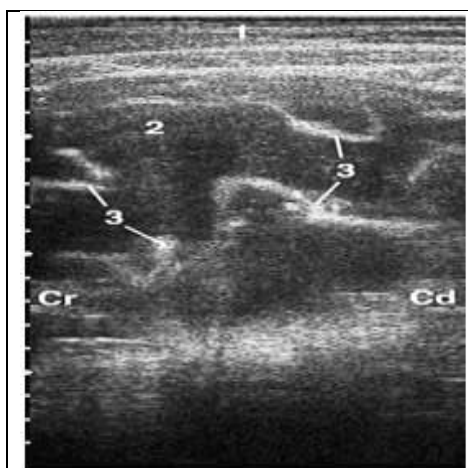


Fig.16: Ultrasonogram of the abomasum dilated secondary to ileus of the jejunum imaged from the right paramedian region. Abdominal wall (1), abomasum (2), abomasal folds (3). Cd, caudal; Cr, cranial.

Abomasitis/Abomasal Ulcers

Focal mucosal abnormalities of the abomasum, including chronic abomasitis, parasitic nodules, erosions, and type 1 ulcers, cannot be reliably visualized ultrasonographically when using a 3.5- or 5.0-MHz transducer. Type 3 abomasal ulcers typically result in ultrasonographic findings resembling those of fibrinous traumatic reticuloperitonitis accompanied by abscess formation. In contrast, type 4 abomasal ulcers are associated with diffuse peritoneal involvement characterized by ascites, fibrinous adhesions, and thickening of adjacent intestinal loops, indicative of generalized peritonitis. To date, direct ultrasonographic visualization of abomasal ulcers has not been documented (Barun, 2009)

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NUTRITIONAL PROPERTIES AND HEALTH BENEFITS OF MILLETS

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Introduction about Millets -

India is an agricultural country and has achieved remarkable growth in food grain production since the Green Revolution. The introduction of high-yielding crop varieties, improved irrigation systems, and modern farming techniques made India self-sufficient in food grain production. However, this success also led many farmers to shift their focus from traditional food crops to cash crops such as sugarcane, cotton, and oilseeds. While this change brought short-term economic benefits, it caused a gradual decline in the cultivation of traditional and nutritious coarse grains, which were once an essential part of rural Indian diets (ICAR, 2022; NABARD, 2023).

In recent years, global climate change has started affecting agriculture in many parts of India. Unpredictable rainfall, frequent droughts, and rising temperatures have made farming difficult and risky. Under such conditions, it has become important to promote crops that can grow in dry and less fertile regions. Coarse grains or millets are naturally suited for these conditions, as they require very little water, minimal fertilizers, and low maintenance. These crops are eco-friendly, drought-resistant, and economically suitable for small and marginal farmers, making them ideal for sustainable agriculture (Gopalakrishnan, 2020; FAO, 2023).

At the same time, the eating habits of people have changed significantly due to urbanisation and the influence of modern lifestyles. Processed and fast foods have replaced traditional grains in daily meals, leading to a rise in lifestyle-related diseases like obesity, diabetes, and heart problems. The nutritional value of traditional grains like millets has been largely forgotten. Recognising this issue, the United Nations declared 2023 as the *International Year of Millets (Coarse Grains)* to raise global awareness about their nutritional, environmental, and economic importance and to encourage their inclusion in everyday diets (United Nations, 2023; IFPRI, 2023).



Various coarse grains

Coarse grains, also called “Nutri-cereals”, include a wide variety of traditional grains such as jowar (sorghum), bajra (pearl millet), nachani (finger millet), kangani (foxtail millet), bhagar or varai (barnyard millet), chena (proso millet), kodra (kodo millet), sava (little millet), kutki (panicum millet), buckwheat (kuttu), amaranth (rajgira), and brown top millet. These grains are rich in essential nutrients like iron, calcium, fibre, protein, and amino acids, and are easy to digest. They are especially beneficial for managing lifestyle diseases such as diabetes, heart problems, and obesity. Because of their high nutritional value and health benefits, millets are now being recognised as superfoods all over the world (Babu & Ramanjaneyulu, 2017; Yadav & Patel, 2018; Ragaee & Abdel-Aal, 2006; Chandrasekara & Shahidi, 2012; Benedict & Charalambous, 2021).

India is the largest producer of coarse grains, contributing around 41% of global production. Apart from their nutritional value, these crops also help improve soil fertility by increasing nitrogen levels, maintaining soil health, and promoting biodiversity. Birds and animals naturally depend on these grains, which helps in conserving the environment. Furthermore, millet cultivation and processing provide new livelihood opportunities for rural communities, especially women and self-help groups (SHGs), who can prepare and market value-added millet-based products like flour, papad, laddus, biscuits, and snacks. This not only supports income generation but also contributes to rural empowerment and local economic growth (IMNET, 2023; Millets for Health Initiative, 2023). Currently, India cultivates about sixteen major types of coarse grains, including jowari (sorghum), bajri (pearl millet), nachani (finger millet), kangani (foxtail millet), bhagar or vari (barnyard millet),

chena or punarva (proso millet), kodra (kodo millet), sava or saanva (little millet), kutki (panicum millet), buckwheat (kuttu), amaranth (rajgira), and brown top millet. It is essential to promote public awareness about the importance of these grains and encourage their regular use in meals. Government programs, public awareness campaigns, school meal schemes, and nutrition drives should work together to revive the use of these traditional crops. By doing so, India can ensure better nutrition, strengthen sustainable agriculture, and preserve its traditional food heritage for the coming generations (Singh & Gupta, 2020; FAO, 2023; NIN, 2019). Some important information about major coarse grains is presented below.

There are many types of coarse grains and approximately sixteen major types of coarse grains are grown and exported in India. Which includes coarse grains like jowari (sorghum), bajri (pearl millet), nachani or nagli (finger millet), kangani or raale (foxtail millet / minor millet), bhagar or vari or varai (barnyard millet), chena/punarva (proso millet), kodra (koda / kodo millet), sava / saanva / jangora (little millet), kutki (korale / panicum millet), buckwheat/kuttu (two pseudo millet), amaranth (Amaranthus) and brown top millet. It is very important to create awareness about the importance of coarse grains and encourage their inclusion in the daily diet. Public campaigns, school nutrition programs, and government schemes should work together to revive these traditional grains. By doing so, India can not only strengthen its agricultural sustainability but also preserve its rich food heritage for generations to come. Some information about some coarse grains given as below,

Types of Different Millets and their nutritional Properties -

1. Sorghum:

This cereal was cultivated in **Egypt** 2,000 years BC. It is cultivated in Africa, India, China, Manchuria, the United States of America and many other countries. Sorghum is the second most important human food crop in India after rice. Maharashtra accounts for 34% of the sorghum area in India.

Nutritional properties-

- The main component of sorghum protein is prolamin which has the property of reducing digestibility during cooking which may be beneficial for health for certain dietary groups.
- Cooked sorghum protein is easier to digest than other cereal proteins.
- Sorghum is rich in protein, fibre, thiamine, riboflavin, folic acid and carotene.

- It is rich in iron, zinc and sodium along with potassium, phosphorus and calcium.



• **Sorghum**

2. Pearl Millet:

This fine cereal is grown in India, Africa and some countries in the Middle East. In India, it is important after sorghum and is cultivated especially in Maharashtra, Rajasthan, Punjab and Madhya Pradesh. Millet has a higher protein content than sorghum.



Pearl Millet

Nutritional properties-

- Pearl millet is rich in protein (12-16%) as well as lipids (4-6%).
- It contains 11.5% dietary fibre. This increases the transit time of food in the intestine. Therefore, the risk of inflammatory bowel disease is reduced.
- Millet has the highest niacin content among all other cereals. It also contains folate, magnesium, iron, copper, zinc and vitamins E and B-complex.
- It has a high energy content compared to other coarse grains.
- It is also rich in calcium and unsaturated fats which are good for health.

3. Finger millet:

This is the most resilient coarse grain of the poor for waterlogged farming. It grows in all types of soils as well as in arable and horticultural crops. It is an important crop in the state of Karnataka and almost as much area is under this crop there. This crop is also cultivated on a large scale in Tamil Nadu, Andhra Pradesh and Maharashtra. It is considered nutritious and nutritious and contains high amounts of protein and calcium. It is considered beneficial for diabetics. The special thing about this grain is that it can be stored for many years without spoiling, unlike other cereals.



Finger millet

Nutritional properties-

- finger millet is the highest source of calcium (300-350 mg/100g)
- finger millet contains the highest amount of minerals. It is low in protein (6-8%) and fat (1.5-2%).
- finger millet proteins are unique due to their sulphur rich amino acid content.
- It has excellent malting properties and is widely used as a dairy product.
- finger millet has high antioxidant activity. This delays ageing.

4. Foxtail millet:

This fine-grained, drought-resistant crop is mainly cultivated in Andhra Pradesh, Karnataka and Maharashtra. It can be cultivated in regions up to 2,000 m. altitude.

Nutritional properties-

- Foxtail millet is high in carbohydrates.
- It contains twice the protein of rice.
- Minerals like copper and iron are abundant.

- It provides a good supply of nutrients. Sweet and delicious and is considered one of the most digestible and non-allergenic grains.



Foxtail millet

5. Amaranth:

It is a plant and coarse grain that is easily found in agriculture in tropical countries and India. It is a plant that usually grows one and a half meters tall. Its leafy vegetables and seeds are also used as a nutritious food and as a fasting food.



Amaranth

Nutritional properties-

- High protein content (13-14%) and carrier of lysine, an amino acid; which is not available in many other grains or is present in negligible amounts.
- Amaranth contains 6 to 9% fat which is higher than other grains. Amaranth oil contains approximately 77% unsaturated fatty acids and is high in linoleic acid.
- It is high in dietary fiber.

- It is high in iron, magnesium, phosphorus, potassium and calcium.
- Amaranth is low in cholesterol and is a rich dietary source of phytosterols.
- It contains lunasin, a peptide like peptide and other bioactive peptides. It prevents two diseases namely cancer and hypertension.

Health Benefits of Coarse millets –

Millets are highly nutritious grains that offer a wide range of health benefits and play a vital role in maintaining overall wellness. They are rich in dietary fibre, high-quality protein, essential minerals like iron, calcium, and magnesium, and important vitamins that help keep the body strong and healthy (Ragae & Abdel-Aal, 2006; NIN, 2019). Regular consumption of millets helps in reducing the risk of heart diseases, controlling diabetes, and improving digestion due to their high fibre content, which supports smooth bowel movement and prevents constipation (Chandrasekara & Shahidi, 2012; Yadav & Patel, 2018). Their low glycaemic index makes them ideal for diabetic individuals, as they release sugar slowly into the blood, helping to maintain steady glucose levels (Benedict & Charalambous, 2021). Millets also help in lowering bad cholesterol (LDL), maintaining healthy blood pressure, and improving cardiovascular health (Singh & Gupta, 2020). They naturally detoxify the body, boost the immune system, and keep the respiratory system healthy. Moreover, millets increase energy levels, strengthen muscles and nerves, and protect against several degenerative diseases such as metabolic syndrome, Parkinson's disease, and certain types of cancer (Chandrasekara & Shahidi, 2012; Babu & Ramanjaneyulu, 2017).

These grains are also a good source of antioxidants like phenolic acids, flavonoids, lignans, avenanthramides, and phytosterols, which reduce oxidative stress, prevent cell damage, and slow down ageing (Yadav & Patel, 2018; FAO, 2023). They enhance brain function, improve memory and concentration, and support mental well-being (Babu & Ramanjaneyulu, 2017). Being gluten-free, millets are suitable for people with celiac disease or gluten intolerance (NIN, 2019). Regular inclusion of millets in the diet aids in weight management, promotes healthy skin and hair, and provides a feeling of fullness that prevents overeating (IFPRI, 2023; IMNET, 2023). With their outstanding nutritional and medicinal value, millets are now recognised as a true superfood that not only provides energy but also helps prevent modern lifestyle diseases such as obesity, hypertension, and diabetes (ICAR, 2022; United Nations, 2023; NABARD, 2023). Therefore, adding millets to our daily diet can lead to better

health, stronger immunity, and a more sustainable lifestyle for both urban and rural populations (Gopalakrishnan, 2020; Millets for Health Initiative, 2023).

Conclusion

Millets are among the most important traditional grains of India, known for their rich nutrition and health benefits. They are rightly called “Nutri-cereals” because they contain good amounts of fibre, protein, vitamins, and minerals like iron, calcium, and zinc, which help keep our body strong and healthy. Eating millets regularly helps in better digestion, maintaining heart health, and controlling blood sugar levels. They are also gluten-free, so they can be safely eaten by people who cannot eat wheat. Millets grow easily in dry and less fertile soils with very little water and almost no chemical fertilizers, which makes them very useful for sustainable and environment-friendly farming. These crops also help in improving the soil and protecting nature. Along with health benefits, millets can also give new income opportunities to farmers and rural women through processing and sale of millet-based foods. Therefore, bringing millets back into our daily diet is very important for good health, food security, and environmental balance. The United Nations declaring 2023 as the International Year of Millets has once again shown the world the true importance of these ancient grains for a healthy and sustainable future.

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AGROFORESTRY FOR ENVIRONMENTAL CONSERVATION AND LIVELIHOOD IMPROVEMENT

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Agroforestry is a dynamic, ecologically based natural resource management system that, through which the integration of trees/woody perennials in farm and rangelands, diversifies and sustains production for increased social, economic and environmental benefits. Agroforestry plays a vital dual role in both environmental conservation and livelihood security by integrating trees with crops and livestock, leading to sustainable ecosystems and resilient communities. It combines agricultural and forestry technologies to create more diverse, productive, profitable, and sustainable land-use systems. In recent decades, agroforestry has emerged as a vital strategy for addressing environmental degradation, enhancing biodiversity, mitigating climate change, and improving rural livelihoods.. It enhances biodiversity, sequesters carbon, improves soil and water health, and simultaneously provides diverse sources of income and food for rural populations. Agroforestry has the potential to contribute to the improvement of rural livelihood, due to the capacity of its various forms to offer multiple alternatives and opportunities to small holders to enhance farm production and income while conserving the biodiversity and environment. The growing food insecurity and deteriorating livelihood situations call for concerted and consorted actions to take advantage of the high potential of agroforestry, among other systems, for promoting livelihood. Various patterns of agroforestry systems are practiced in different agro-ecological regions of india which reflects biophysical and social variations. Trees are planted on the borders or within the field, systemically or at irregular intervals, usually with crops such as rice, wheat, pulse, jute, oilseed, sugarcane, vegetables and others, and farmers also grow shade-tolerant crops such as turmeric, ginger when trees have high canopy coverage . Trees in crop fields work as insurance in case of sudden crop failure or to support crops against environmental hazards and also to provide extra income from trees. Moreover, if there is a failure in one crop, the other crops would supplement the deficit.

1. Agroforestry for Environmental Conservation

Agroforestry contributes significantly to environmental protection and ecological stability through various mechanisms. Agroforestry increases biodiversity and wildlife habitats by mixing plant species and trees, fostering robust ecosystems that are more resilient to pests and climate stresses.

It sequesters significant amounts of carbon dioxide in both trees and soil, making it an essential climate-smart agricultural practice that helps mitigate global warming. Deep-rooted plants in agroforestry systems reduce soil erosion, improve soil structure, conserve water through better infiltration and moisture retention and limit the need for synthetic fertilizers and pesticides. It prevents nutrient run-off and pollution in water bodies, promotes ecological balance, and acts as a buffer against extreme weather events.

1.1 Soil Conservation

Tree roots bind soil particles and prevent erosion. Leaf litter from trees improves soil organic matter and structure. Agroforestry systems reduce surface runoff and enhance water infiltration. Tree litter and root activity enhance soil aggregation, organic matter, and infiltration (Cardinael et al., 2017). Agroforestry systems have been shown to reduce soil loss by 50–90 % compared with bare croplands (Jose, 2009). Riparian buffer strips in agroforestry filter sediments and nutrients, improving downstream water quality (Udawatta et al., 2002).

1.2 Water Conservation

Trees conserve water by reducing surface runoff through interception, slowing down water flow and increasing soil infiltration. They also decrease evaporation by shading the ground, and they promote rainfall through transpiration, which releases moisture into the atmosphere. Furthermore, their root systems improve soil porosity and help replenish groundwater reserves. Trees reduces evapotranspiration and maintains soil moisture. Buffer strips and riparian tree zones prevent water pollution and siltation of rivers.

1.3 Biodiversity Conservation:

Agroforestry acts as a habitat corridor for wildlife. Supports genetic diversity of plants and microorganisms. Reduces deforestation pressure on natural forests.

1. Habitat Heterogeneity and Species Richness

Agroforestry creates habitat mosaics that enhance landscape connectivity and species diversity (Bhagwat et al., 2008). Shade-grown coffee and cocoa systems, for example, maintain up to 60–80 % of forest bird species richness (Perfecto & Vandermeer, 2010). The structural complexity of tree canopies supports diverse taxa, from pollinators to soil microorganisms (Jose, 2009).

b. Functional Diversity and Ecosystem Services

Biodiversity in agroforestry systems underpins essential ecosystem services—pollination, pest regulation, and nutrient cycling (Kremen & Miles, 2012). The presence of multipurpose trees ensures continuous floral resources, sustaining pollinator populations crucial for both wild and cultivated plants (Tscharntke et al., 2011).

1. Agroforestry as a Buffer to Deforestation

In tropical regions, agroforestry provides alternative livelihoods that reduce dependence on forest extraction (Leakey, 2012). By increasing tree cover on farms, it acts as a buffer zone around protected forests, reducing encroachment and promoting landscape connectivity (Schroth & Harvey, 2007).

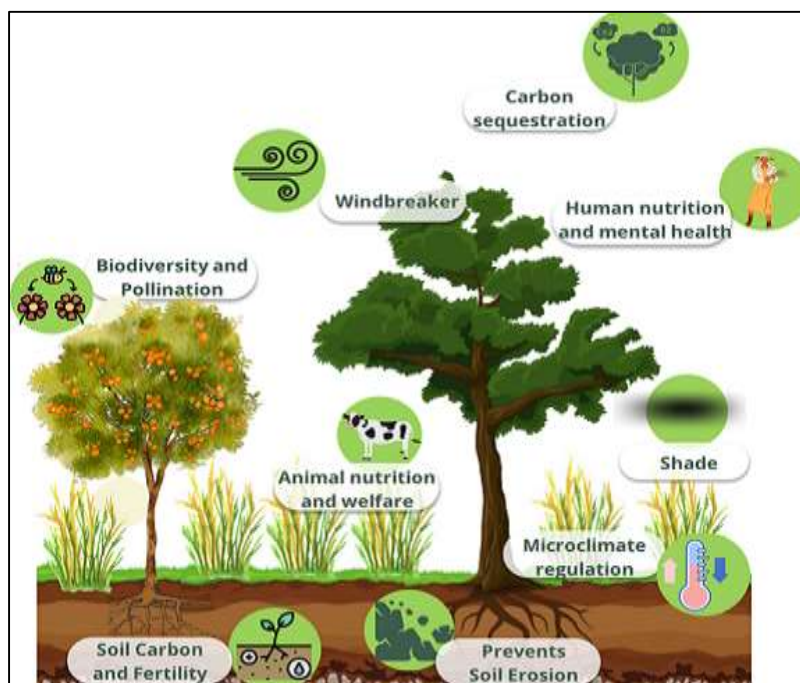


Fig.1 Agroforestry delivers a diverse range of environmental, ecological, economic and social benefits for people and nature. (UKRI Treescapes Programme)

d. Biodiversity Corridors and Climate Refugia

Agroforestry landscapes provide corridors that allow species movement under shifting climate envelopes (Bhagwat et al., 2008). They act as refugia for pollinators and seed dispersers critical for forest regeneration (Tschamntke et al., 2011).

1.4 Landscape Restoration and Land Degradation Neutrality

Agroforestry directly contributes to landscape restoration by re-establishing ecological functions, such as water management and soil conservation, in degraded areas. By supporting the restoration of wastelands and degraded lands, agroforestry helps countries achieve national and international restoration commitments, such as the UN Decade on Ecosystem Restoration and the Bonn Challenge. Agroforestry is central to restoration initiatives such as the Bonn Challenge and the AFR100, targeting millions of hectares of degraded lands (FAO, 2022). Agroforestry plays a crucial role in achieving Land Degradation Neutrality (LDN) by integrating trees with crops and/or livestock to restore degraded lands, improve soil health, and sequester carbon. It contributes to Land Degradation Neutrality (LDN) goals by restoring soil function and vegetative cover (World Bank, 2021).

2. Agroforestry for Livelihood Improvement

Agroforestry systems play a crucial role in supporting rural economies and improving livelihoods through multiple benefits. Agroforestry provides multiple income streams from crops, fruits, nuts, timber, and livestock, making farmers less vulnerable to market or climate shocks. By maximizing land use and protecting crops through natural barriers, it boosts overall yields and profitability, offering employment opportunities and helping alleviate rural poverty. The integration of diverse crops and trees improves food and nutritional security for families, reducing the risks of crop failure and providing a reliable safety net. Agroforestry enhances household consumption and supports rural development through job creation and sustainable resource management. Agroforestry empowering women by providing with nearby food and fuel sources, which reduces the time and effort needed for gathering these resources and can supplement household income.

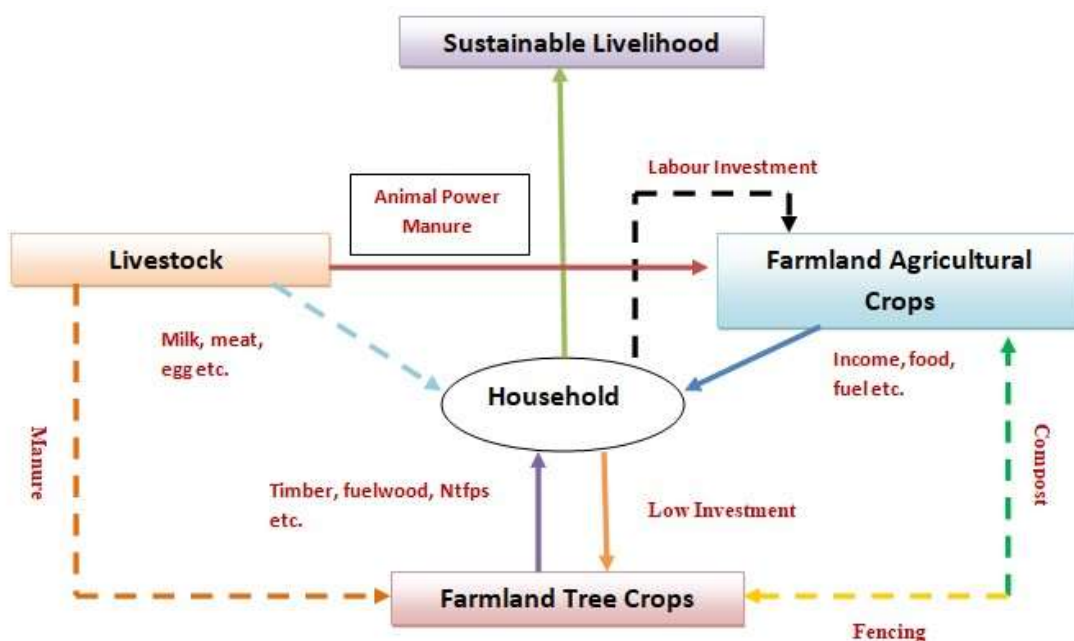


Fig.2 Agroforestry for Livelihood Improvement

2.1 Income Diversification

Agroforestry diversifies income in India by providing multiple revenue streams from products like timber, fruits, and medicinal plants, reducing dependence on a single crop and acting as a buffer against market and weather volatility. It also creates income from non-timber products, carbon credits, and by supporting livestock, ultimately increasing financial stability and improving livelihoods, especially for small farmers.

Farmers gain income from trees (timber, fuelwood, fruits, medicinal plants, etc.) alongside crops and livestock. Reduces dependence on a single income source, minimizing risk. Agroforestry

diversifies income sources through timber, fuelwood, fruits, fodder, and non-timber forest products (Mercer, 2004). For smallholders, this reduces vulnerability to market and climate shocks (Franzel et al., 2001). Empirical data from India and Kenya show agroforestry contributing 25–45 % of household income (Rahman et al., 2020)

2.2 Food and Nutritional Security

Agroforestry systems combine trees with crops and livestock, creating diverse food sources that are more resilient to seasonal hunger and crop failures compared to monocropping. Tree products such as fruits, nuts, and leaves supplement household nutrition. Agroforestry supports year-round food availability. Trees and shrubs provide a direct source of diverse foods, including fruits, nuts, and leafy vegetables, which are vital for a balanced diet and improving the nutritional content of household food intake. Trees enhance food security by directly supplying edible fruits, leaves, and nuts and indirectly through soil fertility improvement that boosts crop yields (Mbow et al., 2014). Tree-based farming systems also provide fodder and shade, improving livestock productivity and overall farm resilience (Bayala et al., 2014).

2.3 Employment Generation

Agroforestry provides year-round employment for farmers and laborers in activities like planting, pruning, and harvesting timber, fruits, nuts, and medicinal plants. A significant number of jobs are created in processing and marketing agroforestry products which encourages small-scale forest-based enterprises (e.g., honey, gum, bamboo crafts). The development of agroforestry also necessitates improvements in rural infrastructure, such as irrigation systems, roads, and transportation networks, creating jobs in construction and logistics.

2.4 Fuelwood and Fodder Supply

Agroforestry systems provide essential resources like fodder for livestock and fuel for cooking, which can be particularly beneficial for women in rural communities thus meets household energy and livestock feed needs and reduces women's burden of collecting fuelwood from distant forests.

2.5 Social and Economic Resilience

Socially, it strengthens community connections and provides essential resources like fuel and fodder, especially for women. Economically, it increases long-term productivity through improved soil and water retention, and offers opportunities like carbon credits and off-farm employment. Enhances farmers' resilience against market fluctuations and climate shocks. Promotes community-based resource management.

2.6 Gender and Equity Considerations

Agroforestry offering opportunities for women's economic empowerment, food security, and leadership, while simultaneously facing challenges like limited land and resource access, patriarchal norms, and a lack of recognition for their critical contributions. To improve equity, it is essential to

ensure women have equal access to resources and training, and that policies are gender-responsive, which can help overcome barriers and fully realize the benefits of agroforestry for both women and communities. Women often play a key role in managing agroforestry species for household consumption and income (Kiptot & Franzel, 2012). Gender-responsive extension and access to credit can enhance adoption rates and equitable benefit distribution (Catacutan et al., 2017). Women can increase their agency and leadership skills through self-help groups and farmer producer organizations involved in agroforestry.

3. Case Studies and Examples

3.1 Taungya System (India)

1. Practiced in forest areas where crops are grown between young tree plantations.
2. Farmers benefit from short-term crops, while the forest department benefits from tree establishment.

3.2 Home Gardens (Kerala, India)

1. Multistorey systems combining coconut, banana, pepper, vegetables, and timber trees.
2. Provide food, income, and ecological stability.

3.3 Silvopastoral Systems (Latin America and India)

1. Integration of fodder trees with grazing lands improves livestock productivity and soil fertility.

3.4 Alley Cropping (Africa and Asia)

1. Leguminous trees planted in rows with crops grown in between.
2. Enhances soil nitrogen and reduces erosion.

4. Policy and Institutional Support

Governments and international agencies promote agroforestry for sustainable development

4.1 National Agroforestry Policy of India (2014):

Encourages integration of trees on farmlands, provides incentives, and promotes research-extension linkages.

4.2 ICRAF (World Agroforestry Centre):

Conducts research and promotes global agroforestry development.

4.3 MGNREGA and Green India Mission:

Support tree planting and agroforestry-based livelihood programs.

5. Challenges in Agroforestry Adoption

1. Limited awareness and extension services
2. Insecure land tenure and tree ownership rights
3. Long gestation period for tree benefits

4. Lack of market access for tree-based products
5. Inadequate credit and insurance support

6. Future Prospects

6.1 Climate-smart agriculture:

Agroforestry aligns with global climate resilience goals.

6.2 Carbon credit markets:

Potential income through carbon sequestration services.

6.3 Agroforestry value chains:

Enhancing processing, packaging, and marketing of non-timber forest products.

6.4 Digital and precision agroforestry:

Use of GIS, remote sensing, and AI for planning and monitoring.

Conclusion

The Agroforestry system is complex and consists of different components such as agriculture, livestock and forest, which together meet the subsistence needs of a typical household. Agroforestry trees contribute several products such as fuelwood, fruits, medicine, fodder and ecosystem services such as soil improvement, biodiversity conservation and water retention and conservation. Therefore to protect the local ecosystem and to promote stability agroforestry practices should be adopted by local communities. The impact of agroforestry adoption on improvement of community livelihoods will increased income through sales of the products, increased crop yield and improved health and nutrition. Therefore, agroforestry is significant in environmental conservation and improvement of rural community livelihoods.

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CANINE BRUCELLOSIS: A STEALTHY ADVERSARY IN KENNELS AND A ZOONOTIC CONCERN

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Abstract

Canine brucellosis, instigated by *Brucella canis*, stands as a formidable bacterial threat to canine reproductive health worldwide. This facultative intracellular pathogen is notorious for triggering late-term abortions, infertility, and chronic infections of the genital tract, often while presenting minimal to no overt clinical signs. Its stealthy nature promotes unchecked circulation, especially within high-density breeding kennels. Diagnosing the disease is complicated by the bacterium's rough lipopolysaccharide structure, which leads to cross-reactions in serological tests. Furthermore, the ability of *B. canis* to persist within host cells renders antibiotic treatments largely ineffective, with relapses being a common outcome. As a zoonotic agent, it poses a tangible risk to humans in close contact with infected dogs. The current lack of a commercial vaccine elevates the importance of disciplined diagnostic screening and robust kennel management protocols. This chapter synthesizes current knowledge on the bacterium's pathogenesis, clinical spectrum, diagnostic challenges, and control strategies, advocating for a unified One Health approach to mitigate its impact.

Keywords: *Brucella canis*, Reproductive Failure, Abortion, Zoonosis, Kennel Management, One Health, Serological Diagnosis

1. Introduction

Reproductive inefficiency and infertility in canine breeding establishments have gained increasing attention in recent years. Although various infectious agents can compromise reproductive health, canine brucellosis remains one of the most significant due to its ability to cause abortion, orchitis, epididymitis, and chronic infertility. The disease first came to prominence in the mid-1960s during an outbreak of abortion in Beagles, which led to the recognition of *Brucella canis* as a distinct pathogen (Hollett, 2006). Despite decades of research, the disease continues to be underdiagnosed because of its non-specific symptoms and limitations of available tests.

A thorough understanding of the pathogen, its epidemiology, and feasible control measures is essential for veterinarians, breeders, and public health professionals.

2. Etiology

The primary etiological agent, *Brucella canis*, is a Gram-negative, non-motile, non-spore-forming, facultative intracellular coccobacillus (Di Bonaventura, 2021). Unlike smooth *Brucella* species, *B. canis* possesses a rough lipopolysaccharide lacking the O-polysaccharide side chain, a characteristic that influences immune interaction and affects serological detection (Hollett, 2006). The organism has a marked affinity for steroid-rich tissues such as the uterus, placenta, epididymis, testicles, and prostate. Its intracellular lifestyle enhances persistence and complicates diagnosis and therapy. Although *B. canis* is the predominant species infecting dogs, exposure to *B. abortus*, *B. melitensis*, or *B. suis* may occur in dogs with close contact to infected livestock (Qureshi et al., 2023). Other species such as *B. ovis* and *B. neotomae* rarely infect dogs, while emerging terrestrial and marine strains including *B. inopinata* and *B. ceti* have uncertain canine pathogenicity (Carmichael & Greene, 2006).

3. Host Range

Domestic dogs serve as the principal host for *B. canis*. Natural infections in wild canids such as foxes and coyotes have been reported, although their role in maintaining the pathogen in nature remains unclear. Experimental infections indicate susceptibility in several mammalian species, but natural spillover events are relatively uncommon (Hollett, 2006).

4. Geographical Distribution

Canine brucellosis is recognized worldwide. Regions with high densities of breeding kennels and inadequate monitoring report more frequent outbreaks. The disease has been documented in many parts of North and South America, Europe, and Asia—including India, Japan, China, Korea, and Malaysia. Sporadic cases occur in Africa and Canada. Increased international trade of breeding dogs continues to facilitate global spread (Wanke et al, 2004).

5. Transmission

Brucella canis is a highly contagious agent whose spread is facilitated by its presence in a wide range of bodily fluids from infected dogs. The primary routes of infection involve mucosal contact with infectious materials.

Direct Contact: The most efficient transmission occurs through:

- **Venereal Spread:** During mating, the bacterium is transmitted via infected semen to the female's reproductive tract, and vice versa through vaginal secretions.
- **Oronasal Exposure:** This is a major route in kennel environments. Dogs become infected by sniffing, licking, or ingesting contaminated materials such as aborted fetuses, placentas, or vaginal discharges, which can contain extremely high bacterial loads (up to 10^{10} organisms per gram).
- **Conjunctival Contact:** Splashes or direct contact with infected fluids with the eye can also lead to infection (Carmichael and Green, 2012).

Shedding and Environmental Contamination: Infected dogs actively shed the bacterium, creating a persistent contamination risk.

- **Males** shed large quantities of *B. canis* in their semen and are also a significant source through urine, with concentrations ranging from 10^3 to 10^6 bacteria per milliliter.
- **Females** shed organisms in vaginal discharges post-abortion and during estrus, as well as in urine and milk.
- Although bacteria can be found in lower concentrations in saliva, nasal secretions, and feces, these are considered less significant sources.

Indirect Transmission: The resilience of *B. canis* in the environment is a critical factor in its spread. The bacterium can survive for several months in cool, moist, and dark conditions, particularly in the presence of organic matter. This allows for indirect transmission via **fomites**, including contaminated kennel floors, bedding, grooming tools, and feeding bowls. Human activity, such as on the clothing or hands of kennel staff, can also mechanically transfer the organism between dogs (Johnson and Walker, 1992).

6. Pathogenesis

After gaining entry through the genital, oronasal, or conjunctival mucosa, *B. canis* rapidly phagocytosed (engulfed) by white blood cells, such as macrophages. Unlike many bacteria, it evades destruction by employing specialized virulence factors, such as its VirB type IV secretion system, which allows it to survive and replicate within these host cells (Chacon-Diaz et al., 2015).

The intracellular bacteria then travel via the lymphatic system and bloodstream, leading to a persistent and intermittent bacteremia that can last for months or even years. This systemic dissemination allows the bacterium to reach and localize in specific target organs (Hollett, 2006).

The pathogen exhibits a marked tropism for reproductive tissues and steroid-rich sites. In females, it invades the placenta and gravid uterus, causing severe placentitis, necrosis of the chorionic villi, and impaired nutrient exchange, which ultimately results in late-term abortion or the birth of non-viable puppies. In males, it establishes infection in the epididymis, testes, and prostate, triggering intense inflammation that leads to orchitis, epididymitis, and ultimately, tissue degeneration and infertility (Carmichael and Joubert, 1988).

The formation of antibody-antigen complexes in the bloodstream can also circulate and deposit in other tissues, such as the intervertebral discs (causing discospondylitis), eyes (causing uveitis), and kidneys (causing glomerulonephritis), explaining the diverse non-reproductive clinical signs associated with chronic infection (Ledbetter et al., 2009; Long et al., 2022).

7. Clinical Manifestations

The clinical presentation of canine brucellosis varies widely, and many infected dogs may remain asymptomatic, contributing substantially to silent transmission within kennels. In females, the most characteristic manifestation is late-term abortion, typically occurring between 45 and 55 days of gestation, often followed by a persistent vaginal discharge. Affected females may also experience repeat breeding failures, early embryonic death, or chronic infertility. In males, the disease commonly presents as epididymitis, orchitis, scrotal swelling, and prostatitis, which can progressively lead to testicular atrophy and markedly reduced semen quality, including oligospermia or azospermia. Scrotal dermatitis may develop secondary to urine scalding. Beyond reproductive signs, infected dogs of either sex may exhibit nonspecific clinical features such as lethargy, weight loss, poor coat condition, uveitis, or musculoskeletal pain associated with diskospondylitis. Because many infected animals show mild or no outward signs, subclinical infections represent a major challenge in controlling the disease within breeding facilities (Kustriz, 2003; Ledbetter et al., 2009).

8. Laboratory Diagnosis

Accurate diagnosis of *Brucella canis* requires a multifaceted approach, combining serology, molecular techniques, and culture. Strict biosafety protocols are mandatory during sample collection and handling to prevent zoonotic transmission.

8.1. Serological Testing

Serology is the cornerstone for screening populations due to its practicality, though interpretation requires care.

- **Antibody Response:** Infected dogs typically develop detectable antibodies within 2-4 weeks post-infection. These antibodies target both cell wall and cytoplasmic proteins of the bacterium (Carmichael and Greene, 2012).
- **Screening Tests:** Common initial tests include the Rapid Slide Agglutination Test (RSAT) and the Tube Agglutination Test (TAT). The Rose Bengal Plate Test (RBPT) is also a sensitive, rapid screening tool effective in early infection; its high negative predictive value makes it excellent for ruling out disease in negative animals (Keid et al., 2009).
- **Confirmatory Tests:** Due to antigenic cross-reactivity, positive screening tests often yield false positives and must be confirmed. More specific confirmatory tests include the Agar Gel Immunodiffusion test using cytoplasmic protein antigens (AGID-CPA) and the 2-Mercaptoethanol RSAT (2ME-RSAT), which helps eliminate non-specific IgM reactions (Carmichael and Greene, 2006). A semi-quantitative TAT titer of 1:200 or greater is generally considered evidence of infection (Nicoletti and Chase, 1987a; Moon et al., 1994). The complement fixation test (CFT), enzyme-linked immune sorbent assay (ELISA), and fluorescence polarisation assay (FPA) can be employed for detecting *B. canis* (Garin Bastuji et al., 2006).

8.2. Molecular Diagnosis (PCR)

Polymerase Chain Reaction (PCR) assays provide a rapid and highly sensitive method for direct detection of bacterial DNA.

- **Advantages:** This technique is not dependent on bacterial viability, is faster than culture, and is less prone to issues related to sample contamination. It is particularly valuable for detecting infection in the window before seroconversion (Wanke, 2004).
- **Sample Choice and Targets:** Whole blood is the preferred sample, though semen, tissue, and vaginal discharges are also suitable. Most diagnostic PCRs are genus-specific, amplifying conserved gene sequences such as *bcs31*, 16S rRNA, and *recA* (Keid et al., 2009).

8.3. Bacterial Culture and Isolation

Culture remains the definitive "gold standard" for confirming a diagnosis.

- **Methodology:** The bacterium can be isolated from blood, semen, vaginal discharge, aborted tissues, and urine. Samples must be collected aseptically, transported in heparinized tubes or sterile containers, kept refrigerated (not frozen), and processed within 24 hours. Specialized

media like Farrell's or modified Thayer-Martin are required for optimal growth (Carmichael and Greene, 2012).

- **Limitations:** Despite its specificity, culture has low diagnostic sensitivity. Challenges include intermittent bacteremia, the fastidious and slow-growing nature of the bacterium, and the potential for inappropriate sample handling. Consequently, a negative culture result cannot definitively rule out infection. While impractical for large-scale screening, a positive culture provides unequivocal confirmation (Wanke, 2004).

9. Treatment

The management of canine brucellosis is notoriously challenging, and treatment is generally discouraged for several key reasons. The intracellular location of *B. canis* shields it from many antibiotics, leading to high relapse rates after therapy is discontinued. It involves prolonged courses of antibiotics that can penetrate cells, typically combining a tetracycline-class drug (e.g., doxycycline, minocycline) with an aminoglycoside (e.g., streptomycin). Regimens such as minocycline with streptomycin or long-acting oxytetracycline with streptomycin have shown some success in suppressing infection. However, even with these intensive protocols, a cure is not assured. The most reliable recommendations for confirmed cases in a breeding environment include permanent isolation from other dogs and neutering to eliminate reproductive shedding, with euthanasia often being the most responsible option to protect public health and prevent kennel-wide outbreaks (Carmichael and Greene, 2012).

10. Control and Prevention

Given the inconsistency and limited success of treatment protocols, prevention remains the most effective strategy for controlling canine brucellosis. All breeding dogs should undergo mandatory testing prior to their introduction into a kennel, followed by a quarantine period of 8–12 weeks with repeat testing to ensure that infected animals are not inadvertently added to breeding populations. Suspected or confirmed cases must be isolated immediately to prevent transmission. Routine screening of kennel dogs every six months is strongly recommended, especially in facilities with high breeding turnover. In addition, strict sanitation practices should be implemented, including appropriate disposal of aborted fetuses and reproductive discharges, coupled with thorough disinfection of contaminated areas using effective agents such as 1% sodium hypochlorite, 70% ethanol, glutaraldehyde, or iodine-based solutions. Adhering to rigorous hygiene, minimizing dog-to-dog contact, and preventing uncontrolled breeding are essential components of a successful control program (Rasool et al., 2023).

11. Public Health Significance

Brucella canis is an important zoonotic pathogen, and humans typically become infected through contact with infected dogs or their reproductive secretions. Individuals with close, frequent exposure to dogs—such as veterinarians, breeders, kennel workers, and laboratory personnel—are particularly at risk. Infected people often develop non-specific clinical signs including undifferentiated fever, fatigue, chills, malaise, lymphadenopathy, and musculoskeletal pain such as arthralgia or back pain. Because these symptoms resemble those of many other febrile illnesses, human brucellosis is frequently misdiagnosed or diagnosed late, especially in regions where specialized tests for *B. canis* are not readily available (Swenson *et al.*, 1972). Laboratory handling of *Brucella* isolates requires Biosafety Level-3 conditions to minimize occupational exposure. Control of *B. canis* is further complicated by several factors, including the absence of any effective or safe vaccine for dogs, the ability of infected animals to remain long-term carriers and shed bacteria intermittently, and the organism's capacity to survive for extended periods in organic material and moist environments. The disease also represents a significant One Health concern, as its management requires coordinated efforts between veterinary and human health sectors, along with strengthened surveillance, breeder education, and kennel biosecurity practices (Lawaczek *et al.*, 2011).

13. Conclusion

Canine brucellosis remains a significant reproductive and zoonotic challenge globally. Its often-subclinical presentation, limited diagnostic sensitivity, and difficulties in achieving complete therapeutic cure hinder effective control. Robust surveillance, improved diagnostic access, stringent kennel biosecurity, and educational outreach are crucial to mitigating its impact. A coordinated One Health approach is essential to protect both canine populations and human health.

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REPRODUCTION OF FUNGI

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Reproduction: Reproduction is the formation of new individuals having all the characteristics of the species.

Types of reproduction:

1. Asexual /non-sexual / vegetative / somatic reproduction
2. Sexual reproduction.

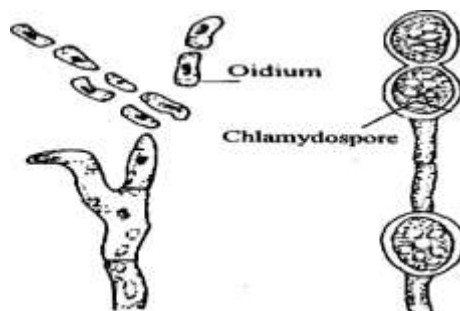
Asexual reproduction : Asexual reproduction stage is also known as imperfect stage and technically called as anamorphic stage. There is no union of nuclei /sex cells/ sex organs. It is repeated several times during the life span of a fungus producing numerous asexual spores. Hence, it is more important for fungi than sexual reproduction. Asexual spores are formed after mitosis, hence also called mitospores.

Methods of asexual reproduction :

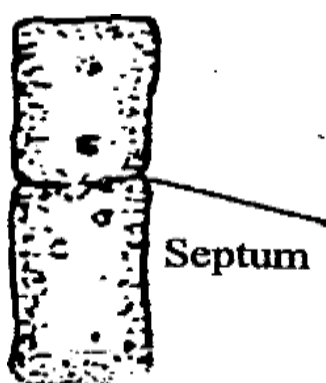
1. Fragmentation
2. Fission.
3. Budding
4. Sporulation (production of spores)

Fragmentation: It is the most common method. Hypha of fungus breaks into small pieces, each broken piece is called a fragment, which function as a propagating unit and grows into a new mycelium. The spores produced by fragmentation are called **arthrospores** (arthron=joint) (spora=seed) or oidia.. Eg. *Oidium*, *Geotrichum*. Sometimes, the contents of intercalary cells or terminal cells of hypha rounded off and surrounded by thick wall and formed as chlamydospores which are thick walled resistant spores produced either singly or in chains. Eg. *Fusarium oxysporum*, *Ustilago tritici*

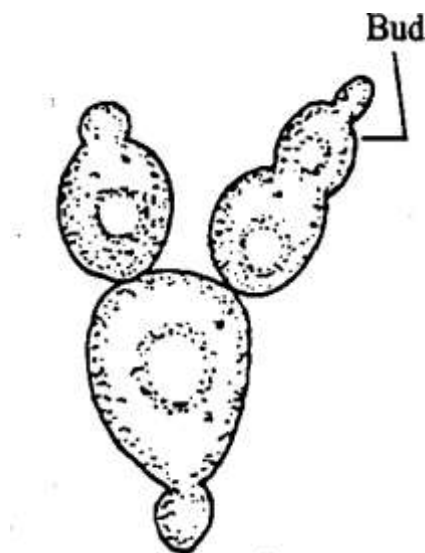
Fission / Transverse fission: The parent cell elongates, nucleus undergo mitotic division and forms



two nuclei, then the contents divide into equal halves by the formation of a transverse septum and separates into two daughter cells. Eg. *Saccharomyces cerevisiae*



Budding: The spores formed through budding are called **blastospores**. The parent cell puts out initially a small out growth called bud / blastos i.e., sprout or out growth which increases in size and nucleus divides, one daughter nucleus accompanied by a portion of cytoplasm migrates into bud and the other nucleus remains in the parent cell. Later, the bud increases in size and a constriction is formed at the base of bud, cutting off completely from parent cell. Bud, when separated from parent cell, can function as an independent propagating unit. Sometimes multiple buds are also seen i.e., bud over bud and looks like pseudomycelium. Eg. *Saccharomyces cerevisiae*.



Sprulation (spores): The process of production of spores is called sporulation. Spore: It is a minute, simple propagating unit of the fungi, functioning as a seed but differs from it in lacking a preformed embryo that serves in the reproduction of same species.

Spores vary in colour, size, number of cells and the way in which they are borne.

There are 2 main types of spores.

1. Sporangiospores

2. Conidia

Sporangiospores: When the asexual spores are produced internally, within the sporangia, such spores are called sporangiospores. The sac like structure which produces sporangiospores is called sporangium. The special hypha bearing sporangium is called sporangiophore which may or may not be distinguishable from hypha. A small sporangium with or without columella containing a few or single spore is called as sporangiolium. Eg. *Choanephora trispora*.

Sporangium which is cylindrical in shape is called as merosporangium. Eg. *Syncephalastrum racemosum*.

Sporangium with columella is called as columellate sporangium. Eg. *Rhizopus stolonifera*.

Sporangiospores are of 2 types.

a. Zoospores /planospores

b. Aplanospores

a. Zoospores /planospores: sporangiospores which are motile by flagella are called zoospores. Also known as planospores. Eg. *Pythium*, *Phytophthora*.

b. Aplanospores: Non-motile asexual spore that is found in some algae and fungi.

Types of flagella: Flagella of zoospores are of 2 types.

a. Whiplash

b. Tinsel

a) Whiplash: A flagellum with long, thick, rigid basal portion and with a short, narrow, flexible, upper portion. It gives a whip like appearance to flagellum.

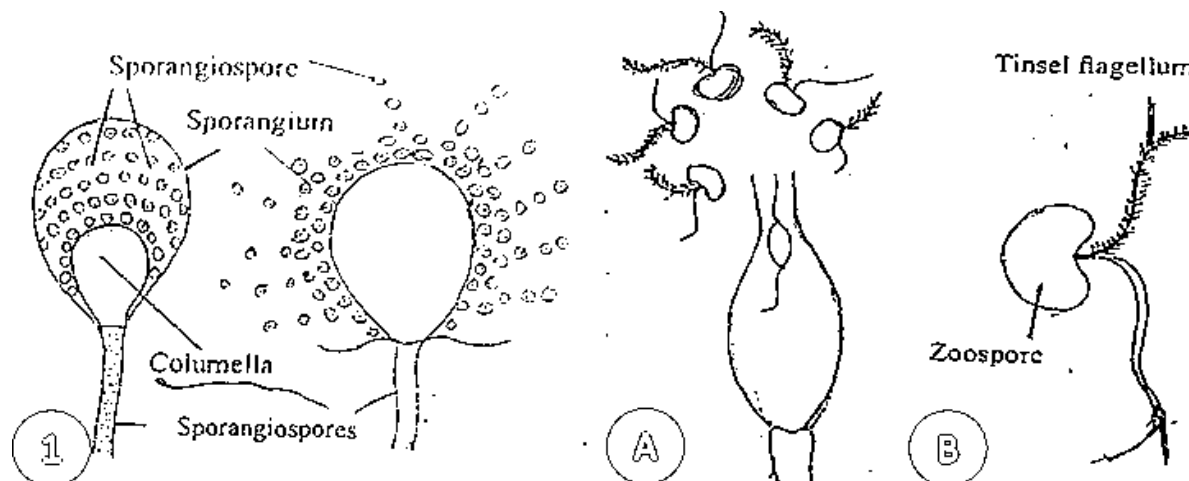
b) Tinsel: It is a feathery structure consisting of a long rachis with lateral hair like projections called mastigonemes or flimmers on all sides along its entire length. The number, position and nature of flagella play an important role in the classification of lower fungi.

Uniflagellate zoospore: A zoospore with a single flagellum, may be placed at anterior or posterior end of spore.

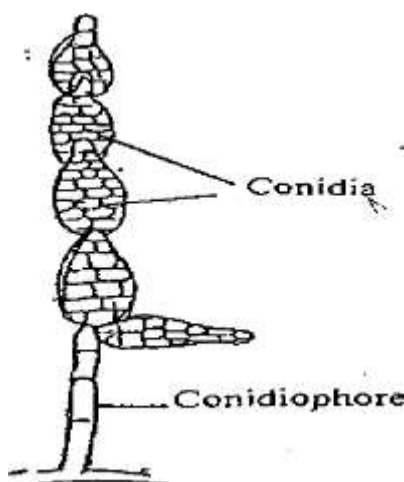
Biflagellate zoospore: A zoospore with two flagella, situated laterally or anteriorly on zoospore.

One whiplash, one tinsel type flagella and equal in size. e.g. *Pythium aphanidermatum*, *Phytophthora infestans*

Both whiplash flagella, unequal in size (heterokont). e.g. *Plasmodiophora brassicae*.



Conidia / Conidiospores: (konis=dust; oides=like) Conidia are non-motile asexual spores which may arise directly from somatic hyphae or from specialized conidiogenous cells (a cell from which conidia are produced) or on conidiophore (hypha which bear conidia). Conidia are produced freely on conidiophore i.e., at the tips or sides of conidiophore or may be produced in specialized asexual fruiting bodies viz., pycnidium, acervulus, sporodochium and synnemata.



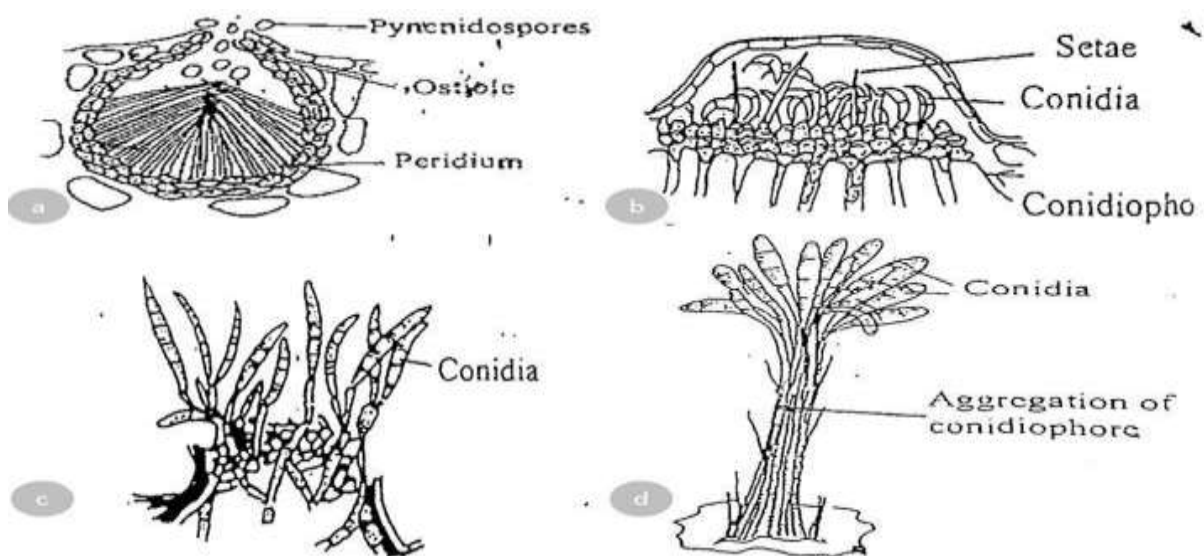
Asexual fruiting bodies :

(a) Pycnidium: (pl. pycnidia) It is a globose or flask shaped fruiting body lined inside with conidiophores which produce conidia. It may be completely closed or may have an opening called ostiole. Pycnidium may be provided with small papillum or long neck. e.g. *Phomopsis*.

(b) Acervulus: (pl. acervuli) A flat or saucer shaped fruiting body with a stromatic mat of hyphae producing conidia on short conidiophores. An acervulus lacks a definite wall structure and not having an ostiole or definite line of dehiscence. e.g. *Colletotrichum*, *Pestalotiopsis*.

(c) Sporodochium: (pl. sporodochia) A cushion shaped asexual fruiting body. Conidiophores arise from a central stroma and they are woven together on a mass of hyphae and produce conidia. e.g. *Fusarium*

(d) Synnemata: (pl. synnema) A group of conidiophores often united at the base and free at the top. Conidia may be formed at its tip or along the length of synnema, resembling a long handled feather duster. e.g. *Graphium*



2. Sexual reproduction: Sexual reproduction involves union of two compatible nuclei or sex cells or sex organs or somatic cells or somatic hyphae for the formation of new individuals. Sexual stage is perfect stage and technically called as teleomorphic stage. Sexual cycle normally occurs once in the life span of the fungus. Sexual spores or sexual structures which contain sexual spores are thick walled, resistant to unfavourable conditions and are viable for longer period and thus these spores help the fungus to perpetuate from one season to another, hence these are called as resting spores. Sexual spores are definite in number.

Sex organs of fungi:

Gametangia: Sex organs of fungi are called gametangia containing gametes or gamete nuclei.

Gametes: Sex cells are called as gametes.

Antheridium: (pl. antheridia) Male gametangium is called as antheridium. Male gametangium is small and club shaped.

Oogonium/Ascogonium:(pl. oogonia/ascogonia): The female gametangium is called Oogonium (oomycetes) or ascogonium (ascomycotina). Female gametangium is large and globose shaped.

Male gametes are called antherozoids or sperm or spermatozoids. Female gametes are called egg or oosphere.

Planogametes: If gametes are motile, they are called planogametes. **Isogametangia:** If gametangia are morphologically similar or identical i.e., indistinguishable as male and female, they are called as isogametangia.

Isogametes: If gametes are similar morphologically, they are called as isogametes. **Heterogametangia:** If gametangia differ morphologically in size and structure, they are called as heterogametangia.

Heterogametes: If gametes differ morphologically, they are called heterogametes. **+ or – signs:** In some sexually undifferentiated fungi, male and female are symbolically designated as ‘+’ (male) and ‘–’ (female).

Classification of fungi based on sex:

1. Monoecious fungi/hermaphroditic fungi: (mono=single,oikos=home) The fungi which produce distinguishable male and female sex organs on the same thallus, which may or may not be compatible are called monoecious/ hermaphroditic fungi.
e.g. *Pythium aphanidermatum*.

2. Dioecious fungi: (di=two, oikos=home) The fungi which produce distinguishable male and female sex organs on two different thalli i.e., there will be separate male and female thalli. e.g. *Phytophthora infestans*.

Classification of fungi based on compatibility;

Homothallic fungi: Fungi in which both sexes occur on same thallus, which can reproduce sexually by it self with out the aid of another thallus ie., self compatible

/ self fertile are called homothallic fungi. e.g. *Pythium aphanidermatum*.

Heterothallic fungi: A fungal species consisting of self sterile (self incompatible) thallus requiring the union of two compatible thalli for sexual reproduction, regardless of the possible presence of both male and female organs on the same thallus. Heterothallic fungi are dioecious e.g. *Phytophthora infestans*.

Phases in Sexual reproduction: There are 3 phases in sexual reproduction.

1. Plasmogamy: union of two protoplasts takes place.As a result of it the two nuclei come

together within the same cell.

2. Karyogamy: union of 2 sexually compatible nuclei brought together by plasmogamy to form a diploid nucleus ($2n$) i.e., zygote.

3. Meiosis: This is reduction division. The number of chromosomes is reduced to haploid (n) i.e., diploid nucleus results into haploid nucleus.

In lower fungi (Phycomycetes -Mastigomycotina and Zygomycotina) plasmogamy, karyogamy and meiosis occurs at regular intervals / sequence i.e., karyogamy follows immediately after plasmogamy. In higher fungi (Ascomycotina, Basidiomycotina), karyogamy is delayed, as a result the nuclei remain in pairs (dikaryotic phase- $n+n$ condition), which may be brief or prolonged.

Dikaryon: A pair of genetically different nuclei, lying side by side without fusion for a considerable period of time is called dikaryon. A cell containing dikaryon is called **dikaryotic cell**. And the process is known as **dikaryogamy**.

Methods of sexual reproduction:

1. Planogametic copulation.
2. Gametangial contact
3. Gametangial copulation
4. Spermatization
5. Somatogamy.

1. Planogametic copulation (gametogamy): This involves the union of 2 naked gametes one or both of which are motile.

a. Isogamy (Isogamous planogametic copulation): If both gametes are motile and similar. e.g. *Plasmodiophora brassicae*.

b. Anisogamy (Anisogamous planogametic copulation) : If both gametes are motile but dissimilar. e.g. *Allomyces macrogynus*

c. Heterogamy: If gametes are dissimilar, one motile, another is non motile. e.g.

Monoblepharis polymorpha.

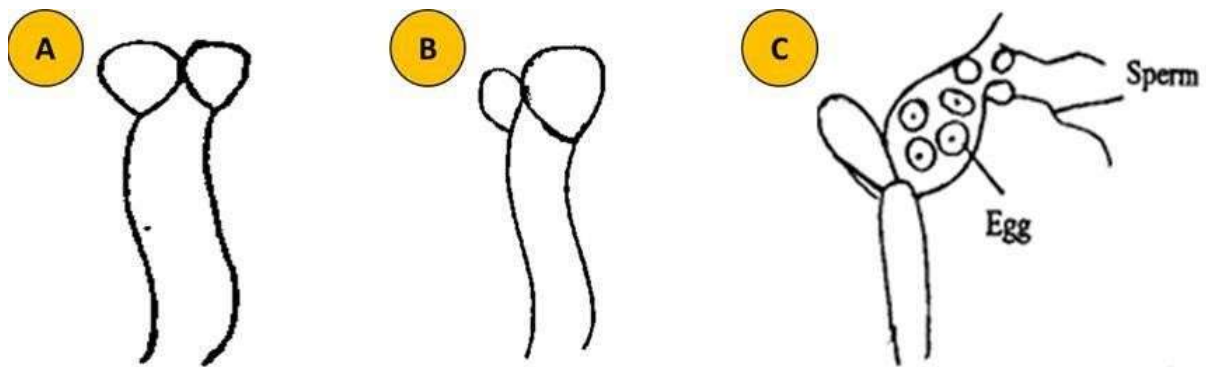
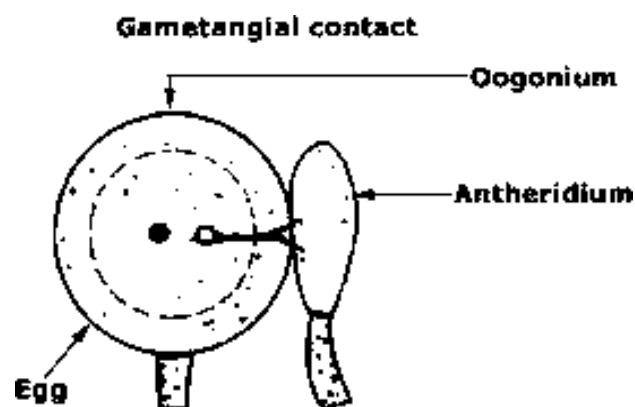


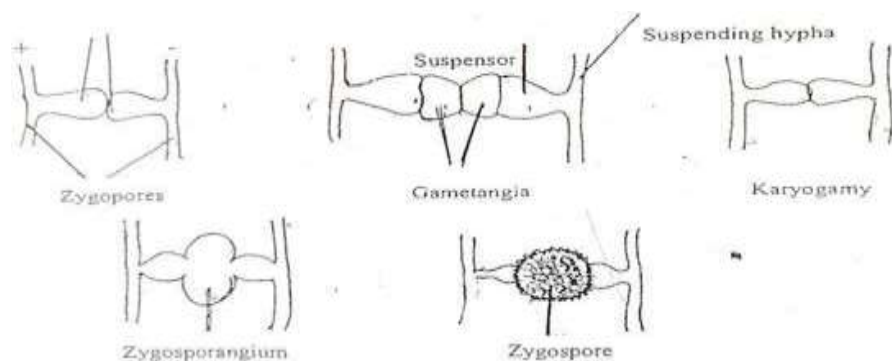
Fig. Planogametic copulation (gametogamy): A) Isogamy B) Anisogamy

C) Heterogamy

2. Gametangial contact (gametangy/oogamy): Male and female gametangia come in contact. At the place of contact, dissolution of wall occurs and a fertilization tube is formed. The contents of male gametangium migrate into female gametangium through a pore or fertilization tube developed at the point of contact. The gametangia do not lose their identity. e.g. *Pythium aphanidermatum*



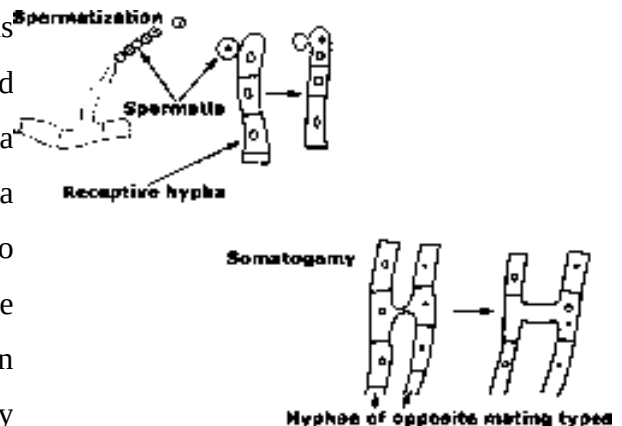
3. Gametangial copulation (gametangiogamy): The isogametangia come in contact, their intervening wall dissolves leading to fusion of entire contents of two contacting gametangia to form a single unit. Gametangia lose their identity. The protoplasts fuse and the unit increases in size. e.g. *Rhizopus stolonifer*.



4. Spermatization: Minute, uninucleate male cells called as spermatia which are produced on spermatophores in a fruiting body (pycnium) are carried to female reproductive structures called receptive hyphae. Spermatia and receptive hyphae come in contact and contents of male spermatium migrate into female receptive hypha, thus making the cell binucleate. This process is called dikaryotization. e.g. *Puccinia graminis tritici*.

5. Somatogamy: Many higher fungi do not produce sex organs. In such cases somatogamy takes place. It is the union of 2 somatic hyphae or somatic cells representing opposite sexes to form sexual spores. e.g. *Agaricus campestri*

Parasexual cycle / Parasexuality: Parasexual cycle is a process in which plasmogamy, karyogamy and haploidisation (non meiotic process) takes place in a sequence but not at specified points in the life cycle of a fungus. It was first discovered in 1952 by Pontecorvo and Roper in *Aspergillus nidulans*, the imperfect stage of *Emericella nidulans*. It is of importance in heterokaryotic fungi (a fungi in which genetically different nuclei are associated in the same protoplast or mycelium). This is one of the methods of producing variability of fungal pathogens. In majority of Deuteromycotina, true sexual cycle is absent but derive many of the benefits of sexuality through this cycle.



Different types of sexual spores: Sexual spores are formed after meiosis, hence also called meiospores.

- 1.Oospores 2.Zygospores
- 3.Ascospores 4.Basidiospores

1. Oospore: A thick walled sexual resting spore produced by the union of two morphologically different gametangia. e.g. *Pythium*, *Phytophthora*, members of class Oomycetes.

2. Zygospore: A thick walled sexual resting spore produced by the fusion of two morphologically similar gametangia. e.g. *Rhizopus*, members of sub-division Zygomycotina.

3. Ascospore: Sexual spore produced in a specialized sac like structure known as ascus. Generally 8 ascospores are formed. e.g. *Erysiphe*, members of sub- division Ascomycotina.

4. Basidiospore: Sexual spore produced on a club shaped structure known as basidium. Generally 4 basidiospores are formed. e.g. *Puccinia*, members of sub- division Basidiomycotina.

Various Life cycle patterns displayed by fungi:

1. Haplobiontic life cycle

2. Diplobiontic life cycle

1. Haplobiontic life cycle: If there is only one free living thallus, which is haploid or diploid in life cycle of a fungus, it is called as haplobiontic life cycle. (long haploid somatic phase and short diploid phase confined to zygote cell, which undergoes meiosis immediately after karyogamy and develop ascospores) e.g. *Schizosaccharomyces octosporus*.

2. Diplobiontic life cycle: If haploid thallus alternates with a diploid thallus, the life cycle is called diplobiontic life cycle, which has a long diploid somatic phase and a very short haploid phase. e.g. *Saccharomyces ludwigii*.

Bioconversion of Agricultural and Food Waste into Bioplastics: Strategies, Challenges, and Future Perspectives

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Abstract:

The growing demand for sustainable materials as an alternative to traditional plastic has driven a significant research interest in biodegradable plastics from agricultural and food waste, which also an urgent environmental concern to take a look it. As such will simultaneously benefit both area like bio-waste act as valuable feedstock rich in polysaccharides, lignin and proteins, serve as low-cost carbon sources for the production of biopolymers such as starch-based plastics, polyhydroxyalkanoates (PHAs), and polylactic acid (PLA). This chapter explore about the conventional plastic, agricultural and fruit waste generation and about the conversion of lignocellulosic and starch-based residues into bioplastics. The encounters of scaling up, cost reduction, and waste heterogeneity are discussed alongside future opportunities such as synthetic biology, nanobiocomposites, and circular bioeconomy integration.

Keywords: Bioplastics, agricultural waste, food waste, cellulose, starch

Introduction:

It is indeed, plastic has become a part of life. It is very difficult to overcome the use of plastic in our day-to-day activities. Hence, a solution to this problem is essential to protect our environment and that is bioplastic extracted from plants, bacteria and fungi as an alternative to the traditional plastics [1].

Conventional Plastics: Production, Usage, and Environmental Challenges

In modern life, plastics have become an indispensable part in every industrial sector from packaging and construction to electronics, textiles, and healthcare (Fig 1). Since their introduction in the mid-20th century, they are driven exponential growth in global plastic production due to their low cost, versatility, and durability. However, this very success has created a persistent environmental challenge due to plastics' resistance to degradation, leading to the accumulation of waste in landfills and natural ecosystems.

According to global data from 2022, the plastic industry was valued at over USD 580 billion, with approximately 367 million metric tons of plastic produced annually worldwide. Despite a slight reduction in output during the COVID-19 pandemic, the long-term trend shows rapid growth: plastic production increased by nearly 32% between 2009 and 2019, adding over 118 million metric tons to the global output [3]. Cumulatively, humanity has produced more than 8.7 billion tons of plastic since the 1950s.

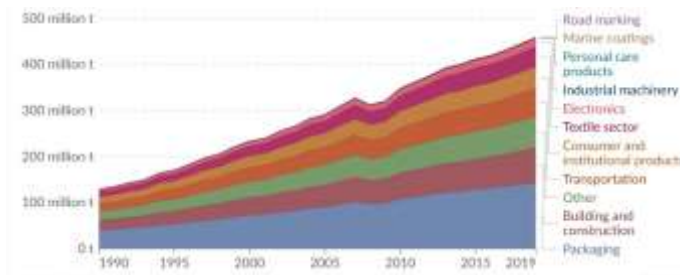


Fig 1: Data showing usage of plastic in different sector.[4]

Global Distribution of Plastic Production

The majority of the world's plastics originate from a few key regions:

- China dominates the global plastic market, contributing around 32% of total production, exporting nearly 14 million tons annually.
- North America (NAFTA region) contributes about 19%, with the United States alone selling 49 million tons of thermoplastic resin in 2020.
- Europe accounts for approximately 16–18% of total production, primarily in packaging and construction applications [5,6].

Within the United States, plastic manufacturing ranks as the 8th largest industrial sector, employing over 1 million people, with production hubs concentrated in states such as California, Michigan, Texas, and Ohio [7].

Moreover, the plastic industry is heavily driven by consumer packaging, which alone accounts for 35% of global plastic use, followed by fig2.

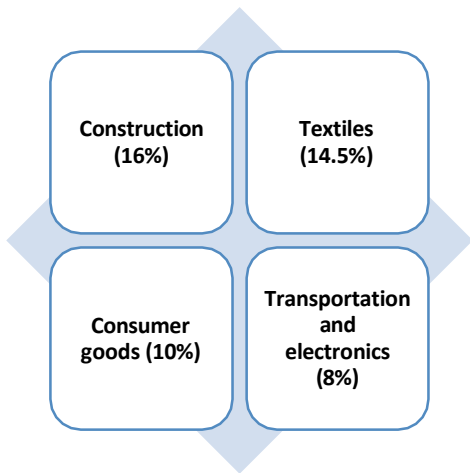


Fig 2: Industrial Sectors and Plastic Utilization

These applications exploit plastics’ flexibility, low density, and resilience to moisture and corrosion. However, most of these products used in packaging materials are single-use, discarded after brief utilization periods, leading to enormous waste volumes.

Common Types of Plastics and Their Properties

Modern plastics are polymers synthesized from petroleum-based hydrocarbons. The Society of the Plastics Industry (SPI) classifies them into seven major categories (Table 1). While each plastic type has specific mechanical advantages—such as PET’s barrier properties or HDPE’s rigidity—most are derived from non-renewable fossil resources and exhibit low recyclability due to composite structures and additive complexity [8-11].

Table 1: Common Types of Plastics and Their Properties

SPI Code	Plastic Type	Common Uses	Global Production (approx.)	Recycling Rate (US)
1	PET (Polyethylene Terephthalate)	Beverage bottles, food packaging	33 million tons/year	29%
2	HDPE (High-Density Polyethylene)	Milk jugs, shampoo bottles, pipes	52 million tons/year	30–35%
3	PVC (Polyvinyl Chloride)	Pipes, cables, windows, medical tubing	38 million tons/year	<1%
4	LDPE (Low-Density Polyethylene)	Plastic bags, films, wraps	64 million tons/year	<10%
5	PP (Polypropylene)	Caps, straws, textiles, auto parts	68 million tons/year	~3%
6	PS (Polystyrene)	Cups, trays, insulation, packaging foam	25 million tons/year	<1%
7	Other (Epoxy, Polycarbonate, Nylon)	Resins, composites, engineering plastics	55 million tons/year (US resins)	Variable

Global distribution and end-use sectors of major plastic types

Environmental Impact and Recycling Challenges

Globally, plastic waste generation has reached 400 million tons per year, with only **9%** effectively recycled. In the United States, the Environmental Protection Agency (EPA) reported that out of 30 million tons of plastic waste generated in 2018, nearly **70%** ended up in landfills, and only 3 million tons were recycled [8-11]. The rest entered the natural environment, contributing to severe pollution problems.

Plastic waste persists for hundreds of years, fragmenting into microplastics (<5 mm) that infiltrate soils, oceans, and food chains. Studies have revealed microplastic particles inside marine organisms, coral tissues, and even human blood, indicating a global contamination crisis. Beyond the ecological risks, recycling remains economically challenging. Sorting and processing mixed plastics is energy-intensive, and the market value of recycled polymers is often lower than that of virgin materials. Consequently, industries often favour new plastic production over recycling, perpetuating the waste cycle.

Energy Consumption and Carbon Footprint

Producing 1 kilogram of plastic requires between 62 and 108 megajoules (MJ) of energy, depending on the polymer type, with silicone-based plastics consuming up to 235 MJ/kg. This high energy demand contributes substantially to greenhouse gas emissions. The cumulative carbon footprint of global plastic production is estimated at 1.8 billion tons of CO₂ annually, nearly 4% of total global [12].

The Path towards Sustainability

In recent years, global awareness has accelerated efforts to mitigate plastic pollution through:

- Bans on single-use plastics (e.g., straws, bags, microbeads);
- Extended producer responsibility (EPR) policies;
- Improved recycling infrastructure and chemical recycling innovations;
- Development of bio-based and biodegradable plastics.

The global plastic recycling market was valued at USD 44.3 billion in 2022 and is projected to reach USD 76.5 billion by 2028, reflecting growing industrial investment and consumer demand for greener alternatives. [13]. However, recycling alone cannot offset the environmental load of traditional plastics. Thus, the focus has shifted toward bioplastics, which utilize renewable biomass feedstocks and offer inherent biodegradability. These sustainable materials have the potential to revolutionize packaging, agriculture, and biomedical industries by aligning with circular bioeconomy principles.

Transition Section: From Plastics to Bioplastics

Conventional plastics have undoubtedly transformed modern life but at the cost of environmental sustainability. Their long degradation times, low recycling rates, and fossil dependency underscore the urgent need for renewable alternatives. Bioplastics, derived from agricultural and food waste, offer a viable solution for all the problems.

Bioplastic

Bioplastics can be broadly categorized based on their origin (bio-based or fossil-based) and degradability (biodegradable or non-biodegradable). These categories determine their chemical composition, environmental impact, and potential applications. Bio-based plastics are derived from renewable biomass such as corn, sugarcane, and vegetable fats, whereas fossil-based plastics rely on petroleum-derived sources. Among these, biodegradable variants like polylactic acid (PLA) and polyhydroxyalkanoates (PHA) have gained significant attention due to their environmental benefits. The classification and applications of various types of bioplastics are summarized in Table 2[14].

Table 2: Classification, feedstocks, and applications of bioplastics based on origin and biodegradability

Type	Feedstock / Source	Major Polymers	Global Production Capacity (%)	Applications
Bio-based, Non-biodegradable	Corn, sugar beet, starch crops	Bio-PE, Bio-PET, Bio-PA (polyamide), Bio-PTT	47.9% total (Bio-PE 12.3%, Bio-PET 2.2%, Bio-PA 18.3%, Bio-PTT 13.5%)	Food packaging, cosmetics, automotive components, toys
Bio-based, Biodegradable	Corn, sugarcane, rice straw, animal fat waste, vegetable fats	Polylactic acid (PLA), Polyhydroxyalkanoates (PHA), Starch blends	49.6% total (PLA 31%, PHA 4.8%, Starch blends 13.8%)	Food packaging, agriculture, biomedical implants, automotive applications
Fossil-based, Biodegradable	Aromatic and aliphatic polyester precursors	Polybutylene adipate terephthalate (PBAT), Polycaprolactone (PCL), Polybutylene succinate (PBS)	5.5% total (PBAT 4.6%, PCL 0.9%)	Packaging, courier bags, cutlery, hygiene products, biomedical applications, industrial composting
Fossil-based, Non-biodegradable	Petroleum derivatives	Polyethylene (PE), Polypropylene (PP), Polystyrene (PS), Polycarbonate (PC), Polyvinyl chloride (PVC), PET	47.9% total	Packaging, construction, automotive, electronics, healthcare

Agricultural and Food Waste as a Biopolymer Source

Agricultural and food waste comprises crop residues, fruit and vegetable peels, husks, shells, and by-products from processing industries. According to the FAO, nearly 1.3 billion tons of food waste is generated globally each year. These wastes, rich in carbohydrates and lignocellulosic material, can serve as feedstock for polymer extraction or microbial fermentation [31].

Table 3: summarize bioplastics and their sources [15-31]

Aspect	Details
Bioplastic Classes	- From biomass and natural resources - Polymers produced by microbes via fermentation of agri-products - Polymers polymerized from oligomers or monomers obtained by fermentation - Polymers derived from fossil fuels
Alternative Raw Materials	Starch, cellulose, wood, sugar, general biomass (used to substitute fossil fuels in bioplastics)
Global Agricultural Waste	Approx. 933 million tonnes produced annually globally; major contributors include agricultural activities, food processing, alcohol, logging, sugar industries. Much waste currently untreated or disposed by incineration/landfilling.
Polysaccharides for Bioplastics	Starch, cellulose, chitin, pectin widely used in bioplastic synthesis. Extraction, processing, application, and disposal differ based on biomass type, plant species, age, etc., affecting polymer composition.
Cellulose Details	Most abundant biopolymer from wood, cotton, lignocellulosic waste. Linear β -D-glucopyranose units (β -1-4 linkages). Crystalline, insoluble, resistant to chemical/mechanical degradation. Extraction must preserve cellulose while removing pectin, hemicellulose, lignin. Examples: Mango leaves, mangosteen peel CNC fillers, Parthenium hysterophorus cellulose acetate films. Often combined with other fibers (e.g. cocoa pod, bagasse).
Starch Details	Biodegradable polysaccharide of amylose and amylopectin. Second largest biomass. Used as filler or main polymer but requires plasticizers (glycerol, sorbitol, etc.) for thermoplasticity. Thermoplastic starch (TPS) has biodegradability and similar transparency to conventional plastics, though mechanical strength varies with amylose content. Reinforcements: dialdehyde lignocellulose, cellulose nanocrystals.
Lignin Details	Found in woody plants' cell walls, slow degradation (200–500 °C). Used as polymer matrix reinforcement to reduce cost, improve UV resistance, water absorption, mechanical stability due to antioxidant effects. Applications include composites with polyethylene-vinyl acetate; improved impact/thermal resistance in PLA-based composites; lignin nanoparticles enhance mechanical properties.
Chitin/Chitosan Details	From fungal cell walls and arthropod exoskeletons. Chitosan used to enhance antimicrobial properties, improve biodegradation. Combined with PLA or starch matrices, improves mechanical and antimicrobial properties though

	may reduce tensile strength. Can be formulated with bioactive agents and nanoparticles for edible and active packaging applications.
Edible Bioplastics	Composed of cellulose, starch, dextran, inulin, alginate, carrageenan, pectin, chitosan, mucilage. These bio-moieties enable films with mechanical strength, oxygen barriers, and appealing appearance/scent. Bioactive additives (antioxidants, antimicrobials) improve shelf life and functionality. Protein- based bioplastics from albumin, soy, whey add antibacterial benefits.
Sustainability Considerations	Agro-industrial waste use offers biodegradability benefits and reduces pollution but processing may cause untreated waste. Extraction methods require energy; land use changes, food resource competition, high production costs, water demand, and fertilizer use are environmental concerns. Thus, agro waste bioplastics are promising but not a standalone solution for replacing conventional plastics fully.

Bioconversion of Agricultural and Food Waste into Bioplastics

The growing accumulation of agricultural residues and food waste has emerged as a global environmental challenge. Bioconversion provides an eco-friendly strategy to transform these wastes into value added products such as bioplastics, thereby contributing to the circular bio-economy. This sustainable approach utilizes microbial, enzymatic, and physicochemical processes to convert organic matter rich in carbohydrates, lipids, and proteins into biodegradable polymers such as polylactic acid (PLA), polyhydroxyalkanoates (PHAs), and starch-based bioplastics [32-36].

2. Feedstock Selection and Pre-treatment

Agricultural and food wastes such as corn, sugarcane bagasse, rice husk, fruit peels, potato starch residues, and whey are rich in cellulose, hemicellulose, starch, and sugars, making them suitable carbon sources for microbial fermentation. However, these complex polymers require pre-treatment to release fermentable sugars.

Common pre-treatment methods include [32]:

- Physical methods: milling, extrusion, and steam explosion to break lignocellulosic structure.
- Chemical methods: acid or alkaline hydrolysis to solubilize hemicellulose and lignin.
- Enzymatic hydrolysis: cellulases, amylases, and pectinases convert polysaccharides into glucose, fructose, and xylose.

For example, rice straw and sugarcane bagasse hydrolysates are widely used for lactic acid and PHA production due to their high sugar yield and minimal inhibitor formation.

3. Microbial Fermentation and Polymer Formation

Once hydrolyzed, the released simple sugars and fatty acids serve as substrates for microbial fermentation [33, 34]. The process involves two main stages:

- a. Fermentation: Microorganisms such as *Lactobacillus sp.*, *Cupriavidus necator*, and *Bacillus*

megaterium metabolize sugars into intermediates like lactic acid, hydroxyalkanoic acids, or succinic acid.

- b. Polymerization: These intermediates are then polymerized either biologically (intracellularly) or chemically (via condensation reactions) to form biodegradable polymers.

Bioplastic pathway:

1. Polylactic Acid (PLA):

- Produced by fermenting sugars into lactic acid using *Lactobacillus delbrueckii*.
- Lactic acid undergoes condensation polymerization or ring-opening polymerization (ROP) to yield PLA [35].
- PLA is widely used in packaging, biomedical devices, and textiles.

2. Polyhydroxyalkanoates (PHAs):

- Synthesized intracellularly by bacteria such as *Cupriavidus necator* and *Pseudomonas putida* under nutrient-limited but carbon-rich conditions.
- Agricultural oils, molasses, and food waste serve as carbon sources.
- PHAs are thermoplastic, biodegradable, and biocompatible, ideal for biomedical and agricultural films [35].

3. Starch and Protein-Based Bioplastics [35]:

- Derived directly from food waste rich in starch (e.g., potato, cassava) or proteins (e.g., soybean, whey).
- Modified through plasticization (using glycerol or sorbitol) and blending with natural fibers or biopolymers to improve flexibility and water resistance.

4. Downstream Processing and Purification

After fermentation, bioplastics are extracted and purified through:

- Cell lysis (chemical or enzymatic) to release intracellular PHAs.
- Solvent extraction and precipitation for polymer recovery.
- Drying and pelletization, followed by extrusion or molding into films, coatings, or composites.

The choice of purification process affects yield, cost, and polymer properties. Sustainable alternatives such as supercritical CO₂ extraction and solvent-free recovery are being explored to reduce environmental impact [35].

5. Advantages of Bioconversion Approach

- Sustainability: Utilizes renewable waste biomass, reducing dependence on fossil resources.
- Waste valorization: Converts low-value residues into high-value materials.
- Carbon neutrality: Reduces greenhouse gas emissions through closed-loop resource cycles.
- Scalability potential: Integrates into existing agro-industrial systems, enabling decentralized

production [35].

Processing of Bioplastics and Biocomposites

The development of bioplastics and biocomposites relies heavily on identifying and optimizing suitable processing methods that ensure high-quality material performance. Unlike conventional petroleum-based plastics, bioplastics often have limitations such as lower thermal stability, reduced mechanical strength, and sensitivity to processing

conditions. Therefore, continuous innovation in their processing techniques is essential to improve their functionality and scalability [37-43].

3. Casting Techniques

Among laboratory-scale methods, solution casting is the most widely adopted for research due to its simplicity, low cost, and ability to produce uniform films with desirable mechanical and thermal properties. In this process, biopolymers such as starch, gelatin, or soy protein are dissolved or dispersed in a solvent, plasticized (often with glycerol or vegetable oils), and cast into thin films [37-39, 14].

- Example: Edible cassava-based bioplastics have been produced using glycerol and modified vegetable oils to enhance flexibility and water resistance.
- Similarly, gelatin–corn starch–papaya composites have been developed for food packaging, demonstrating good biodegradability. However, the casting method is not easily scalable since it requires large volumes of solvent and energy for solvent recovery. Furthermore, as a batch process, it may produce variations in film uniformity.

4. Conventional Plastic Processing Techniques

To industrialize bioplastic production, established plastic manufacturing technologies have been adapted [37-39, 14]. These include:

- Extrusion: Widely used due to its high productivity and cost-effectiveness. It is suitable for producing continuous products such as films and filaments but has drawbacks like uneven raw material distribution and high maintenance costs.
- Compression Molding: Suitable for high-strength bioplastics with advantages like low cost, dimensional accuracy, and short cycle times. However, it has longer processing durations and limited design flexibility.

- **Injection Molding:** The most efficient for high-volume production, offering precision, reproducibility, and adaptability to complex shapes. It is also compatible with biodegradable materials such as PLA (polylactic acid) and PBS (polybutylene-succinate). These techniques are essential to transition bioplastics from laboratory to industrial scale, minimizing the need for new equipment investments while maintaining productivity and quality standards.

5. Challenges in Processing Bioplastics

Processing bio-based polymers introduces issues such as rheological instability, polymer degradation, and thermal instability. For instance, PLA is prone to chain scission at high temperatures, reducing molecular weight and melt strength, leading to brittle or defective parts. The Melt Flow Index (MFI) is often used to monitor processability of higher MFI indicates lower viscosity and easier flow, but excessive degradation can lower product quality. To overcome these challenges, strategies such as:

- Addition of chain extenders (e.g., maleic anhydride, epoxidized additives) to enhance molecular weight,
- Copolymerization and blending (e.g., PLA with PBSA or natural rubber to improve toughness), and
- Reinforcement with natural fibers or nanoparticles are being implemented.

6. Natural Fiber-Reinforced Biocomposites

Reinforcing bioplastics with natural fibers (e.g., cellulose, rice husk, agave bagasse, or chitosan) improves their strength, stiffness, and thermal stability. Processing typically involves [14,40-42]:

4. Extrusion compounding to disperse fibers, followed by
5. Molding (injection or compression) to form final products. Pretreatment of fibers to remove moisture or enhance surface compatibility is critical to avoid degradation and improve interfacial bonding. Studies show that cellulose nanofiber (CNF) and microcrystalline cellulose (MCC) reinforcements significantly increase tensile strength and modulus while maintaining biodegradability.

7. Additive Manufacturing (3D Printing)

Recently, fused deposition modeling (FDM) using PLA-based filaments has gained attention for producing customized bioplastic components. FDM is advantageous due to low material waste,

flexibility, and reduced environmental footprint. Reinforcement with agricultural waste fibers (e.g., coir, hemp, or rice husk) enhances mechanical strength and biodegradability. Optimization of printing parameters (temperature, infill density, and layer orientation) further improves structural performance and stability [14, 43].

Role of Nanotechnology in Bioplastics

Nanotechnology has emerged as a powerful tool in improving the structural and functional properties of bioplastics derived from agricultural and food waste. The incorporation of nanomaterials such as nanocellulose, nanoclay, graphene oxide, and metal oxide nanoparticles (ZnO, TiO₂, Ag, CuO) significantly enhances the mechanical strength, thermal stability, and barrier properties of biopolymer matrices like PLA, starch, and PHA. These nanoparticles form strong interfacial interactions with polymer chains, reducing gas and moisture permeability while increasing flexibility and durability. In addition, metallic nanoparticles impart antimicrobial and UV-resistant characteristics, making nanobioplastics suitable for food packaging and biomedical applications. Nanocellulose and lignin nanoparticles, often derived from agricultural residues, also promote biodegradability and sustainability, reinforcing the eco-friendly nature of these materials. Furthermore, nanotechnology enables the development of smart bioplastics that can respond to environmental stimuli or indicate spoilage, expanding their functionality. Overall, the integration of nanotechnology into bioplastic production bridges the performance gap between biodegradable and petroleum-based plastics, paving the way for next-generation sustainable materials with enhanced efficiency, safety, and versatility [44-46].

Table 4: Usage of nanomaterials in the bioplastic production

Nanomaterial	Polymer Matrix	Improvement Observed	Applications
Nanocellulose	Starch, PLA, PHA	Enhanced tensile strength, flexibility	Food packaging, biodegradable films
Nanoclay (MMT)	PLA, PCL, starch	Better barrier and mechanical strength	Packaging, coatings
Silver nanoparticles (AgNPs)	Chitosan, PLA	Antimicrobial activity, surface stability	Medical and food packaging
ZnO / TiO ₂ nanoparticles	Starch, PHA	UV resistance, thermal stability	Agricultural films, medical materials
Graphene oxide	PLA, PBAT	Improved conductivity, barrier effect	Smart sensors, electronic packaging
Lignin nanoparticles	Starch, PHB	Increased antioxidant capacity	Active packaging
SiO ₂ nanoparticles	PLA, PBS	Surface hardness, optical clarity	Biodegradable utensils, containers

Characterization of Bioplastics

The characterization of bioplastics is an essential step in understanding their structural, thermal, mechanical, and functional properties, which determine their suitability for various applications such as packaging, biomedical use, and agriculture. Since bioplastics are derived from renewable sources like agricultural and food waste, their properties depend largely on the type of feedstock, the polymerization process, and the inclusion of plasticizers or reinforcing agents [46-56]. A comprehensive characterization ensures the quality, performance, and biodegradability of the final product (Table 5).

Table 5: Characterization techniques for bioplastics.

Characterization Technique	Purpose / Measurement	Key Findings / Interpretation	Typical Observation in Bioplastics
Scanning Electron Microscopy (SEM)[46-56]	Examines surface morphology and microstructure	Identifies cracks, roughness, pores, and filler dispersion	Smooth surfaces for pure polymers; rough/irregular for fiber- or nanoparticle-reinforced bioplastics
Atomic Force Microscopy (AFM)[46-56]	Studies nanoscale topography and roughness	Determines uniformity, phase separation, and coating quality	Surface roughness increases with addition of cellulose or starch fillers
Fourier Transform Infrared Spectroscopy (FTIR)[46-56]	Identifies functional groups and bonding patterns	Detects O–H, C=O, and C–O–C vibrations confirming polymerization and crosslinking	Broad O–H stretch ($\sim 3300\text{ cm}^{-1}$); C=O at $1700\text{--}1750\text{ cm}^{-1}$ indicating esters or carboxylic acids
X-Ray Diffraction (XRD)[46-56]	Determines crystalline vs. amorphous structure	Evaluates crystallinity index (CI) and polymer ordering	PLA, PHB show semi-crystalline peaks; starch-based films mostly amorphous
Nuclear Magnetic Resonance (NMR)[46-56]	Confirms chemical composition and monomer arrangement	Detects carbon and hydrogen environments in the polymer backbone	Distinct ^1H and ^{13}C peaks confirm successful polymerization and copolymer formation
Differential Scanning Calorimetry (DSC)[46-56]	Measures melting (T_m), glass transition (T_g), and crystallization (T_c) temperatures	Determines thermal transitions and stability	PLA: $T_g \sim 55\text{--}65^\circ\text{C}$; starch-based bioplastics $T_g \sim 40^\circ\text{C}$; additives modify crystallinity
Thermogravimetric Analysis (TGA)[46-56]	Evaluates thermal degradation profile	Determines degradation temperature and residue percentage	Decomposition stages: moisture loss ($50\text{--}150^\circ\text{C}$), polymer degradation ($200\text{--}350^\circ\text{C}$)

Dynamic Mechanical Analysis (DMA)[46-56]	Measures viscoelastic behavior (E' , E'' , $\tan \delta$)	Evaluates stiffness and temperature-dependent elasticity	Increased storage modulus with fiber/nanoparticle reinforcement
Tensile Testing (UTM) [46-56]	Assesses mechanical strength and elongation	Determines tensile strength, elasticity, and flexibility	PLA: high strength, low elongation; starch/glycerol films: low strength, high elongation
Young's Modulus Measurement [46-56]	Evaluates stiffness and rigidity	High modulus = rigid structure; low modulus = flexible	Reinforcement increases modulus; plasticizers reduce it
Water Vapor Transmission Rate (WVTR) [46-56]	Measures water barrier efficiency	Assesses permeability in packaging applications	Low WVTR = better barrier; hydrophobic additives improve resistance
Oxygen Permeability Test [46-56]	Evaluates gas barrier properties	Determines suitability for food packaging	Lower oxygen permeability indicates longer shelf life
Biodegradation/Composability Tests [46-56]	Determines environmental degradation behaviour	Monitors weight loss, CO_2 evolution, and microbial action	Bioplastics degrade within 30–90 days depending on composition
UV–Visible Spectroscopy (Transparency)[46-56]	Measures light transmittance and opacity	Evaluates film clarity and UV protection ability	High transparency (>80%) for neat films; reduced clarity with fillers

Conclusion and Future Perspectives

Numerous global dependence on conventional plastic has resulted in unembellished environmental challenges and increases the carbon food print. It is the time of thinking of an alternate solution for the conventional plastic though it is cheap and alternate that is bioplastics from different biological sources are on the expensive side. However, biodegradable polymers such as PLA, PHA, and starch-based composites is a sustainable alternative and methodology of converting such as microbial fermentation, enzymatic hydrolysis, and nanotechnology incorporation in the bioplastic production can reduce waste accumulation but also contribute to a circular bio-economy.

Moreover, the future of bioplastic are in the hands of biotechnology, nanoscience and artificial intelligence to create smarter, more efficient production system to track and monitor and alter the degradation properties of bioplastics.

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STEM CELLS AND REGENERATIVE MEDICINE: ADVANCES AND APPLICATIONS

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Abstract

Stem cells are unique cells that can restore the structure and function of damaged tissues, developing foundation of regenerative medicines. Embryonic, adult, induced pluripotent stem cells have notable therapeutic potential through self-renewal and controlled differentiation. Tissue engineering integrates stem cells with complex molecular signalling mechanisms and specialized niche environments that coordinate repair of damaged tissues. Current advances in tissue engineering such as biomimetic scaffolds, 3D bioprinting, and organoid systems, give new opportunity in personalized therapies for neurological, cardiovascular, musculoskeletal, and cutaneous disorders. Although persistent challenges in cell survival, immune responses, and safety, novel technologies offer safer and durable outcomes.

Keywords: Stem cells, Regenerative medicine, Tissue engineering, Molecular signalling, Personalized therapies.

1. Introduction

Stem cells are unique cells present in various tissues and organs of the body throughout every developmental stage, starting from an early embryonic growth to adulthood capable of undergoing specialized and developmental process. These cells are undifferentiated cells with the distinctive ability to self-renew, arise clonally from a single cell, and differentiate into specialized cell types. Their potency varies upon their origin: pluripotent stem cells such as embryonic stem cells derived from the inner cell mass of the blastocyst and induced pluripotent stem cells generated by the reprogramming of somatic cells. Multipotent stem cells include mesenchymal stem cells, which form a restricted range of cell type and oligopotent tissue-resident stem cells are known to drive the renewal of specific organ systems. Due to the tissue-regenerating and repairing capabilities of stem cells, they form the foundation of regenerative medicine, which is rapidly advancing to restore or replace damaged cells, tissues and organs. Their application in cell-based therapies, tissue engineering, and disease modelling heralds new therapeutic strategies for degenerative and chronic ailments (Appasani & Appasani, 2010; Kolios & Moodley, 2012).

2. Types and Sources of Stem Cells

Stem cells are undifferentiated cells with properties of self-renewal and lineage-specific differentiation. They are divided into embryonic stem cells, adult (somatic) stem cells, and induced pluripotent stem cells based on their origin and potency and each possessing distinctive advantages and translational barriers (Kolios & Moodley, 2012). ESCs are derived from the inner cell mass of the blastocyst and have full pluripotency, giving rise to all three major germ layers. It has a strong ability to differentiate and have been demonstrated that ESCs can proliferate into neural and cardiac lineages with functional integration in preclinical models. However, the derivation of ESCs necessitates the destruction of embryos, which significantly raises ethical and regulatory concerns (Trounson, 2002).

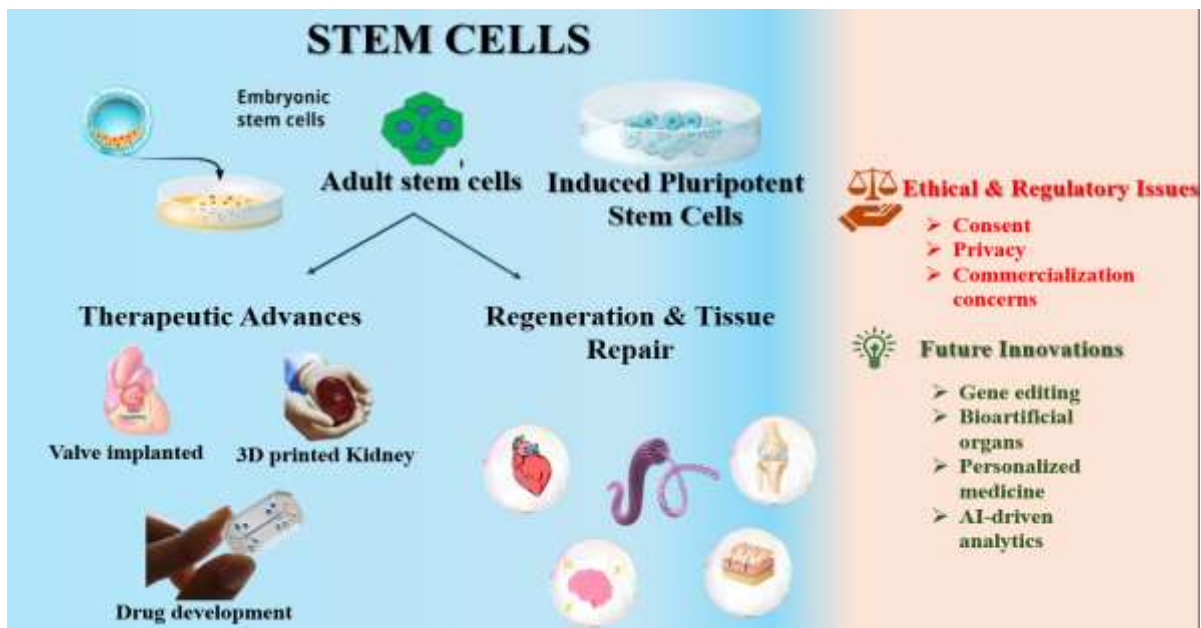


Figure 1: Stem cells combined with tissue engineering enable target tissue repair and effective regenerative therapy

Adult stem cells represent multipotent progenitors that reside in tissue, including bone marrow, skin, and muscle, and sustain homeostasis and repair. Their potential to differentiate is usually limited to cell types of their resident organ, and their number and regenerative efficacy decrease with age. In any case, adult stem cells such as hematopoietic and mesenchymal stem cells, have found widespread applications in clinical therapy due to fewer ethical concerns and high biocompatibility in autologous settings (Gurusamy, 2018; Kolios & Moodley, 2012). Induced pluripotent stem cells (iPSCs) are somatic cells that have regained pluripotent properties by the expression of transcription factors. iPSCs generally bear a close resemblance to ESCs in self-renewal and differentiating capacities without destroying embryos, which makes them an attractive cell source for personalized regenerative therapies, disease modelling, and drug development. iPSCs introduce newer challenges in respect to genetic privacy, consent, and governance of gene-edited cell products (Lo & Parham, 2009; Walia et al., 2012).

3. Cellular and Molecular Basis of Regeneration

Regeneration is a fundamental biological process through which organisms restore the structural and functional integrity of tissues following injury. Coordinated interplay between stem cell activity, molecular signalling networks, and the surrounding tissue microenvironment are driven for tissue regeneration. This is also regulated by epigenetically, transcriptionally, and extracellularly. Additionally activating or suppressing of pluripotency-association genes, histone acetylation and RNA modifications (those mediated by Mettl3-dependent m6A methylation), promote lineage commitment. Furthermore, structural and biochemical signals provided by the surrounding matrix, including heparan sulfate proteoglycans, further favor neural, hepatic, and osteogenic phenotypes. Multiple signalling pathways orchestrate stem cell fate in the process of regeneration. Hedgehog, Wnt/ β -catenin, Notch, and TGF- β pathways maintain a critical balance between self-renewal and differentiation, while aberrant activation of these pathways is closely associated with cancer stem cells' persistence and resistance to therapy. MicroRNAs add another layer of post-transcriptional regulation, modulating oxidative stress responses and crosstalk between pro-regenerative and oncogenic signalling networks (Tanabe, 2015). The stem cell niche is a specialized anatomical and molecular microenvironment that preserve stem-cells quiescence, survival, and controlled activate them following injury. To orchestrate repair, cellular constituents which include stromal and endothelial cells, niche-specific extracellular matrix, and gradients of nutrients and morphogens-integrate systemic physiological cues. For instance, rapid epithelial renewal in the intestinal crypt contrast with tightly regulated quiescence of hematopoietic or neural stem cells (Morrison & Spradling, 2008). Ultimately, the integration of these molecular signals and niche interaction direct the mechanism of tissue repair and regeneration. Tissue repair and regeneration are highly ordered processes involving hemostasis, inflammation, proliferation, and remodelling, to which immune cells, stromal cells, the extracellular matrix, and resident stem/progenitor cells make critical contributions. Following injury, platelets initiate clot formation and release growth factors that recruit inflammatory cells and activate the local stem cells. Macrophages removing the waste materials while producing cytokines that stimulate stem cell proliferation and differentiation. In the proliferative phase, angiogenesis restores blood supply, and stem cells generate new epithelial and connective tissues to rebuild structure and function. Finally, in the remodelling phase, strengthening and reorganization of the ECM take place to complete the restoration and functional integration of regenerative tissue (Eming, Martin, & Tomic-Canic, 2014).

4. Tissue Engineering and Regenerative Approaches

Tissue engineering integrates biological principles with engineering strategies to develop functional tissue replacements, utilizing three essential components such as stem cells, biocompatible scaffolds, and bioactive growth factors. Cells themselves are the sources of regeneration stem cells, primary cells,

or progenitors. Biodegradable scaffolds are temporary extracellular matrices and provide signal for cell adhesion, proliferation, and matrix deposition. These scaffolds are embedded with growth factors and bioactive molecules that regulate cellular recruitment, lineage commitment, and tissue morphogenesis. Biodegradation and nanoscale signals must be considered in an effective design to stimulate tissue specific regeneration, scaffold pores, stiffness (Howard et al., 2008). Natural polymers (such as collagen, alginate, hyaluronic acid) and synthetic materials (such as PEG, PCL) are combined in bioinks, balancing bioactivity with mechanical stability to support cell viability. Moreover, recent advances in 3D bioprinting have enabled the prompt organization of cells and biomaterials for the fabrication of specific patient. Tissue engineering also helps for personalized disease modelling and testing of drugs by organoid bioprinting and microfluidic organ-on-chip systems (Rawal et al., 2021; Murphy & Atala, 2014). Clinical translation is most advanced in musculoskeletal and skin repair. When grafting is a limited option, mesenchymal stem cells and osteo/chondrogenic growth factors used to create bone and cartilage that have demonstrated enhanced functional integration. In the case of burns and chronic ulcers, bioengineered skin grafts, hydrogel dressings, and stem cell-based therapies are healing through improved vascularization and lowered scar generation. Beyond these tissues, advancement in bioprinted models of liver, kidney, and heart suggest to future potential for whole-organ regeneration. Despite these developments, several significant challenges persist with areas vascularization, immune capability, long-term stability, and regulatory approval in clinical applications. Future innovations in biomaterial science, smart scaffolds, computational modelling, and personalized regenerative techniques will help to accelerate translation toward more effective and durable patient outcomes (Zhou, 2024; Dutra Alves, 2025).

5. Clinical Applications and Therapeutic Advances

Stem cell-based regenerative medicine is a rapidly developing field across clinical disciplines characterized by limited natural repair. For neurological conditions such as Parkinson's disease, stroke, spinal cord injury, and Alzheimer's disease, early-phase trials have shown improved motor functions, mobility and partial recovery of sensation when it is incorporated with stem-cell based regenerative therapy. Alzheimer's diseases studies are aimed at slowing down cognitive decline. Overall, these observations support both neural replacement and neuroprotective effects, through challenges persist regarding long-term integration and controlled differentiation (Nguyen, 2019). In cardiovascular diseases stem-cell based regenerative medicine shows improvements in ventricular function, scar reduction, and exercise tolerance after myocardial infarction and heart failure. For example, in peripheral ischemia, enhanced blood flow to the limb and reduced amputation risk shows therapeutic angiogenesis. However, cell survival and immune compatibility need optimization (Terashvili & Bosnjak, 2019). Orthopedic applications of bone marrow and adipose derived stem cells, either alone

or on scaffolds, report promising healing in case of osteonecrosis, large bone defects, and cartilage lesions. However, evidence mostly from small cohorts, and ongoing trials are required to standardize the efficacy (Im, 2017). Regenerative wound healing strategies, including skin substitutes comprising MSCs and bioengineered grafts, hasten the closure of diabetic and venous ulcers while minimizing scar formation following serious burns, thus favoring wider clinical acceptance (Mahajan, Soker & Murphy, 2024). In transplantation medicine, the infusion of MSC in kidney and liver graft recipients has reduced inflammation and elevated early graft function, while ex-vivo regenerative perfusion seeks rehabilitate donor organ before implantation (Hoogduij et al., 2020). All these case studies and active clinical trials suggest promising functional recovery in neurological, cardiovascular, orthopaedic, dermatologic, and transplant conditions. Continued refinement of delivery systems, biomaterial integration, and long-term safety evaluation remains critical for routine clinical translation.

6. Challenges, Ethics, and Future opportunities

Stem-cell based treatments have a lot of clinical promise, even though still there are some ethical, technological and legal issues to overcome. Ethical disputes continue over the moral status of the embryo in the use of the embryonic stem cells, whereas alternative sources also raise concerns about consent, donor privacy, genetic alteration, and commercialization. There is a need for stringent global regulatory oversight to avoid exploitation and ensure transparent and safe clinical translation in the face of increasing “stem cell tourism” (Lo & Parham, 2009). Therapeutic efficiency is also diminished by poor engraftment, host immunological rejection and chances of mutant genes introduced by in vitro cell. These safety concerns slowly adopted in clinical practice (Leventhal et al., 2011). Emerging technologies will influence regenerative medicine. However, gene editing through different methods like CRISPR and base editing give new ways to correct inherited mutations and develop universal donor cells, but it also raises risks of off-target effects and ethical concerns around heritable modification. Bioartificial organs, with supporting advances in tissue engineering, decellularized scaffolds, and 3D bioprinting, potentially solve issues of organ shortages but still present major challenges in scaling and regulation hurdles (Deneault, 2024; De Bartolo & Mantovani, 2022).

Recently developed personalized medicine precision approaches, such as multi-omics profiling, AI-driven analytics, organoids, and organ-on-chip are focused on enhancing therapeutic response while minimizing adverse side effects and ensuring long-term functional success (Ho et al., 2020). Harmonized ethical governance, improved safety strategies, and equitable access to emerging therapies are the basis of further progress in the futuristic regenerative medicine. Stem cell science is well positioned to go progress from experimental promise to reliable, personalized clinical solutions, however, further interdisciplinary innovations are still required.

7. Conclusion

Stem cell and regenerative medicine are growing advancing fields with the potential to restore injured tissues and treat previously incurable illness. Fig.1 describe about the biology of the stem cells and tissue regeneration has encouraged the development of the bioengineered scaffolds, organoids, and bioprinted tissues, leading to significant clinical advances in neurological, cardiovascular, orthopedic, and transplant therapies. Immune rejection, safety, ethics, and inconsistent regulation, remain major challenges to wide-scale implementation. These barriers are overcome by some promising emerging approaches, such as gene editing, personalized therapy, the production of bioartificial organs and AI driven analytics. Yet, scientific, ethical, and clinical improvement will continue to be crucial for transformational possibilities of regenerative medicine.

Conflict of interests

The authors declare that they have no conflict of interest, financial or otherwise.

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BIOTECHNOLOGICAL ADVANCEMENTS IN AQUACULTURE: ENHANCING PRODUCTIVITY, SUSTAINABILITY AND HEALTH

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Abstract

The use of biotechnological tools in aquaculture aims to ensure that the sector benefits from the application of molecular, genetic, and cellular technologies, thereby enhancing its productivity, sustainability, and, ultimately, its health status. The present chapter offers a brief account of the main fields in which biotechnology and genetics are employed, such as marker-assisted selection, genome editing, and cryopreservation, as well as other biotechnological developments that impact feed, health management, and the rehabilitation of ecosystems. The arrival of CRISPR, probiotics, nutrigenomics, and biofloc technology is already improving disease resistance, feed conversion, and waste treatment. On the whole, due to these biotechnological applications, change has begun, and aquaculture, which used to be operated in isolation, is gradually becoming a science-informed, eco-friendly, and indispensable global food supplier that protects aquatic flora and fauna.

Keywords: Fishery Biotechnology, Genetic Improvement, Nutrigenomics, Aquaculture Sustainability, and Health Management.

1. Introduction Fishery biotechnology means the application of molecular, cellular and genetic tools for the fish's increase in productivity, disease resistance, and sustainability. It includes newer methods of genetic improvement, disease diagnosis, and feed technologies, as well as environmental management (Matthew et al., 2025). The rise in molecular markers, Genomic sequencing, and bioprocess engineering has given a different dimension to the traditionally corrupt aquaculture. By using biotechnology economically, aquaculture could meet the fast-growing global demand for protein while maintaining ecological balance. Biotechnology in fisheries supports not only food security but also conservation of aquatic biodiversity and ecosystem health.

With global aquaculture supplying more than 60% of the world's fish consumption in 2030, this suggests that biotechnology can ensure productivity without harming the environment (Msangi et al., 2013). Emerging alongside AI, nanotechnology, and bioinformatics, precision aquaculture heralds a new era of product lines in which data, biology, and automation converge to craft resilient and sustainable fish systems (Fig. 1).

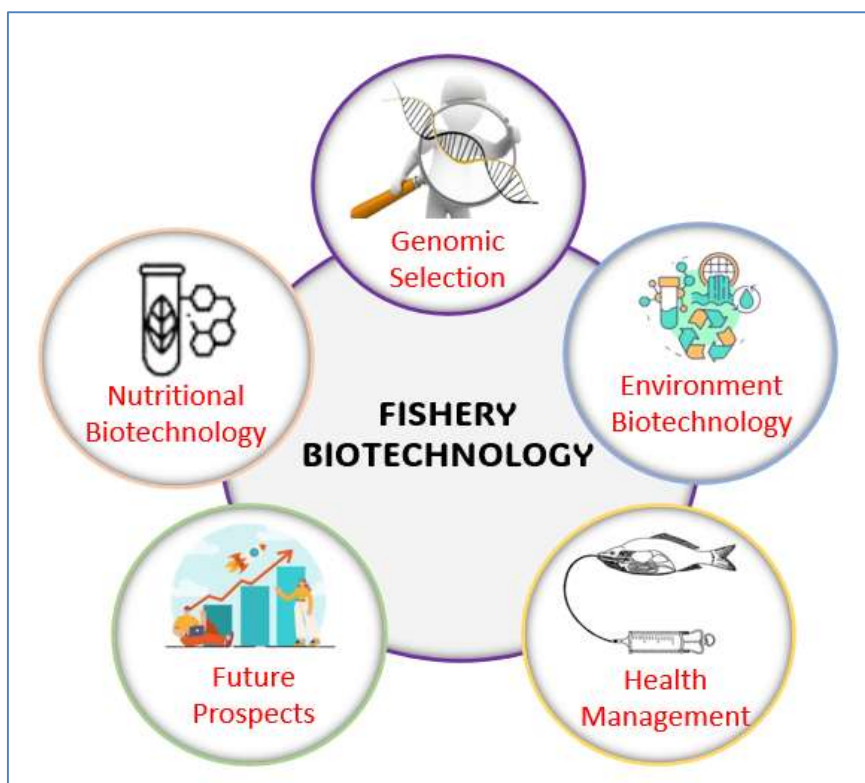


Figure 1. Fishery Biotechnology's mainstays — combining genetic selection, environmental and nutritional advancements, health management, and future outlooks for eco-friendly aquaculture.

2. Genetic Improvement and Breeding Technologies

Genomic selection helps improve growth rate and feed conversion rate, and confers tolerance to stressful stimuli, as molecular tools can be used to modify one or more genetic traits. Using molecular instruments that assess genomic selection increases the accuracy and efficiency of trait selection (Nadeem et al., 2018). These genomic tools can nonetheless be employed in combination with traditional breeding techniques. This method is one of the oldest and most successful procedures for improvement in aquaculture species. Conventional techniques are often inefficient and environmentally detrimental; consequently, systematic selection is employed to accelerate growth, improve feed utilization, or maintain multiple desirable traits (Sonea et al., 2023). Marker-assisted selection contributes to the process via molecular markers linked to quantitative trait loci to identify superior genotypes at an early stage. For example, in Atlantic salmon, QTL for resistance against Infectious Pancreatic Necrosis Virus (IPNV) were incorporated successfully into selection lines, thus drastically reducing mortality. The same MAS principle has been applied to carps and tilapia for growth and salinity adaptation (Buchmann, 2022). Selecting for a given trait using MAS is faster than using phenotypic selection in its direct form. Fish genome sequencing technologies have the ability to create detailed, precise, and accurate maps. Currently, molecular markers such as microsatellites, SNPs, and AFLPs are used in population studies, DNA fingerprinting, and QTL mapping to map

growth rate, feed conversion, and stress tolerance in several species (Sharma et al., 2024). CRISPR-Cas9 and TALENs are the genetic tools that have changed the genetic research in aquaculture significantly. These types of tools help to change the coding regions (introns) of the targeted gene, which will be helpful for economic traits. Such as knocking out myostatin in tilapia using CRISPR, resulting in increased muscle and body development, while catfish undergo target knockouts of reproductive genes to make them sterile, which would also serve as examples (Coogan, 2021).

Transgenic Fish Issues of Ethics

Transgenesis is the introduction of foreign genes to fish genomes to improve growth, temperature resistance, and disease tolerance (Wang et al., 2025). The most famous example is an AquAdvantage®-engineered salmon that grows twice as fast as regular salmon. Public acceptance remains limited because potential risks have mainly driven concerns about biosafety, the environment, and the ethics of applying such technologies (Blix, 2023). Other issues include gene escape in wild populations and its effects on biodiversity. Hence, there needs to be strict regulations and containment arrangements for any commercial-scale prospection of transgenic fish.

Cryopreservation and Germplasm Conservation

Cryogenic storage is also carried out in support of genomic resource banks for selective breeding and restoration programs. Cryoprotectants, such as DMSO and Glycerol, have resulted in successful fertilization of fishlike carp, salmon, and catfish after the thawing process. Many germplasm repositories (Gene banks) are built to preserve the genetic material of the three species and high-quality broodstocks (Aljaser, 2022).

3. Innovative Tools in Biotechnology for Feed and Nutrition

Nutrition is the largest expense of aquaculture, making up 50-70% of the total cost; therefore, the nutritional biotechnology serves as the core of the sustainable production through microbial fermentation, enzyme engineering, feed efficiency, gut health, and environmental sustainability as its support pillars.

Development of Bioengineered and Microbial Protein Sources

Single-cell proteins from yeast, bacteria, and fungi emerge today as sustainable alternatives to fish meal. The bioengineered proteins presented here are highly digestible and, more importantly, maintain a balanced amino acid profile and do not harm the environment. For instance, microbial protein production is presently used for salmonid and shrimp feeding by employing methanotrophic bacteria (*Methylococcus capsulatus*) (Chen et al., 2021). The inclusion of these types of proteins in feed formulations reduces reliance on wild-caught fisheries resources and thus aligns the activities with those of a circular bioeconomy. The antibiotics-assisted gut microbiota in fish was significantly changed after they had received certain prebiotics, such as inulin, which also made them more resistant

to a certain pathogen. The strategy of using beneficial insects and those feeding on organic waste as well as algae could be integrated and generate a system that could ultimately reach full circularity for several nutrients. Enzymes such as phytase, protease, and cellulase are added to feed to eliminate anti-nutritional factors and, thus, increase its utilization by the fish (Kırkpınar et al., 2025). These protein sources, including microalgae such as spirulina and chlorella, along with insect meals like those from black soldier fly larvae, are superior and provide the best type of proteins and omega-3 fatty acids. Insects raised on organic waste are a solution for a permanent protein source and, at the same time, produce colorful biomass containing pigments, antioxidants, and vitamins (Chavez, 2021).

Nutrigenomics and Metabolic Optimization

Nutrigenomics deals with the interaction between diet and genes. A very careful and detailed transcriptomic analysis has revealed the complex ways in which nutrient availability affects fish metabolism and immunity. These data provide a corporate feeding strategy of the species and life stages (Song et al., 2023). Given these studies, the amino acid profile and immune gene expression in stressed tilapia may offer some hope for diet-based stress resistance (Wang et al., 2024).

4. Health Management and Disease Control

Apart from the positive aspects of aquatic health management, there is always an unwanted problem with disease in the health menu of aquaculture. Several biosolutions can be envisioned, ranging from ultra-specific biotechnological interventions and detailed molecular diagnostics to in vivo recombinant vaccines, all of which help overcome problems in pathogen control and boost fish health and welfare. With advances in modern molecular tools such as PCR, qPCR, and LAMP, pathogen detection has become rapid and specific. Nanotechnology biosensors, therefore, are able to pick white blood cells and microbes within minutes, allowing health monitoring to be carried out on-site (Rathore et al., 2025). Consequently, for example, there are instantaneous detections of *Vibrio* and Koi herpesvirus through CRISPR-based diagnostic platforms.

Vaccine Development and Immunostimulants

Recombinant DNA and subunit vaccines have significantly changed the defense against diseases in marine animal farming. They are particularly vulnerable to such diseases as those caused by the bacterium *Aeromonas hydrophila* and Viral Nervous Necrosis (VNN). Immunostimulants such as β -glucans, chitosan, and herbal extracts help achieve a prophylactic effect by strengthening the innate immune system and antioxidant status (Akbar Ali et al., 2022). That would be the less invasive, modern way of vaccinating on a mass scale, tailored to each antidote, since modern vaccines are composed of oral and intramuscular formulations produced through different biotechnology processes.

Probiotics and Antimicrobial Peptides

By themselves, probiotics increase feed conversion and, at the same time, act as a competitive barrier against gut pathogens colonizing. Genetic engineering for antimicrobial peptide (AMP) production could generate a universal immune response distinct from conventional antibiotics (AL-Azzawi et al., 2024).

Biocontrol and Bioremediation for Aquaculture

Biofloc technology and microbial consortia are increasingly considered a weapon against pathogenic and competing microbes that impede nutrient cycling in the system (Li and Dai, 2025). Beneficial microorganisms introduced into water bodies can also keep the system free of organic pollutants and ammonia (if present).

5. Environment and Ecosystem Biotechnology

The field of environmental biotechnology applies microbial and molecular methods to the recycling of waste, biodiversity monitoring, and, on the other hand, the restoration of degraded aquatic ecosystems. The use of microbial biofilters and biofilms is necessary for the effective removal of nitrogenous and phosphorus wastes. Also, a custom microbial consortium can degrade organic matter effectively while maintaining a stable dissolved oxygen level. Using all these processes under the Recirculating Aquaculture System (RAS) makes it more sustainable (Velmurugan and Pandian, 2024). Biofloc technology (BFT) changes waste nutrients into microbial biomass, which can then be used as a feed complement. It also reduces the need for water changes and improves nutrient retention (Nisar et al., 2024). The microorganisms in the biofloc serve as both a biocontrol system and a nutrient source, thereby making closed-loop aquaculture systems even more effective. Among the methods for managing aquaculture biotechnological waste residues, such as anaerobic digestion or composting, the residues are sometimes used for biofertilizer or biogas production, thereby integrating them with agriculture. A study was conducted by Kurniawan et al. 2025, which showed that recycling plays a significant role in resource conservation and carbon emission reduction. Environmental DNA and metagenomic sequencing are highly effective techniques for biodiversity monitoring and assessing pollution. Non-invasive methods assess the presence and abundance of species; consequently, this information is used to study ecosystem dynamics and anthropogenic impacts (Stephenson et al., 2022).

6. Future Prospects and Ethical Considerations

Advancements in various fields are increasingly influencing the development of aquaculture biotechnology. The combined use of AI, omics, and nanobiotechnology could be sufficient to enable the development of predictive, data-driven, and sustainable aquaculture frameworks. Image analysis with AI and machine learning algorithms can identify disease outbreaks, plan feeding schedules accordingly, and observe fish behavior. Tools like genomics, proteomics, and metabolomics provide a detailed understanding of fish physiology and its interaction with the environment (Kayode-Edwards

et al., 2024). For drug delivery, biosensing, and precise nutrient release, nanobiotechnology is used. Public opinion, being convinced of biomedicine on the market, will be very decisive. Environmental and genetic containment of modified organisms must be secured adequately through ethical and biosafety frameworks. International organizations such as the FAO and the OECD are calling for open communication on risk assessments and labeling, arguing that these are critical to building consumer trust (Stankus, 2021). Increasing reliance on multi-omics data, smart sensors, and circular economy models will characterize the future of aquaculture. Transitioning to ecosystem-based management and nature-positive production will set a new sustainability benchmark and ensure that biotechnology is a factor in the well-being of both humans and the planet (Ferri and Arnés García, 2023).

7. Conclusion

Biotechnology within the fishing industry is an innovative and revolutionary science that implements new technical methods in aquaculture, particularly in genetics, nutrition, health care, and environmental management. It makes the environment more pollutant-free. Introducing genetic tools, artificial intelligence, and the latest developments in ecology will surely assist this new breed of species in prospering and protecting their territories.

Conflict of interests

The authors declare that they have no conflict of interest, financial or otherwise.

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APPLICATIONS OF BIOTECHNOLOGY IN ENVIRONMENTAL AND INDUSTRIAL SECTORS

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ABSTRACT

Biotechnology replaces chemical intensive methods with eco-friendly biological processes in order to protect the environment and contribute to sustainable industrial development. Microbial and enzymatic systems contribute to waste degradation, production of biofertilizers, and control of pollution through bioremediation and biosorption. Industrial biotechnology makes use of biocatalysts for manufacturing in the food, textile, and pharmaceutical industries, thereby contributing to clean production and reduction of emissions. The development of new fields such as synthetic biology, nanotechnology and biosensors has enhanced efficiency, monitoring of pollutants and synthesis of bioproducts. Together these support circular bioeconomy goals and help in the transformation of waste into valuable resources, aligning with global development objectives of environmental sustainability.

KEYWORDS: Biocatalysts, bioremediation, biosensor, biotechnology & sustainability

1. INTRODUCTION

Biotechnology offers tools to improve resource efficiency, environmental protection, systemic integration and economic benefits (Barragan - Ocana et al., 2020). It promotes environmental protection by substituting chemical processes with biological methods. Selective breeding and fermentation are examples of traditional methods, while modern biotechnology employs complementary methods like genetic modification gene mapping and CRISPR to accurately change organisms at the genetic level, resulting in an expanded variety of applications in enzymes are acting as biocatalysts. They are essential for decomposition, nutrient cycling and the breakdown of pollutants through bioremediation (Ezeonu et al., 2012). Industrially, they are used in food processing, textiles and for the creation of pharmaceuticals and detergents (Khan, 2025). The innovation of biofuels, creation of biodegradable plastics, microbial and enzymatic processes significantly help in the mitigation of climate change. Biotechnology serves a pivotal function in the attainment of the United Nations Sustainable Development Goals (SDGs) by integrating technological advancement with principles of environmental sustainability (Donato et al., 2023).

2. MICROBIAL AND ENZYMATIC PROCESSES IN WASTE MANAGEMENT

Major global concerns are waste generation from domestic, agricultural and industrial sources. Biodegradation refers to the natural process in which living microorganisms such as bacteria and fungi break down organic materials into simpler compounds like carbon dioxide, water and biomass. Conversion of compost in a controlled aerobic biodegradation process converts organic waste into nutrient rich compost (Manea et al., 2024). *Pseudomonas*, *Bacillus* and *Aspergillus* species are degrading plastics, gaining prominence as an alternative to mechanical recycling.

In the absence of oxygen, microbial decomposition of organic matter generates biogas and provides renewable energy (Adeleke et al., 2023). Waste biomass is utilized for the production of biofertilizers, organic acids, enzymes and pigments after microbial fermentation (Mahish et al., 2024). Also, food waste and lignocellulosic biomass can be converted into bioethanol and organic acids such as lactic acid through microbial bioprocesses. During this process, cellulases and amylases are used to improve the efficiency of enzymatic conversion. The process is significantly used in wastewater treatment and it facilitates, agricultural waste management systems (Kavya et al., 2024).

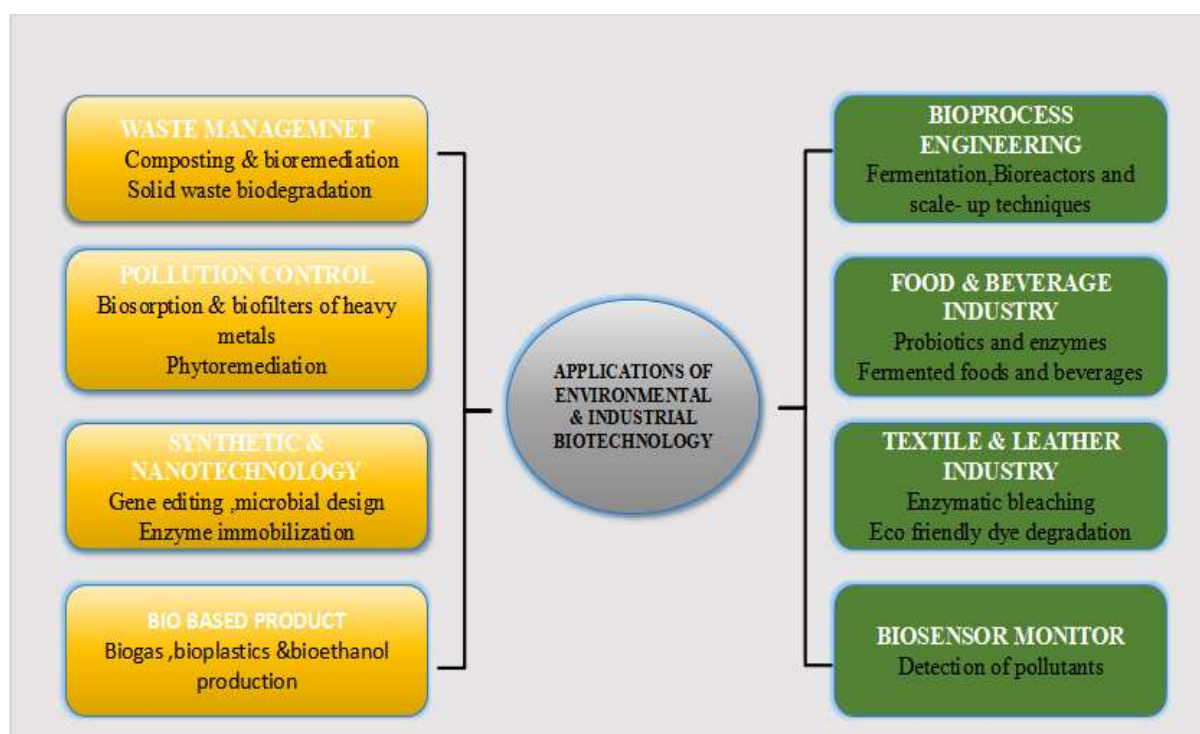


Figure 1. Overview of the applications of environmental and industrial biotechnology

3. BIOTECHNOLOGY FOR POLLUTION CONTROL

Bioremediation is removing contaminants by employing bacteria, fungi, and algae to detoxify polluted environments (Dubey et al.,2025). Hydrocarbon-degrading bacteria such as *Alcanivorax borkumensis* and *Pseudomonas putida* are extensively used in oil spill remediation (Gong et al., 2018). Algae generating biomass that can be valorized as biofuel feedstock (Rawat et al., 2013).The process of heavy metal ion (lead, cadmium and mercury) binding to bacterial cell walls can be metabolism dependent or independent (Bonner,2018). Fungal species like *Aspergillus niger* & *Rhizopus arrhizal* shows remarkable biosorption capacities. Biofiltration systems are used to purify contaminated air and water streams (Naha et al.,2022). Genetically modified microorganisms has the capabilities of degrading pollutants. Modified strains of *Pseudomonas putida* are capable of effectively breaking down aromatic compounds. Genetically engineered microorganisms are playing a major role in sustainable development (Adetunji et al.,2024).

4. INDUSTRIAL BIOTECHNOLOGY AND GREEN MANUFACTURING

Industrial biotechnology, also known as "white biotechnology". Biocatalysts are enzymes that catalyze the rate of chemical reactions. Bioethanol, biodiesel, biobutanol, bioplastics such as polyhydroxyalkanoates (PHA), polylactic acid (PLA) & biogas is a hallmark of industrial biotechnology (Naser et al., 2021). By reducing waste production and greenhouse gas emissions, industrial biotechnology helps to promote greener production. Enzyme consuming less energy & it boosts product yield and quality (Chapman et al., 2018).

TABLE 1: This table highlights major industries and their biotechnological functions.

Textile industry	Cellulases and laccases are used for dye degradation and fabric finishing.
Paper industry	xylanases and lignin peroxidases replace toxic chlorine-based bleaching agents, reducing effluent toxicity (Gangwar et al., 2014).
Food industry	Proteases and lipases are used in the food sector to improve flavor and change the fat content.
Pharmaceutical industry	Enzymes help create stereospecific drugs (Singh et al.,2016).

5. EMERGING TECHNOLOGIES AND INNOVATIONS

Companies utilize synthetic biology and genetic engineering with computational and systems biology to scale production, reducing costs and increasing outputs. Synthetic DNA building blocks and custom microbes are created for specific industrial applications (Clarke et al.,2020). They degrade pollutants and enhance crop resilience. This field addresses climate change and food security by creating sustainable biological solutions. Metabolic engineering systems increase the yield of desirable products such as bioethanol, organic acids, and enzymes. For example, *Escherichia coli* strains have been engineered to improve the polyhydroxybutyrate (PHB) synthesis (Lin et al., 2025), and

cyanobacteria are being modified to fix atmospheric CO₂ into renewable fuels. It exemplifies the convergence of industrial and environmental goals (Agarwal et al., 2022). Nanobiotechnology is amalgamation of science, engineering and technology to create a new material at the size of 1 to 100 nanometers. Plant materials are utilized for nanoparticle synthesis (Osman et al., 2024). It's helping address environmental challenges by improving sustainability and reducing pollution using these technologies, solar panels, and hydrogen storage. Nano-material filters are removing pathogens and contaminants from water (Ojha, 2020). It's more effective than traditional methods. Nano air filters are removing harmful gases from the air. Enzyme immobilization on nanocarriers enhances enzyme stability, reusability, and catalytic performance (Singh et al., 2025).

MICROBIAL CONSORTIA AND BIOFACTORIES

Two or more species breaking pollutants. It offers robustness and efficiency. Bio factories facilitate complex transformations such as lignocellulose breakdown and biohydrogen production. Microbial consortia are useful in the pharmaceutical and food industries (Rodriguez et al., 2023). Biosensors detect biological agents such as bacteria, viruses, and toxins. It's very sensitive. Air quality biosensors also detect pollutants like nitrogen dioxide in industrial and indoor environments. Water quality biosensors can detect heavy metals. Enzyme-based biosensors, microbial biosensors, and immunosensors are commonly used to assess water quality in rivers, lakes, and groundwater. Soil sensors detecting specific parameters like nutrient levels, pH, and presence of contaminants (Sharma & Sharma, 2023).

6. FUTURE PERSPECTIVES & CHALLENGES

Due to increased transportation, there is a rise in greenhouse gas emissions, air pollution and a greater risk of oil spills. Globalization is leading to deforestation and the depletion of nonrenewable resources like fossil fuel. Also, global shipping can disrupt local systems. Invasive species are being transported to new environments. In the future, artificial intelligence will play a new role in reducing industrial waste (Demirel et al., 2024). We should focus on bioplastic and biodegradable materials, advances in resource efficiency, and innovations in green energy industries (Rosenboom et al., 2022; Wei et al., 2022).

7. CONCLUSION

Environmental and industrial biotechnology are integral pillars of modern biotechnology. An overview of the applications of environmental and industrial biotechnology. The use of microbes, enzymes, and bio-based processes has changed waste into wealth, reduced pollution, and promoted the growth of green industries. Inventions in synthetic biology, nanobiotechnology, and biosensor design are enabling precise, efficient, and across numerous industries. Nowadays, biological alternatives are replacing chemical-intensive methods. Biotechnology promotes a cleaner environment and supports

sustainable development. The future of biotechnology lies in the integration of bioprocessing and computational approaches with policies aimed at establishing a circular bioeconomy.

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BIOTECHNOLOGY IN WASTE MANAGEMENT AND ENVIRONMENTAL PROTECTION

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Abstract

Environmental biotechnology incorporates the use of bacteria, plants, and enzymes, along with technological innovations like genetic engineering, nanobiotechnology, artificial intelligence and omics-based approach to address and solve environmental problems. This chapter provides an overview of biotechnology in waste management and environmental pollution, to manage the damage caused by rising population, industrialization, and urbanization by finding eco-friendly ways to protect nature, reduce waste, and build a sustainable, greener world for the welfare of human beings in the future. On the other hand, there are serious limitations involved in implementing the technologies because of their high cost, time consumption, and biosafety concerns that have to be addressed through a regulatory framework, proper planning, and public engagement.

Keywords: Bioremediation, Environmental biotechnology, Green technology, Phytoremediation, Synthetic biology.

1. Introduction

Environmental biotechnology aims to remove pollutants from the environment and restore them by means of the utilization of microbes, plant-based systems, enzymes, and metabolic intermediates, using principles from genetics, life sciences, chemistry, and engineering as strategies (Sminova et al., 2023). Methods like enhancing the genetics of microbes with higher abilities to digest or neutralize toxic substances, the use of recombinant DNA technology, and synthetic biology are also marking their promising advantage (Lutzer et al., 2024). Modern digital technologies like big data analytics, artificial intelligence (AI), and industry 4.0 frameworks like automation, Internet of Things (IoT), and smart systems in industries help in real-time monitoring of the environment, its optimization, and control. An overview of the integrated role of biotechnology in addressing environmental challenges through six major approaches is illustrated below in **Fig.1**. Beyond conventional waste management, environmental biotechnology also supports the conversion of waste into valuable resources by the development of biodegradable materials and biofuels, thereby minimizing industrial pollution and reducing dependency on non-renewable resources (Bano et al., 2020).

2. Microbial and Enzymatic Approaches to Waste Treatment

Microorganisms serve as the foundation of biological waste treatment, especially agro-industrial waste. They degrade and convert complex organic and inorganic pollutants into simpler, non-toxic compounds by specialized enzymatic pathways, thereby facilitating the reutilization of organic residues (Wang et al., 2022). Solid-state fermentation and submerged fermentation use microorganisms that enhance the conversion of agricultural by-products into bioactive compounds, including enzymes, antioxidants, and natural flavors (Kieliszek et al., 2020).



Figure 1: Overview of biotechnological approaches in environmental biotechnology

Microbial waste treatment methods remove 90% of wastes and are much cost cost-effective when compared to conventional physiochemical methods (Ezemba et al., 2021). Microorganisms produce specialized proteins, which are known as microbial enzymes, offering high specificity and efficiency to convert waste into valuable resources and support a sustainable circular economy. They are available either as a purified enzyme or a whole-cell biocatalyst in live or immobilized microorganism form (Orlando et al., 2023). Oxidoreductases, peroxidases, laccases, and hydrolases are some of the enzymes used for remediation. Recent innovations like enzymes derived directly from organic waste, like fruit residues, are gaining attention as sustainable biocatalysts for soil improvement, wastewater remediation, landfill leachate, and composting. Research using advanced energy-based computer models has proved how microbial enzymes break down food waste and wastewater more quickly and efficiently (Sayin & Kaban, 2024).

Oxidoreductases and Oxygenases, deployed by hydrocarbon-degrading bacteria and fungi dismantle hydrocarbons and ultimately yield water and carbon dioxide. This biocatalytic route offers an alternative, to the dispersants that are traditionally used in oil-spill remediation (Wang et al., 2022). A promising frontier in biological waste management is the breakdown of waste: microbial enzymes such as esterases, cutinases and hydrolases attack the sturdy polymer chains of materials, like polyethylene terephthalate (PET) fragmenting them into simpler components. Bacterial PET degradation has opened avenues, for upcycling with microbes fermenting depolymerized intermediates such as ethylene glycol, into higher-value chemicals (Orlando et al.,2023; Lee et al.,2023; Balola et al.,2024). Similarly, in terms of wastewater treatment, enzyme-based systems efficiently remove hazardous pollutants and organic carbon by minimizing the generation of sludge and its operating expenses (Wang et al.,2022). Thanks to bioreactor engineering, strategic enzyme supplementation, and the deployment of microbial consortia, through which this can now be achieved (Ezeji for et al., 2014).

3. Bioremediation and Phytoremediation Technologies

Bioremediation consists in entrusting the environmental degradation processes of selected living organisms. They remove and decompose them to excessively healthier forms. Decomposition can take place in situ or following different methods like biological absorption, bioaccumulation, biotransformation, and biomineralization. It is a purely innovative and sustainable option in comparison to old waste management processes. Raklami et al. (2022) defines phytoremediation as a technique predicated on the natural capability of plants to remove, stabilize, dispose of, or decompose contaminants in water and soil. Such pollutants are heavy metals, organic chemicals, radioactive substances, herbicides, solvents, and hydrocarbons. In situ bioremediation applies biological agents directly to contaminated environments to enable pollutant degradation without excavation, thus minimizing ecosystem disturbance. While ex situ bioremediation treats excavated materials under controlled conditions for greater efficiency in highly polluted sites (Mosa et al., 2016). For the successful implementation of both approaches, understanding the microbial–pollutant interactions and site-specific environmental factors that control biodegradation is vital (Gu, 2020). Microbial systems such as the hydrocarbon-degrading *Pseudomonas putida* and *Alcanivorax borkumensis* effectively metabolize oil hydrocarbons ,while *Bacillus subtilis* helps with heavy metal detoxification via biosorption and bioprecipitation, revealing the versatility of microbial bioremediation across environmental applications (Vázquez-Núñez et al., 2020).

GEMs are designed with the expectation of improving bioremediation through superior breakdown of resistant pollutants, when compared to native strains, by transferring specific genes that code for pollutant-degrading enzymes (Hassan & Saleh, 2023). Engineered GEMs may utilize precise genome editing and CRISPR-Cas systems to further optimize pathways for the degradation of targeted

pollutants (Patil & Desai, 2024). Beyond the generation of GEMs, recombinant strains of *Pseudomonas* and *Escherichia coli*, which demonstrate greater degradation of chlorinated chemicals and aromatic hydrocarbons, have been developed using advances in synthetic biology and genetic engineering (Joutey et al., 2013). A further approach that combines principles of genetic engineering with nanotechnology, enhances microbial tolerance, stability of enzymes, and uptake of pollutants, thus creating a novel and targeted approach to environmental clean-up (Ebah et al., 2022; Firincă et al., 2025).

As discussed in the earlier section, plants can be used as natural cleansers of polluted environments by undertaking different types of phytoremediation methods like Phyto stabilization (plant stabilizes pollutants in the soil itself), Phytoextraction (plants extract pollutants from soil or water, especially heavy metals), Rhizofiltration (root filtration), Phytodegradation or Phyto transformation (plants break down pollutants into less harmful substances) over using chemicals or machines (Raklami et al., 2022). Aquatic plants like *Lemna minor* and *Eichhornia crassipes* aid large-scale purification by absorbing heavy metals and nutrients from wastewater (Khellaf et al., 2022). The utilization of plant–microbe systems increases pollution contaminant removal by enhancing plant growth, metal uptake, and degradation of organic contaminants, while genetic engineering and nanotechnology advances continue to establish these sustainable detoxification approaches (Zeppilli & Maturro, 2025).

4. Industrial and Environmental Applications

Industrial biotechnology can provide sustainable waste management options by converting waste to valuable bioproducts through microbial fermentation, composting, or enzymatic degradation (Fan et al., 2022). Fungi such as oyster mushrooms, international agricultural by-products are important substrates for either microbial or fungal activity in the conversion of agricultural wastes into useable organic materials (such as biofertilizers, animal feed, biodiesel and bioenergy). Using yeast-based bioproducts (β -glucans, vitamins, or carotenoids), or producing microbial biosurfactants or other bioproducts from renewable resources is an illustration of waste valorisation for waste streams which reduces waste disposal costs while also promoting sustainability and the circular economy (Kieliszek et al., 2020).

Biotechnology is integrated into green production processes in order to use more eco-efficient production methods, thereby reducing waste at the source itself. Green bioprocesses utilize biological (microbial, enzyme, and plant-based) systems that have shown to improve the use of raw materials, reduce the uptake of hazardous materials, and energy efficient Struszczyk et al., 2025). With advances in genetic technologies, even bacteria are developing the capacity to degrade hazardous pollutants and polymers to non-harmful products (Semwal et al., 2025). Anaerobic digestion, nutrient recovery,

biofilm reactors, activated sludge, and other microbial technologies have been successful in the treatment of industrial effluents while recovering functional materials (Palani et al., 2021). Altogether, the above-discussed methods could provide a sustainable and recycled bioeconomy system by generating renewable energy from wastes and mitigating its harmful environmental impacts.

Case Studies: Pulp and Paper, Textile, and Mining Industries

Traditionally harmful bleaching agents are used in the pulp and paper sector to remove lignin. Some of the agents used are Chlorine (Cl₂), Chlorine dioxide (ClO₂), Hypochlorite (NaOCl), and Hydrogen peroxide (H₂O₂), which led to the release of toxic chlorinated compounds like dioxins, furans, and phenols, which are toxic to aquatic life and carcinogenic to humans. So, to eliminate chlorine usage, reduce toxic wastewater accumulation, and promote environmental sustainability, microbial enzymes like lignin peroxidase, xylanases, and laccases can be used to remove lignin without creating any harmful byproducts. They have also improved paper quality and processing time. The enzymatic process has now appeared as a green alternative and substitute for sustainable chemical methodologies in the textile industry for dye removal. Enhancement of the fiber recovery and acrylic degradation done by the specific strain (*Gordonia alkanivorans* S7) (Struszczyk et al., 2025). In mining industrial effluents are detoxified by microbial bioreactors, while bioleaching with *Acidithiobacillus ferrooxidans* absorbs metals from low-grade ores, thereby reduce environmental toxin by allowing for the sustainable recovery of minerals (Semwal et al., 2025).

5. Future Prospects and Challenges

Genetic modification and Synthetic biology produce custom-made microorganisms that not only have enhanced metabolic competence but also do degradation of organic pollutants, toxic materials, and plastics very fast. All these specially designed systems together with artificial intelligence and data modelling support the analysis of the bioremediation process by temperature, enzyme activity, and pH, thus making the process more precise, faster, and through the whole range more efficient (Patil and Desai, 2024).

Nanobiotechnology uses nanomaterials and bio-based nanocomposites to improve the metabolic pathways of enzyme stability and the uptake of toxic pollutants, simultaneously enhancing biocompatibility of biological systems in harsh environmental conditions (Firincă et al., 2025). Omics-based approaches (metagenomics and metabolomics) also provide a more complete understanding of microbial community dynamics, like understanding the functional genes and metabolic pathways within microbial communities (Das et al., 2023). Even though these methods have shown acceptable promise in laboratory conditions yet the ability to scale from laboratory settings to actual field application remains a challenge due to microbial competition, ecological variability, and geographical differences. The incorporation of immobilization carriers like biochar as well as digital technologies,

including AI, big-data analytics, and sensor networks, is anticipated to overcome these challenges and greatly enhance the capacity for flexible, energy-efficient, and sustainable remediation systems scalable to principles of a circular bioeconomy (Lutzer et al., 2024).

6. Conclusion

With the intervention of Biotechnology, environmental protection by waste management and pollution prevention is easier, and they can be transformed into eco-efficient and resource-recovering systems. Besides, omics technologies open up a truly detailed view of the whole microbial ecosystem. We are able to see microbial interaction, how they react to pollutants or reproduction, distinguish how their population changes along with time, and even see the exact chains of genes and actions, realizing degradation. We are talking about metagenomics, meta-transcriptomics, and metabolomics anyway. The tech advances, on the other hand, especially correlate with bioremediation and a really practical, green way to restore polluted areas. Microbial one, enzymatic, and phytoremediation are illustrative here, as all these techniques then detoxify the harmed ecosystems and let them operate on them in terms of the circular economy in a while. At the same time, new tools such as synthetic biology, nanobiotechnology, and artificial intelligence are facilitating systems design for adapting to dynamic conditions and dealing with complex and recalcitrant hard-to-degrade pollutants. Despite ongoing challenges of optimizing processes and the heterogeneity of environmental conditions, from other concepts and studies, the emergence of these multidisciplinary innovations reinforces the importance of and reliance on biotechnology to meet global sustainability goals, protect the ecological balance of our planet, and provide a more resilient and cleaner society for future generations.

Conflict of interests

The authors declare that they have no conflict of interest, financial or otherwise.

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ADVANCES IN HUMAN DIAGNOSTIC BIOTECHNOLOGY: ACCELERATING PRECISION AND PERSONALIZED CARE

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Abstract

The rapid innovations in molecular techniques, biosensors, imaging systems, point-of-care testing and AI have transformed diagnostics that resulted in improved detection and monitoring of diseases with faster, accurate and personalized care. Molecular diagnostic methods such as PCR, NGS, and CRISPR improve sensitivity and specificity while biosensors and lab-on-a-chip systems are portable and cost-effective. Imaging systems and their integration improves real-time analysis and accuracy of diagnosis. Point-of-care and wearable devices provides easier access to patients while AI technologies also assist and have potential to improve further in future. Despite challenges across these techniques, continuous advancements provide solutions and promotes efficient, accessible and patient-centred diagnostics.

Keywords: Molecular diagnostics, Biosensors, Point-of-care device, Artificial intelligence (AI), Personalized medicine.

1. Introduction to Modern Diagnostics

Diagnostic methods have advanced and offer accurate and timely strategies to detect diseases and monitor patients' health. Techniques involving molecular methods, biomarkers, imaging systems, point-of-care testing, artificial intelligence (AI) have developed efficiently and offer multiple applications in the diagnostic field. A key factor is all of these methods are integrated to improve the accuracy of diagnostics for a long term. Utilizing AI in diagnostics are enhancing its precision and efficiency yet still relies on gradual improvement in traditional medical tools and clinical expertise (Shipilov et al., 2023).

2. Molecular Diagnostic Techniques

Molecular diagnostic techniques detect and analyse diseases by targeting disease-specific biomolecules such as nucleic acids, carbohydrates, proteins and lipids. It also involves the analysis of pathogen homology and drug resistance genes. These techniques offer rapid detection, low detection time, increased automation, high sensitivity and specificity (Liu et al., 2023; Rolando, Melkonian & Walt, 2024). These methods don't require specialized personnels and some are commercially available for non-laboratory testing (Relich et al., 2025).

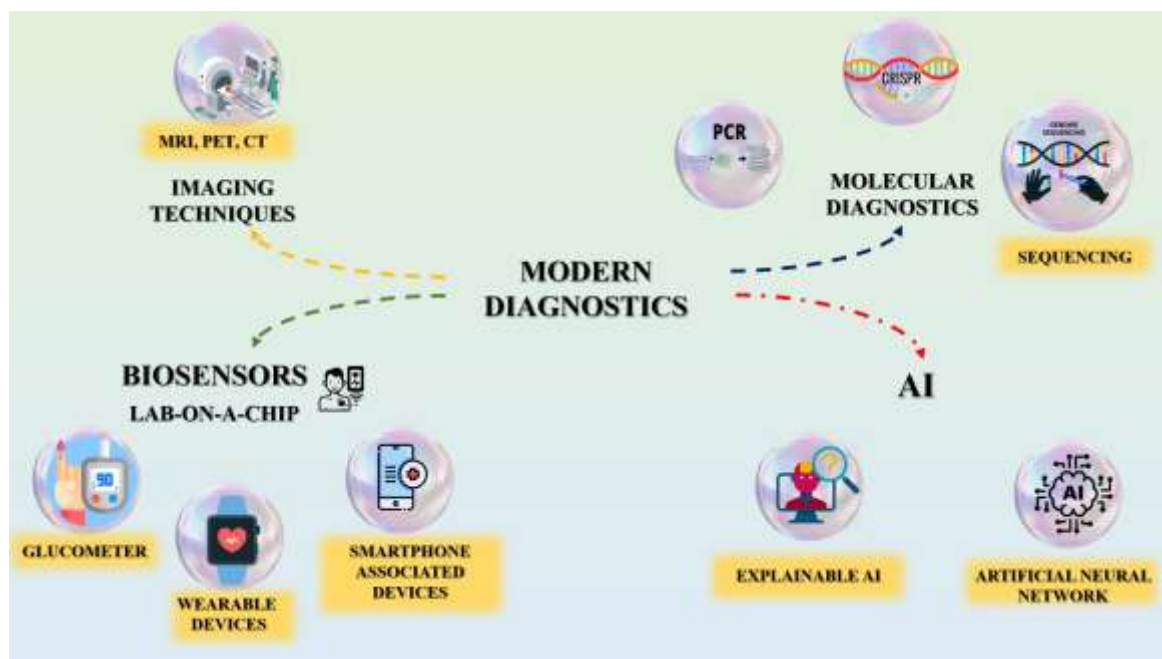


Figure 1: Modern diagnostic methods and its tools

Molecular diagnostics has a great impact on personalized medicine, oncology, infectious diseases and other medicinal areas (Algusheri et al., 2024). Polymerase chain reaction (PCR) is highly sensitive and specific technique in diagnosis as it is used to determine the presence of DNA sequence in samples. It is used in diagnosis of cancer and infectious diseases. There are many variants for PCR that involved in diagnostics. Digital PCR (dPCR) quantifies viruses such as BK polyomavirus, HRV, HIV, HPV, and MTB, and detects drug-resistant mutations (Liu et al., 2023) whereas droplet digital PCR (ddPCR) detects cancers like melanoma and colorectal and lung carcinomas. Quantitative PCR (qPCR) detects bacteria including pneumococci and *H. pylori* while qRT-PCR (Quantitative reverse transcriptase PCR) assists in leukemia diagnosis and detects miRNA. Multiplex qPCR (MqPCR) detects multiple viruses like adenovirus, rotavirus, and SARS-CoV-2. SSP-PCR (Sequence-specific primer PCR) detects human platelet or leukocyte antigens (Rolando, Melkonian & Walt, 2024). Gene sequencing has the highest accuracy in pathogen detection among other molecular techniques. It can be used both for diagnosing known pathogens and identification of unknown pathogens. Next generation sequencing (NGS) detects microbial species and sequences even if there is no clear etiological evidence from laboratory tests. Multiple pathogens can be detected and identified using NGS accurately and simultaneously. Other than identification, NGS also detects drug resistant, virulence genes and analyse host immune responses. Within a short period, it allows researchers, healthcare providers to gather pathogen genomic information, track evolution and transmission and provide valuable insights during infectious diseases epidemics (Liu et al., 2023). Apart from infectious diseases, NGS plays crucial role in cancer diagnostics, genetical disorders, prenatal testing and epidemiological applications (Algusheri et al., 2024; Rolando, Melkonian & Walt, 2024). Third-

generation sequencing (TGS) detects changes in DNA/RNA in a simple and cost-efficient way. TGS can detect SARS-CoV-2 and other respiratory viruses simultaneously in a short time. This method incorporates nanopore system which can be used on-site and out-of-hospital detection of coronavirus, ebola virus, adenovirus and other viruses (Liu et al., 2023). Isothermal amplification is a value added and recently advanced technique in the molecular diagnostics. Techniques like recombinase polymerase amplification (RPA) and loop-mediated isothermal amplification (LAMP) possess various advantages. RPA is simple but yields faster results when compared to PCR or NGS. It is also tolerant to PCR inhibitors. LAMP, on other hand generates nucleic acid from self-priming compounds in a very short time compared to other molecular techniques. CRISPR based diagnostic tests involve isothermal amplification and Cas enzyme for a high specific detection of target compounds. These tests are rapid, less instrumental and lyophilization-friendly. CRISPR has higher specificity even in presence of non-specific amplification or background nucleic acids (Algusheri et al., 2024; Rolando, Melkonian & Walt, 2024).

3. Biosensors and Lab-on-a-Chip Technologies

Biosensors are emerging tools of modern diagnostics with rapid, accurate and cost-efficient detection and monitoring of various diseases (Hayati & Karim, 2024). It comprises of physiochemical transducer and a biological element like enzymes, nucleic acids, aptamers and antibodies. These biological components bind to complementary target/analyte that induces a chemical reaction which will be detected and converted to electric signal by transducer (Lina et al., 2022; Hayati & Karim, 2024). Also, there is a case where the target or analyte converted to a product that releases heat, gas, electrons or hydrogen ions (Srivastava & Sharma, 2022). Samples such as human tissues and body fluids such as blood, urine, saliva and cerebrospinal fluid can be used for detection and monitoring of analyte. Recently, non-invasive liquid biopsies have be recognized as valuable sample source as it is easy, risk-less while also assists in monitoring (Lina et al., 2022). Biosensors are classified based on transducer element such as;

Electrochemical biosensors: These sensors are developed and already commercialized as they are cost-efficient and easier to use (Lina et al., 2022). These detect electrical properties such as current, potential or impedance when analyte binds to biological element in the device. In diagnostics, these sensors detect diseases such as sepsis, SARS-CoV-2, hepatitis C, malaria, melioidosis, HPV, HIV and other bacterial or viral diseases (Parihar et al., 2025).

Optical biosensors: These sensors detect changes in light absorption, fluorescence and surface plasmon resonance during interactions between analytes and biological materials. They are widely used in pathogen detection, cancer diagnosis and drug discovery due to the sensitivity and selectivity

(Lina et al., 2022; Hayati & Karim, 2024). In diagnostics, they detect SARS-CoV-2, hepatitis B, malaria, Ebola, *E. coli*, HIV-1 and tuberculosis (Parihar et al., 2025).

Piezoelectric biosensors: These are mass-based biosensors and detects electric signal when mechanical stress applied. Quartz crystal microbalance (QCM) is a commonly used type for DNA-based sensing (Lina et al., 2022; Srivastava & Sharma, 2022). In diagnostics, they detect SARS-CoV-2, hepatitis B, dengue, tularemia, *Salmonella spp.*, HIV and tuberculosis (Parihar et al., 2025).

Biosensors development in recent years has upgraded the diagnostics field efficiently. Nowadays, they offer continuous glucose monitoring and assists in monitoring chronic diseases like diabetes. Biosensors are integrated to wearable devices which results in enhancement of tracking multiple physiological parameters such as heart rate, oxygen saturation and metabolic markers which can be helpful in conditions like cardiovascular diseases and sepsis (Hayati & Karim, 2024). Also, biosensors are involved in real-time monitoring of amino acids uptake and its levels during physical exercise while assessing signs of metabolic syndromes (Wang et al., 2022). Biomarkers associated with metabolic disorders such as obesity have also been under research (Ozcan & Sezginturk, 2021).

4. Imaging-Based Diagnostic Technologies

Imaging systems have developed rapidly by innovations in methodology and technology that resulted in improving diagnostics and patient care (Rong & Liu, 2024). Image processing techniques improve image quality through noise reduction and artifact removal and highlight abnormalities like tumor, fractures and hemorrhages and assist in computer-aided detection and diagnosis. They made a huge impact in analysing tissue characteristics, disease progression, response to treatment and prognostic predictions (Wang, 2024). PET & SPECT are used in oncology, neurology and cardiology (Kang, Zurutuza & Zaman, 2023; Rong & Liu, 2024) with SPECT involves also in bones. MRI imaging is applied in neurology, cardiology and musculoskeletal systems along with real-time analysis. Along with PET, MRI shown promise for molecular imaging in cancer (Hellebust & Richards-Kortum, 2012). CT imaging is used in diagnosis of cancer, trauma and angiography. There are other imaging techniques like Ultrasound, OCT, MPI, PA are also involved in various diagnostic processes (Kang, Zurutuza & Zaman, 2023). With already existing systems, hybrid imaging techniques like PET/CT, PET/MRI and SPECT/CT have also evolved and integrates the anatomic and functional data for accurate detection and monitoring. In cancer imaging, PET/CT is used to detect metabolic activity and structural details (Rong & Liu, 2024). Fluorescent dyes are used as contrast agents and used in molecular imaging techniques. There are fluorophores that targets antibody, peptides, metabolic biomarkers for molecular imaging process (Hellebust & Richards-Kortum, 2012). Nano-based contrast agents which tagged with either fluorescent dyes or radioactive isotopes are used along with imaging techniques like CT, MRI, PET to visualize cancer cells whereas Quantum dots are

used for long term imaging as they possess better stability (Wu et al., 2024). Integration of AI in imaging systems improves image quality, real-time analysis, early diagnosis, automation and predicts disease progression and its biomarkers (Rong and Liu et al., 2024; Yilmazer, 2024). It also improves diagnosis by enhancing interpretation of images with accuracy, rapid and consistent. AI detects brain tumours, breast cancer, pneumonia, tuberculosis, pulmonary nodules, musculoskeletal abnormalities, rheumatoid arthritis while also in diseases such as neurodegenerative, intestinal and cardiovascular diseases (Bhandari, 2024).

5. Rapid and Point-of-Care Testing

Point-of-care testing is a rapid testing tool in diagnostics that is used to easily assess disease condition by healthcare professionals and patients. These types of testing reduce inconvenience of patient's long commute, easy and rapid testing (Larkins, Michael, and Aparna Thombare, 2023). Lateral flow assays (LFA) are portable and affordable analytical devices detects illnesses faster. With electrochemical sensing, fluorescence, nanotechnology and SERS, LFAs are improved with specificity and sensitivity, rapid and early diagnosis and further enhancement would be with next generation chips that combined with amplification and detection of DNA, as well as CRISPR and nano/quantum materials (Kakkar et al., 2024). Screen-printer technology is another technique seen in portable devices to perform electrochemical screening of pathogen antigens. POC devices are integrated even with smartphones to detect pathogens. Techniques like Mie scattering, hot embossing method, slipchip method, ELISA reader are integrated with smartphones by using cameras or sensor apps to detect or monitor processes (Roy et al., 2022). Also, there are portable imaging devices such as X-ray machines, ultrasound devices, CT scanners, MRI devices for faster and easier diagnosis (Saez Armenta, 2025). Wearable devices such as thermometers, blood pressure cuffs/monitors, pulse oximeters, ECG, EEG, EMG, EOG, EGG, PPG devices are easy to use and real-time monitoring. Also, there are sound-detecting devices integrated with AI that can detect cough variations in COVID-19 patients (Lippi, Pigghi, & Mattiuzzi, 2024).

6. Future Trends and Challenges

Treatment strategies in precision medicine aims to tailor interventions based on patient's unique genetic, molecular, clinical, behavioral and environmental profile (Monani et al., 2025). Researches in personalized medicine and treatment are underway especially in molecular imaging for improvement of selective and proper treatments (Signore et al., 2023). Personalized precision medicine has targeted various diseases conditions such as communicable diseases such as HIV, COVID-19, *Mycobacterium tuberculosis*, & non-communicable diseases such as cancer, diabetes and pulmonary conditions. However, precision treatments are expensive and privacy concerns of medical data (Sekaran et al., 2025). AI models assist in deciding which type of cancer therapy benefit the patient

which will eventually avoid unnecessary therapies and risky treatments. Deep learning can be used to improve specialized and effective treatment, along with a guarantee that future patients will also be treated with personalized care (Monani et al., 2025). Artificial neural networks (ANN), a development of AI, focus on improving accuracy of diagnosis, defining novel preventions and treatments, decision support, postprocessing and quality control in clinical settings (Göndöcs & Dörfler, 2024). Explainable AI shows great potential to improve transparency and enhance understanding and validation of predictive analysis which also assists in personalized health monitoring (Vani et al., 2025). Despite all the advantages, AI still lacks standardization in diagnostic protocols. Also, a single unknown noise can make a big risk of prediction. Data quality can be sensitive in AI incorporation. There are also challenges such as generalization, limited data, bias, interpretability. AI has potential to replace healthcare workers which is an ethical challenge that must be managed properly to ensure its integration into the field. There also should be legal and ethical regulations necessary as it is related to privacy, data security and informed consent (Shah & Gautam, 2023).

7. Conclusion

As mentioned above, modern diagnostics have evolved a lot and still have potential to evolve even more accurate, faster and personalized disease detection. The integration of all diagnostic methods has enhanced prediction and accessibility. Despite of challenges seen in diagnostic methods, there are continuous advancement and progress towards rectifying those challenges in the field. Overall, the future of diagnostics lies in integrating rapid, accurate and patient-centred technologies to improve the healthcare systems.

Competing Interests

The authors declare no competing interests.

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BIOTECHNOLOGICAL INNOVATIONS IN PLANT SECONDARY METABOLITE PRODUCTION: IN VITRO STRATEGIES AND METABOLIC ENGINEERING

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1. Introduction

Plants possess an extraordinary biochemical capacity, synthesizing a vast and diverse array of organic compounds known as secondary metabolites (SMs). These molecules extend beyond the basic requirements of growth and reproduction (primary metabolism) to fulfill critical roles in the plant's long-term survival, including ecological interactions, defense mechanisms, and adaptation to environmental stresses. The immense commercial and therapeutic value of SMs is underscored by their diverse applications such as pharmaceuticals, agrochemicals, nutraceuticals, and industrial additives. The totality of biochemical processes within an organism is known as its metabolism, and metabolites are the small molecule intermediates and products. While primary metabolites are essential for immediate survival, the absence of SMs typically results in long-term impairment of an organism's survivability or fecundity. In modern industry, SMs serve as invaluable sources for a wide range of products, including pharmaceuticals, flavors, fragrances, and pesticides.

In recent years, the escalating global demand for Plant-Derived Medicinal Compounds (PDMCs) has prompted a critical shift in production methods. Conventional sourcing relies on extraction from native plant tissues, a process plagued by several disadvantages: low yields in native tissues, tissue-specific localization, seasonal and environmental dependency, and significant sustainability concerns leading to the overexploitation of wild populations. In response, in vitro culture technologies have emerged as controlled, reproducible, scalable, and sustainable platforms for the efficient biosynthesis of desired secondary metabolites.

2. Classification

Chemical Diversity of Secondary Metabolites

The classification of SMs can be ambiguous, as many intermediates overlap with primary metabolism, such as amino acids and sterols. However, SMs are broadly categorized into five major classes: alkaloids, terpenoids and steroids, fatty acid-derived compounds and polyketides, non-ribosomal polypeptides, and enzyme cofactors.

They are systematically classified based on criteria such as:

- **Chemical Structure:** Featuring distinct groups like heterocyclic rings (alkaloids), phenyl groups (flavonoids), or isoprene units (terpenoids).
- **Biosynthetic Pathway:** Originating from specific routes like the mevalonate pathway (terpenoids), the acetate pathway (polyketides), and the shikimate pathway (flavonoids/phenolics), with amino acids contributing to alkaloids.
- **Molecular Composition:** Containing components that include nitrogenous, sulfur-containing, aromatic, or glycosidic groups, which often dictate their pharmacological activity.

3. Commercial relevance of secondary metabolite production

The commercial significance of Plant-Derived Medicinal Compounds (PDMCs) is soaring, making the efficient production of secondary metabolites (SMs) crucial for the pharmaceutical sector. Conventional methods, which rely on extraction from native tissues, face critical limitations such as low yields, seasonal dependence, overexploitation issues, and the requirement for complex purification. The adoption of the *in vitro* system (cell and tissue culture) provides a superior alternative, offering a controlled, scalable, and sustainable platform for the reliable, year-round biosynthesis of these valuable compounds for medicinal and other commercial products.

4. *IN VITRO* CULTURE TECHNOLOGY FOR SECONDARY METABOLITE PRODUCTION

The core commercial relevance of secondary metabolite production lies in the pharmaceutical industry. The disadvantages inherent in conventional extraction methods—such as the necessity for complex purification and the vulnerability to seasonal variation—make them less efficient for large-scale production. *In vitro* systems overcome these limitations by allowing for constant, year-round production in a controlled environment, thus offering a reliable platform for the modern medicinal market. The term "*in vitro*" (Latin for "in glass") denotes biological material cultured outside its natural environment. Plant cell and tissue cultures are powerful tools for the controlled production of SMs, provided the culture parameters are rigorously optimized.

Table – Baseline Conditions for Plant Tissue Culture Systems

PARAMETER	OPTIMAL RANGE / CONDITION
Temperature	25 ± 2°C
Light Intensity	40–60 μmol m ⁻² s ⁻¹
Photoperiod	16 h light / 8 h dark
Relative Humidity	~50–70% (room); near 100% inside vessel
Culture Medium	MS (Murashige & Skoog) most common
pH (pre-autoclaving)	5.6–5.8

4.1. Callus Culture

Callus is an unorganized, pluripotent cell mass typically induced from explants (e.g., leaf segments, root tips) on a solid growth medium supplemented with auxins and cytokinins. This tissue is formed when cell differentiation is reversed, causing the explant to produce new, unspecialized meristematic cells. The most common media include Murashige and Skoog (MS) medium, with Gamborg's B5 and Linsmaier and Skoog (LS) media also frequently employed. Sterilization is a crucial practice, often involving a sequence of treatments like 70% ethanol and 0.1% sodium hypochlorite (NaOCl).

4.2. Suspension Culture

A cell suspension culture is initiated by placing friable callus fragments into a sterile liquid medium under continuous agitation. This system promotes uniform cell growth and enhanced metabolite production, often exhibiting higher rates of cell division than callus culture.

Suspension cultures operate primarily in two modes:

1. **Batch Culture:** Cells are grown in a fixed volume of medium until growth ceases due to nutrient depletion or waste accumulation. The growth curve follows predictable phases:
 - **Lag Phase:** Initial adaptation with minimal development.
 - **Log Phase:** Exponential growth and rapid cell division (doubling every 20–50 hours, depending on the species).
 - **Stationary Phase:** Cell number stabilizes, and growth stops as nutrients run out.
2. **Continuous Culture:** A dynamic system where fresh medium is continuously added and spent medium is removed, maintaining the cells in a steady, productive growth phase (an "open system"). This is highly beneficial for constant productivity.

4.3. Organ Culture and Hairy Root Technology

Organ cultures involve culturing a whole or part of a plant organ (e.g., shoot or root) to preserve its specific architecture and function, which is often crucial since SMs accumulate in specific organs (e.g., alkaloids in roots, flavonoids in leaves). *In vitro* organ cultures, particularly root and hairy root systems, replicate the natural biosynthetic environment more closely than undifferentiated callus or cell suspensions, often resulting in higher metabolite yields.

Hairy Root (HR) Culture is a specialized and highly effective organ culture technique. It is induced by infecting a plant explant with the soil-borne bacterium *Agrobacterium rhizogenes*, which causes hairy root disease. The bacterium transfers a segment of its Root-Inducing (Ri) plasmid's T-DNA into the plant genome. This T-DNA carries genes (*iaaM*, *iaaH*, *ipt*) that code for enzymes controlling auxin and cytokinin biosynthesis, which in turn induces the formation of rapidly growing, lateral, and *Agrobacterium*-free HRs. HR cultures are favored because of their:

- Genetic and Biochemical Stability: They maintain a consistent metabolite profile over long-term subcultures.
- High Growth Rate: Rapid proliferation without the need for exogenous plant growth regulators.
- Capacity for Biosynthesis: They can synthesize a wide diversity of SMs, often at levels comparable to, or exceeding, the native plant.

5. Advanced Strategies to Enhance Metabolite Yield

To maximize the output of valuable secondary compounds, several advanced strategies are integrated with *in vitro* culture protocols.

5.1. Elicitation: Stress-Induced Biosynthesis

Elicitation is a strategy where small quantities of stress factors, known as elicitors, are applied to a living system to induce or improve the biosynthesis of specific compounds. This process taps into the plant's natural defense mechanisms to redirect metabolic flux.

Elicitors are classified based on their origin:

- Abiotic Elicitors: Non-biological factors, including physical stresses (light, salinity, temperature), chemical agents (heavy metals like Ni, Ag, Co), and hormones, which modulate key metabolic pathways.
- Biotic Elicitors: Substances of biological origin, such as polysaccharides from plant cell walls (chitin, pectin) or microorganisms.

The success of elicitation hinges on optimizing parameters like elicitor concentration, duration of exposure, and culture age. For instance, a study on ajmalicine production in *Catharanthus roseus* showed that while higher concentrations (5.0%) of an elicitor extract enhanced accumulation, a further increase (10.0%) adversely affected the yield, demonstrating the need for precise dose-response optimization.

A notable example of biotic elicitation involves the co-cultivation of *Centella asiatica* with the endophytic fungus *Colletotrichum gloeosporioides*. This symbiotic co-cultivation resulted in a marked enhancement in root growth and biomass, including a 1.65-fold increase in root number. Crucially, the content of the key triterpenoid, asiaticoside, reached 4.08 mg/g in co-cultivated plants, substantially surpassing the 2.63 mg/g found in control plants. This enhancement is attributed to the fungal elicitor activating the Methylerythritol Phosphate (MEP) pathway, boosting the levels of the terpenoid precursor, squalene, and subsequently stimulating secondary metabolism.

5.2. Precursor Feeding

Precursor feeding involves the exogenous addition of specific biosynthetic intermediates or metabolic precursors to the culture medium. The underlying principle is that supplying these key substrates can

bypass rate-limiting steps in the plant's endogenous biosynthetic pathways, thereby increasing the final metabolite yield.

This strategic approach is widely favored for industrial production due to its capacity for providing reproducible results in controlled *in vitro* environments. It also plays a role in conserving wild plant populations by shifting metabolite sourcing to the laboratory.

Despite its efficacy, precursor feeding presents challenges:

- **Cytotoxicity:** High concentrations of precursors can be toxic to plant cells, compromising viability and productivity.
- **Uptake Limitations:** Inefficient uptake and transport across the cell membrane can restrict the precursor's bioavailability.
- **Metabolic Diversion:** The fed precursors may be rerouted into unintended endogenous pathways, reducing the yield of the target compound.

Table -Successful examples for precursor feeding

Medicinal Plant	Precursor Supplemented	Increased Compound(s)	Reference
<i>Coptis japonica</i>	L-tyrosine	Berberine	Ikuta <i>et al.</i> , 1999
<i>Plumbago zeylanica</i>	Acetate	Plumbagin	Mali <i>et al.</i> , 2012
<i>Artemisia annua</i>	Mevalonic acid lactone	Artemisinin	Zhang <i>et al.</i> , 2011
<i>Catharanthus roseus</i>	Tryptophan	Ajmalicine, Catharanthine	Jaleel <i>et al.</i> , 2007
<i>Panax ginseng</i>	Squalene	Ginsenosides	Kim <i>et al.</i> , 2004
<i>Withania somnifera</i>	Sodium acetate	Withanolide A	Sharada <i>et al.</i> , 2007
<i>Taxus wallichiana</i>	Phenylalanine	Taxol	Wang <i>et al.</i> , 2007

A case study on *Sphagneticola calendulacea* demonstrated the power of this technique in transgenic hairy roots. Supplementing the culture with 0.5 mM phenylalanine led to a significant elevation in the coumestan compound, wedelolactone. The precursor-fed system yielded 440.33 µg/g DW of wedelolactone, compared to 422.01 µg/g DW in non-fed transgenic roots and only 294.44 µg/g DW in the wild-type plant. Furthermore, this strategy dramatically increased other phenolic compounds, with ferulic acid and rutin increasing up to 8.11-fold and 33.77-fold, respectively, over controls.

5.3. Genetic Transformation and Metabolic Engineering

Genetic transformation is the process of introducing foreign DNA into an organism's genome to achieve the stable integration and expression of desired genes. *Agrobacterium* -mediated transformation, as used in hairy root culture, is highly efficient and is one of the best techniques for enhancing SM production *in vitro*.

Metabolic engineering is a sophisticated extension of genetic transformation used to redesign cellular pathways for improved biosynthesis. Researchers can manipulate the flow of metabolites by:

- Introducing heterologous genes.
- Blocking competing pathways.
- Altering genes that encode rate-limiting enzymes.

Table - functional analysis of metabolic engineering tools for secondary metabolite enhancement in plants

Tool	What It Does	How It Works	Role in Metabolite Enhancement	Example
CRISPR/Cas9 Genome Editing	Modifies plant genes very precisely	Cuts DNA at targeted sites and edits via repair mechanisms	Silences undesired genes, introduces new biosynthetic pathways	Boosted artemisinin in <i>A. annua</i> (Li <i>et al.</i> , 2020)
Agrobacterium-Mediated Transformation	Transfers new genes into plant cells	Uses <i>A. tumefaciens</i> or <i>A. rhizogenes</i> to deliver DNA into plant tissue	Creates hairy roots with stable expression; boosts production platforms	Enhanced phenolics in <i>Momordica dioica</i> (Thiruvengadam <i>et al.</i> , 2016)
Transcription Factor Modulation	Controls entire biosynthetic pathways	Overexpresses or silences key TFs like MYB, WRKY, bHLH	Turns on multiple metabolite-related genes simultaneously	Flavonols boosted in tobacco by <i>AtMYB12</i> (Grotewold <i>et al.</i> , 2000)
RNAi Silencing	Switches off unwanted or competing genes	Uses siRNA to target and degrade specific mRNA transcripts	Prevents metabolic cross-talk; redirects precursor flow toward desired products	Increased vinblastine in <i>C. roseus</i> (Chen <i>et al.</i> , 2018)

A study on hairy root cultures of *Momordica dioica*, induced using *A. rhizogenes*, successfully demonstrated enhanced phenolic production. Analysis revealed that transgenic roots produced significantly higher levels of flavonols, hydroxycinnamic acids, and hydroxybenzoic acids compared to non-transgenic roots. Specifically, levels of catechin, rutin, quercetin, and gentisic acid were markedly higher, correlating with stronger antioxidant, antibacterial, and antiviral activities. This work validates HR culture as a stable and reliable platform for the overproduction of valuable phenolic compounds.

6. BIOTRANSFORMATION

Biotransformation is a chemical modification of some compounds by biological systems, typically involving enzymatic reactions that convert substrates into bioactive and extractable forms. It is widely used to enhance or diversify metabolite profiles. More precisely, it refers to the enzymatic conversion of exogenously supplied precursors into structurally modified or functionally enhanced metabolites by plant cells, tissues, or enzymes. (Yashavardhan *et al.*, 2020)

Table- CORE CHEMICAL REACTIONS IN SECONDARY METABOLITE BIOTRANSFORMATION

Reaction	What It Does	Relevance in SM Enhancement
Hydroxylation	Introduces hydroxyl (-OH) groups into a molecule	Increases solubility and bioactivity of compounds (Khirwadkar <i>et al.</i> , 2013)
Glycosylation	Attaches sugar moieties to a molecule	Enhances stability, solubility, and pharmacokinetics (Yashavardhan <i>et al.</i> , 2020)
Oxidation	Adds oxygen or removes hydrogen, increasing polarity	Converts precursors into more reactive forms (Bajaj <i>et al.</i> , 2020)
Reduction	Removes oxygen or adds hydrogen, lowering oxidation state	Balances oxidative reactions, especially in polyphenol transformations
Methylation	Adds methyl (-CH ₃) groups, often affecting activity and mobility	Modifies antimicrobial or anticancer properties of metabolites

This is a multi-stepped process involving a living production platform. It typically initiates with substrate selection, followed by biocatalyst preparation, which involves plant cell cultures, microbial strains, or isolated enzymes . Following enzymatic activation, the transformation reaction is initiated, usually involving hydroxylation, glycosylation, or methylation and the products undergo **isolation**, **purification**, and **characterization** to conform structural integrity and bioactivity. After that optimization and scale up strategies are employed to enhance yield and productivity

7. Challenges and Future Prospects

Despite considerable progress, several challenges must be addressed to ensure the widespread industrial adoption of *in vitro* metabolite production systems.

7.1. Overcoming Key Challenges

- **Low Yield and Metabolic Bottlenecks:** Low yields remain a primary limitation, often due to rate-limiting enzymes and feedback inhibition in biosynthetic pathways. Overcoming this requires a deeper understanding of pathway regulation and the application of robust metabolic engineering.
- **Cytotoxicity and Uptake:** The cytotoxicity caused by high concentrations of elicitors or precursors can compromise cell viability. Furthermore, the lack of efficient membrane transporters can restrict the cellular bioavailability of these compounds, posing a hurdle for standardization.
- **Culture Stability and Scale-Up:** Long-term culture stability is a concern, as genetic and epigenetic drift can lead to the loss of biosynthetic capacity over successive subcultures. Finally, scaling up

laboratory protocols to industrial production requires cost-effective, reproducible methods and rigorous quality control frameworks.

7.2. Avenues for Future Advancement

The future of *in vitro* SM production is focused on integration and precision.

- **Scalable Bioreactor Platforms:** Hairy root and cell suspension cultures, when integrated with advanced bioreactor technologies, enable large-scale, high-yield production under fully reproducible conditions, meeting the needs of pharmaceutical and nutraceutical industries.
- **Precision Genetic Engineering:** Tools such as CRISPR/Cas9 and transcription factor modulation offer unprecedented precision for manipulating biosynthetic pathways. These tools allow for the targeted enhancement of rate-limiting steps, suppression of competing pathways, and the activation of 'silent' gene clusters, which promises to dramatically improve metabolite yield and diversity.
- **Synergistic Approaches:** The simultaneous application of advanced strategies, such as the synergistic use of elicitation (e.g., microbial endophytes like *C. gloeosporioides* in *C. asiatica*), precursor feeding, and genetic transformation, has been shown to result in substantial improvements in productivity.

The integration of these *in vitro* culture systems with cutting-edge biotechnological strategies represents a transformative shift in the production of medicinal compounds. By enabling year-round, controlled production, these techniques not only satisfy the growing demand for plant-derived bioactives but also offer a vital and sustainable alternative to conventional harvesting, supporting the conservation of endangered medicinal plant species.

HARNESSING AI AND BIOINFORMATICS TOOLS FOR GENOMIC PREDICTION IN CROP BREEDING

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1. Introduction

Crop improvement is becoming increasingly challenging due to rapid population growth, evolving nutritional requirements and unpredictable climate patterns that intensify the exposure of crops to diverse biotic and abiotic stresses. Traditional phenotype-based breeding has delivered great gains but is slow, resource-intensive and unable to meet the speed required for resilience and nutritional goals. Genomic prediction (GP) uses genome-wide marker information to predict complex trait values and offers a transformational route to accelerate genetic gain by enabling early selection, reducing generation time, and increasing selection intensity without exhaustive phenotyping for every genotype. Over the last decade GP has moved from proof-of-concept to routine application in several major crops and breeding programs aided by cheaper genotyping, standardized bioinformatics pipelines, and the application of modern machine learning (ML) and artificial intelligence (AI) methods that can exploit large, multimodal datasets.

Traditional phenotype-based breeding has driven significant genetic gains over the past century; however, its dependence on multi-season, multi-location evaluation makes it comparatively slow, resource-intensive, and insufficient to meet the urgency of modern breeding targets. The emergence of genomic prediction (GP) has transformed this landscape by enabling the use of genome-wide marker information to estimate breeding values early in the breeding cycle, thereby reducing reliance on exhaustive phenotyping. Over the past decade, GP has matured from a conceptual framework to an operational tool in numerous crop improvement programs, largely due to the availability of low-cost genotyping, advances in bioinformatics, and the incorporation of machine learning (ML) and artificial intelligence (AI) methodologies capable of handling large and heterogeneous biological datasets. These developments have laid the foundation for integrating multi-omics information and high-throughput phenotyping, positioning GP as a central pillar in modern, data-driven crop breeding.

2. Fundamentals of Genomic Prediction

At its core, genomic prediction aims to estimate the genetic value (Genomic Estimated Breeding Value) of individuals using genome-wide markers and a model trained on a population with both genotypes and phenotypes. Building a reliable GP pipeline requires attention to three interdependent elements:

- (A) population design and phenotyping strategy
- (B) genomic data processing and representation and
- (C) model evaluation and validation.

A. Population design and phenotyping strategy.

The accuracy of GP depends critically on the relationship between the training population (individuals used to fit the model) and the selection candidates, as well as the heritability and genetic architecture of the trait. Well-designed training sets capture the genetic diversity and linkage disequilibrium (LD) structure relevant to prediction targets. Practical strategies include using historical multi-environment trial (MET) data, designing training sets that maximize relatedness to selection candidates or assembling multi-parent populations to broaden applicability. Sparse phenotyping strategies, where not every genotype is measured in every environment can reduce costs while preserving predictive performance when combined with appropriate genomic models.

B. Genomic data processing and representation.

Accurate marker data is the foundation of GP. Typical steps are variant calling, quality filtering (minor allele frequency, missingness, Hardy–Weinberg considerations) and imputation to fill sporadic missing calls. Simple biallelic SNP codings are widely used but genomic relationship matrices (GRMs) that summarize realized similarity between individuals are the workhorse input for BLUP-based methods. When working with very large marker sets, dimensionality reduction (e.g., principal component analysis, marker pruning, or marker pre-selection) or genomic feature engineering (e.g., haplotype blocks, functional annotations) can improve computational efficiency and sometimes accuracy.

C. Model evaluation and validation.

A rigorous validation framework is essential to estimate real-world predictive performance. Cross-validation remains the most common tool, but breeders must choose CV schemes that mimic practical selection decisions like random CV (random splits), family-wise CV (leave-family-out), and environment-wise CV (leave-environment-out) provide different perspectives on expected accuracy when predicting new individuals, new families, or new environments, respectively. Evaluation metrics typically include Pearson correlation between observed and predicted values, mean squared error, and rank-based measures when selection rank is primary. Transparent reporting of training/test splits,

hyperparameter tuning, and environmental structure avoids optimistic bias and improves reproducibility.

Integration of additional data modalities i.e., high-throughput phenomics (UAV imagery), transcriptomics, and environmental covariates is increasingly routine and can raise accuracy for complex G×E traits when models are designed to leverage multimodal inputs. However, integration requires careful preprocessing, alignment of samples across modalities and model architectures able to combine heterogeneous data without overfitting.

3. Bioinformatics Tools and Pipelines for GP

Advance in science and modern technologies have taken studies on plant biology and crop improvement to greater heights (Schutser *et. al.*, 2007). Conventionally, decisions in plant breeding depends on the phenotyping, therefore requires extensive phenotyping across seasons and locations. Crop selection and breeding decisions are now supported with huge genotypic and phenotypic data generated using low-cost high-throughput genotyping and phenotyping respectively. In addition to generation of huge data, handling the same for genomic prediction is equally important. For which, bioinformatics tools support the generation and handling of massive genomic datasets.

3.1 Preprocessing of Genomic Data

Bioinformatics plays a crucial role in transforming the raw genomic data into curated high-quality data sets ready for downstream analysis like genomic prediction and association studies. After the data acquisition, quality of the sequence should be assessed and contaminations are screened using tools like FastQC (Clark and Lillard, 2024). FastQC offers quality control checks on raw sequence data to identify adapters and low quality reads. These adapter sequences can be filtered and trimmed using tools such as Trimmomatic or Cutadapt. Trimmomatic is used to trim paired-end data whereas cutadapt locates helps in locating and removing adapter sequences, primers, poly-A tails etc (Martin, 2011).

3.2 SNP calling

Process of identifying genetic variations from genomic sequence data, which is later used to predict an individual's traits. Genomic prediction uses SNPs to establish associations between markers and phenotypes (Wu *et al.*, 2023). Next-generation sequencing reads are aligned onto a reference genome available, further variable sites are identified and genotypes at those sites are determined.

3.3 Imputation methods

Missing data can potentially bias the downstream analyses in genomic prediction. Algorithms infer missing genotypes by detecting shared haplotypes, linkage disequilibrium between markers, referring to known reference panels or population data. Individuals of a species/population share segments of DNA with others due to ancestry, linkage disequilibrium (LD), and recombination patterns called haplotype blocks.

Family-based imputation uses the known pedigree relationships. Used for structured crosses where pedigree information is available and reliable. Tools like MERLIN, Alphalmpu are used for family-based imputation. Crops with diverse germplasm where large reference panel is available, LD patterns and haplotype frequencies of the population are used for the imputation. Tools like IMPUTE2, Beagle, Minimac etc., are used.

For high-dimensional datasets where the data is large and complex machine-learning methods are used for imputation of the missing data. Machine-learning methods such as k-Nearest Neighbours (KNN), random forests, deep learning etc., based imputations are operated.

3.1.3 Quality filtering

Quality filtering eliminates biologically unrealistic and redundant markers and ensures only reliable SNP-markers are used for genomic prediction or calculating the GEBVs. At sample level filtering population outliers far from the main cluster are removed. Duplicates or identity-by-descent individuals and individuals with unusual heterozygosity due to contamination or inbreeding are also removed. At marker-level filtering SNPs with minor allele frequency which contributes for little variance pose reduction in prediction accuracy. Markers showing segregation distortion (deviating from Mendelian ratios in biparental mapping population) and monomorphic markers are also removed.

3.2 Genetic Relationship Matrices (GRMs)

Since most of the economically important traits are polygenic in nature, precise estimation of genetic value (Jiang *et al.*, 2019). Best linear unbiased estimate (BLUP) is proposed by Henderson to estimate genetic value, which takes advantage of inheritance similarities among individuals based on pedigree information. With the availability of genome wide SNPs, similarities among individuals are calculated (Meuwissen *et al.*, 2001), where the pairwise kinship among individuals are usually described with a matrix called genomic relationship matrix (GRM). Instead of pedigree data GRM is used to estimate the breeding value called genomic best linear unbiased estimate (GBLUP) (Jiang *et al.*, 2019).

Commonly used bioinformatics software

TASSEL: general linear model and mixed linear model are implemented in TASSEL (Trait Analysis by aSSociation, Evolution and Linkage) approaches for controlling population and family structure (Bradbury *et al.*, 2007). GBLUP is used for genomic prediction in TASSEL, which incorporates kinship matrix to account for relatedness between individuals to estimate the genetic value.

GAPIT: Genome Association and Prediction Integrated Tool is a freely available R package program which performs a genome-wide association studies and genomic prediction. It uses unified mixed model, EMMA, the compressed mixed linear model, and P3D/EMMAx for statistical analysis of genetic data. To decrease computational time without compromising statistical power Population

Parameters Previously Determined (P3D), or Emma has been developed and incorporated (Zhang *et al.*, 2010)

rrBLUP: R package which is effective for predicting the performance of individual genotypes based on high-density marker data. It combines Ridge Regression with Best Linear Unbiased Prediction. It is a widely used model in plant breeding for estimating the breeding values by fitting a mixed model where genetic markers are used as random effects.

4. Classical Statistical Models in Genomic Prediction

Classical models form the methodological backbone of genomic prediction and provide interpretability, computational efficiency, and in many cases robust baseline performance. The most widely used families, their rationale, and practical considerations are summarised below;

4.1 GBLUP / rrBLUP (Best linear unbiased prediction with genomic relationships).

GBLUP models treat the collective effect of markers as a random effect with covariance proportional to the genomic relationship matrix (G). In practice this is equivalent to ridge regression on markers (rrBLUP) and assumes many small, additive marker effects drawn from a common normal distribution. GBLUP is computationally efficient, scales to large numbers of markers, and often serves as the benchmark because it performs reliably across many polygenic traits. Its main limitation is the assumption of homogeneous shrinkage, large-effect loci are shrunk the same as small-effect loci which can be suboptimal when genetic architecture is sparse or contains major QTLs.

4.2 Bayesian alphabet methods (BayesA, BayesB, BayesC π , Bayesian LASSO, etc.).

Bayesian methods relax the homogeneous-shrinkage assumption by allowing marker-specific variances or mixture priors that place some markers near zero and a few with larger effects. BayesA assumes t-distributed marker effects (heavy tails), BayesB introduces a point mass at zero (variable selection) and BayesC π estimates the proportion of nonzero markers. These methods can outperform GBLUP when trait architecture includes large-effect loci, but they are often more computationally intensive and require careful MCMC/EM tuning. Empirical comparisons show mixed results: Bayes methods sometimes improve accuracy for traits with oligogenic control but may not outperform GBLUP for highly polygenic traits.

4.3 Kernel methods and RKHS.

Reproducing Kernel Hilbert Space (RKHS) regression models map marker genotypes into a high-dimensional kernel space, capturing non-linear patterns and epistatic interactions without needing explicit interaction terms. RKHS can outperform linear methods for traits with non-additive genetic variance, but kernel choice and tuning (bandwidth parameters) strongly influence results. RKHS strikes a pragmatic balance: it extends linear BLUP methodology while remaining interpretable and typically less data-hungry than large neural networks.

4.4 Practical comparisons and best practices. No single model universally dominates; model choice should reflect trait architecture, training set size, computational resources, and the prediction target (within-population vs across-population vs new environments). Best practice is to (a) use GBLUP/rrBLUP as a robust baseline, (b) test Bayesian and RKHS methods for traits suspected to have large effect loci or non-additive variance, and (c) include appropriate CV schemes that reflect breeding decisions. Ensemble approaches (weighted model averages) often combine complementary strengths and provide stable gains in accuracy. Importantly, interpretability and integration with breeding workflows (selection index construction, expected genetic gain calculations) remain crucial evaluation dimensions beyond raw predictive correlation.

5. Integration of multiomics data for genomic prediction

Development of a observable traits is a result of a complex interplay between its genotype, environment, and several molecular layers of regulation such as transcriptomes, metabolomes, and epigenomes. With the advent of modern sequencing platforms and next-generation genomic technologies, the integration of high-density genotypic data with conventional phenotypic information has transformed contemporary crop improvement programs. Furthermore, advances in molecular biology have enabled the incorporation of multi-omics datasets including transcriptomics, metabolomics, epigenomics, and high-throughput phenotyping into breeding pipelines. This comprehensive data integration has substantially enhanced the precision, efficiency, and predictive power of crop improvement strategies.

The integration of multi-omics datasets with pan-omics platforms and systems biology approaches enables more accurate prediction of complex traits and a deeper understanding of the regulatory networks underlying crop performance. Such multiset-data-driven predictive models are making crop breeding more efficient (Clark and Lillard, 2024).

5.1 Transcriptomics: It is a complete set of all RNA molecules in a cell or organism. Transcriptomic data can be used to investigate how genes are expressed and regulated at various levels. Transcriptomic data along with genotypic and phenotypic data can be incorporated for genomic prediction through mixed linear models (MLMs) and machine learning approaches.

Some of the MLM models are;

5.1.1 Genomic and Transcriptomic Best Linear Unbiased Predictor (GTBLUP): This model treats genomic and transcriptomic data as two separate data sets, and as random effects.

5.1.2 GTCBLUP/GTCBLUPi: This is an extension of GTBLUP, which addresses redundant information between genomic and transcriptomic data (Haas *et. al.*, 2025). This model ensures that any phenotypic variation associated with both genome and transcriptome which is attributed to only one which yields a more accurate partitioning of variation.

5.2 Weighted Multiple Kernel Ridge Regression (WMKRR): Multiple kernel learning approach is used to assign and integrate different weightages to genomic and transcriptomic data. Predictive ability of transcriptome-based Gaussian kernel (GRBLUP) is significantly higher than the predictive ability of GBLUP (Li *et al.*, 2019).

5.3 Machine learning approaches: Complex, non-linear complex interactions both within and between multi-omics layers are incorporated and their redundancies are managed and predictions are made using ML methods such as Gradient Boosting Machines or Deep learning approaches.

5.4 Epigenomics: Gene expression is also regulated by chemical modification of DNA and histones called epigenetic regulation which either makes gene accessible for transcription or condense to prevent transcription. PUMICE (Prediction Using Models Informed by Chromatin conformations and Epigenomics) is a method which integrates 3D genomic and epigenomic data with eQTL (expression quantitative trait loci) to increase the efficiency of prediction of gene expression. EpiXan method attempts to incorporate the epigenomic data to improve the prediction accuracy (Khunsriraksakul *et al.*, 2022).

6. Genomic Prediction Pipelines: Integrating Bioinformatics and AI

Modern GP pipelines combine high-density genomic markers with phenotypic and environmental data to generate accurate estimates of breeding values. A typical workflow begins with genotyping using GBS or SNP arrays, followed by quality control to remove markers with missing data, low minor allele frequency, or technical errors. Genotype imputation methods ensure dense and complete datasets suitable for downstream analysis.

Once the genomic dataset is curated, statistical and machine-learning methods such as GBLUP, Bayesian regressions, and deep learning are trained using available genotype–phenotype information (Meuwissen *et al.* 2001; de los Campos *et al.* 2013). These models learn the relationships between DNA marker variation and trait expression, enabling prediction of genomic estimated breeding values (GEBVs) for untested lines. Cross-validation ensures reliability, while independent test sets provide unbiased estimates of prediction accuracy. AI enhances GP by learning nonlinear patterns, integrating heterogeneous datasets, and capturing subtle marker–environment interactions. Deep neural networks and ensemble models deliver improved predictive performance in complex traits, especially under variable environmental conditions (Montesinos-Lopez *et al.*, 2018). The integration of genomic, environmental, and high-throughput phenomic data into unified AI-driven pipelines is transforming GP into a scalable framework suitable for national breeding programs.

7. Multi-Environment Trials and G×E Interaction

Genotype × environment (G×E) interaction is a central challenge in plant breeding because it influences both the stability and predictability of genotype performance across diverse production

environments. A genotype may perform exceptionally well in one environment but poorly in another, due to differential sensitivity to climate, soil characteristics, disease pressure, or management practices. Such variations lead to changes in marker effects and genetic correlations among environments, which limit the accuracy of genomic prediction (GP) when a single environment-agnostic model is used. This makes the modeling of G×E indispensable for developing climate-resilient and widely adapted crop varieties.

Multi-environment trials (METs) form the foundation for studying and quantifying G×E. These trials evaluate breeding lines across diverse agro-ecological zones, seasons, and management regimes. METs generate phenotypic and environmental datasets that allow breeders to estimate stability parameters, identify broadly adapted lines, and detect specific adaptation to stress-prone or favorable environments. The integration of METs into GP pipelines helps capture the environmental variance structure, ensuring that prediction models remain robust when deployed across different target environments.

To model G×E statistically, several advanced approaches have been developed. Mixed models incorporating genotype × environment interaction terms account for differential performance of genotypes across environments. Reaction norm models further allow the slope and intercept of each genotype to vary with environmental gradients, such as drought intensity, temperature fluctuations, or soil fertility. Factor-analytic models, widely used in MET analysis, efficiently capture shared environmental covariances by estimating latent factors that drive environmental similarity (Jarquin *et al.* 2014). These models improve prediction accuracy by modeling environment-specific marker effects rather than assuming constant effects across all testing conditions.

Artificial intelligence (AI) and machine-learning approaches have extended the capacity to model G×E by integrating genomic markers with environmental covariates and phenomic variables. Neural networks, convolutional models, and ensemble learning methods can detect nonlinear patterns and complex dependencies between genotype performance and environmental characteristics. By incorporating environmental descriptors such as rainfall, temperature profiles, soil organic matter, and cropping system management, AI-based GP models produce more robust predictions, especially when genotypes encounter diverse or unpredictable growing conditions. The emergence of high-throughput UAV-based phenotyping has revolutionized MET data collection. UAVs equipped with thermal, multispectral and hyperspectral sensors can capture canopy temperature, vegetation indices, biomass proxies, and pigment composition with high spatial and temporal resolution. These measurements are strongly correlated with drought response, heat tolerance, nitrogen-use efficiency, and overall physiological performance. UAV-derived phenomic traits provide continuous monitoring across the crop cycle, reducing reliance on destructive sampling and enabling early detection of stress responses

that may not be visible to the naked eye. Integrating UAV-derived traits into GP models enhances their predictive power by enriching the model with dynamic phenotypic information that reflects real-time plant–environment interactions.

Collectively, METs, advanced statistical models, AI-driven environmental integration, and UAV-based phenotyping form a comprehensive framework for addressing G×E. This integrated approach ensures that genomic prediction models are not only accurate but also environmentally informed, enabling the development of stable, high-performing crop varieties suited to diverse and climate-changing agro-ecosystems.

8. Applications in Modern Crop Breeding

Genomic prediction now plays a central role in breeding programs worldwide. Its key applications include:

8.1 Biotic stress resistance

GP enables prediction of complex disease resistance traits such as rusts, viral diseases, and bacterial blights, reducing reliance on artificial screening and enabling early selection.

8.2 Abiotic stress tolerance

Traits like drought, heat, salinity, and submergence tolerance exhibit polygenic inheritance. GP, combined with environmental covariates, improves selection under climate variability and stress-prone conditions.

8.3 Yield and grain quality

Low-heritability traits such as grain protein, oil content, and kernel weight benefit from GP because it leverages genome-wide marker information and accelerates selection across generations.

8.4 Integration with speed breeding and DH systems

Combining GP with speed breeding and doubled haploid production significantly shortens breeding cycles. Early genomic selection followed by rapid generation advancement enables faster development of elite lines with desirable trait combinations. Overall, GP is reshaping breeding pipelines by enabling simultaneous improvement of complex traits and reducing the cost and time traditionally required for multi-year field evaluations.

9. Challenges and Ethical Considerations

9.1 Data limitations

Accurate GP requires large, high-quality, multi-environment phenotyping datasets. Data scarcity, inconsistent experimental designs, and poor metadata reduce model accuracy and portability. The adoption of standard metadata frameworks and FAIR data principles is essential for long-term utility.

9.2 Model overfitting and generalization

AI and deep-learning models are highly flexible but risk overfitting small datasets. Careful cross-validation, regularization, and appropriate feature selection are necessary to ensure robust predictions.

9.3 Computational and operational constraints

GP demands significant computational capacity for model training and data handling. While cloud-based and local HPC systems offer solutions, technical expertise and recurrent costs pose limitations for small breeding programs.

9.4 Ethical and data governance concerns

The use of large genomic datasets raises important issues related to data ownership, privacy, farmer consent, and equitable access. Breeding programs must establish clear guidelines to ensure fair use and responsible data sharing, especially when working with local landraces or indigenous genetic resources.

9.5 Need for standardized protocols

Standardized guidelines for genotyping QC, phenotyping protocols, and data annotation are critical for reproducible GP. Without harmonized methods, model transferability across breeding programs remains limited.

Conclusion:

Genomic prediction has emerged as one of the most powerful innovations shaping the future of crop breeding, offering a shift from conventional phenotype-driven selection to a predictive, data-centric approach. By integrating genome-wide molecular markers, high-resolution phenotypic measurements and increasingly diverse multi-omics datasets, GP enables breeders to estimate genetic merit with higher accuracy and at earlier stages than ever before. This accelerates genetic gain, reduces breeding cycle duration, and enhances the development of varieties tailored to emerging climatic, ecological, and nutritional challenges. The rapid advancement of bioinformatics tools has played a pivotal role in operationalizing GP, providing scalable solutions for data preprocessing, genotype imputation, relationship matrix construction, and model bench-marking. Classical statistical frameworks such as GBLUP, Bayesian methods, and RKHS remain foundational due to their interpretability and robustness, while AI and ML approaches extend predictive capacity by capturing nonlinear relationships, gene–environment interactions, and complex trait architectures. The synergy between these statistical and computational paradigms strengthens the reliability and applicability of GP across diverse crops and breeding schemes. The growing use of AI, deep learning, multi-environment trial (MET) analytics, and high-throughput UAV-based phenomics underscores a shift toward holistic, environmentally informed prediction systems. Such integrative pipelines not only model genotype performance across varied environments but also support the development of climate-resilient and resource-efficient cultivars. As breeding programs transition toward data-driven decision

making, the incorporation of multi-omics layers such as transcriptomics, metabolomics, epigenomics will further enhance biological interpretability and predictive precision.

Despite its transformative potential, GP faces challenges related to data quality, model transferability, computational requirements, ethical considerations, and the need for standardized protocols. Addressing these obstacles will require coordinated efforts in capacity building, data governance, and infrastructure development. Nevertheless, the trajectory of GP clearly demonstrates that AI-enhanced, bioinformatics-supported prediction is set to become an indispensable component of next-generation breeding pipelines.

Overall, harnessing AI and bioinformatics tools for genomic prediction represents not merely a technological enhancement but a paradigm shift toward smarter, faster, and more resilient crop improvement strategies capable of meeting global agricultural demands in a changing world.

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