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Methods of Breaking Seed Dormancy and Hybrid Seed Production

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ARTICLE ID: 37

Introduction

Seed has a pivotal role in human and animal nutrition and life. Many of the seeds, after distribution of mother plants or harvest do not germinate in optimal conditions due to a period of dormancy. Seed dormancy is a physiological phenomenon in wild and crop plants, and is more common in wild plants than the crop plants (Farahani *et al.* 2011).Seed is said to be dormant when it is not germinating even though it has the capacity to germinate under favourable environmental conditions (moisture, temperature, light) (Baskin and Baskin 2004). Seed dormancy is defined as the condition of the seed when it fails to germinate even though the favourable environmental conditions are present. Dormancy is defined as any phase in the life cycle of a plant in which the active growth is temporary suspended. According to him, dormancy can be due to unfavourable environmental factors called imposed dormancy or quiescence, while the dormancy due to conditions within the dormant plant or organ is called innate dormancy or rest. Seed dormancy is induced in the seed in the seed development and maturation phase. The environmental conditions (temperature, light, and soil nitrate) faced by the mother plant during seed maturation stage influence the severity of dormancy (He et al. 2014).

The level of seed dormancy is high in freshly harvested seeds and is reduced after a period of dry storage (after ripening) (Née et al. 2017).The main reason behind these conditions is that they require a period of rest before being capable of germination. These conditions may vary from days to months and even years. These conditions are the combination of light, water, heat, gases, seed coats and hormone structures. Lack of seed germination at certain times and suitable conditions are a big problem for seed researchers, botanists, and farmers. Seed dormancy in agricultural ecosystems due to use of inputs and energy is a negative feature. Thus, seed dormancy is a factor for non-uniform seedbed and reduced yield. Seed consumption for sowing also increased, with seed dormancy. There are certain major causes for the seed dormancy. Listed below are the few reasons for the seed dormancy.

- Light
- Temperature
- Hard Seed Coat
- Period after ripening
- Germination inhibitors
- Immaturity of the seed embryo
- Impermeability of seed coat to water
- Impermeability of seed coat to oxygen
- Mechanically resistant seed coat
- Presence of high concentrate solute

Advantages of seed dormancy

1. Dormancy allows the seed to remain in suspended animation without any harm during drought, cold or high summer temperature.
2. The dormant seeds can remain alive in the soil for several years. They provide a continuous source of new plants, even when all the mature plants of the area have died down due to landslides, earth quake, floods or continued drought.
3. It helps the seed to get dispersed over long distances through unfavorable environment.
4. The small seeds with impermeable seed coat belonging to edible fruits come out of the alimentary canal of birds and other animals uninjured e.g., *Guava.*
5. Dormancy induced by the inhibitors present in seed coats is highly useful to desert plants. The seeds germinate only after a good rainfall which dissolves the inhibitors. The rainfall ensures the seed a proper supply of water during the germination.
6. It allows storage of seeds for later use by animals and men.

Causes of dormancy: -

Dormancy can be due to single or combination of many different factors. The various factors of the seed dormancy are:

1. Seed coat induced dormancy: -Seed coat is formed from integuments of the ovule. Chemically it is composed of complex mixture of polysaccharides, hemicelluloses, fats, waxes and proteins. During seed ripening, the seed coat become dehydrated and forms a tough and hard protective covering of the embryo. The seed coat induced dormancy may be due to following causes

- **Water impermeability: -**Impermeability seed coat to water is created by genetic and environmental factors; however, most researchers have shown that this feature is largely hereditary. Seed permeability also affects by environment conditions. Environmental conditions during seed development and maturation, is involved on seed impermeability. In general, climate and soil conditions have a significant impact on the final stages of seed maturation. In Seeds impermeable to water (hard-coated seeds), impenetrability may be due to a cuticle layer or developed a layer of strong epidermal cells. In the seeds permeability of the coat increases slowly in dry stage, due to action of microorganisms like Bacteria and Fungi.
- **Gas impermeability: -**The seed coats of certain seeds are impermeable to gases such as O_2 and CO_2 e.g. ,*Xanthium.* Since O_2 is required for early respiratory activity in germinating seeds, the seed fails to germinate. This is also found in many grasses. Such dormancy of the seed is reduced by storing them for long time. Oxygen is required by the germinating seed for metabolism. Oxygen is used in aerobic respiration, the main source of the seedling's energy until it grows leaves. Some seeds have impermeable seed coats that prevent oxygen from entering the seed, causing a type of physical dormancy which is broken when the seed coat is worn away enough to allow gas exchange and water uptake from the environment (Raven *et al.* 2005; Siegel and Rosen, 1962).
- **Mechanical resistance of seed coat to the growth of embryo: -**The seeds of some common weeds e.g.: *Allisma, Amaranthus and Capsella* etc have such hard and tough seed coats that it prevents any appreciable expansion of embryo. In these seeds water penetrates the seed but enlargement of embryo is limited by mechanical strength of seed coat. As long as the seed coat of *Amaranthus* are saturated with water, the dormancy in *Amaranthus* may persist for about 30 years, however if seed coats

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become dry and then again become saturated with water, they are no longer able to resist the expansion of embryo. At about 40^0C temperature these seeds germinate.

2. Embryo induced dormancy: -It may be due to two reasons:

- **Rudimentary and poorly developed embryo: -**Rudimentary or immature embryo are found in *Fraxinus, Ginkgo, Orchidaceae, Orobanche* etc. at the time of shedding of their seeds. So, these seeds will need a time gap for the development of embryo, then only they germinate.
- **Embryo fully developed but unable to resume growth: -** In many species do not germinate even though their embryos are fully developed and conditions for germination are favorable. Even if seed coats are removed in these seeds still these do not germinate. Dormancy of such seeds is due to physiological conditions of embryo. In these seeds germination is induced if stored in moist well aerated and low temperature conditions called stratification or after ripening.

3. Dormancy due to specific light requirement: - Seeds of certain plants such as *Nicotiana*,*Lactuca* need a specific light treatment for germination. In imbibed *Lactuca sativa,* seed germination is stimulated by red light of 660nm wavelength while it is inhibited by far red light of 730 nm wavelength.In *Rumex* seeds germination promotes with increase in intensity and exposures to light duration of more than 12 hours.These light sensitive seeds are called photoblastic seeds.

4. Dormancy due to germination inhibitors: -A number of chemicals such as Phenolics like Alkaloids, Cyanogenic chemicals (Cyanide releasing substances),Coumarin (unsaturated lactones) Ferulic acid and Abscisin II etc present in seed coat, endosperm, embryos, juice or pulp of fruits (Tomatoes) inhibit seed germination. If these inhibitors are leached out of the seed the germination of seeds takes place.

Methods of breaking seed dormancy: - The dormancy of seeds can be broken and dormant seeds can be induced to germinate by one or combination of more than one method described below:

1. **Scarification: -** Scarification is the process of breaking, scratching, mechanically altering or softening the seed covering to make it permeable to water and gases. Three types of treatments are commonly used as scarification treatments. These include mechanical, chemical and hot water treatments.

a) Mechanical scarification:

- Mechanical scarification is a method for overcoming the effect of an impermeable seed coat. Mechanical scarification can be done by rubbing seeds between two pieces of sandpaper, abrasive, sand or with a severe shaking. Heating, cooling, extreme changes in temperature, dipping seeds in boiling water for a short period of time, piercing the coat of seeds by needle, placing the seed exposed to certain and intermittent wavelengths are other techniques that cause seeds become permeable to air and water.
- It is simple and effective if suitable equipment is available
- For large scale, mechanical scarifiers are used
- Seeds can be tumbled in drums lined with sand paper or in concrete mixers containing coarse sand or gravel. The sand gravel should be of a different size than the seed to facilitate subsequent separation
- Scarification should not proceed to the point at which the seeds are injured and inner parts of seed are exposed

b) Chemical scarification: Seed coat may be removing by treated with chemical materials. Acid treatments are often used to break down especially thick impermeable seed coats. Dry seeds are placed in containers and covered with concentrated Sulphuric acid (H_2SO_4) or HCl in the ratio of one part of seed to two parts of acid.The number of seed treated at any time should be restricted to not more than 10kg to avoid uncontrollable heating. The seeds should be immersed in acid in a glass, china, or earthenware container, and should be stirred occasionally with a glass rod; however, too much stirring will cause the acid to heat undesirably. The seeds must be removed from the acid just before any acid penetrates the seed coats. When the allotted time is finished, the seeds should be removed promptly and washed thoroughly in several changes of water to neutralize completely all remaining acid. For some species the duration of the acid bath depends on the specific batch of seeds and can only be determined empirically. After treatment and a thorough washing, the seeds may be sown or dried and stored for several months (Emery, 1988; Mabundza *et al.* 2010). In new techniques, selected enzymes in the seed coat, such as Cellulase and Pectinase are used for eliminate the seed coat. Also, organic solvents such as alcohol and acetone are used to eliminate seed dormancy.

c) **Hot water scarification**: Drop the seeds into 4-5 times their volume of hot water with temperature ranging from 77 to 100°C. The heat source is immediately removed, and the seeds soaked in the gradually cooking water for 12 to 24 hours. Following this the unswollen seeds may be separated from the swollen seeds by suitable screens. The seed should be sown immediately after hot water treatment.

1. **Stratification: -** This method is employed in the seeds in which dormancy is due to conditions of embryo. In this process the seeds are exposed to well aeriated moist conditions under low temperature (0 to 10°C) for weeks to months.During the stratification some changes occur in seed which are necessary for seed germination.

- The concentration of nitrogen and phosphorous are shifted to the various parts of the seed.
- Various constituent aminoacids, organic acids and enzymes are also shifted.
- The concentration of various growth regulators is changed.

It is done by two methods i.e. cold stratification and warm stratification.

- **Cold stratification:** In this method, seeds are imbibed at low temp (3-5˚C) for a period of time (48 h) and then draining of water and seeds are mixed with moist (sand, sand peat mixture etc.) 2-4 times of their volume and stored at cold temperature 3-5˚C. Biochemical activity due to high moisture and low temp, transforms complicated food material to simple forms that is taken by embryo and micro-organisms activity is also reduced. It is done to break physiological and morphological dormancy. Study shows that cold stratification induces a response to gibberellic acid and initiated cell division resulting in release of physiological dormancy and subsequent germination in *Taxus chinensis* seeds Zhang et al (2013).
- **Warm stratification:** In this method, seeds are imbibed in cold water at 3-5˚C for a period of time (48 h) and then draining of water and seeds are mixed with moist or water retained medium (sand, sand peat mixture etc.) 2-4 times of their volume and stored at warm temperature 20- 25˚C. Partial digestion of the outer layer and final weakening of inner layer results in increased oxygen uptake and energy to embryonic growth, cell expansion, increase GA_3 and deceased ABA level. It is done to break physical, mechanical and morphological dormancy.

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2. **Alternating temperature:-**In some seeds *Poa pratensis* the dormancy is broken by the treatment of an alternating low and high temperatures. The difference between the alternating temperatures should be more than 10 – 20 ^{0}C. This method is done in the seeds where embryo is immature.
3. **Light:-** Light sensitive seeds are called photoblastic. The dormancy of positive photoblastic seeds can be broken by exposing them to red light (660 nm). For red light (730 nm) inhibits the seed germination indicating the involvement photoreversible pigment phytochrome in the proces of seed germination. This pigment occurs in two forms one red absorbing and other far red absorbing. Both these forms are photochemically interconvertible. The red absorbing form (PR) is converted into far red (PFR) after absorbing the red light. The far red form absorbs the far red light and is converted back into red absorbing form of the pigment. It is supposed that in positive photoblastic seeds, the far red absorbing form of the pigment is stimulating to seed germination while red absorbing form is inhibitory to seed germination.
4. **Pressure :-** Seed germination in certain plants like *Melilotus alba* and *Medicago sativa* can be greatly improved after being subjected to hydraulic pressure of about 2000 atmosphere at 10^{o}C for about 15-20 minutes. The pressure changes the permeability of seed coat to water resulting in to seed germination.
5. **Growth regulators:-** Growth regualtors are most widely used to hasten the development of roots in cuttings and to increase the number of roots. Kinetins and gibberellins have been used to induce germination in positively photoblastic seeds like lettuce and tobacco etc. Besides a number of chemcials such as KNO_3, thiourea and ethylene etc have also the capacity to promote seed germination. Many plant regulators are linked with seed germination enhancement and seedling growth in favorable conditions, but the key hormones are gibberellic acid, Indole-3- Acetic-Acid and Kinetin etc. (Faridi et al., 2000). Growth hormones increase in growth potential of embryo, weaken the structure covering the embryo through cell wall hydrolases, counteracts the effect of ABA, reduces photo-D, promotes germination in thermo-D through ABA metabolism, initiation of enzymes, mobilize food substances which

leads to cell division, promotes germination in viable seeds through cell division, cell elongation and embryo growth (Pandiya, 1989).

6. **Seed priming:** It is a pre-sowing treatment of seed for uniform and rapid germination with reduction in germination time. It is applied in annual as well as in perennial crops. In the last 25 years, seed priming is commonly applied in different vegetable and flower species to increase the rate and uniformity of seedlings. Seed treatment also removes or decreases the level of dormancy in dormant seeds.

Seed priming also plays a significant role in improvement of seed germination as well as uniformity of heterogeneously matured seed lots. Priming comprises different categories such as solid matrix priming, hydropriming and osmopriming for breakage of seed dormancy and to enhance seed vigor in many horticultural as well as agronomic crops (Neamatollahi and Darban 2010). Currently, numerous researchers recommended that seeds priming be considered as a useful strategy for good germination and healthier seedling growth resulting in higher yields (Naba'ee et al. 2013). Most widely used seed priming procedures are:

- **Hydropriming Effects on Seed Dormancy and Vigor:-** Hydropriming is more helpful for the breakage of seed dormancy, improvement of seed germination, and seedlings emergence which ultimately results in higher yield. Previous research work confirmed that hydro seed priming greatly depends on type of plant species, water potentiality, priming duration, seed vigor, temperature, humidity and storage conditions and many others. Hydropriming is a very simple, easy, very low cost, less time required and environmental-friendly type of priming (Ghassemi-Golezani et al. 2008). Hydropriming is found to be more useful and efficient type of priming in decreasing the possibility of poor stand establishment in an extensive variation of ecological circumstances.
- **Osmopriming Effects on Seed Dormancy and Vigor**:- Osmopriming is famous as one of the most special types of seed priming which can be used to improve the growth and yield of horticultural and field crops (Ghassemi- Golezani et al. 2008; Neamatollahi and Darban 2010). In osmopriming, seeds were soaking in aerated little water prospective solutions which enhance pre-germination accomplishments to remain continue before actual germination. Seed priming with NaCl increases the morpho-physical, biochemical, and molecular changes in plants which are cultivated

under high salinity stress. The seeds treated with NaCl during sowing showed significant positive effects on seed germination, plant growth, and development of tomatoes. Tomato seeds were primed or treated with PEG-6000 and were found to have significant improvement in germination rates. Moreover, it has been concluded that osmopriming is necessary for better seed germination, which resulted in good plant phenology with excellent crop yield (Khan et al. 2008).

- **Solid Matrix Priming Effects on Seed Dormancy and Vigor:-** Solid matrix seed priming can be successfully used for acceleration of seed germination as well as acts as delivery system for selective fungicides to control insects-pests and soil- borne diseases (Mercado and Fernandez 2002). Okra is a vegetable crop and found to be very more delicate under cool temperature. Solid matrix seed priming has diverse applications for okra seeds. This priming can be more efficiently used for priming of seeds before sowing which have greater capability to enhance seed germination cultivated under cold soils. Seed priming might be used to provide the highest membrane reliability in the embryo and the emerging seedling decreasing seepage with the membranes. Conclusively, solid matrix priming can be used to attain improved seed germination. Numerous types of constituents can be efficiently used for solid matrix priming. However, optimum circumstances for moisture content and seed priming period might be evaluated for every matrix (Mereddy 2015).

Conclusion

Seed dormancy is a prerequisite for preservation and cultivation of crops. Nowadays, seed dormancy and vigor are very much important in crop production. There are many types of dormancy which resist the seed for not germinating under favorable conditions. Seed dormancy occurs due to many factors like environmental factors and hormonal regulations. To overcome this different methods can be potentially used in crop production to increase the uniformity of germination along with better growth and developments.

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