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Biofortification in Millets - A Potential Approach for Nutritional Security

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ARTICLE ID: 06

Introduction

In the present world of ever-increasing population, increased dependencies on cereal based diets has made mankind more vulnerable to increased malnutrition, food scarcity and henceforth nutritional insecurity. Cereal based food systems dominated by rice and wheat are deficient in a number of micronutrients which are otherwise essential requirements for a proper balance diet. In this context millets offer themselves as potential way-out to solve the problems of hunger and malnutrition. In semi-arid tropics and drought-prone regions of Asia and Africa, millets are the potential source of energy after cereals. Millets have better nutritional properties as they contain high amount of proteins, minerals, essential amino acids, and vitamins. Millets are generally known as "small seeded grasses" including pearl millet [*Pennisetum glaucum* (L.) R. Br.], foxtail millet [*Setaria italica* (L.) Beauv], finger millet [*Eleusine coracana* (L.) Gaertn], proso millet [*Panicum miliaceum* L.], kodo millet [*Paspalum scrobiculatum*], barnyard millet [*Echinochloa* spp.], and little millet [*Panicum sumatrense*]. Among these small seeded grasses, pearl millet has major contribution (95 per cent) in production, followed by foxtail millet, which is famous for food in semi-arid tropics of Asia and as forage in Europe, North Africa, North America, and Australia. Finger millet serves as the primary food for rural populations of Africa and Asia and proso millet is cultivated in drier regions of the world. Among the millets, barnyard millet has the shortest harvesting period of six weeks. To address the threat of micronutrient malnutrition, biofortification of millets provides an economically feasible alternative. Biofortification is an effective and sustainable food-based approach which is used to overcome the malnutrition by producing nutrient-rich crops easily available to poor population. However, the presence of antinutrients (phytic acid, polyphenols, and tannins) still hampers the biofortification in millets. But, modern technologies, such as, RNA interference and genome editing tools

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[clustered regularly interspaced short palindromic repeats (CRISPR), zinc finger nucleases (ZFNs), and transcription activator-like effector nucleases (TALENs)],can be used to reduce these antinutrients in millets.

Nutritional Significance of Millets

Millets are nutritionally superior to cereals due to the presence of high amount of essential amino acids,proteins and minerals like iron (Fe), zinc (Zn), phosphorus, calcium (Ca), potassium (K), dietary fibers, and vitamin B. About 80 per cent of millets are used for edible food products as flour, cakes, porridges, *etc*. Other uses of millets involve fodder for animals and in brewing industry for alcoholic products. Due to its high nutritive value, millets are generally considered best for the well-being of lactating mothers, infants, elderly, and convalescents. For instance,pearl millet is a rich source of Fe, Zn, and lysine (17–65 mg/g of protein) compared to other millets. Foxtail millet has a high content of protein (11 per cent), fat (4 per cent) and antioxidants (phenols, phenolic acids, and carotenoids), and is thus considered as an ideal food for diabetic patients. Finger millet grains are rich source of minerals like calcium, magnesium, and potassium and amino acids like methionine, lysine and tryptophan and polyphenols. Barnyard millet is the richest source of crude fiber (13.6 per cent) and iron (186 mg/kg dry matter) meanwhileproso millet has the highest level of proteins (12.5 per cent). Kodo millet is rich in magnesium content (1.1 g/kg dry matter). Therefore, millets serve as multi-grains containing multiple nutrients which are best for collective health benefits.

Starch

Starch in grains of millets is typically made up of two polymers, namely, amylose (15 to 30 per cent) and amylopectin (70 to 85 per cent). Depending on the amylose content, millet genotypes are grouped into two main phenotypes,*i.e.*, waxy and non-waxy. Waxy grains are glutinous in nature withalmost 100 per cent amylopectin and no amylose content, so are easily digestible and hence a great option as food for infants. Waxy proso millet varieties are majorly found in China, East Asia and Japan. The waxy proso millet varietieshave better fermentation efficiency for bioethanol production than non-waxy proso millet varieties. Foxtail millet with waxy grains isgrown in parts of Taiwan, Japan, Indonesia, and Philippines. To transform non-waxy elite varieties into waxy phenotypes, recent technology of genome editing with site-specific nucleases for inducing genetic mutations is a lucrative approach.

Essential Amino Acids

As compared tothe estimated average requirement for lysine (5 per cent) and tryptophan (1.1 per cent), the existing diet based on cereal proteins have very low levels of lysine (1.5–2 per cent)and tryptophan (0.25–0.5 per cent). Millets, on the other hand, such as finger millet, are rich sources of essential amino acids. Therefore, millets can be suitable model crops to study and investigate the genetic control of protein quality.

Minerals

Main focus for mineral biofortification has been on iron and zinc, which are the key limiting mineral micronutrients of people in developing countries with plant-based diets. To develop iron and zinc biofortified pearl millet, a project is being carried out at ICRISAT through selection and conventional breeding. In cultivation trials, a significant correlation among pearl millet iron and zinc contents has been confirmed. However, significant negative correlation between iron content and seed yield has also been found. Underperformance of the F_1 generations of crosses between parents with high zinc and iron contents through lack of realization of expected yields is also a major concern. Finger millet with about five to thirty times more calcium than wheat and rice can be used as a model system to examine seed calcium accumulation.

Vitamins

In developing countries, Vitamin A deficiency in children and pregnant women is a serious threat. Millets have low genetic variation for beta carotene content, precursor of vitamin A. Therefore, traditional breeding is not feasible for enhancing provitamin A content in millets and transgenic technology favouring genetic engineering of vitamin biosynthesis pathways in plants is a successful approach. Research on genetically modified (GM) millets enriched with vitamins is gaining popularity for commercial production due to revised regulatory issues related with the GM crops.

Antinutrients

Bioavailability of minerals in millets is hampered by antinutrients (phytic acid, polyphenols, and tannins) present in grains. High phytate content and polyphenols decreases the bioavailability of iron in millets. Generally, the processes, such as, decortication, fermentation, malting, flaking, roasting, and grinding can remove antinutrients from grains. Reduction in antinutrients during plant growth and development is therefore a promising strategy to improve the bioavailability of minerals from nutrient-rich millets. For producing low phytate crops, three enzymes *viz*., myo-inositol-3-phosphate synthase (MIPS),myo-inositol-3-phosphate 5/6-kinase(MIK), and inositol 1,3,4,5,6- pentakisphosphate 2-kinase (IPK1) are the molecular targets which are expressed in different levels of the biosynthetic pathway. Thus, the modern tools of genome editing have enormous scope in producing biofortified millets with precise alteration of phytic acid biosynthetic pathway.

Breeding Approaches for Biofortification

The micronutrients traits are mostly governed by additive genes and hence, biofortification of crops with conventional breeding approaches is not an easy task. High heritability of micronutrients and less influence of environment on these traits is beneficial for breeding true-to-type lines. Pedigree method is the most basic method in millet breeding which involves the progenies produced from bi-parental crosses and it also uses composites as a base population with the potential to widen the genetic base of cultivars and increase the genetic gains for the yield in hybrids. In case of pearl millet, A1, A4, and A5cytoplasmic sterility systems (CMS) are widely used. The standard three-line system is used to produce hybrids through cytoplasmic genetic male sterility. In India, currently, A1 and A4 are being used widely for commercial biofortification breeding programs in pearl millet to diversify the new cultivar base. At ICRISAT, breeding program for pearl millet biofortification has been followed with three breeding phases-I, II and III. In first phase, genetics of traits, screening of germplasm and creation of genetic variability is done in a short period. Then in second phase, validation of high iron and zinc breeding lines along with hybrid parents is performed to create fast-track biofortified hybrids or varieties. The third phase deals with long-term objective of developing high iron and zinc breeding lines and hybrid parents along with the proper genetic diversification of these lines at NARS and ICRISAT breeding programs. The pearl millet biofortification is currently transitioning between fast-track breeding phase and genetic diversification and mainstreaming phase. This fast-track breeding approach utilizes existing breeding lines with moderate to high content of iron and zinc.

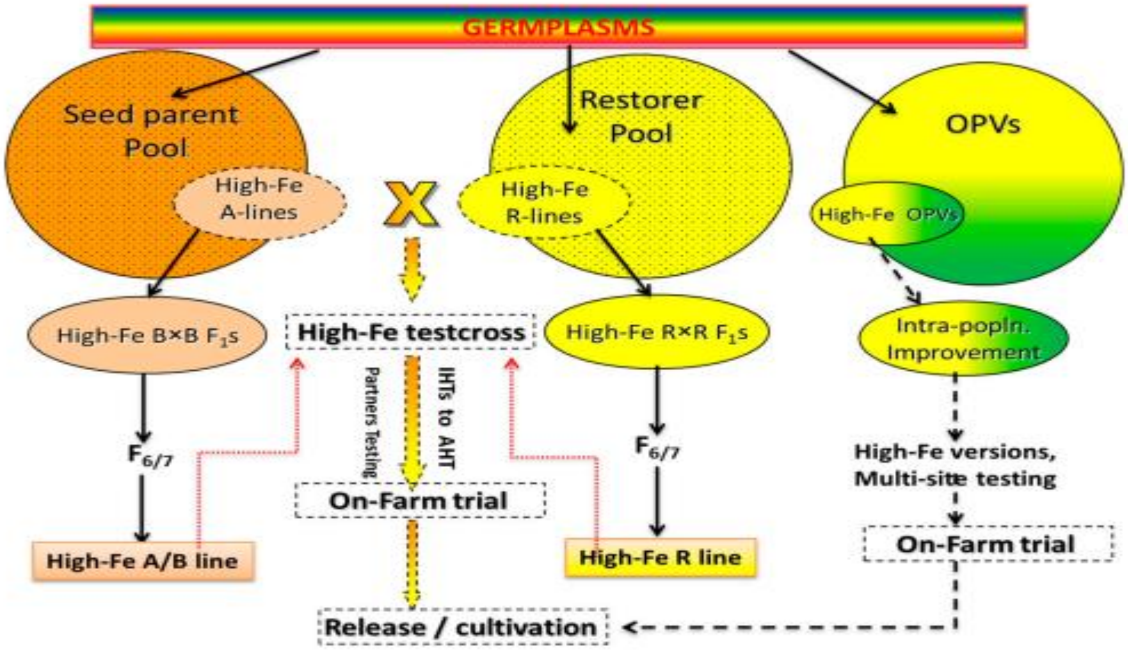

Figure 1. Development of biofortified fast-track hybrids at ICRISAT in India and OPV development in West Africa (Source: Govindaraj *et al*., 2019)

Conclusions

Biofortification is a potential approach to ameliorate the problems of nutritional paucity through the production of nutritionally enriched crops. Shifting the focus of national and international breeding programmes towards production of more nutritionally rich cultivars of major crops with better performance is the need of the hour. Public and private partnerships should create opportunities for elevating the importance ofnutri-cereals and millets on a global scale. Prospectively, genetic biofortification would be a sustainable and cost-effective strategy onwards the enhancement of nutritional value in staple crops. Biofortified cultivars are also being produced by conventional breeding methods, hence foods and grains derived from such cultivars are free from the certain challenges of food regulations and consumer acceptance.Due to the lack of appropriate regulatory mechanism to support biofortification of crops, markets are the only backbone for enhancing processed foods and creating opportunities for farmers. Although, wheat and rice would continue to play significant roles in mitigating the threat of food security,biofortified millets hold inherent potential to overcome the food-cum-nutritional insecurity in dryland poor households.