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Breeding Biofortified Pearl Millet Cultivars

with high iron density

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INDIA has alarmingly high level of micronutrient malnutrition, arising from widespread iron (Fe) and zinc (Zn) deficiencies. About 80% of the pregnant women, 52% of the non-pregnant women, and 74% of the children in the 6-35 months age group suffer from iron deficiency-induced anemia. About 52% of the children below 5 years are zinc deficient. Various interventions such as supplementation, fortification, dietary diversification and biofortification have been advocated to address this problem. Crop biofortification, which refers to the breeding of cultivars with high levels of micronutrients, is increasingly being recognized as a cost-effective and sustainable approach.

Pearl millet biofortification research has shown large genetic variability for Fe and Zn densities in breeding lines and germplasm, and identified several lines and accessions with 90-100 mg/kg Fe density and 70-80 mg/kg Zn density, indicating good prospects of their genetic enhancement. A non-destructive, cost-effective and rapid X-ray Fluorescence Spectroscopy (XRF) screening method has been standardized. Highly significant and very high positive correlations of XRF estimates with those of Inductively Coupled Plasma Optical Emission Spectroscopy (ICP) method for Fe density ($r=0.69$ to 0.97) and Zn density ($r=0.54$ to 0.98) enable reliable mass screening of breeding lines. It has been observed that Fe and Zn densities are predominantly under additive genetic control. Also, there is highly significant and very high positive correlation between these micronutrients (often $r \geq 0.80$), indicating that simultaneous effective selection can be made for both micronutrients. Both micronutrients, in general, have been found uncorrelated with grain weight and flowering time, indicating that pearl millet cultivars with high Fe and Zn densities can be effectively bred with large grain size and in a range of maturity. Fe and Zn densities have been found either uncorrelated with grain yield or show low to moderate negative correlation ($r = 0.29$ to 0.26), but not always significant.

Pearl millet biofortificaion research at the International Crops Research Institute for the Semi-arid Tropics (ICRISAT), conducted in alliance with HarvestPlus and in partnerships with the All India Coordinated Pearl Millet Improvement Project, State Agricultural Universities and private seed companies, prioritized its research in three key areas. First, considering the larger variability for Fe density than Zn deficiency, greater seriousness of Fe deficiency than Zn deficiency in certain populations, and highly significant and positive association between these two micronutrients, major emphasis is on Fe density, with Zn density being improved as an associated trait. Second, since all the public sector programs are predominantly engaged in hybrid development, and seed companies breed only hybrids, the emphasis has been on breeding high-Fe hybrids, with high-Fe open-pollinated varieties (OPVs) as a low priority. Third, since the hybrid breeding India, is largely confined to the A zone of northern India and B zone of peninsular India, receiving more than 400 mm annual rainfall, the biofortificattion research has initially confined its efforts targeted to these two zones.

ICRISAT has developed and disseminated a large number and diverse range of breeding lines and populations over the last four decades, which have been extensively used by breeders, both in public and private sector, for breeding OPVs

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HarvestPlus Biofortification Program of the Consultative Group on International Agricultural Research (CGIAR) has initiated the development and dissemination of improved crop cultivars with elevated levels of these micronutrients in several crops, including pearl millet. Pearl millet, grown on 8-9 million ha in India, is major dryland cereal in the arid and semi-arid regions. It serves as a significant source of dietary energy and contributes 19-63% of the Fe and 16-56% of the Zn intake from all food sources to a vast population in parts of some of the major pearl millet growing states of India.

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and hybrids. As a short and medium term objective, released OPVs, and released and/or commercial, were evaluated to examine the variability for Fe and Zn density, and for possible identification of those with high levels of these micronutrients. In a 7-location trial of 18 released OPVs developed at 9 public sector research organizations, including ICRISAT, the Fe density in these OPVs varied from 42 mg kg to 67 mg kg, and Zn density from 37 mg kg to 52 mg kg with ICTP 8203 having the highest Fe density (67 mg kg) followed by ICMV 221 (61 mg kg) and AIMP 92901 (56 mg kg). While ICTP 8203 had also the highest level of Zn density (52 mg kg), ICMV 221 and AIMP 92901 had 45-46 mg kg Zn density. In a 2-location trial of 122 released/commercial hybrids from 44 research organizations (including seed companies), the Fe density varied from 31 to 61 mg kg and Zn density varied from 32 to 52 mg kg (73 mg kg Fe and 55 mg kg Zn in ICTP 8203 used as a control). Fifteen hybrids with high Fe identified from this trial were re-evaluated at 10 locations to identify those with confirmed high level of Fe density as well as to re-define the base level of Fe for hybrids, based on extensive multilocation data. The Fe density in hybrids varied from 46 mg kg to 56 mg kg and Zn density from 37 mg kg to 44 mg kg. Four hybrids viz., Ajeet 38, Proagro XL 51, PAC 903 and 86M86 had the highest Fe density of 55-56 mg kg and 39-41 mg kg Zn density. Since HarvestPlus has fixed the incremental target of 30 mg kg for Fe density, any hybrid to be called biofortified must have at least 50% of this incremental target above the base level of 42 mg kg, which would be 67 mg kg. These four high-Fe hybrids were close to be what would be called biofortified. Based on the continuing cultivation, 86M86, Ajeet 38 and XL 51 were included in the Nutri-Farm Pilot Program of the Government of India, launched in 2014.

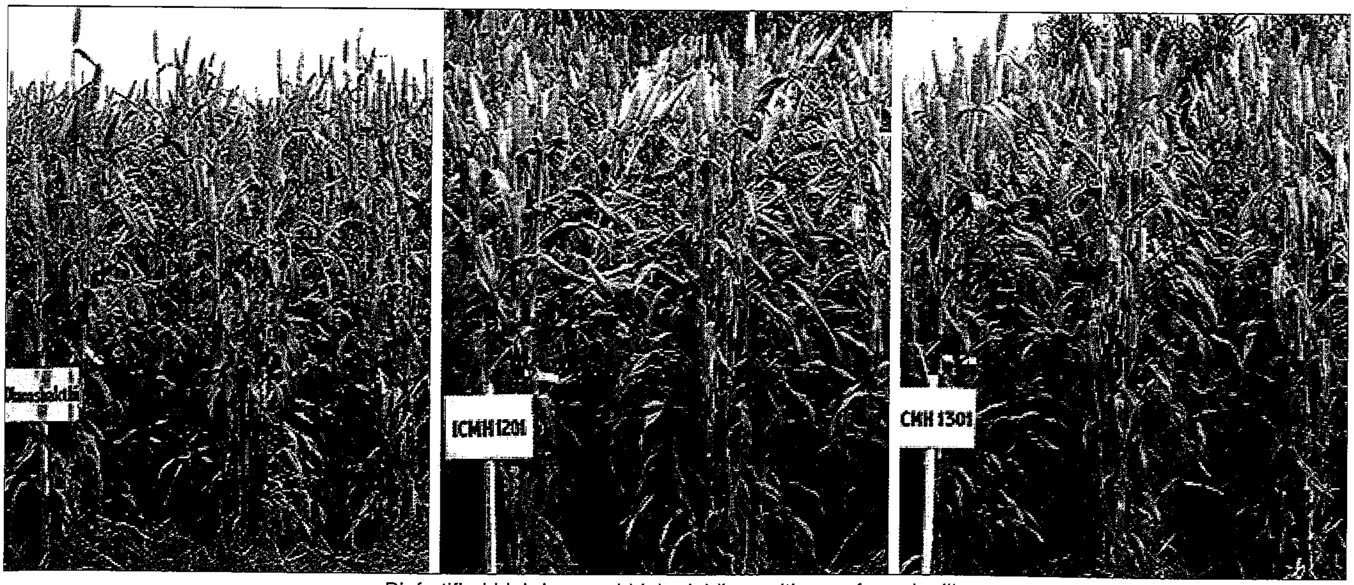
Table 1. Performance of biofortified pearl millet cultivars for Fe and Zn density, grain yield and time to 50% flower in multi-location trials.

Set	Cultivar	Fe density (mg/kg)	Zn density (mg/kg)	Grain yield (t/ha)	Time to 50% bloom (days)
OPV set 1[a]	Dhanashakti	71	40	2.20	45
	ICTP 8203*	65	40	1.97	45
OPV set 2[b]	ICMV 221 Fe 11-2	70	58	4.24	43
	ICMV 221*	63	53	3.98	39
Hybrid set 1[c]	ICMH 1201	75	39	3.58	48
	86M86*	56	37	4.37	54
	ICTP 8203*	71	43	2.58	45
Hybrid set 2[d]	ICMH 1301	77	42	3.26	52
	86M86*	58	37	4.10	54
	ICTP 8203*	75	44	2.46	45

*[a] data from 44 trials during 2010-2011; [b] data from 3 station trials during 2012-2013; [c]data from 48 trials during 2011-2013 and [d] data from 29 trials during 2012-2014; *control cultivar*

ICTP 8203, cultivated as the most popular OPV, was identified having the highest level of both Fe and Zn densities in a series of trials over the years. It was also observed that there was large variability for Fe and Zn densities within this OPV. Utilizing this variability for Fe density, an improved version of ICTP 8203, designated as ICTP 8203 Fe10-2 and released as 'Dhanashakti', was developed that had 71 mg/kg Fe density (9% more than ICTP 8203), and 2.2 t/ha of grain yield (11% more than ICTP 8203), with no changes in Zn density, seed size, flowering time and other traits. This indicates that high-Fe OPVs identified in this study can be further improved for Fe density. Variability for Fe and Zn density was observed in another popular and high-Fe OPV (ICMV 221), and utilization of this variability led to the development of its -Fe version (ICMV 221 Fe-11-2), which had 70 mg/kg Fe density (11% higher than ICMV 221). This improved version also 58 mg/kg Zn density (9% higher than ICMV 221), and 4.24 t/ha grain yield (7% higher than ICMV 221), but it flowered 4 days later than ICMV 221.

ICRISAT has developed and



Biofortified high-iron and high-yielding cultivars of pearl millet disseminated more than 180 seed parents (A/B pairs) so far, which had not been bred for Fe density as a target trait. Among these, 12 A/B pairs have been identified having high Fe density (64-77 mg/kg), and ICMA 98222 and ICMA 99222 were found to particularly promising with respect to Fe density, yield potential and other agronomic traits. Utilizing, these two A-lines and advanced high-Fe breeding lines as potential restorers, high-Fe hybrids have been developed with good yield potential. Two of these (ICMH 1201 developed on ICMA 98222 and ICMH 1301 developed on ICMA 99222) have been most widely tested (Table 1). Based on the performance over 48 field trials, ICMH 1201 had 75 mg/kg Fe density (similar to ICTP 8203) but had 3.6 t/ha grain yields (38% higher than ICTP 8203). ICMH 1201 flowered only 3 days later than ICTP 8203. ICMH 1301 tested in 32 trials had 77 mg kg Fe density (similar to ICTP 8203) and 3.3 t ha grain yield (33% higher than ICTP 8203). It is significant to note that almost all the high-Fe cultivars (both OPVs, and hybrids) has shown >40 mg/kg of Zn density, exceeding 37 mg/kg of target level fixed for biofortified high-Zn wheat cultivar development.

As compared to the most popular and highest yielding hybrid 86 M 86, which also had the highest Fe among the commercial hybrids, both these biofortified hybrids had 33-34% higher Fe density. The biofortified hybrids had 18-20% less grain yield compared to 86 M 86, but these flowered 2-6 days earlier than 86M 86. Dhanashakti has now been adopted by more than 60,000 farmers, mostly in Maharashtra. Truthfully labeled seed (TLS) production of ICMH 1201 was undertaken by Shaktivardhak Seed Company in 2014 for commercialization under its brand name Shakti-1201 and it has been adopted by 35,000 farmers, mostly in Maharashtra and Rajasthan. Much greater progress in breeding high-Fe hybrids with high-grain yield is expected in the near future by utilizing A- and R-lines that are being developed through targeted breeding for high-Fe density.

Pearl millet studies have shown that Fe and Zn densities are either not correlated with grain yield, or that there are negative and moderate to low correlations (not always significant). These results considered along with the relative performance of improved higher versions of ICTP 8203 and ICMV 221 with their original versions, and hybrid performance for grain yield and Fe density presented above shown that pearl millet cultivars that combine high Fe density and high grain yield can be developed. A question generally asked is about the bioavailability of biofortified high-Fe cultivars compared those with low Fe density. Three efficacy feeding trials including children below three years of age from Benin (Africa) and Karnataka (India) and 12–16 years age from Maharashtra in India, and non-pregnant non-lactating (NPNL) women in 17-35 year age group in Benin (Africa), have shown that the bioavailability of Fe in pearl millet is 7.0-7.5%, regardless of their density levels in grains. Thus, the consumption of whole grain products made from, say Dhanashakti and the two biofortified hybrids would provide 17 mg day^{-1} of Fe which would fully meet the recommended daily allowance (RDA) in adult men and 80% of the 21 mg RDA in the NPNL women. Considering 7% bioavailability, this Fe intake, in turn, would make 1.2 mg Fe bioavailable in men (far above the daily requirement of 0.84 mg), and would meet 72% of the 1.65 mg daily requirement of NPNL women.

SUMMARY

Iron and zinc deficiencies are widespread and serious public health problems worldwide, including India. Biofortification is a cost-effective and sustainable agricultural strategy to address this problem. Research on pearl millet has shown that large genetic variability available in this crop can be effectively utilized to develop high-yielding cultivars with high iron and zinc densities. Both Open–pollinated varieties and hybrids of pearl millet with high grain yield and high levels of iron and zinc densities have been developed. While some of these have been adopted and are under cultivation, others are at various stages of testing in the station and national trials.

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