



Nutritional quality improvement in maize (*Zea mays*): Progress and challenges

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ABSTRACT

Malnutrition is one of the important problems which affect the overall human productivity costing huge economic losses to the nations. Addressing malnutrition problem is one of the important components of sustainable development goals. In this context, biofortification of staple food crops could be one of the most practical, environment friendly, cost effective and sustainable approaches in the long run. Maize (*Zea mays* L.) being staple food crop for more than 900 million populations across the globe, enhancing the nutrient content along with yield is of paramount importance. Maize display large genetic diversity for all the quality parameters and several mutants are available each of the quality traits. Across the globe, several efforts have been made to identify new gene(s) and QTLs for different quality traits and their mobilization to develop new and improved biofortified cultivars. However, any technology or product remains meaningless unless it reaches the main stakeholders. The main stakeholders are the poorest of poor of the society who are most affected due to malnutrition. In order to make biofortification a success story, there is need to address several challenges like appreciated support price for the produce, dedicated production zones, value addition and supply chain development. The policy intervention with respect to sensitization on importance of nutrient rich cultivars and their acceptance by farmers, traders and consumers are the key to success.

Key words: Genes, Maize, Mutant, Policy, Quality

Maize (*Zea mays* L.) is the most imperative cereal crop worldwide with the highest global production of 1060 million tonnes (FAOSTAT 2017). It is used as food in humans, feed for poultry and livestock, and raw material for an array of industrial- and processed-products (Yadav *et al.* 2015). More than 900 million people depend on maize for their staple food around the world particularly in the Latin America, Africa and Asia including India (Shiferaw *et al.* 2011). Maize provides 62% of the proteins from all cereals in Meso America, while it is 43% in Eastern and Southern Africa, 28% in Andean Region, 22% in West and Central Africa and 4% in South Asia (Hossain *et al.* 2018).

Malnutrition has emerged as one of the major problems especially in under-developed and developing countries of the world (Bouis and Seltzman 2017). The resource-poor suffers the most from 'hidden hunger', a term more often

used to describe malnutrition. Approximately two billion people are being short of essential micronutrient like iron and vitamin A in their daily diet at global level (Global Nutrition Report 2017). Nearly 45% of deaths of children under age of five are linked to malnutrition (Black *et al.* 2013). Malnutrition contributes to global burden of disease, and loss in annual GDP in Asia and Africa to the extent of 11% (IFPRI 2016).

Considering the paramount importance of balanced nutrition, global community has set 'Sustainable Development Goals' (SDGs) to chart a path towards meeting current human needs without compromising the ability of future generations to meet their needs. Of the 17 goals, 12 contain indicators that are highly relevant to nutrition, reflecting central role of nutrition in sustainable development. Improved nutrition is the platform for progress in health, education, employment, female empowerment, and poverty elimination. It has been estimated that alleviating malnutrition is one of the most cost-effective steps with every \$1 invested in proven nutrition programme offers benefits worth \$16 (IFPRI 2016). Thus, efforts directed towards providing the balanced and nutritious food assumes great significance (Zunjare *et al.* 2018, Sarika *et al.* 2018).

Agricultural systems have traditionally focussed mostly on increasing productivity. However, now research policies must focus that not only provide enough calories to

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meet the energy needs of the poor, but also deliver all the essential nutrients needed for adequate nutritional health. 'Biofortification' a process in which micronutrient density in crops is increased through plant breeding, is proposed as a sustainable and cost-effective mean for providing the required levels of nutrition in natural form to alleviate malnutrition in humans (Gupta *et al.* 2015). Among various micronutrients, lysine, tryptophan, vitamin A, vitamin E, iron (Fe) and zinc (Zn) have remained deficient in endosperm among traditional maize varieties.

Maize is also considered as a crop of industrial importance due to its wide utilisation in industry as raw material and also its role in the world economy and trade (Yadav *et al.* 2015). Maize grains are rich source of carbohydrate which possesses diverse usage as an industrial raw material. Sticky maize or high amylopectin maize is a popular choice in South-East Asian countries (Devi *et al.* 2017). Corn oil is also gaining popularity due to desirable fatty acid composition; rich source of linoleic acid (18:2), oleic acid (18:1), palmitic acid (16:2), steric acid (18:0), small amounts of linolenic acid (18:3), and trace amount of other fatty acids. Further, as compared to other edible oils, maize oil has the advantage of being low in the proportion of mono-saturated fatty acids (Rakshit *et al.* 2003). Besides, specialty corns like sweet corn, baby corn and pop corn have become popular choice worldwide (Mehta *et al.* 2017, Yadav *et al.* 2015). Thus, development of biofortified maize cultivars has huge potential in alleviating malnutrition problem at global level as huge natural variation in the form of mutants or otherwise, existed in maize for several nutritional traits like provitamin A, vitamin E, high-lysine and -tryptophan etc. . In this review, we report availability of different mutants, their effects on target traits, utilization in the breeding programme followed by the challenges for their dissemination.

Genetic variation for nutrient content in maize

The carbohydrates, proteins, fats, vitamins, minerals, fiber and water are the main nutrients required to fulfil daily needs of human body (Welch and Graham 2004). Considerable variations are reported in maize germplasm for different nutrients. These nutritional components are essential for growth, development, immunity, reproduction, metabolism and other physiological functions.

The important source of energy which converts the glucose into energy is starch, which varies from 59.60 - 74.40% in maize kernel (Cook *et al.* 2012, Guo *et al.* 2013, and Yangcheng *et al.* 2013). The kernel energy density is about 365 kcal/100 g, which is close to rice (360 kcal/100g) and wheat (340 kcal/100g) (USDA Natl. Nutrient Database, <https://ndb.nal.usda.gov/ndb/>). Assuming 90% energy availability, an average male requires a daily energy requirement of 2800 kcal (Mertz 1970, Brown *et al.* 1988). That means if total energy requirement is to be met from maize nearly 600 g is needed per day. The starch is composed of amylase and amylopectin and the variations for amylose and amylopectin have been reported upto a maximum

of about 80 and 100%, respectively (Hallauer 2000). Considerable variations are observed in maize germplasm for different nutrients (Table 1). The protein which plays a major role in enzymatic and hormonal activities, content in maize kernel ranges from 4.50-13.24% (Enyisi *et al.* 2014, Ai and Jane 2016, Pedersen *et al.* 2014, Cong *et al.* 2015, Butts-Wilmsmeyer *et al.* 2017). Essential amino acid lysine required for proper growth and muscles development range from 0.16-0.86% in maize kernel (Tang *et al.* 2013, Reddy *et al.* 2013, Cong *et al.* 2015, Bjarnason and Vasal 1992, Vivek *et al.* 2008). Similarly, other two essential amino acids, tryptophan and methionine range from 0.02-0.074% (Cong *et al.* 2015, Bjarnason and Vasal 1992, Vivek *et al.* 2008) and from 0.15-0.37% (Tang *et al.* 2013, Lai and Messing 2002), respectively. Corn oil having major role in improving the availability of fat soluble vitamins and carotenes, varies between 1.4 and 6.0%. In high oil Illinois lines oil content up to 15% is reported (Lambert *et al.* 1998, Enyisi *et al.* 2014, Tang *et al.* 2013, Cong *et al.* 2015, Ai and Jane 2016).

β-carotene, a precursor of vitamin-A having significance in terms of vision, immunity and reproduction, ranges between 3.4-21.7 mg/kg in maize kernel ((Muthusamy *et al.* 2014, Pillay *et al.* 2014). Vitamine-E plays important role as antioxidant, improving immune responsiveness and prevention of oxidation of polyunsaturated fatty acids (PUFA) which is present in maize kernel approximately in the range of 4.6-30 mg/kg (Li *et al.* 2012). The iron, which plays an important role as a catalyst in transporting the oxygen to red blood cells (Fe) content varies from 11.28-83.35 mg/kg in maize kernel (Agrawal *et al.* 2012, Prasanna *et al.* 2011, Mallikarjuna *et al.* 2014, Chakraborti *et al.* 2011b). Zn, an integral part of different enzymes involved in synthesis and degradation of carbohydrates, protein and lipids, range from 3.81-52.95 mg/kg in maize kernel (Chakraborti *et al.* 2011a, Prasanna *et al.* 2011, Guleria *et al.* 2013, Mallikarjuna *et al.* 2014). The anti-nutritional factor, phytic acid is a strong chelator of Fe²⁺ and Zn²⁺ *in-vivo* and lead to an insufficient bio-availability of Fe and Zn (Hunt 2003). Konietzny and Greiner (2003) reported the range of phytic acid from 0.68-14.2 mg/g in maize kernel on the dry weight basis.

Discovery of mutants influencing maize quality traits

In maize, the *waxy1*, the first gene influencing kernel quality was identified by Collins and Kempton in 1913. It is recessive in nature and influences kernel type with specific phenotype of dull and waxy-like appearance. Subsequently many additional genes modifying kernel appearance, including *ae1*, *su2*, *fl1*, *fl2*, *du2*, *o2* and *bt1* were identified through genetic analysis (Hutchinson 1921, Mangelsdorf 1923, 1926). During the initial era of maize genetics, genes were identified based on the distinct morphological characters conditioned by the mutant genotype. Allelic relationship has been determined using classical genetic complementation experiment and new mutants have been assigned as novel ones or allelic to existing mutant (Coe

Table 1. Breeding target and genetic variation for different nutrients in traditional maize

Nutrients	Breeding Target	Range	References
Protein	9-11%	4.50-9.87% 6.0-12% 7.5-9.1% 5.0-10.8% 8.34-13.24%	Enyisi <i>et al.</i> (2014) Ai and Jane (2016) Pedersen <i>et al.</i> (2014) Cong <i>et al.</i> (2015) Butts-Wilmsmeyer <i>et al.</i> (2017)
Lysine	2.5%	0.73-0.86% 0.38-0.58% 0.21-0.38% 0.16-0.26%	Tang <i>et al.</i> (2013) Reddy <i>et al.</i> (2013) Cong <i>et al.</i> (2015) Bjarnason and Vasal, (1992), Vivek <i>et al.</i> (2008)
Tryptophan	0.60%	0.036-0.074% 0.02-0.06%	Cong <i>et al.</i> (2015) Bjarnason and Vasal (1992), Vivek <i>et al.</i> (2008)
Methionine	0.5%	0.15-0.17% 0.20-0.37%	Tang <i>et al.</i> (2013) Lai and Messing (2002)
Oil	6%	2.17-4.43% 4.93-5.62% 1.4-5.0% 3.0-6.0%	Enyisi <i>et al.</i> (2014) Tang <i>et al.</i> (2013) Cong <i>et al.</i> (2015) Ai and Jane (2016)
Starch	73%	67.10-74.40% 59.60-73.00% 66.6-74.1%	Guo <i>et al.</i> (2013) Cook <i>et al.</i> (2012) Yangcheng <i>et al.</i> (2013)
Provitamin-A	15 mg/kg	3.4-21.7 mg/kg	Pillay <i>et al.</i> (2014), Muthusamy <i>et al.</i> (2014), Choudhary <i>et al.</i> (2014)
Tocopherol	15 mg/kg	4.6-14.8 mg/kg	McDonald <i>et al.</i> (1998), Li <i>et al.</i> (2012), Egesel <i>et al.</i> (2003)
Fe	60 mg/kg	12.5 -19.7 mg/kg 20.38-54.29 mg/kg 11.28-60.11 mg/kg 16.61-83.35 mg/kg 13.95-39.31 mg/kg	Tang <i>et al.</i> (2013) Agrawal <i>et al.</i> (2012) Prasanna <i>et al.</i> (2011) Mallikarjuna <i>et al.</i> (2014) Chakraborti <i>et al.</i> (2011b)
Zn	38 mg/kg	12.5-20.9 mg/kg 17.57-49.14 mg/kg 21.85-40.91 mg/kg 15.14-52.95 mg/kg 3.81-35.83 mg/kg 14.27-53.20 mg/kg	Tang <i>et al.</i> (2013) Chakraborti <i>et al.</i> (2011a) Chakraborti <i>et al.</i> (2011b) Prasanna <i>et al.</i> (2011) Guleria <i>et al.</i> (2013) Mallikarjuna <i>et al.</i> (2014)
Phytic acid (antinutritional factor)	1.1 mg/g	11.5-14.2 mg/g dry weight 0.68-1.5 mg/g	Konietzny and Greiner (2003) Cong <i>et al.</i> (2015)

1985, Jha *et al.* 2016). Genetic analysis using these mutants led to development of detailed genetic maps, which were further enriched with biochemical and molecular markers for identification and localization of genes governing quality traits in the linkage maps (Yang *et al.* 2005, Yan *et al.* 2010). Different endosperm mutants with enhanced quality traits as reported by the researchers have been presented in Table 2.

Maize endosperm is constituted primarily of starch rich tissues that support embryo at germination and hence determines its nutritional quality (Balconi *et al.* 2007). The protein is mainly stored in maize endosperm as a group of prolamins, known as zeins. Zeins are synthesized on rough endoplasmic reticulum (ER) membranes and accumulate in the ER as insoluble accretions called protein bodies (Larkins and Hurkman 1978). Certain mutants altering zein synthesis lead to protein bodies with abnormal morphology, size,

number and result in kernels with a soft and starchy texture. Mutations reducing α -zein synthesis, such as *opaque2* (*o2*) (Mertz *et al.* 1964), results in small unexpanded protein bodies (Geetha *et al.* 1991). Mertz *et al.* (1964) reported that the maize endosperms homozygous for the *o2* mutant recorded different amino acid pattern than the normal maize kernel and have 69% more lysine. Than *o2* mutant was identified in W22 inbred and located on chromosome 7L. Another important mutant *opaque16* (*o16*) was identified in China. The QCL3024 (*o16*) and QCL3021 (*o16*) lysine mutant lines having opaque endosperm were derived from a self-cross population isolated from Robertson's Mutator stock. Two F_{2:3} populations were developed, one from a cross between QCL3024 and QCL3010 (a wild type line) and another from a cross between Qi205 (*o2*) and QCL3021 and evaluated for lysine content. The distributions indicate

Table 2 Details of different mutants/genes for enhancement of quality traits

Mutants/Genes/ QTLs	Inheritance pattern	Phenotypic effect	Enzymatic activity/Mode of action	Chr.	Source	References
<i>Protein</i>						
<i>Opaque-1 (o1)</i>	Recessive	The amount of non-zein protein in <i>o1</i> was nearly identical to that in the wild type, no significant increase in lysine content.	Encodes a Myosin XI Motor Protein that affects protein body formation by disrupting ER morphology and motility	4L	W64A <i>o1</i>	Hunter <i>et al.</i> 2002
<i>Opaque-2 (o2)</i>	Recessive	Different amino acid pattern, higher lysine per cent, reduced zein to glutenin ratio	Encodes a defective basic-domain-leucine-zipper transcription factor, regulates several endosperm-expressed genes, particularly in 22-kDa α -zeins.	7L	W22	Mertz <i>et al.</i> 1964; Schmidt <i>et al.</i> 1990
<i>Opaque-5 (o5)</i>	Recessive	The amount of non-zein protein was nearly identical to that in the wild type, nearly 1. 4% higher lysine per cent.	-	7L	W64A <i>o5</i>	Hunter <i>et al.</i> 2002
<i>Opaque-6 (o6)</i>		Relatively high in lysine content, showed shift in zein to glutenin ratio, increase in non-protein nitrogen albumins and insoluble proteins	-	8L	<i>O6</i> mutant	Ma and Nelson 1975
<i>Opaque-7 (o7)</i>	Recessive	Reduction in α -zein protein synthesis and the formation of protein bodies that are significantly smaller than normal	Encodes an acyl-activating enzyme-like protein that influences amino acid and zein protein synthesis	10L	W22	Wang <i>et al.</i> 2011
<i>Opaque-9 (o9)</i>	Recessive	Nearly 1. 4% higher lysine content	-	5L	W64A <i>o9</i>	Hunter <i>et al.</i> 2002
<i>Opaque-10 (o10)</i>	Recessive	Controls Protein Bodies morphology in maize endosperm.	Encodes a novel cereal-specific Protein Bodies protein, is essential for the ring-shaped distribution of 22-kD and 16-kD zeins		<i>o10-N1356</i>	Yao <i>et al.</i> 2016
<i>Opaque-11 (o11)</i>	Recessive	Increased non-zein protein, 1. 8 times higher lysine	-		W64A	Hunter <i>et al.</i> 2002
<i>Opaque-12 (o12)</i>	Recessive	Thin, varied size, scarred mutant kernels that produce chlorophyll-deficient plants.	-	4S	ox-7638	Nelson 1981
<i>Opaque-13 (o13)</i>	Recessive	The mutant kernels are etched and may have a thin rim of corneous starch on the abgerminal side	-	1S	ox-7729	Nelson 1981
<i>Opaque-15 (o15)</i>	Recessive	2- to 3-fold reduction in γ -zein mRNA and protein synthesis	Alters the ratio of mRNAs encoded by the A and B γ -zein genes	7L	MGN-25:969-5	Dannenhoffer <i>et al.</i> 1995
<i>Opaque-16 (o16)</i>	Recessive	<i>o16</i> along with <i>o2</i> increases lysine by 30% over <i>o2o2</i> or <i>o16o16</i> alone	-	8L	Robertson's Mutator stocks	Yang <i>et al.</i> 2005
Mutator-tagged <i>opaque-140 (mto140)</i>	Recessive	mto140/arodh-1 seeds shows a general reduction in zein storage protein accumulation and an elevated lysine due to disruption in amino acid biosynthesis.	-		W64A (BC6)	Holding <i>et al.</i> 2010
<i>Floury-1 (fl-1)</i>	Semi-dominant	Mutant fl-1 do not manifest significant increase in Lysine as <i>o2</i> but increase the methionine content.	Encodes an ER membrane protein involved in facilitating the localization of 22-kD α -zein in the protein bodies	2L	W64Afl1-Mu1	Hays and East 1915

Contd.

Table 2. (Continued)

Mutants/Genes/ QTLs	Inheritance pattern	Phenotypic effect	Enzymatic activity/Mode of action	Chr.	Source	References
<i>Floury-2 (fl-2)</i>	Semi-dominant	Formation of a soft, starchy endosperm with a reduced amount of prolamin (zein) proteins and twice the lysine content of the wild type.	Associated with small, irregularly shaped protein bodies, elevated levels of a 70-kDa chaperone in ER and a novel 24-kDa polypeptide in zein fraction.	4S	W64A/fl2	Nelson <i>et al.</i> 1965; Coleman <i>et al.</i> 1997
<i>Floury-3 (fl3)</i>	Semi-dominant	Relatively high in lysine content, showed shift in zein to glutenin ratio.	-	8L	fl-3	Ma and Nelson 1975
<i>Floury-4 (fl4)</i>	Semi-dominant	Reduction in α -zein protein synthesis and the formation of protein bodies that are significantly smaller than normal	Defective signal peptide in a z1A 19-kD α -zein	4S	5512G mutant x W22	Wang <i>et al.</i> 2014
<i>Mucronate (Mc)</i>	Dominant	1. 4 times higher lysine.	Mc encodes a 16-kD γ -zein with a frame shift mutation	2L	B37Mc W64A	Salamini 1981; Hunter <i>et al.</i> 2002
Defective endosperm B30 (<i>De-B30</i>)	Dominant	Increased non-zein protein, 1. 8 times higher lysine	De-B30 is a 19-kD α -zein with a single amino acid replacement resulting in a defective signal peptide	7S	B37De *-30 W64A	Salamini 1981; Hunter <i>et al.</i> 2002
<i>Methionine</i>						
<i>Zps10(22)</i>	-	<i>Zps10(22)</i> is structural gene and <i>Zpr10(22)</i> is regulatory gene	Overproduction of the zein protein by trans-acting mechanism	9 & 4	BSSS53, W23 and B73	Benner <i>et al.</i> 1989
<i>dze1</i>		30% phenotypic variability explained	Overproduction of a methionine-rich, 10-K zein	4	(MO17 $\times$ BSSS53) $\times$ MO17	Chaudhuri and Messing 1995
<i>Starch</i>						
amylose extender1 (<i>ae1</i>)	Recessive	66% high amylose than normal endosperm	Branching enzyme II	5	A636 $\times$ B73	Vineyard and Bear 1952
brittle1 (<i>bt1</i>)	Recessive	Shrunken endosperm of brittle texture, slightly darker than normal	Adenylate translocator	5	Improved Learning and Sanford white	Mangesdorf 1926
brittle-2 (<i>bt2</i>)	Recessive	Shrunken endosperm of brittle texture, slightly darker than normal	ADP-Glc pyrophosphorylase	4	-	Teas and Teas 1953
dull1 (<i>dul1</i>)	Recessive	42% high amylose than normal endosperm	Starch synthase II	10	Surcropper	Mangesdorf 1947
shrunken1 (<i>sh1</i>)	Recessive	Highly collapsed, opaque and brittle and have a weight of 75% that of normal endosperms	Sucrose synthase	9	Kansas	Hutchison 1921
shrunken-2 (<i>sh2</i>)	Recessive	Shrunken, opaque to translucent	ADP-Glc pyrophosphorylase	3	Natural mutant	Mains 1949

Contd.

Table 2. (Concluded)

Mutants/Genes/ QTLs	Inheritance pattern	Phenotypic effect	Enzymatic activity/Mode of action	Chr.	Source	References
shrunken-4 (<i>sh4</i>)	Recessive	Shrunken and opaque	-	5	Sh4/Sh4	Tsai & Nelson 1969
sugary1 (<i>su1</i>)	Recessive	Increased amounts of amylose and intermediate fractions compared with normal starch.	Isoamylase	4	Natural mutant	Correns 1901
sugary-2 (<i>su2</i>)	Recessive	Slightly tarnished to tarnished	-	6	-	Eyster 1934
waxy-1 (<i>wx1</i>)	Recessive	Increases sugars and water-soluble polysaccharides	Starch granule bound starch synthase	9	Sanford white	Collins 1909
<i>Provitamin-A</i>						
<i>Y1 (P_{sy1})</i>	Dominant	conversion of white maize to yellow/orange	<i>Y1</i> gene encodes phytoene synthase which is synthesizes the carotenoids	6L	Q60	Buckner <i>et al.</i> 1990
<i>crtRB1</i>	Recessive	7-27% phenotypic variability explained	Hydroxylation of α - and β -carotene into non-provitamin A carotenoids	10	B73 $\times$ BY804	Yan <i>et al.</i> 2010
<i>lcyE</i>	Recessive	-	Converts more lycopene to the β , ϵ branch and produce α -carotene and lutein	8	Q $\times$ 47	Harjes <i>et al.</i> 2008
<i>Vitamin-E</i>						
<i>VTE4</i>	Recessive	Increase 3. 2 fold vit. E compare to normal maize	Encodes γ -tocopherol methyltransferase which is involved in the rate limiting conversion of γ T to α T	6L	Q60	Li <i>et al.</i> 2012
<i>Low phytic acid</i>						
<i>lpa1-1</i>	Recessive	66% reduction in seed phytic acid phosphorus	Reduced phytate matched solely by increased inorganic P. Viable	1S	M ₂ progeny of 90046-13	Raboy <i>et al.</i> 2001
<i>lpa2-1</i>	Recessive	50% reduction in seed phytic acid phosphorus	Reduced phytate matched by increased inorganic P and increases in other Inositol phosphates. Viable but plant and seed effects.	1S	M ₂ progeny of 90041-1, 90041-4, and 90041-12	Raboy <i>et al.</i> 2001
<i>lpa241</i>	Recessive	90% reduction of phytic acid and about a tenfold increase in seed-free phosphate content	“Ins-supply” pathway and the later Ins phosphate/Ptd Ins-phosphate pathway that converts Ins to InsP6	1S	ACR seed stock (mutant)	Pilu <i>et al.</i> 2003
<i>lpa3</i>	Recessive	50% less phytic acid than the corresponding wild-type	Role in myo-inositol and MIK in phytic acid biosynthesis in developing seeds	1S	M ₂ progeny lpa-061326	Shi <i>et al.</i> 2005

that the lysine content in the two populations is regulated by the major gene of *o16* and genes of *o2* and *o16* (double mutant), respectively. The *o16* alone possesses lysine and tryptophan that are as high as *o2* and it does have influence on opacity (Sarika *et al.* 2016). The pyramiding of *o16* mutant with *o2* led to higher accumulation of lysine and tryptophan (Yang *et al.* 2005, Sarika *et al.* 2018). A new mutant, *opaque15* (*o15*), reported 2-3 fold reduction in γ -zein mRNA. On phenotype basis, *o15* appears to be a mutant of an *o2* modifier gene (Dannenhoffer *et al.* 1995). The mutant floury-2 (*fl2*) identified by Nelson *et al.* (1965) had lysine content equal to mutant *o2*. But it also had enhanced methionine concentration than in any other stock tested. Likewise several mutants have been identified that can alter the zein synthesis and increase the protein in maize. The major reason for these changes is the synthesis of proteins with a greater content of basic amino acids in the acid-soluble fraction of the mutant endosperm. This is accompanied by a reduction in the ratio of zein to glutelin.

In contrast to above mentioned recessive mutations, a dominant mutation DeB-30 influencing protein quality is also reported in maize (Salamini *et al.* 1979). It contains 50% more lysine than the normal maize but linked with reduction of seed weight limiting its practical utilization in breeding for quality improvement. Another dominant mutant *Mc* (Salamini *et al.* 1983) interferes with the synthesis of storage proteins in the endosperm and results in enhanced level of methionine. Several endosperm mutants (at least 18 such mutants) effecting kernel phenotypes (brittle texture) and grain quality (susceptibility to insect pests, and inferior functional characteristics of products made from their flour) by altering maize starch have been identified, but only for very few mutants, molecular basis of the mutation is well characterized (Hunter *et al.* 2002).

Several mutants affecting starch synthesis pathway can alter the level of amylose and amylopectin as well as sugar content in the kernel. The mutants, *viz.* *sugary 1* (*su1*), *shrunk 1* (*sh1*), *amylose extender 1* (*ae1*), *brittle 1* (*bt1*) and *waxy 1* (*wx1*) were discovered by Correns (1901), Hutchison (1921), Vineyard and Bear (1952), Mangesdorf (1926) and Collins (1909), respectively. Among these, *su1* and *sh1* have been extensively used worldwide for development of sweet corn cultivars. Mutant *su1* is a mutated version of gene influencing starch debranching enzyme. This enzyme is responsible for enhancing the water soluble polysaccharides, reducing sugars and sucrose in milky to ripening stage and decrease starch accumulation in mature kernels, resulting in sweet kernels (East and Hayes 1911, Dinges *et al.* 2001). Recessive mutant, *se 1* is a modifier of *su 1* and enhances sugar level in maize kernels (Ferguson *et al.* 1978). Another gene for sweetcorn, *sh2* as identified by Mains (1949), encodes the large subunit of the starch biosynthetic gene, adenosine diphosphate glucose pyrophosphorylase (AGPase). This enzyme plays a crucial role in starch biosynthesis (Hannah and Nelson 1976, Bhave *et al.* 1990, Lee *et al.* 2009, Hannah *et al.* 2012). Among the genes influencing sugar content *su1* and *sh2* have been used

quite extensive in sweet corn breeding, however relatively few studies have been conducted on combining *su1* and *sh2* in a single genetic background. A study conducted at ICAR-Indian Agricultural Research Institute (IARI), New Delhi led to the development of array of diverse sweet corn inbreds in the genetic background of *su1su1*, *sh2sh2* and *su1su1/sh2sh2* but it resulted in generation of few promising sweet corn hybrids (Hossain *et al.* 2013).

Pro-vitamin A is another important nutrient element of human diet for which considerable natural variation is present in maize germplasm. Based on combined approach of association analysis, linkage mapping, expression analysis and mutagenesis, it has been found that the favourable alleles of *lcyE* locus alter flux down carotene versus β -carotene branches of the carotenoid pathway and can enhance pro-vitamin A content up to three-fold in maize endosperm (Harjesh *et al.* 2008). Another major QTL for pro-vitamin A, *viz.* *crtRB1* has been mapped which significantly enhances beta-carotene content by blocking its conversion to abscisic acid (ABA) (Yan *et al.* 2010).

The genes governing the level of anti-nutritional factor phytic acid have also been identified and *lpa1-1* was the first mutant allele identified in M2 segregating generation of 90046-13, which reduces the phytic acid by 50 to 60% in seed but total phosphorous is unaltered (Raboy *et al.* 2001). The decrease in phytic acid in mature *lpa1-1* seeds is resultant of corresponding increase in inorganic phosphate (Pi). In the mature *lpa2-1* seed it is accompanied by increases in Pi and at least three other myo-inositol (Ins) phosphates. In both cases the sum of seed Pi and Ins phosphates is constant and similar to that observed in normal seeds. Homozygosity for either mutant results in a seed dry weight loss, ranging from 4 to 23% (Pilu *et al.* 2003).

There are no specific single genes discovered for high oil, methionine and micronutrients (Fe & Zn) but several minor effect QTLs have been reported by different researchers (Table 3).

Classical to molecular approaches for quality breeding in maize: In classical plant breeding relying on phenotypic selection for quality trait remained effective historically. However, for quality traits, indirect selections based on morphological traits have remained largely ineffective because of lack of definite correlation between quality traits and morphological characteristics. Most of the quality traits in maize are governed by recessive genes (Mertz, *et al.* 1964). Hence, stringent control of pollination is required while handling quality related breeding material and the selection can be made on the basis of biochemical evaluation rather than phenotypic selection. Further, recessive genes can be selected for only in homozygous state because every backcrossing calls for one cycle of selecting to select the desirable segregants. Hence, backcross breeding turns out to be time taking. Further, presence of modifiers as in the case of *o2*, the task become further complicated. A combination of more than one nutritional trait i. e. pyramiding nutritional traits is a

Table 3. Detail of QTLs influencing different quality traits in maize

QTL/Gene	Mapping population (size)	Parentage	Linked markers	Marker type	Chromosome location	PVE (%)	Ref
<i>Protein, oil and starch concentration/composition</i>							
74 loci (SNPs) for oil biosynthesis (including 26 for oil concentration) regular and 35 high-oil lines)	508 (AM508, 473 regular and 35 high-oil lines)	Diverse Panel	SNPG/T Indel_8 Indel_146/472 Indel_2000 Indel_20	SNPs (Illumina Maize SNP50 Bead Chip)	1,2,4,6,8 and 9	83 (26 loci for high oil)	Li et al.2012
15 QTLs (palmitic, stearic, oleic, linoleic, and linolenic acids) qOIL06-1 and qPRO06-1	BC ₁ S ₁ (150)	IHO × B73	umc1006	SSRs	6	10.9-39.6	Wassom et al.2008b
Several QTLs for kernel oil, protein, and starch	BC ₁ S ₁ (150)	IHO × B73	-	SSRs	6	36.7 (Oil)	Wassom et al.2008a
11 QTLs (kernel oil concentration) and 10 QTLs (kernel protein concentration)	F _{2:4} (165)	Zheng58 (low oil and protein) × B73 (high oil and protein)	umc1904- phi100175 umc1272- bnlgl839 umc1019-umc2038 umc219-umc2243	SSRs	1, 2, 3, 4, 5, 6, 7, 8 and 10	4.6 to 11.1 (Oil) 4.2 to 11.4 (Protein)	Yang et al.2016
17 QTLs (6, 6 and 5 for kernel oil, protein and starch concentrations, respectively)	F _{2:3} (298)	By804 × B73	bnlg2086-umc1122 phi96100-umc1422 umc1562-umc1960	SSRs	1, 2, 4, 5, 6, 7, 8 and 9	4.3- 13.1 (Oil) 5.19-6.66 (Protein) 4.1-7.9 (Starch)	Zhang et al.2008
12 and 6 QTLs (4 and 2 QTLs for starch, 4 and 3 QTLs for protein and 4 and 1 QTL for oil) in F _{2:3} and BC ₂ F ₂ populations, respectively	F _{2:3} (259) BC ₂ F ₂ (220)	Dan232 (dent corn) × N04 (popcorn)	umc1269-umc1948 umc1112-umc1936 umc1403-phi001	SSRs	1, 3, 4, 5, 6, 7 and 8	5.2-10.6 (Starch) 5-14.3 (Protein) 6.2-8.5 (Oil)	Yan Yang et al.2008
16 single-population and 19 joint-population QTLs for protein and 21 QTLs for protein-oil	RILs (282) RILs (263)	GY220 × 8984 GY220 × 8622	umc2075-bnlgl2046 bnlg1325-bnlgl1523	SSRs	3, 5, 6, 7, 8 and 9	4.4-13.4	Yang et al. 2014
12 and 14 QTLs for oil in two populations, respectively	RILs (282) RILs (263)	GY220 × 8984 GY220 × 8622	umc2316-umc1979	SSRs	1, 3, 4, 5, 6, 8 and 10	3.93-14.47	Yang et al.2012
58 QTLs (kernel oil content, embryo quality related traits)	RILs (245)	By804 × B73	Q8-umc1979	SSRs, InDels & Candidate gene markers	1, 2, 3, 4, 5, 6, 7, 8, 9 and 10	1.1-20.5	Yang et al.2012

Contd.

Table 3 (Concluded)

QTL/Gene	Mapping population (size)	Parentage	Linked markers	Marker type	Chromosome location	PVE (%)	Ref
<i>Carotenoids (Vitamin A and E)</i>							
3 Major QTLs for carotenoids (β-carotene, β-cryptoxanthin)	F _{2:3} (200) Testcross families (185)	W64a × A632 F _{2:3} families × AE335	<i>y1SSR-bnlg249</i> <i>zdsRFLP-phi034</i> <i>bmc1176-bmc1599</i>	SSR	6, 7 and 8	11.8-25.4	Wong <i>et al.</i> 2003
31 QTLs for carotenoids	RILs (203)	By804 × B73	YIsrr-umc1595 umc2313-YIsrr	SSR, STS and CAPS	1, 3, 5, 6, 7, 8 & 10	6.6-27.2	Chander <i>et al.</i> 2008
Several QTLs for γT, αT, TT and α/γ	F _{2:4}	W64a × A632 F _{2:4} families × AE335	<i>phi085</i>	SSR	1 and 5	-	Wong <i>et al.</i> 2003
31 putative QTL for tocopherol content and composition	RILs (208)	By804 × B73	umc1075-umc1304 umc1598-bnlg1811 bnlg1792-phi091	SSR, STS, CAPS, and AC-PCR	1, 2, 3, 5, 6, 7, 8, 9 and 10	29.32 (γ-T) 15.54 (α-T) 18.40 (δ-T)	Chander <i>et al.</i> 2008
30 QTLs with 3 major QTLs (qd1-1, qc5-1/qd5-1 and qc5-2)	F _{2:4} (237) F _{2:4} (218)	K22 × C17 K22 × Dan340	PZA02117.1-PHM4926.16 PZA00352.23-PZA02060.1	SNPs (GoldenGate assay)	1 and 5	53 (αT) 30 (αT) 25 (αT)	Shutu <i>et al.</i> 2012
Major QTL <i>ZmVTE4</i> (α-tocopherol variation) Two InDels and one SNP	Association panel (543 inbred lines)	Diverse panel	<i>InDel17</i> , <i>InDel118</i> , <i>SNP25815</i> <i>bnlg1237</i> & <i>phi085</i>	SNPs (Illumina)	5	33	Li <i>et al.</i> 2012
Major QTL <i>ZmVTE2</i>	F _{2:3}	N6xNC296	SNPs <i>Fe and Zn</i>	SNPs (GBS)	9	22	Fenton <i>et al.</i> 2018
27 QTLs	RILs	B73×Mol17	mmp 144, rz87 ay110452, ay110625 and mmp 125	SSRs	9 & 10	4-46	Baxter <i>et al.</i> 2013
3 and 10 QTLs for grain Fe concentration and bioavailability, respectively	RILs	B73×Mol17	mmp 144, rz87 and sh1 (Fe concentration) psr754b, php20528 and umc 2134 (Fe Bioavailability)	SSRs	3, 6 and 9	26 and 54 respectively	Lunga'ho <i>et al.</i> 2011
3 co-localized QTLs for Fe, Zn, Mg and P	RILs	B84×OS6-2	<i>bnlg1456</i>	SSRs	3	24.10 and 22.40 for Fe and Zn respectively	Simic <i>et al.</i> 2011

more desirable strategy for quality enhancement than to improve individual trait in separate genetic backgrounds. Deployment of phenotypic selection for multiple quality trait improvement simultaneously, possesses financial and operational challenges as it is in terms of cost, time and labour. Furthermore, plant breeders need to combine a suite of traits in a single cultivar, which may limit gains from phenotypic selection.

These constraints of conventional breeding can be overcome through molecular breeding that helps to study genetic diversity, characterize genetic architecture of germplasm and thereby enhancing the efficacy of selection (Moose and Mumm 2008). Molecular markers have also been successfully harnessed for mapping of QTLs for quality traits in maize. Various studies on identification or mapping of major genes/QTLs and minor QTLs have been carried out in recent past (Table 4). Desirable mutants having major effects have proved vital role in the nutritional improvement programmes.

The protein quality attributing genes, viz. , *o2* and *o16* genes for high lysine content were mapped through molecular markers by Babu *et al.* (2005) and Yang *et al.* (2005) respectively on chromosome 7S and 8L. Markers Assisted Selection (MAS) can help to combine the genes for quality traits through marker assisted gene pyramiding approach thereby providing more acceptable alternative of

normal maize in the form of multi-nutri-maize (multiple nutrient rich maize). Zhang *et al.* (2013) introgressed two genes, viz. , *o2* and *o16* for increasing lysine content in waxy line QCL5019 from 0. 28% to more than double 0. 62% in introgressed families. The maize hybrid, Vivek QPM 9 released in 2008 having enhanced lysine and tryptophan as per its non-QPM version of Vivek 9 is the first successful example of commercial release of MAS-derived maize hybrid in India (Gupta *et al.* 2013). Similarly, further effort for utilizing MAS for enhancing provitamin A levels in Vivek QPM 9 resulted into provitamin A maize hybrid Pusa Vivek QPM9 Improved (Muthusamy *et al.* 2014). It possesses both *o2* and *crtRB1* genes and therefore multnutrient rich. Zunjare *et al.* (2018) further successfully combined both *crtRB1* and *lcyE* in the genetic background of four popular QPM hybrids, HQPM-1, HQPM-4, HQPM-5 and HQPM-7. The products (inbreds/hybrids) with enhanced quality and developed through MAS has been detailed in the Table 4.

There are different genotypes or cultivars developed based on sweet corn mutants, viz. , Boston, Bonus and Jublee (*su1*-based), Anava, Champ and Dallas (*se*-based) and Candle, challenger and Sheba (*sh2*-based). Other than these the genotypes developed based on the different combinations of *sh2*, *su1* and *se* genes have been developed, viz. , IL27a, I453 and P39 (*su1su1/Se1Se1/Sh2Sh2*), IL677a (*su1su1/se1se1/Sh2Sh2*) and EPS18 (*Su1Su1/Se1Se1/sh2sh2*)

Table 4 Details of MAS undertaken for various quality traits in maize

Trait(s) improved	Gene(s) introgressed	Marker name	Marker type	Inbred/ hybrid	Country	Reference
QPM	<i>opaque2</i>	phi057, phi112 and umc1066	SSR	inbred	India	Babu <i>et al.</i> 2005
QPM	<i>opaque2</i>	phi057, phi112 and umc1066	SSR	inbred	Uganda	Manna <i>et al.</i> 2005
QPM	<i>opaque2</i>	phi057, phi112 and umc1066	SSR	inbred	Kenya	Danson <i>et al.</i> 2006
QPM	<i>opaque2</i>	phi057	SSR	Inbred & hybrid	Thailand	Jompuk <i>et al.</i> 2011
QPM	<i>opaque2</i>	phi057, phi112 and umc1066	SSR	Inbred	India	Gupta <i>et al.</i> 2013
QPM	<i>Opaque2</i>	umc1066 and phi057	SSR	Hybrid	India	Hossain <i>et al.</i> 2018
QPM	<i>opaque2</i>	phi057 and umc1066	SSR	Inbred	Serbia	Kostadinovic <i>et al.</i> 2014
QPM	<i>opaque2</i>	Phi057	SSR	Inbred	Philippines	Magulama and Sales 2009
QPM	<i>opaque16</i>	umc1141 and umc1121	SSR	Inbred	China	Yang <i>et al.</i> 2013, Zhang <i>et al.</i> 2010
QPM	<i>opaque16</i>	umc1141 and umc1149	SSR	Inbred	India	Sarika <i>et al.</i> 2016
QPM	<i>opaque2</i> & <i>opaque16</i>	(phi057, phi112 and umc1066) (phi027 and phi112)	SSR	Inbred	China	Zhang <i>et al.</i> 2013, Sarika <i>et al.</i> 2018
ProA	<i>crtRB1</i>	umc1066 <i>crtRB1</i> -3'TE-F, <i>crtRB1</i> -3'TE-R1 and <i>crtRB1</i> -3'TE-R2	SSR	Inbred & hybrids	India	Muthusamy <i>et al.</i> 2014
ProA	<i>crtRB1</i>	crtRB1-5'TE-2 and crtRB1-3'TE-1	SSR	Inbred	China	Liu <i>et al.</i> 2015
ProA	<i>crtRB1</i> & <i>lcyE</i>	Phi057 and InDel 3'TE and 5'TE	SSR	Inbred & hybrid	India	Zunjare <i>et al.</i> 2018
Vitamin-E	<i>VTE4</i>	InDel7 and InDel118	Functional markers	Inbred	China	Feng <i>et al.</i> 2015
Low phytate	<i>lpa2</i>	umc2230	SSR	Inbred	India	Tamilkumar <i>et al.</i> 2014, Sureshkumar <i>et al.</i> 2014

based) have been developed (Revilla *et al.* 2006, Szymanek *et al.* 2015).

The high oil populations IHO, SHO, DHO, ALHO, ASK, ALEX synthetic, KYHO and hybrids, *viz.* Illinois 6021, 6052, 6001 and Burr white have been developed (Hopkins 1899, Wang *et al.* 2009). These have been developed through cyclic selection of high oil lines.

Amylose is a linear macromolecule which contains glucose units with α -1, 4 linkages in which each macromolecule contained one reducing end and one non-reducing end. Amylose of high-amylase corn starch has a high degree of polymerization (Takeda *et al.* 1989). High amylose containing maize commonly known as amylo-maize possesses more than 50% amylose contents. Amylo-maize lines, *viz.* H99ae, OH43ae, B89ae, B84ae and GEMS-0067 lines have been reported (Li *et al.* 2008). The branched component of starch is amylopectin. The iodine uptake by the branched amylopectin in high amylopectin lines (waxy lines) increase at low temperature (Banks and Greenwood 1975). For example, the iodine binding capacity of waxy maize amylopectin is 0.17 at 20°C, and 0.15% at 1.5°C. Waxy corn is a popular choice in the entire South-East Asia

(Devi *et al.* 2017). Several landrace accessions with high amylopectin are available and used as part of food (Park *et al.* 2008, Liet and Thinh 2009, Bao *et al.* 2012, Zheng *et al.* 2013). Besides several hybrids with high amylopectin have been developed (Zhang *et al.* 2013, Yang *et al.* 2013).

Challenges and future perspectives: Biofortification of maize with quality traits is an essential feature to address nutritional severity. Genomics and marker assisted selection (MAS) technology has opened new avenues for improvement of complex quality traits. Following this rate a wide array of biofortified maize with high lysine, tryptophan and provitamin A has been developed. However, biofortification process and biofortified maize are associated with number of issues; some of the issues are discussed here.

First of all, enhanced protein and its content is negatively associated with grain yield (Bjarnason and Vasal 1992). Modifications of other quality traits are as such not associated with yield loss. The biggest challenge in developing cultivars with improved quality traits lies with modification of the quality through alteration of genes involved in multiple

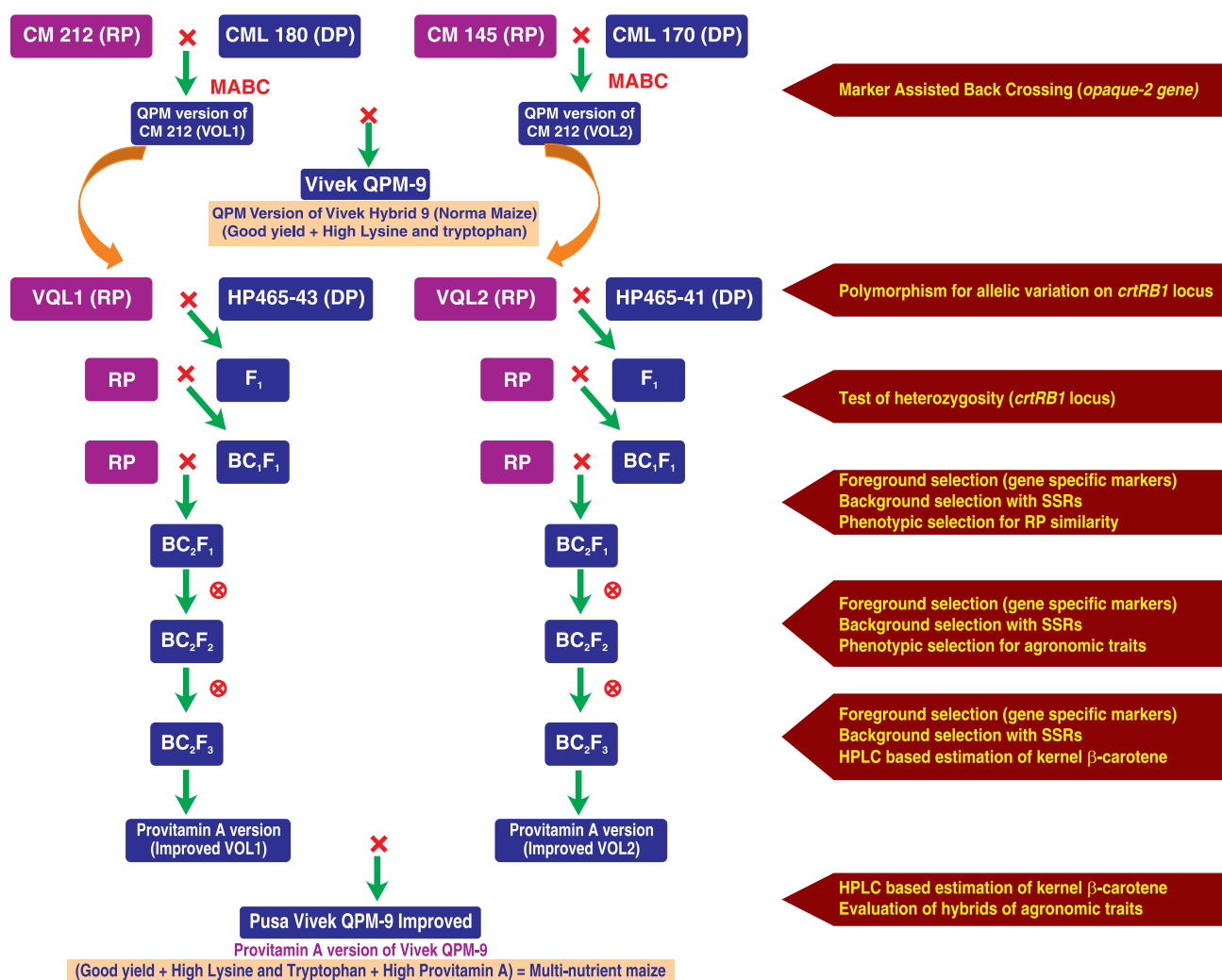


Fig 1 MAS strategy for utilization of recessive genes for development of multi-nutrient maize.

metabolic pathways without compromise with grain yield (Collard and Mackill 2008). Hence, this is not applicable in case of sweet corn as the grain as such is not the end use. Earlier there was several QPM based hybrids released in India, viz. , Shaktiman series and Protina but major drawback of these hybrids yield was less as compared to normal hybrids. QPM version of Vivek hybrid 9, Vivek QPM-9 possesses similar grain yield potential as that of the original hybrid (Gupta *et al.* 2013). Hossain *et al.* (2018) also reported similar grain yield potential of Pusa HM4 Improved, Pusa HM8 Improved and Pusa HM9 Improved with their original non-QPM version, viz. , HM4, HM8 and HM9. In other study, MAS-derived versions of HM-4, HM-8, Vivek QPM-9 and Vivek Hybrid-27 for β -carotene evaluated by Muthusamy *et al.* (2014) at two different locations of India found that β -carotene-rich version of original hybrids were similar for grain yield potential. Hence, it may be concluded that, quality enhanced as maize cultivars can be developed without any yield penalty. Thus, plant breeders need to add nutrition as an objective to their breeding programs.

Second challenge comes in terms of commercialization of biofortified products. There are two aspects in this regards convincing the farmers. Firstly, the farmers need to be convinced of the benefits of growing and consumption of the products, and secondly the economic benefits associated with growing such products. To convince the farmers a strong extension service is needed. This can be addressed by launching ground level awareness campaign about health benefits related to its consumption. The perception of people about low yield potential of quality fortified maize is to be changed. Food processing industries needs to be linked with quality maize cultivation to harness its good benefit. Policy intervention is needed to encourage quality maize cultivation at appreciable support price. Nutrition education campaigns that effectively empower caregivers with knowledge about the importance of nutrients in health would help in a great way. Provitamin A-biofortified sweet potato projects in Kenya and Mozambique have documented the effectiveness of appropriate nutrition education. They employed community theatre, group demonstration sessions, and radio programs, in creating demand for such fortified products (De Groote, *et al.* 2010). In Zambia, the HarvestPlus project is working closely with the Ministry of Health, the National Food and Nutrition Commission, the Ministry of Agriculture and Cooperatives (extension service), and others to develop nutrition education strategies to create lasting demand for provitamin A biofortified maize and other sources of vitamin A (De Groote *et al.* 2010). Such an integrated awareness campaign is certainly needed to harness the benefits of biofortified maize.

Public awareness campaigns exploiting the power of the media and national public health experts to highlight micronutrient deficiencies and promote adoption of nutritious crops, would help in the dissemination of the technology. It will also be important to communicate not only with allies or those who are undecided, but also with opponents of biofortification to ensure that they are well

informed before influencing their constituents. Groote *et al.* (2010) reported that the adoption of QPM cultivars by the farmers varied a lot among East African countries with 70% adoption in Uganda and 30% adoption in Tanzania, while Kenya reported none. Besides the knowledge of nutritional benefit of QPM, the response of farmers' participation in extension activities and reliable supply of good quality seeds were the important factors for the successful adoption. A study in Zimbabwe by Stevens *et al.* (2008) revealed that ~94% of the respondent agreed to consume yellow maize instead of traditional white maize, if educated on health benefits.

The third challenge is since the most of gene(s) conferring quality enhancement are of recessive in nature, maintenance of quality attributes under farmers' field is difficult due to out-crossing with non-fortified maize pollen from neighboring fields. Therefore, it is needed to implement 'biofortified maize village concept' on the pattern of seed village concept to ensure and optimize nutritive advantages associated with recessive gene-based biofortified maize (Groote *et al.* 2010; Gupta *et al.* 2015). The biofortified maize technology propagated through a village concept can only pass on the benefit of the technology consumers and farmers. This could be accepted by the next generation of farmers through strong policy interventions. Such interventions will also strengthen community-based seed production. Seed village concept would help to produce quality seeds by mitigating the outcrossing and enrich availability of quality seed at local level.

Fourthly, the effects of different micronutrients, viz. , lysine, tryptophan, provitamin A, Fe and Zn are phenotypically invisible on grains. In this case it is difficult to convince the traders about the quality standards of the farm produce of fortified maize grains. Considering the lack of availability of rapid detection kits, there is need to develop such portable rapid detection kits to detect the quality of produce and thereby assuring good price to the farmers based on the extent of the quality. A new method using a 'proprietary formulation' developed at Indian Institute of Maize Research, Ludhiana completes hydrolysis of maize endosperm proteins in 30 minutes (unpublished results/ personal communication). The new methodology needs to be converted into a kit which is expected to drastically shorten the time required for tryptophan estimation, and can be used for estimating other amino acids as well. Conscious observation of food habit of people, industry is coming up with newer food products frequently. Increased awareness of people through internet is pushing industry to provide novel options to cater their needs. Corn flakes, as a nutritionally rich breakfast is a classical example. Collaboration with industries is thus vital to develop such products from biofortified maize grains that can benefit both industry and consumer. Maize is the main component of maize-soybean feed mixture which is the major poultry feed across the globe. A deficit of essential amino acids in such feed mix has long been supplemented through addition of synthetic amino acids which raises the cost of feed making

the profits sensitive to price fluctuation of final produce. Synthetic supplements are continuously being used despite the availability of maize cultivars with balanced quality of protein. A systematic approach need to be adopted to sensitise entrepreneurs engaged in feed sector to adopt cost saving biofortified cultivars.

Finally the focus on the enhancement of yield in normal maize with less emphasis on quality from the beginning of maize breeding has resulted into the narrow genetic base germplasm for quality improvement. In the past although the recurrent selection programmes resulted into development of high oil and high protein population but progress was at very slow rate. The problem of malnutrition has sensitized plant breeders to focus on biofortified varieties. The molecular marker technology has also boosted the quality improvement programme through rapid introgression of favorable gene(s) into high yielding commercial cultivars. The maize breeding programmes needs to be strengthened through developing heterotic pools for quality traits and thereby developing high yielding nutritious maize hybrids. The new breeding technologies, viz. marker assisted recurrent selection (MARS) and genomic selection (GS) open the door to assist for enhancement and confirming the quality nutrients in maize with short period in current era.

The worldwide significant impact of biofortification, recognized by public officials is key to the success of biofortified crops. The declaration of remunerative price through minimum support price and/ or premium price for biofortified maize grains in the market will encourage the farmers to grow more biofortified maize. Easy loan and subsidy to village level entrepreneurs to initiate small-scale enterprises for the development of biofortified maize-based processed food products would help in their greater dissemination. An integrated approach involving promising cultivars, extension agencies, products value addition, policy support would be important key to success of biofortified maize cultivation.

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