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Action of *o2* modifiers for endosperm modification and amino acids on *o2* maize

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ABSTRACT

QPM (Quality Protein Maize) with o2 gene alone cause to improve tryptophan and lysine content but negative correlation with normal maize phenotype. QPM lines reported low yield, susceptible to ear rot disease and storage pest due to insignificant variation in kernel for modification. Also amino acid imbalance in QPM lines with o2 genes which could not supplement daily intake of protein supplements. This malnutrition deficiency leads to disorders like improper growth, kwashiorkor and Pellegra. To mitigate this problem, breeding of QPM with o2 modifiers for endosperm modification and amino acid modification was focussed and found to be effective with sufficient nutrient supplements. Endosperm modifiers act on 27-kDa γ-zein to modify into vitreous endosperm which could be resistance to ear rot disease. Further amino acid modifiers act on maintenance in balancing amino acid content of both zein and non zein protein. By the action of these o2 and o2 modifiers, modern Quality Protein Maize (QPM) developed with increased lysine and tryptophan and also with good agronomic traits.

1. Introduction

Maize one of the important cereal crop in the world after rice and wheat. Due to diverse functionality and high yield among cereals, maize is also called as "Queen of Cereals". It is mainly used as a livestock feed but the normal maize protein is of poor nutritional quality due to a deficiency in two essential amino acids (lysine and tryptophan). Discovery of opaque2 (o2) mutants which has effect on increase in tryptophan and lysine content, indeed development of QPM was focussed on kernel modification and increased amount of essential amino acids. But o2 mutant associated with low seed density and soft kernel texture and inferior agronomic traits which are all reduce the market price of o2 maize. This problem was mitigated by discovery of another set of genes which acts on endosperm modification and some genes modifiers amino acid content.

opaque2 modifiers for endosperm modification

Due to undesirable agronomic characters associated with o2 mutants, researchers focussed on to develop maize with modifier genes which came to known as modern QPM with vitreous endosperm. This modified endosperm structure associated with change in zein and non-zein protein ratios. In QPM kernel decrease in α-zein amount negatively correlated with γ-zein which led to endosperm modification (Prasanna *et al.,* 2001). also this modification has positive correlation with nutrient enrichment (2% higher protein) and 13 % improvement in yield due to vitreous in nature, resistance to ear rot disease, storage pest, kernel and kernel cracking. Multi location trials of QPM revealed that Genotypes X Environment Interaction (GEI) has effects on yield and yield contributing traits and endosperm modification character by difference in yield and other agronomic traits (Pixley and Bjarnason, 2002). In addition to that endosperm modification positively correlated with methionine and opacity and negatively correlated with amino acid content due to effect of environment and inverse association in some QPM lines like B73o2 (Gutierrez-Rojaset *et al.,* 2008).

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In development of QPM, selection of lines with hard endosperm was performed by conventional and molecular approach. In conventional approach those modifiers were selected by opacity of kernel could be assessed by light table which is custom-made box to differentiate hard endosperm maize from soft endosperm maize. In QPM due to segregation of o2 and o2 modifiers after every selfing generation varying degrees of opacity were expressed which could help to select genotype of o2 bearing cultivar. Based on the selection of kernels with soft endosperm with o2o2 constitution which had significant variation against hard endosperm kernels with O2O2 and O2o2 genotypes success of breeding programme would be defined for development of QPM (Vivek *et al.,* 2008)

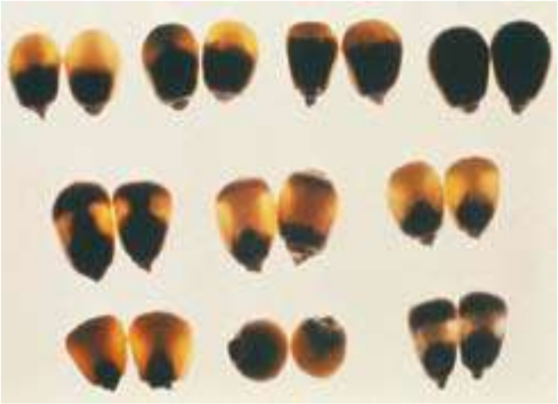

Figure 1. Varying degrees of kernel opacity indicate varying levels of endosperm modification Source: Vivek *et al.* (2008)

Triploid nature of endosperm has effect on degree of vitreousness, hardness and opacity of kernel, especially when action of multiple loci different dosage effect in kernel modification. Molecular studies and Quantitative Trait Loci studies showed that modifiers associated with chromosome 7 in which bin number 7.02 (*umc1016, umc1929, umc1393* and 27-kDa γ-zein and bnlg1022a) and on chromosome 9 at bin number 9.03 (*phi022*, *bnlg127*, and *bnlg1714*) and 9.05 (*umc1078*) (Holding *et al*., 2008). In QPM improvement in endosperm modification positively correlated with increase in 27-kDa γ-zein which was located in chromosome 7 (bin no. 7.02) implied that essential role of 27-kDa γ-zein in endosperm modification. In QPM, α zeins are responsible for starchy endosperm which regulated by transcription factor from *o2* gene which could be modified by RNA interference (RNAi) technology which results in increased amount of protein. RNAi on RNAi/+ genotype showed *o2* maize phenotype with reduced expression of 27 and 16-kDa γ-zeins led to affect endosperm modification (Wu *et al*., 2011).

opaque2 modifiers for amino acid content modification

Amino acid modifiers acts on balance in zein and non zein protein content and their related amino acid contents. Inbred lines with 0.05 to 0.11 % of tryptophan and 0.7% Quality Index generally defines QPM which could sufficiently discriminate QPM inbred lines from normal maize lines (Vivek *et al.,* 2008). Studies on QPM with modifiers about phenotypic characterization and amino acid content estimation revealed endosperm texture and amino acid contents were found to be negatively correlated (Gutierrez-Rojas *et al.,* 2008). Also degree of endosperm modification and amount of tryptophan and lysine was found to be more in QPM x QPM crosses with o2 gene and o2 modifiers than the crosses between o2 maize lines (Ignjatovic-Micic *et al.,* 2010).

In QPM maize kernels major quality traits like lysine content in maize and protein quality was under the control of additive effects with partial-dominance in both normal as well as in drought stress conditions. QPM lines with amino acid modifiers contained less protein than their wild type counterpart and reduction in the starch content result from early termination of the starch biosynthesis in the o2 mutant followed by faster hydrolysis by α-amylases than dry-ground wild type maize to provide energy for animal growth and to produce glucose for alcohol fermentation (Hasjim *et al.,* 2009). Availability of the amino acid modifiers could identify through estimation of tryptophan (Vivek *et al.,* 2008) by Hopkins-Cole reaction which could revealed the amount of lysine. This could be validated by molecular analysis with reported SSR markers for o2 modifiers *viz.* bnlg1179, bnlg1643, bnlh1633, bmc2136 and umc1357 (Krishna *et al.,* 2012). Besides o2 gene, breeding for development of QPM with modifier genes for endosperm modification and amino acid improvement could accelerate to develop and to introgress multigenic traits into maize and also which could be efficiently used as tool to mitigate nutrient deficiency by supplementary of protein supplements.

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