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The Maize Journal is published half yearly by the Maize Technologists Association of India. The Journal publishes papers based on the results of original research on maize and related issues in the following areas:

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## Screening methods of maize germplasm for resistance against maize stem borers: A review

Pradyumn Kumar<sup>1</sup> · Harshita Kaushik<sup>2</sup> · S. B. Suby<sup>3</sup> · Sachin Suroshe<sup>4</sup> · Jaswinder Kaur<sup>5</sup> · Jawala Jindal<sup>6</sup>

**Abstract:** Screening of maize germplasm for resistance against stem borers is pre-requisite for the development of resistant varieties/hybrids. Leaf injury rating, larval development period, larval weight and stem tunnel length are some of the parameters used for measuring antibiosis, while ovipositional preference, number of egg masses/eggs per plant are the parameters for antixenosis. Leaf injury rating after artificial infestation of plants by laboratory reared pest has been in use for screening the germplasm world over. Recently, several studies have been conducted in which a large number of germplasms are screened for resistance using their antibiotic and antixenotic traits and a robust susceptibility index is developed utilizing their results.

**Keywords:** Antibiosis · Antixenosis · Germplasm · Resistance · Screening methods

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### Introduction

Maize (*Zea mays* L.) is widely grown throughout the world and has the highest production of all the cereals with 1.162 billion tons produced in 2020. It serves as a staple food for millions of people in different parts of the tropical world. The average yield of temperate maize is 7.0 tons/ha, whereas global average is 4.2 tons/ha. The earlier literature cites over 160 insect and mite species which attack maize crop in India Fletcher (1914, 1917), Ayyar (1940), Bhutani (1961), Pant and Kalode (1964) and Mathur (1987) observed over 250 species of pests associated with maize in field and storage conditions. The pyralid *Chilo partellus* and the noctuid *Sesamia inferens* and muscids, *Atherigona* spp. are important pests of maize ecosystem. A robust germplasm screening technique is prerequisite for developing resistant hybrids against these pests. Parameters of antibiosis of germplasm were extensively used by studying leaf injury level, tunnel length caused by stem borers, larval development period, larval weight, etc. Attempts are now being made to make the screening more efficient by incorporating antixenotic trait, the oviposition preference.

### *Germplasm screening techniques for resistance against stem borers*

Screening of maize germplasm against resistance is an integral component of breeding program for developing resistant varieties/hybrids. Germplasm with broad genetic background; efficient technique of mass rear healthy insects for artificial infestation of plants; an efficient technique to infest plants uniformly and an accurate technique to evaluate insect damage in the field are some

of the important aspects of maize germplasm screening for resistance. CIMMYT and its research partners around the world have worked towards developing screening techniques and breeding protocols which have enabled the development and release of resistant maize germplasm Mihm (1984). Techniques have been standardized to mass rear the stem borers on artificial diet and infest maize plants efficiently and rapidly. Recording leaf injury rating on artificial infested plants was developed by Sarup (1983) which is now used conventionally to screen maize germplasm.

#### *Method used to study role of antibiosis in imparting resistance*

Maize germplasms to be tested in fields are sown in rows separated by non-experimental maize. Twelve days after germination, the plants were infested by black-headed stage of eggs or neonate larvae from laboratory culture of stem borer. Kaur *et al.* (2014) found that 12 days after germination plants are most suitable for the establishment of stem borer larvae on them. The symptoms of feeding were visible by number of leaves bearing holes. After 25 days of infestation, the plants were observed for leaf injury rating on 1–9 scale. In another set of infested rows, the plants were dissected after 7, 14 and 21 days of infestation to observe the tunnel length and recovery of larvae/pupae. The larvae recovered were reared on baby corn till pupation in the laboratory. The larval/pupal weight and larval development period were recorded.

#### *Method used to study the role of antixenosis in imparting resistance*

Oviposition is the first interaction of stem borer females with maize plants. To study the oviposition preference, the experiment was conducted in multi-choice test and no choiced test. In multi-choice test, the germplasm to be tested were planted in pots and kept in a walk-in cage. The adult pairs were released at the rate of one pair per four plants. The females prefer to lay more eggs on the plants of their choice germplasm. After five days of release, the plants were examined for the number of egg masses per plant, number of eggs per egg mass, the number of eggs per plant and number of plants oviposited. These observations were recorded. In no-choice test for each germplasm, four plants were kept in a versatile

collapsible cage developed by Kumar *et al.* (2019) and a single pair of adults was released in the cage. The gravid female had no other germplasm to choose for oviposition. After five days of release, the parameters observed in case of multi-choice test were observed and recorded in no-choice test as well.

#### *Review of antibiosis and antixenosis in imparting resistance*

In antibiosis type of resistance, the biology of the insect is affected leading to reduced longevity and reproduction, and increased insect mortality. Antibiosis decreases larval development as well as the number of larvae per plant, thereby decreasing the stem damage levels (Pimentel, 2002). The leaf injury rating scale to evaluate the damage caused by stem borer which is used for screening maize germplasm was first developed by Sarup (1983). Durbey and Sarup (1984) showed the adverse effect of rearing *Chilo partellus* on different maize germplasm. Maximum antibiosis resulted from rearing on resistant Antigua Gr. I, Mex-17 and tolerant Ganga 5. The expression of antibiosis due to tolerant Ganga-5 revealed its intermediate behavior towards *Chilo partellus*. The average larval and pupal weights were significantly lower on resistant varieties (Antigua Gp. I and Mex-17) as against susceptible varieties (Basi Local and Vijay composite). Chamarthi (2008) opined that resistance factors which can be quantified or monitored in plant can be used as marker traits for screening germplasm against stem borers. Leaf feeding damage, which is the first larval feeding symptom, is an important marker trait which has been used by various workers in order to distinguish resistant from susceptible genotypes (Kumar, 1994). Singh *et al.* (2011) observed that the direct effect of stem tunneling on loss in maize grain yield was greater than the effect of leaf feeding. To achieve an overall improvement in the level of genotypic resistance that protects all stages of plant growth, resistance to more than one damage variable is required. Cholla *et al.* (2018) found a significant correlation between leaf injury ratings and stem tunnel length. Based on the selection index developed using these two parameters, he identified WNZPBTL2 and PFSR 51016/1 as the resistance sources of *Chilo partellus*.

Morphological traits such as leaf toughness, stem penetrometer resistance, trichome density; biochemical traits such as stem sugar content and leaf injury, number

of exit holes and stem tunnel length were used to develop selection index. Based on this index, 120 maize inbred were categorized into resistant, moderately resistant, moderately susceptible and susceptible germplasm (Shelmith *et al.*, 2013). Secondary metabolism, which involves specialized, often complex and species-specific biosynthetic pathways, is thought to provide compounds which are accumulated and stored, so that when attacked, the plant is already equipped with the means to deter or kill herbivores. Plants may possess constitutive defenses that can act as a physical barrier, as in lignification or resin production, or can act as a biochemical signal perceived by the herbivore, as deterrents of feeding or oviposition, or can act as a toxin. Toxic compounds e.g., alkaloids, terpenoids, phenolics, forcing specialists to invest resources in detoxification mechanisms that in turn incur growth and development costs. Plant parts that are of high fitness value or that are under a high risk of attack may be best protected by constitutive defenses, whereas others may be better defended by induced responses (Wittstock and Gershenson, 2002). All the three components of resistance have been identified in stem borer resistant maize.

Gundappa *et al.* (2013) studied effects of two phenolic acids on *Chilo partellus* in maize inbred lines. Both the phenolic acids were negatively correlated with leaf injury rating and tunnel length at all the plant ages. The *p*-coumaric acid is predominant phenolic acid in maize inbred lines compared to ferulic acid. These phenolic acids were quantified in 17 Indian maize inbred lines. The *p*-coumaric acid and ferulic acid in leaves range from 1.3 to 3.9 mg/g and 1.5 to 4.7 mg/g, respectively. Higher quantities of these acids were found in inbred lines HKI 577, HKI 323, HKI 1105. The higher quantities of these acids were recorded in plants at 10 and 25 days after germination. Bioassay of neonate larvae of *C. partellus* done by diet incorporating with phenolic acids resulted in increased mortality by *p*-coumaric acid (41.5 per cent) than ferulic acid (17.7 per cent) over control (Gundappa, 2012). Antixenotic mechanism of resistance, which is employed by the host plants, deters the insects from oviposition (Painter, 1951; Valencia, 1984; Afzal *et al.*, 2009), feeding and seeking shelter (Dabrowski and Kidiavai, 1983; Woodhead and Taneja, 1987; Sharma and Nwanze, 1997; Dhaliwal and Arora, 2003). This mechanism renders the plants undesirable or, in other words, to be bad hosts for an easy invasion of insects (Shoonhoven *et al.*, 1998). Oviposition of many

lepidopterans is a critical step in their life cycle because of the limited mobility of first instars larvae (Feeny *et al.*, 1983; Showler and Moran, 2003). Plant volatiles, especially herbivore-induced volatile components, such as green leaf volatiles, terpenes, alkenes, 14 carboxylic acids and alcohols (Holopainen, 2004) play important role in mediating behavior such as host plant searching and acceptance in phytophagous insects; in attracting parasitoids or predators of pest insects; in directing insect oviposition as well as in influencing insect-plant interactions (Durbey and Sarup, 1982; Anderson and Alborn, 1999; Wink, 2003; Lonstantopoulou *et al.*, 2004; Steinbauer *et al.*, 2004; Rassmann *et al.*, 2005; Ruther and Kleier, 2005; Carroll *et al.*, 2006; Raguso, 2008). It is hypothesized that ovipositing females have evolved to lay eggs on hosts that elicit the best performance of the offspring (Thompson and Pellmyr, 1991). Scheirs and De Bruyn (2002) reported that egg-laying females choose oviposition sites that enhance their own long-term fitness at the expense of their offspring. Females when confronted with an array of potential hosts, exhibit a hierarchy in their preferences (Courtney *et al.*, 1989). When a number of potential hosts are available, female will lay most eggs on her most preferred plant, fewer on the next preferred and so on. Durbey and Sarup (1982) studied different plant parameters viz. percentage of plants oviposited, plant height, percentage of leaves oviposited and number of egg masses per plant for ovipositional responses of *Chilo partellus* amongst resistant (Antigua Gr. I, Mex-17, Ganga-5) and susceptible (Basi local and Vijay composite) maize germplasm under caged conditions. They found that the moths ensured greater survival of the freshly hatched larvae to continue the progeny on susceptible germplasm than to get eliminated on resistant ones.

## Conclusion

Oviposition preference was studied in multi-choice test and confirmed by no-choice test method in 20 germplasm. Germplasm, significantly varied on oviposition preference. The adult preference for oviposition and larval performance was studied on *Sesamia inferens*. The correlation between parameters of preference and performance of *Sesamia* was poor (0.19). Thus, it is important to develop approaches that eventually improve the efficiency of selecting borer-resistant genotypes

keeping both antibiotic and antixenotic parameters in view in a high yielding background. The selection criteria should consider measuring the combined effect of different components of host plant resistance, an approach that requires the use of appropriate indices that result in selection for resistance as well as grain yield performance.

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## ***In vitro* regeneration in maize (*Zea mays* L.)**

**Krishan Kumar<sup>1</sup> · Alok Abhishek<sup>1</sup> · Abhishek Kumar Jha<sup>1</sup> · Bhupender Kumar<sup>1</sup> · Sujay Rakshit<sup>2</sup>**

**Abstract:** Maize is a versatile cereal crop having the highest genetic potential, production, and productivity. In the past few decades, plant tissue culture and transformation approaches have played an important role in maize improvement via introducing beneficial transgene(s) or modulating the expression of the endogenous gene(s), etc. However, the capability of *in vitro* regeneration in maize is highly influenced by genotypes, type of explants, media compositions among others. Some genotypes are more amenable to tissue culture producing embryogenic calli, while others are recalcitrant to tissue culture. Genotypic differences in morphogenesis and organogenesis are generally reported that might be possible due to differences in endogenous hormone levels. The *in vitro* regeneration potential in maize is usually decreased during the channelized path of tissue maturation, therefore embryogenic callus is mostly achieved from immature zygotic embryos. The present article aimed to provide the current state of the art in maize somatic embryogenesis. Further, the article describes the procedure for maize whole plant regeneration from embryogenic callus.

**Keywords:** Maize · Somatic embryogenesis · Whole plant regeneration

### **Introduction**

Conventional plant breeding has contributed exceptionally to crop improvement. Nonetheless, it has a major limitation to introduce desired traits into crop plants through genetic crossing due to requirements of species compatibility and the dearth of germplasm diversity within the species. However, the advent of plant tissue culture and transformation methods have made it possible to insert beneficial foreign gene(s) into crop plants i.e. into sexual incompatibility species. Plant tissue culture techniques are useful to multiply and propagate plants under *in vitro* conditions on a rapid and large scale, irrespective of season, with less space requirement and in a shorter time frame. These techniques have been utilized for micro-propagation, genetic transformation, double haploid line production, storage of plant cells and organs, biosynthesis of secondary metabolites, etc. Thus, plant tissue culture and transformation methods have played a pivotal role in gene-function studies and crop improvement (Agarwal *et al.*, 2018; Kumar *et al.*, 2018).

Maize (*Zea mays* L.) is the most widely grown cereal crop in the world with wide adaptability under diverse ecologies. It is consumed as feed and food, and also has myriads industrial applications, including the production of bioethanol and starch. It is one of the most extensively studied crops. The successful development of a tissue culture and transformation system for maize helped to develop genetically modified (GM) or transgenic maize having one or more novel traits that may not occur naturally in the species (Kumar *et al.*, 2020). To date, nearly 250 transgenic events in maize have been commercialized (ISAAA database 2019; <https://www.isaaa.org/>), hence tissue culture and transformation played a major role in the genetic improvement of maize in the past two decades. The *in vitro* regeneration in maize is highly

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dependent on genotypes and influenced by many factors like media, hormone composition, temperature, and light influx.

#### *In vitro* regeneration

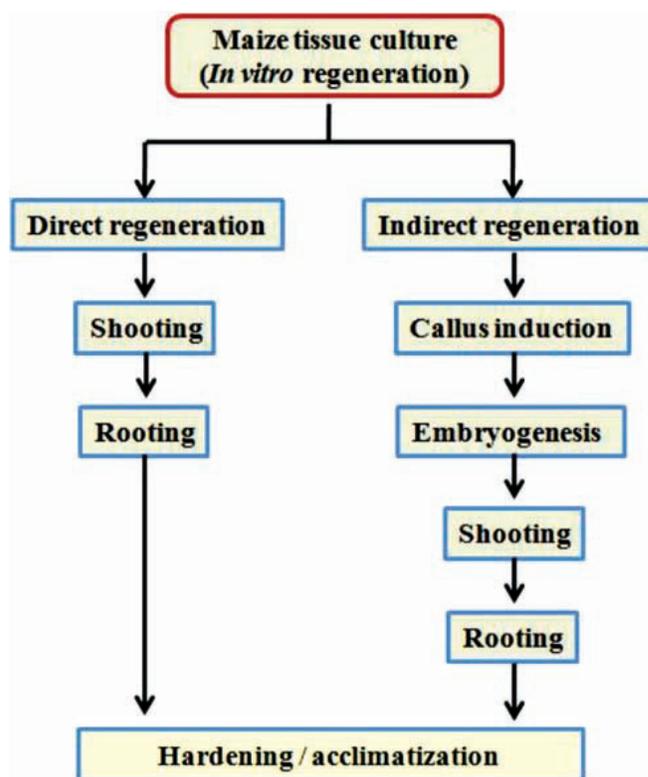
The regeneration ability in maize is highly dependent on the genotype as only a few genotypes are amenable for tissue culture and transformation. *In vitro* regeneration in maize was first reported by Green and Phillips (1975) using immature embryos as explants. Generally, regeneration can be achieved directly (without callus formation) or indirectly (with callus formation followed by somatic embryogenesis) (Figure 1). It has been shown that callus induction and *in vitro* regeneration is highly challenging in maize as compared to other cereal crops (Jia *et al.*, 2008; Zhao *et al.*, 2008; Martinez and Wang, 2009; Anami *et al.*, 2010; Rakshit *et al.*, 2010; Chu *et al.*, 2011; Yadava *et al.*, 2017). Many factors influence callus production in maize such as genotype, source and stage of explant, and growth medium components. Several reports are available in temperate, subtropical, and tropical maize germplasm for embryogenesis and indirect organogenesis (Armstrong, 1999; Ahmadabadi *et al.*, 2007; Aguado-Santacruz *et al.*, 2007; Rakshit *et al.*, 2010; Tiwari *et al.*, 2015). In comparison to indirect regeneration, relatively lesser research has been carried out on *in vitro* regeneration via the direct organogenesis pathway. Factors affecting whole plant regeneration in maize are described below:

#### *Genotype and explant*

Different tissues are used as a source of explants for maize regeneration by different groups globally. The regeneration in temperate, subtropical, and tropical maize germplasm has been achieved using different explants such as immature embryo, nodal culture, leaf, and mature embryo, etc. (Armstrong, 1999; Bohorova *et al.*, 1999; Ahmadabadi *et al.*, 2007; Aguado-Santacruz *et al.*, 2007; Rakshit *et al.*, 2010; Malini *et al.*, 2015; Tiwari *et al.*, 2015). However, immature embryos are the most preferred explants for producing embryogenic callus and hence indirect regeneration in maize (Armstrong, 1999; Aguado-Santacruz *et al.*, 2007; Yadava *et al.*, 2017; Agarwal *et al.*, 2018). This is due to the higher callusing and regeneration efficiency of immature embryos as the

regeneration capacity is reduced as tissue mature. However, immature embryos are usually not available throughout the year, and culturing them is also laborious. The age of explants like immature embryos also influences the efficiency of callus induction and hence regeneration capacity (Abhishek *et al.*, 2014).

Apart from explants type, the genotype of the explants is also shown to affect the regeneration potentiality in maize. The nuclear genes have been implicated in controlling regeneration capability (Tomes and Smith, 1985; Hodges *et al.*, 1986). Further, it has been proposed that at least one gene or a block of genes control the somatic embryogenesis of maize in tissue culture (Willman *et al.*, 1989). Bohorova *et al.* (1995) showed the effect of various maize genotypes on somatic embryogenesis. Abhishek *et al.* (2014) reported the effect of different genotypes on embryogenic type II callus production and whole plant regeneration in tropical maize. Thus, significant differences have been reported in callus induction, embryogenic type II calli production, and regeneration potential between various explants and genotypes in maize.



**Figure 1.** Schematic representation of various routes of maize tissue culture procedure

### *Media composition and hormone concentration*

Optimization of tissue culture medium components is of utmost importance for the establishment of a reproducible *in vitro* regeneration system. To date, a range of basal nutrient media has been utilized for tissue culture in various cereal crops and most of the cases. Murashige and Skoog (MS)-based medium (Murashige and Skoog, 1962) is found superior, as it promotes rapid callus growth, embryogenic callus development, and whole plant regeneration (Hanzel *et al.*, 1985; Luhrs and Lorz, 1987; Bregitzer 1992; Rakshit *et al.*, 2010). The first somatic embryos in maize were produced in the year 1975 by Green and Phillips from embryo scutellar tissues. In maize mostly, N6 (Chu), MS or Linsmaier and Skoog (LS)-based culture media have been utilized for *in vitro* regeneration and transformation at different stages. Optimization of various media components, *viz.*, carbon source, vitamins, amino acids and concentration of plant hormones (growth regulators) in the tissue culture medium is imperative for using these media. Depending upon maize genotypes and explants stage, various reducing and non-reducing carbon sources such as glucose, fructose, galactose, and sucrose have been tested in the tissue culture media. However, sucrose is the most widely used carbon source (Yadava *et al.*, 2017). Amino acids such as *L*-asparagine, *L*-glutamine, *L*-arginine, *L*-proline, and *L*-cysteine, etc. being a source of organic nitrogen have been used in tissue cultures to augment somatic embryogenesis and regeneration (Armstrong and Green, 1985; Claparols *et al.*, 1993; Kim and Moon, 2007). Various vitamins such as thiamine, ascorbic acid, pyridoxine, riboflavin, myo-inositol, folic acid, niacin, pantothenic acid, biotin, etc. have also been tested in tissue culture. Vitamins, in combination with other media components, have been shown to improve callus growth, somatic growth, embryonic development, and rooting via affecting secondary metabolite production and cell signaling pathway (Abrahamian and Kantharajah, 2011). Plant growth regulators, mainly cytokinins [6-benzyloaminopurine (BAP), kinetin and Zeatin etc. and auxins like indole-3-acetic acid (IAA), indole-3-butyric acid (IBA), 2,4-dichlorophenoxy-acetic acid (2,4-D), *Dicamba* (3,6-dichloro-2-methoxybenzoic acid) and naphthalene-acetic acid (NAA)] are used in different combinations and concentrations during maize tissue culture by researchers as they played a very critical role

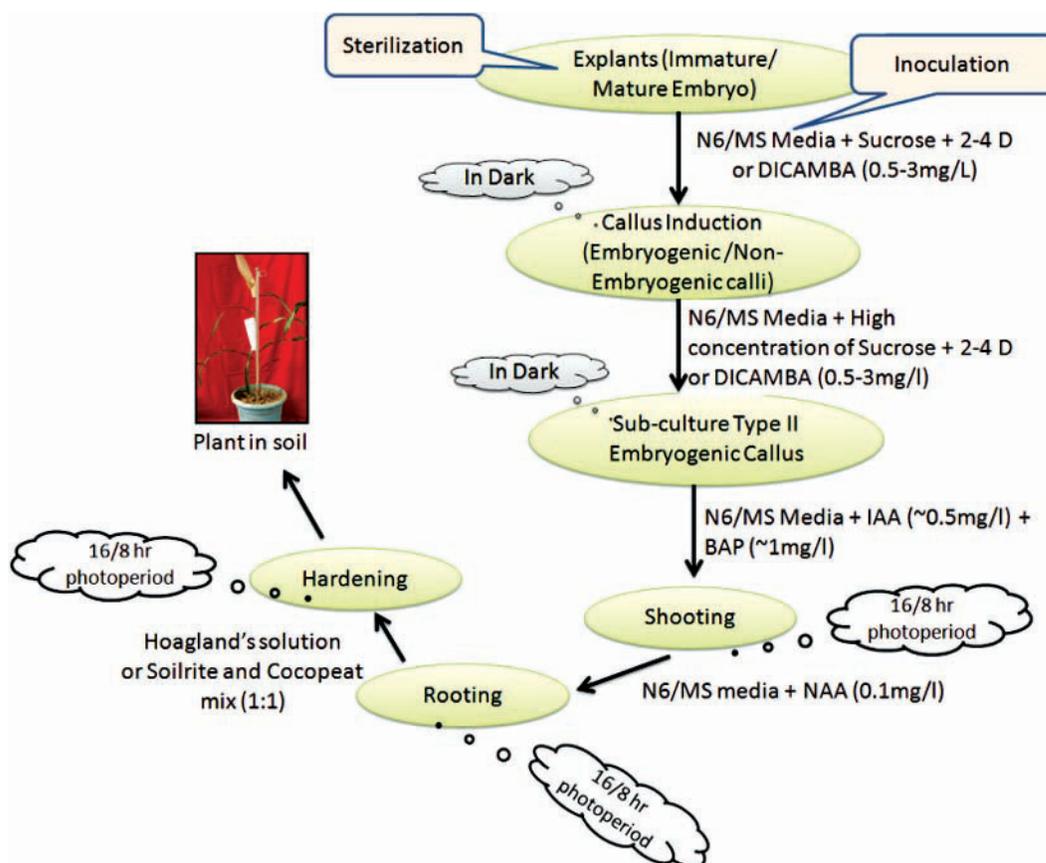
in callus induction, multiplication, shoot development and root development (Bohorova *et al.*, 1999; Frame *et al.*, 2000; Rakshit *et al.*, 2010; Abhishek *et al.*, 2014; Tiwari *et al.*, 2015; Muppala *et al.*, 2020).

### *Callus induction and embryogenic callus production*

In tropical maize factors like genotypes, media, source of auxin and their concentrations significantly affect callus induction frequency (Rakshit *et al.*, 2010). Proline (a source of nitrogen supply and osmoprotectant) and casein hydrolysate are considered very essential amino acids for callus formation but their high concentration may adversely affect callus proliferation (Zhao *et al.*, 2008; Joshi *et al.*, 2010; Wang *et al.*, 2012). The callus obtained from immature and mature embryos were supplemented generally with 2,4-D or *Dicamba* in MS or N6 basal media for the production of regenerable callus (Furini and Jewell, 1994; Rooz 2002; Huang and Wei, 2004; Rakshit *et al.*, 2010) (Figure 2). In maize, two types of embryogenic callus *i.e.* Type I and type II are obtained by *in vitro* culture of different explants. Former is slow-growing, compact/non-friable, harder with characteristic yellow color while later is fast-growing, friable, soft with characteristic white color (Carvalho *et al.*, 1997). Type II callus is more regenerable and best for obtaining a large number of calli via sub-culturing due to its friable nature (Omer *et al.*, 2008; Manivannan *et al.*, 2010).

MS salts have a higher concentration of inorganic nitrogen, but a lower ratio of nitrate to ammonium ( $\text{NH}_4^+$ ) compared to N6 salts (Armstrong *et al.*, 1991; Elkomin and Pakhomova, 2000). It has been shown that the use of MS salts as callus induction media leads to compact Type I callus due to lower nitrate and high  $\text{NH}_4^+$  levels, whereas, use of N6 salts lead to friable Type II callus due to high nitrate level and low  $\text{NH}_4^+$  level (Elkonin and Pakhomova, 2000; Yadava *et al.*, 2017). The callus induction media having 2,4-D, proline, and casein hydrolysate when supplemented with  $\text{AgNO}_3$  have shown to induction of type II callus from immature embryos (Armstrong *et al.*, 1991; Songstad *et al.*, 1991 and El-itriby *et al.*, 2003). The regeneration ability of the callus is highly genotype-specific and also requires specialized skills to visually select the regenerable portion of the callus. A generalized flow diagram of the indirect embryogenesis pathway of maize tissue culture is given in Figure 2.

**Figure 2.** Flow diagram depicting the indirect embryogenesis pathway of maize tissue culture



### Organogenesis and regeneration

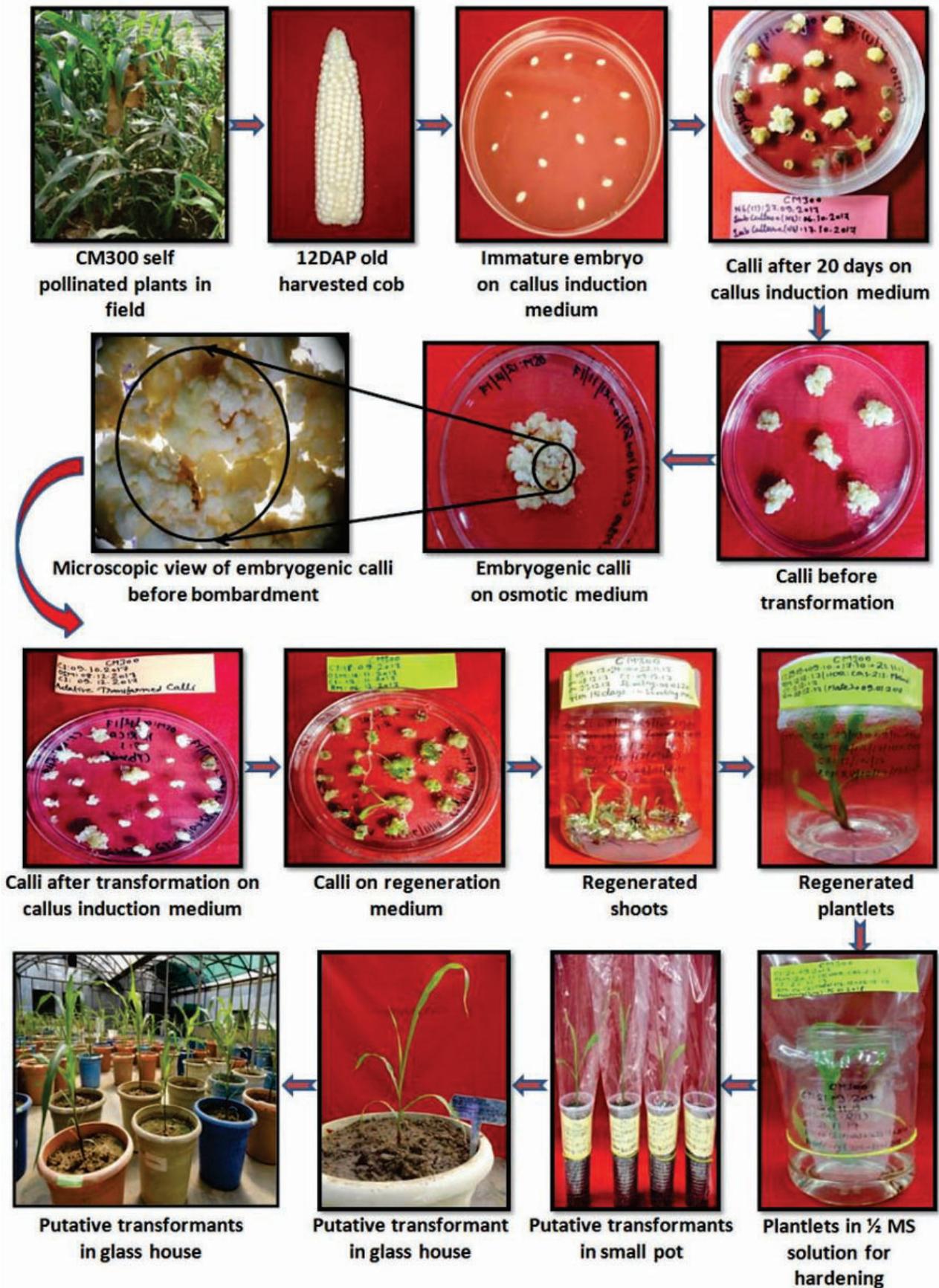
Phytohormones play a crucial role *in vitro* organogenesis during tissue culture. Organogenesis involves three distinct phases – in the first phase, plant cells are de-differentiated to attain organogenic competence; in the second phase, de-differentiated cells are determined for specific organ formation, and in the third phase organ morphogenesis continue. In cereals, the initiation of somatic embryogenesis has been shown to depend on cytokinins. The development of a white, nodulated embryogenic callus in somatic embryogenesis and the formation of green buds during organogenesis suggests divergent modes of differentiation during morphogenesis.

In maize, usually, the application of auxins and cytokinins are essential for successful shoot regeneration and hence for the successful development of plantlets. It has been shown that the phytohormones such as kinetin and BAP are important along with 2,4-D during *in vitro* regeneration for an efficient shoot regeneration/shooting from immature and mature embryos derived calli (Ozcan, 2002; Wang *et al.*, 2006; Jia *et al.*, 2008; Zhao *et al.*, 2008; Rakshit *et al.*, 2010; Wang *et al.*, 2012; Tiwari *et*

*al.*, 2015; Muppala *et al.*, 2020). Depending upon genotype and explants, root regeneration can be done either in a hormone-free medium or by applying auxins (mostly NAA) (Figure 2). Diagrammatic representation of various stages during embryogenic calli formation and *in vitro* regeneration in maize by utilizing immature embryos as explant source is given in Figure 3.

### Conclusion

*In vitro* callusing and regeneration is highly challenging in maize compared to other crops like rice. Among the various explants, the immature embryo is preferred for regeneration in maize due to its better regeneration potential and reproducibility than other explants. However, the availability of immature embryos throughout the year is a major limitation in the rapid development of improved maize cultivars via genetic transformation. Further, *in vitro* regeneration is highly dependent on genotypes and also is influenced by different media components such as phytohormones (mainly auxins and cytokinins), vitamins, amino acids (mainly proline and casein hydrolysate), etc. Despite all these challenges, maize is the prime target crop



**Figure 3.** A pictorial representation of embryogenic calli formation and *in vitro* regeneration in CM300 inbred maize line using immature embryos as explant source

for tissue culture and transformation and hence the maximum number of genetically modified (transgenic) events belonging to various traits have been approved and commercialized in maize globally. Therefore, tissue culture to develop transgenic maize having desired useful trait(s) is a forefront technology for maize improvement.

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# Quality protein maize for food and feed purpose: A nutritive approach

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**Abstract:** Protein energy under nutrition (PEU) and micronutrient deficiency is common in countries where population mainly depends upon cereal grains including maize to derive most of their nutritional requirements. So, proper nutrition through different ways such as nutrient supplementation and biofortification is essential for improvement and maintenance of human health. In spite of important uses of maize, it is deficient in two essential amino acid i.e. lysine and tryptophan along with moderate deficiency of isoleucine. These deficiencies have been addressed through the development of nutritionally improved maize known as Quality Protein Maize (QPM) which possesses the desired concentration of lysine and tryptophan as recommended by Food and Agricultural Organization. Development of QPM holds a great promise to signify its importance not only as quality food but also as quality feed for poultry, piggery and animal sectors.

**Keywords:** Food · Feed · Normal maize · Quality protein maize

## Introduction

Maize (*Zea mays* L.) is gaining popularity among all the cereal grains, due to its increased utilization as food (23 per cent) and livestock feed (63 per cent). It is known as ‘Queen of Cereals’ and ranks third in production after wheat and rice. Most of the population residing in the

developing countries including Africa and Latin America derive almost half of their protein and energy requirements from maize. Maize is consumed in different forms such as corn flakes, cornmeal, porridge, chapattis, flat breads, flour, tortillas, and breakfast cereals etc. India produces various specialty maize such as Quality Protein Maize, pop corn, sweet corn and baby corn along with high yielding maize genotypes (flint and dent corn). Although it contains about 10 per cent of protein, but the nutritional quality of maize protein is poor due to its higher prolamine fraction and lower tryptophan, lysine and threonine content than required for proper nutrition (Carrillo *et al.*, 2007). The low protein quality in the diet increases the risk of Protein Energy Under-nutrition (PEU) which adversely affects pregnant women and children. PEU results in development of kwashiorkor or pellagra also termed as “weaning disease”. Pellagra emerges from the chronic protein imbalance which increases susceptibility to diseases like gastroenteritis and tuberculosis. So, it becomes important to supplement food with an alternative protein source rich in lysine and tryptophan (Liu, 2007; Ullah *et al.*, 2010).

Lysine and tryptophan play an important role in the growth of tissues and protein synthesis. In the body, tryptophan converts to niacin and reduces the chances of pellagra. Biofortification, bacterial fermentation and nutrient supplementation are major strategies to lessen the risk of protein energy under nutrition. The requirement to ameliorate the nutritional profile of maize has been recognized by using conventional breeding techniques with development of QPM. The opaque-2 mutation decreased the zein fraction and increases the non-zein protein fraction which is rich source of lysine and tryptophan. QPM possesses the required potential to improve the nutritional status of the poor populations and to serve as a vehicle for cost savings from expensive infant formulas and corn chips etc. Incorporation of Quality Protein Maize in food

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and livestock feed is the most economical way to increase their protein quality as the amino acid composition of quality protein maize is superior than normal maize (Adom and Liu, 2002; Ruiz-Ruiz *et al.*, 2008; Carrillo *et al.*, 2007).

### Maize Composition

The maize kernel is comprised of starch (@ 70 per cent), protein (@ 10 per cent), oil (@ 2–4 per cent) and vitamins and micronutrient. Maize occupied a well-deserved position as being a ‘poor man’s nutria-cereal due to high proportion of carbohydrates, proteins, fats and micronutrients. In the developing countries particularly in Africa and Latin America, several million people derive half of protein and approx. 56 per cent of energy requirements from maize because the protein from animal source is miserable and expensive for huge population. Maize germ oil obtained through wet milling possess’ potential application in salads dressing and cooking. Roasted maize kernels also possess application as substitute of coffee. Maize is also rich in phytochemicals or bioactive compounds mainly carotenoids (lutein, xanthophylls, zeaxanthin), phytosterols (sitosterol, stigmasterol, campestral) and phenolic compounds (ferulic acid and anthocyanin). Anthocyanins are water soluble pigments imparts reddish to purple colour to maize (Abdel-Aal *et al.*, 2006). Maize is considered as good raw material for the development of various industrial products such as starch, porridge, weaning foods, ethanol etc.

### Quality Protein Maize

The quality of maize protein is poor as about 70 per cent maize endosperm protein (zein) is devoid of lysine and tryptophan. The protein quality of maize kernel was significantly improved with the development of a maize cultivar opaque-2. In 1963, Researcher at Purdue University (USA) found that maize mutants opaque-2 (op2) possess almost twice endosperm protein than present in normal maize. It is followed by the introduction of another mutant named *floury-2* (*fl2*). These mutants derive their names from endosperm characters like soft, opaque or floury (Carrillo *et al.*, 2004). The superior protein quality of high lysine mutant (*o2* and *fl-2*) recognized with undesirable traits as being susceptible to stresses and ear rot diseases. The chalky and soft

endosperm prompt the inconsistency of grain which holds water content in it and causes harvest loss. These are the main limiting factors which restrict the adoption and utilization of mutants. By using conventional breeding techniques International Maize and Wheat Improvement Center (CIMMYT) improved the nutritional profile of maize through constant efforts and overcame the original opaque 2 defect. The new normal looking and tasting opaque 2 mutant were renamed as quality protein maize, a biofortified foods (Milind *et al.*, 2013). The amount of lysine and tryptophan in the maize endosperm protein is almost double in QPM varieties than in non-QPM or normal maize. Furthermore, it also raises the amount of amino acids like aspartic acid, glycine, arginine and histidine along with a decrease in the concentration of alanine, glutamic acid and leucine. A reduction in leucine concentration maintains the leucine and isoleucine ratio which helps in the liberation of tryptophan required for the synthesis of niacin. The comparative evaluation of nutritional components in QPM and normal maize is presented in Table 1.

Due to its better amino acid composition it is more economical to use diets incorporating QPM as it can lead to progressive reduction in the use of fishmeal and synthetic lysine additives. The amino acid composition of

**Table 1.** Comparison between chemical composition (a) and amino acid profile (b) of common and quality protein maize (Abiose and Victor, 2014)

#### A. Chemical composition

| Components           | Common maize | Quality protein maize |
|----------------------|--------------|-----------------------|
| Protein (%)          | 9.80         | 9.72                  |
| Fat (%)              | 4.50         | 4.85                  |
| Ash (%)              | 1.62         | 1.50                  |
| Carbohydrate (%)     | 73.83        | 73.98                 |
| Sodium (mg/100 g)    | 61.65        | 43.88                 |
| Magnesium (mg/100 g) | 141.30       | 137.10                |
| Potassium (mg/100 g) | 77.23        | 79.24                 |

#### B. Amino acid profile of quality protein maize and normal maize

| Amino acid (g/100 g of endosperm protein) | Ganga-5 (Normal) | Opaque-2 |        |         |
|---|------------------|----------|--------|---------|
|   |                  | Shakti   | Rattan | Protina |
| Lysine                                    | 1.88             | 4.07     | 3.76   | 3.68    |
| Tryptophan                                | 0.35             | 0.92     | 0.93   | 0.90    |
| Leucine                                   | 14.76            | 9.19     | 9.72   | 9.26    |
| Protein (%)                               | 8.50             | 9.20     | 9.00   | 6.10    |

the meals from normal and quality protein maize shows that quality protein maize is superior to normal maize in terms of threonine (3.20 g/100 g), methionine (1.20 g/100 g), glutamic acid (7.50 g/100 g), isoleucine (2.74 g/100 g). The level of lysine in QPM (2.64 g/100 g) was higher than the level of lysine in common maize. Due to the nutritional benefits of QPM including high tryptophan and lysine levels, higher niacin bioavailability, calcium and carotene content and lower leucine content, its utilization can be explored for novel and conventional food products without the onset of their quality deterioration.

### Utilization of quality protein maize for food purpose

Quality protein maize having high lysine and tryptophan content has been used in human nutrition and for feeding to monogastric animals which decreases the supplementation or fortification of feed and food material, respectively. Maize is the main staple food in Ghana where it has been reported that children fed with high lysine maize porridge gain weight and become healthier than children fed with common maize porridge (Ignjatovic-Micic *et al.*, 2008). The high level of lysine and tryptophan content increases biological value of QPM to almost double than normal maize. The nitrogen balance index for opaque 2 maize proteins and skim milk protein is 0.72 and 0.80, respectively, which describes that the protein quality of QPM is 90 per cent of that of milk protein. Research studies were conducted to evaluate the nutritional effect of QPM through feeding programs on human and animals (De Valenca *et al.*, 2017). It was revealed that consumption of QPM by infants as well as children increased the rate of weight gain and height by 12 per cent and 9 per cent, respectively (Emerson *et al.*, 1935). Although QPM is a nutritionally improved commodity, but its adaptability in the market is less, due to the lack of remunerative market price and interest in the maize food processors to popularize QPM products. QPM can be processed for the synthesis of a number of food products as follows:

#### *Nixtamalized flour from quality protein maize*

Alkaline treatment (lime) of maize kernel followed by steeping and washing to remove excess alkali is employed to obtain the product termed as Nixtamal and process is

known as Nixtamalization. Nixtamal is ground to produce dough named Masa, a base ingredient to produce tortilla. It is an important step in the production of several maize products such as maize tortilla, tortilla chips, maize chips, flour, snacks and taco shells etc. Nixtamalization improves the nutritional quality particularly partial lipid saponification and starch gelatinization of product.

Carrillo *et al.* (2004) produced nixtamalized maize flour possessing acceptable physico-chemical and functional properties from QPM V-537 and determine the best nixtamalization process combination selected from nixtamalization variable that is time (20–85 min.), time of steeping (8 to 16 hours) and concentration of lime (3.3–6.7 g Ca(OH)<sub>2</sub>/l). The optimum lime concentration, cooking and steeping time of nixtamalization to prepare nixtamalized flour were found to be 5.4 g, 31 min and 8.1 hours, respectively.

#### *Temph flour*

Solid state fermentation of boiled soybean, corn, wheat and groundnut using different *Rhizopus* spp. is employed to produce and improve the nutritional quality as well as palatability of traditional food namely Temph. These strains work by synthesizing enzymes to hydrolyze food constituents, reduce or eliminate anti-nutritional factor and to produce product with desired flavor and textural properties.

Rodrigue *et al.* (2004) evaluated and compared the nutritional and physico-chemical properties of unfermented QPM and QPM temph flour, soaking in acetic acid solution at 25°C for 16 hr followed by cooking at 90°C for 30 min and cooling at 25°C can be employed. Researchers reported that with solid state fermentation process, amino acid composition of product was improved as the content of histidine (0.81 g/100 g protein), isoleucine (0.52 g/100 g protein), leucine (1.46 g/100g protein), and tryptophan was increased (p<0.05). Fermented QPM flour with improved protein digestibility, protein efficiency ratio (2.10), PDCAAS (0.83), can be further found application for the fortification purpose to fortify different food products such as tortillas, bread, cookies etc. Inoculation of cooled grains can be done with *Rhizopus oligosporus*. This process also results in the reduction of antinutritional factors and increase in amino acid availability, protein digestibility.

### ***Instant flour***

Extrusion cooking is employed for the development of instant flour from QPM with little effluent problem as production of instant flour by nixtamalization process is time consuming and leads to high effluent waste. In extrusion cooking, various process variables such as screw speed, feed rate, feed moisture content, temperature, die shape and size, affect the properties of product. During extrusion cooking, over-gelatinization of starch reduces.

Moreno *et al.* (2003) developed instant flour with desirable functional properties using single screw extruder by mixing of maize grits (V 537) with alkali and to obtain moisture content up to 28 per cent. The process variables were temperature of extrusion process (70–100°C), screw speed and concentration of lime that varied from 0.1–0.3 per cent and 30–80 rpm, respectively. The optimum extrusion conditions for the preparation of instant flour can be temperature 79.4°C, lime concentration 0.24/100 g and screw velocity 73.4 rpm having highly acceptable physicochemical and functional properties.

### ***Weaning food***

Carrillo *et al.* (2007) determined the best proportion of chickpea and quality protein maize for preparation of weaning food and assessed the nutritional quality of developed product. The extrusion cooking of nixtamalized QPM and chickpea was employed at 79.4°C and 150.5°C at 73.5 rpm and 190.5 rpm, respectively. True protein and available lysine content of developed product was evaluated. It was reported that the most suitable combination of nixtamalized quality protein maize flour and chickpea flour was 21.2 per cent and 78.8 per cent, respectively. The final product contained 20.07 per cent protein content, 71.14 per cent carbohydrate content and covers amino acid profile except tryptophan, suitable for growth of infant.

### ***Extruded Instant porridge***

Extruded products with various shapes, flavours, textures and colours are developed in the food industry. The most widely consumed extruded products are made primarily with cereals/grains but they tend to have low protein quality and many other nutrients. Therefore, it is necessary to

optimize the extrusion conditions for the development of nutritious extruded products. Kaur *et al.* (2019) optimized the extrusion process variables to develop and compare functional properties of normal maize and QPM based instant porridge. Independent variables such as moisture content, barrel temperature and screw speed were varied from 14–18 per cent, 125–175 °C, and 400–550 rpm, respectively. Carbohydrate, Protein solubility, hydration power and milk absorption capacity (MAC) were assessed using response surface methodology (RSM). It was reported that screw speed exhibited non-significant ( $p \leq 0.01$ ;  $p \leq 0.05$ ) affects on HP and CS while feed moisture and barrel temperature exhibited significant affect ( $p \leq 0.01$ ;  $p \leq 0.05$ ) on PS, HP and MAC. Optimum extrusion processing variables for development of QPM based porridge were ranged from 411.61–466.50 rpm screw speed, 14.19–15.36 per cent feed moisture at 150°C. Study showed that extruded product can be developed from quality protein maize instead of normal maize without affecting functional properties of extrudates negatively.

### ***Quality protein maize for feed purpose***

For monogastric animals, complete protein sources are required for their growth and QPM plays an important in poultry feed and piggery. Utilization of QPM for feed purpose reduces the demand of protein supplementation up to 50 per cent and decrease the requirement to incorporate synthetic lysine. Maize is mainly used to as livestock feed because of its high energy, fibres and easy digestibility. But maize varieties are low in lysine and tryptophan content. So supplementations from other protein sources become necessary. The inclusion of QPM in the diet of monogastric animals and weaner pigs increases their weight and growth performance. Addition of QPM also reduces the cost of feed formulation by 5 per cent, this is very significant saving for developing countries (Groote *et al.*, 2010).

### ***Weaner pigs***

Quality protein maize incorporation in feed formulation indicates an increase in weight of weaner pigs by 29.8 per cent (De-Quan and Shi-Huang, 1994) as compared to normal maize based feed. Addition of quality protein also reduces the cost of feed formulation by 5 per cent, this is very significant saving for developing countries.

Piglets at the initial stage of weaning requires more lysine and tryptophan as compare to growers and this situation leads to expensive supplementation by using commercial grade amino acids especially from soyabean meal.

## Conclusion

Consumption of food with low nutritious value in terms of protein quality is mainly responsible for protein energy under nutrition which leads to morbidity and mortality of children. Advancement in technology continuously efforts which leads to the development of biofortified crops like QPM to reduce malnutrition in target countries. QPM possess a potential to reduce protein energy under nutrition in countries where population is solely depends on maize for their protein and energy requirements. But QPM has inbuilt drawback in terms of agronomic performance as compared to non-QPM varieties. Hence, there is need to introduce initiatives for improvement and adaptability of quality protein maize as potential food component.

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## Multi-environment field testing to identify better performing and stable genotypes for water logging stress tolerance in tropical maize

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**Abstract:** Maize is the third most important cereal crop after wheat and rice in the world. Nearly 80 per cent of maize in India is grown under rainfed ecology. In the era of climate change, its production and productivity are limited by several biotic and abiotic stresses. Amongst the abiotic stresses, drought and water logging stresses are the major one. Considering it, the current study was carried out to evaluate and select water logging tolerant genotypes suitable for stress-prone ecology. A set of 70 maize varieties consisting of 61 single cross hybrids and 09 composites of late-medium duration and 13 single cross hybrids of early duration were evaluated in two separate trials at three different environments. Two same sets were constituted in each trial for evaluation one under water logging stress and another as control set where no water stress was imposed. The overall grain yield under stress conditions in different environments was varied from 4806.7 to 1715.4 kg/ha in late-medium trials and 3727.2 to 1418.3 kg/ha in early. Similarly, the yield under non-stress was varied from 5949.3 to 2926.3 kg/ha in late-medium and 5324.0 to 2873.2 in early duration trials. The analyses done using the Additive Main effect and Multiplicative Interaction (AMMI) model, has differentiated

the test genotypes based on their performance and interaction with the target environments. Based on AMMI selection, in the late-medium group, three hybrids, such as IMH 1527, DMRH 1419 and CMH-08-292 (yielded > 5000 kg/ha) and in the early, VMH 51 and IMH 1533 (> 4000 kg/ha) were repeatedly found in first four selection in more than one environments. Further, tolerant hybrids of medium duration were validated by planting in large plots size under water logged and control conditions with respect to the susceptible one. The non-significant yield reduction ranging from 5.7 per cent to 12.8 per cent was observed in the tolerant hybrids, however in susceptible, it was highly significant ranging from 64.9 per cent to 82.1 per cent. The anatomical study showed a sufficient number of well-developed aerenchymatous cells in the roots tissues of tolerant hybrids under water stress conditions. The identified tolerant hybrids can be explored in stress-prone ecologies and can be used as source germplasm for further diversification of water logging stress breeding programme.

**Keywords:** Anatomical study · Genotype stability · Maize · Multi-environments · Water logging stress

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### Introduction

Maize (*Zea mays* L.) is one of the diverse cereal crops that can be cultivated in a wide range of ecologies and seasons. In India, maize growing ecology has been categorized in five different zones for effective evaluation and its cultivation. The harvest of maize has diverse uses such as it can be used as feed, food, fodder as well as to prepare industrial by-products. In India, more than 1000

products and in the USA and other countries nearly 3500 products are being developed from maize. Maize is being grown as a rainfed crop in India where nearly 80 per cent of the total area is under rainfed and the remaining 20 per cent is irrigated (Kumar *et al.*, 2016). Being a rainfed crop, it has to face several abiotic and biotic stresses due to the country's unpredictable and extremely changing weather patterns. The prominent abiotic stresses in the rainfed ecology are drought and water logging stress (Kumar *et al.*, 2016). Now due to climate change, the plant has to face both of these stresses very frequently in the same growing season (Kumar *et al.*, 2020). Around 15 per cent of the maize-growing areas are affected by flooding in Southeast Asia, resulting in a yearly production loss of 25–30 per cent (Chen *et al.*, 2014).

Maize is susceptible to water logging especially at its early vegetative stages. Water logging stress at early growth stages causes severe reductions in plant height, dry matter accumulation, and ultimately affects yield (Mukhtar *et al.*, 1990; Rosenzweig *et al.*, 2002; Rao and Li, 2003; Liu *et al.*, 2010). In water logged conditions, the root's ability to supply nutrients and water for plant growth and development is severely inhibited (Simone *et al.*, 2002; Smethurst and Shabala, 2003).

Yield stability is one of the prime objectives in the development of plant varieties that have high yield potential in combination with the wider adaptability over different agro-climatic conditions (Kumar *et al.*, 2016 & 2017). Genotypes which yield better over different agro-climatic conditions are more adaptive and stable. Thus, a comprehensive study of the adaptability and stability of genotypes is a major aim in plant breeding programs (Das *et al.*, 2011). The evaluation of the genotypic performance of hybrid maize cultivar candidates in many environments generates valuable data to determine how stable and adapted genotypes are (Crossa, 1990). To increase accuracy and to study the genotype  $\times$  environment interaction (GEI), the different statistical analyses like Additive Main effect and Multiplicative Interaction (AMMI) and Genotypic & Genotypic-by-Environment (GGE) are being used worldwide for interpreting Multiple Environment Trial (MET) (Choudhary *et al.*, 2020). In AMMI analyses we considered the combination of ANOVA and principal component analysis (PCA) together for interpreting G  $\times$  E and stability of the genotypes (Sadeghi *et al.*, 2011; Choudhary *et al.*, 2020). Measurement of the performance of the genotypes and their stability over

several environments particularly in terms of their yields in normal and stress-prone ecology is the primary objective in any maize breeding programme. Considering the facts, the current study was carried out to evaluate the different maize hybrids and varieties in multiple environments under water logging stress as well as in normal (non-stress) conditions to identify high yielding and stable tolerant genotypes for high moisture stress. The development of genotypes having the ability to withstand waterlogged stress conditions could be beneficial and suitable for maize-growing farmers of the country.

## Materials and methods

### Experimentation

In the current study, 70 maize varieties (9 OPVs and 61 hybrids) of medium-late and 13 hybrids of early maturity were used as experimental materials which were evaluated for water logging stress in three different environments as Begusarai, Delhi and Ludhiana during *Kharif* season of 2018. The maize genotypes were evaluated using randomized block design with two rows of 3 m length in two different replications. Two sets of 70 genotypes were constituted for each location to evaluate one under water logging stress and another as control where no stress was applied. The water logging stress was imposed at the seedling stage i.e. 25 days after sowing by stagnating water up to 5–8 cm above the soil surface in the stress plots continuous for ten days. Remaining, all packages of practices were adopted as followed in the control plots. In the control plots, no stress was imposed and normal packages of practices were followed to raise the healthy crop. The hand weeding was done to keep the field weed-free. To further supplement the managed water logging stress, the sowing was taken up in the first week of July, so that it gets coincide with more rainfall at the critical plant growth stage. The observations were recorded for aerial roots development, per cent death of the seedlings, lodging, days to anthesis and silking, and plots yield. The tolerant hybrids identified based on the multiple environments evaluation were further validated under water logging conditions with the susceptible checks on large plots size (45.5 m<sup>2</sup>) to confirm their performance. The trial for validation was conducted during *kharif* season of 2019 at the Delhi location.

### Statistical analysis

The data recorded on grain yield was subjected to AMMI analysis which combines Analysis of Variance (ANOVA) with additive and multiplicative parameters into a single model (Gauch, 1988) using GenStat 17<sup>th</sup> Ed. (2014). The output was used for analyzing the yield performance and stability of the genotypes in the water logging and normal conditions in different environments. The simultaneous study of the genotype plus genotype-environment interaction was also performed.

### Results and discussion

The grain yield under stress conditions in different environments was varied from 4806.7 to 1715.4 kg/ha in late-medium trials and 3727.2 to 1418.3 kg/ha in early maturing trials (Table 1). Similarly, the yield under non-stress was varied from 5949.3 to 2926.3 kg/ha in late-medium and 5324.0 to 2873.2 in early (Table 1). The highest and lowest yielding environments in both the trials were Delhi and Begusarai, respectively.

The Begusarai center is in the north east plain zone in which generally, heavy rains occurred during *khari* season and hence resulting in more yield loss. The sowing

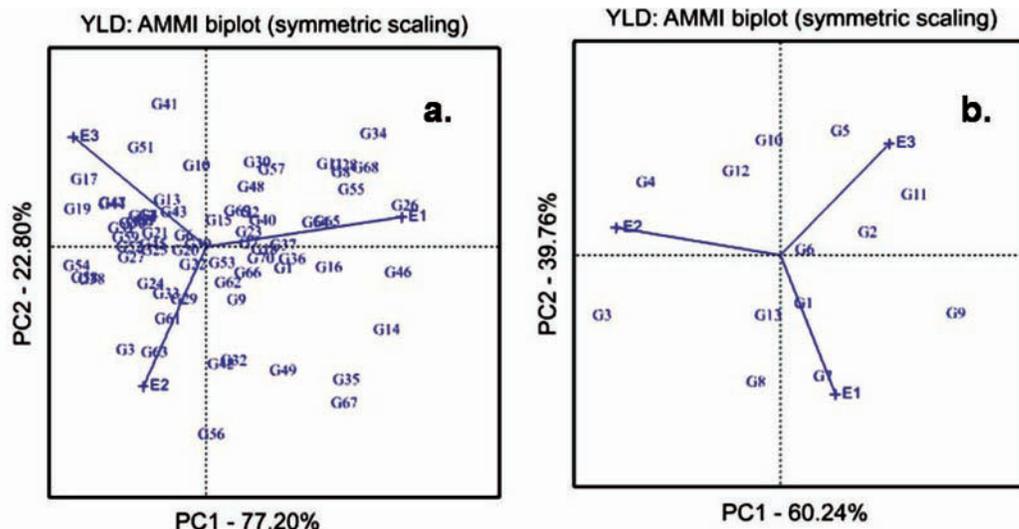
was taken up in the first week of July that also has further contributed to face heavy rainfall by the plants at their critical growth stage. The AMMI analysis shows that all three environments were different and clearly categorized the test entries with respect to their interaction with the target environment (Figure 1).

This also indicated that these testing sites can be considered to provide three different testing environments for the evaluation of genotypes under water logging stress (Baraki *et al.*, 2014). The genotype main effect plus genotype by environment interaction (GGE) biplot used principal component comprised of a set of elite genotypes scores multiplied by environment scores which gives a two-dimensional biplot (Ding *et al.*, 2008; Sadeghi *et al.*, 2011; Ayed *et al.*, 2016). The best four genotypes were selected using AMMI selection based on their performance under water logging stress. The details of the first four genotypes selected based on AMMI selection are given in Table 2. In the late-medium group, three hybrids, such as IMH 1527, DMRH 1419 and CMH-08-292 of the medium maturity group were repeatedly found in first four performing genotypes under water logging stress in more than one environment (Table 2). They were yielding, more than 5.0 t/ha under water logging stress. Similarly, in the early maturity group, two hybrids such as VMH 51 and

**Table 1.** Overall mean yield observed at various locations for different maturity trials under water logging stress and non-stress conditions

| Locations      | Late-medium maturing trial |                   | Early maturing trial |                   |
|----------------|----------------------------|-------------------|----------------------|-------------------|
|                | Non-stress (kg/ha)         | Stress WL (kg/ha) | Non-stress (kg/ha)   | Stress WL (kg/ha) |
| Delhi (E1)     | 5949.3                     | 4806.7            | 5324.0               | 3727.2            |
| Ludhiana (E2)  | 5447.2                     | 3724.0            | 4732.3               | 2177.2            |
| Begusarai (E3) | 2926.3                     | 1715.4            | 2873.2               | 1418.3            |

**Figure 1.** The AMMI biplot graphs, **a.** Late-medium group and **b.** Early group showing the interaction of genotypes in different environments (E1: Delhi; E2: Ludhiana; E3: Begusarai).



IMH 1533 were repeatedly found in the first four performing genotypes under water logging stress in more than one environment (Table 2). The grain yield of these hybrids under water logging stress was more than 4.0 t/ha. The IMH 1527 in the medium group and VMH 51 in early were the hybrids that repeatedly found a place in the first four in all three environments. These genotypes were also performing relatively better in non-stress conditions, hence can be considered for cultivation in water logging prone maize ecologies of the country.

To further validate the performance of water logging stress-tolerant hybrids, during *kharif* season of 2019, medium-duration hybrids such as IMH 1527, DMRH 1419 and CMH-08-292 were planted along with three susceptible checks *viz.*, SMH 1, SMC 6 and SMC 4 at Delhi location. All three tolerant hybrids have yielded > 6.0 t/ha grain yield, however, the susceptible one was yielding ranging from 2.0 to 3.2 t/ha. In the tolerant hybrids, there was a non-significant yield reduction observed from their control treatment (Figure 2). The yield reduction was ranging from 5.7 per cent, in the case of IMH 1527, to 12.8 per cent in the case of DMRH 1419. Similarly, in the case of

the susceptible entries, a significant yield reduction was observed ranging from 64.9 per cent in the case of SMC 4 to 82.1 per cent in the case of SMH 1 (Figure 2). IMH 1527 was yielded the highest amongst the tolerant ones with a yield of 6871 kg/ha.

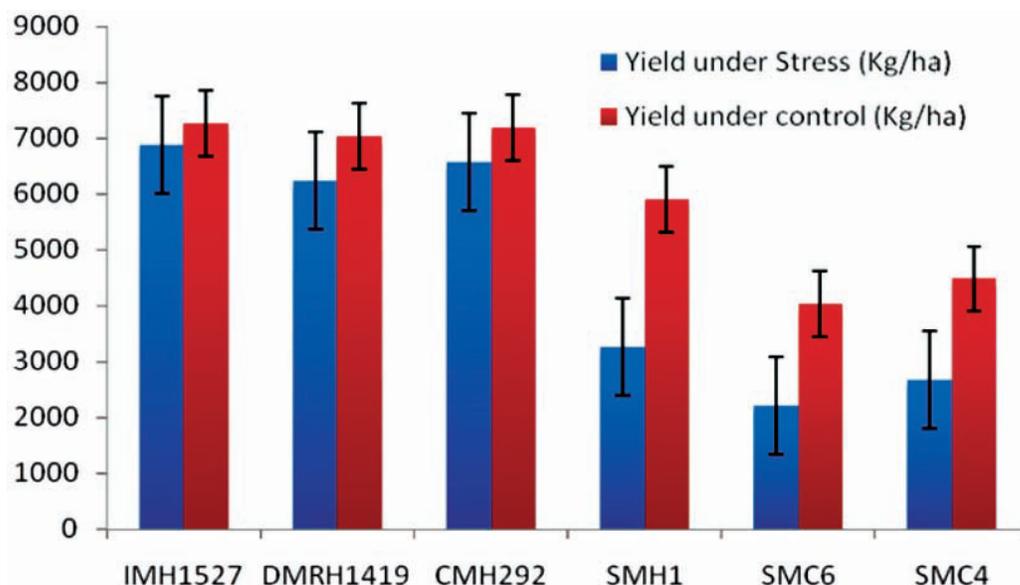
The roots tissues were studied for aerenchyma development for these selected tolerant and susceptible hybrids under water logging stress and control conditions. The anatomical study showed a sufficient number of well-developed aerenchymatous cells in the intercellular regions of tolerant genotypes as compared to the susceptible one (Figure 3).

Aerenchyma can facilitate the movement of various gases (O<sub>2</sub>, CO<sub>2</sub>, ethylene, and methane) in and out of tissues, and move oxygen from the stem to the root in plants exposed to flooding conditions which reduce hypoxic stress. The identified water logging tolerant hybrids in the current study can be explored in stress-prone maize growing ecologies of the country. These hybrids further can be utilized in breeding programme to develop new germplasm for water logging stress tolerance.

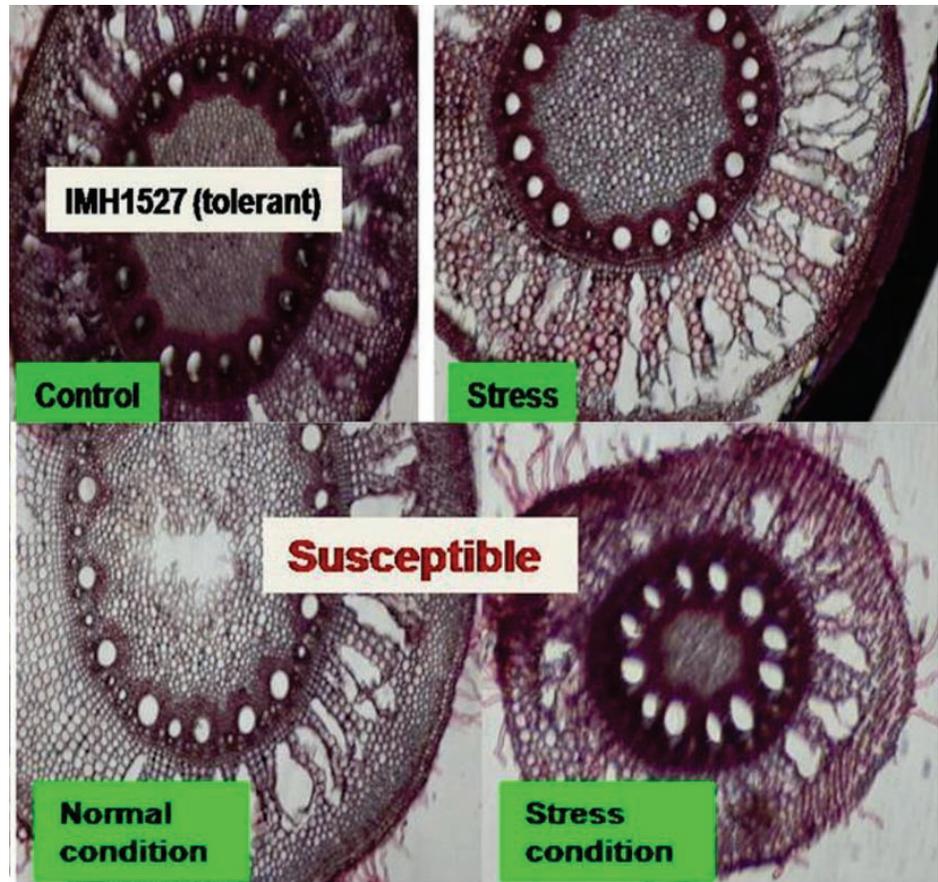
**Table 2.** Best four maize performing genotypes under water logging stress in different locations selected based on AMMI analysis

| Locations | Late-medium maturing |                 |                 |                 | Early maturing  |                 |                 |                 |
|-----------|----------------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|
|           | 1 <sup>st</sup>      | 2 <sup>nd</sup> | 3 <sup>rd</sup> | 4 <sup>th</sup> | 1 <sup>st</sup> | 2 <sup>nd</sup> | 3 <sup>rd</sup> | 4 <sup>th</sup> |
| Delhi     | IMH1527              | DMRH1419        | CO(H)M9         | CMH08-292       | PMH2            | VMH39           | VMH51           | IMH1533         |
| Ludhiana  | CMH-08-287           | CMH08-292       | DMRH 1384       | IMH1527         | IMH1533         | PMH5            | VHM51           | VMH47           |
| Begusarai | DMRH1419             | IMH1527         | DMRH 1415       | KMH22168        | LQMh1           | VHM51           | IMH1529         | VQPM9           |

**Figure 2.** Performance of the selected water logging tolerant and susceptible maize hybrids and varieties on large plots size under high moisture stress and control conditions at Delhi location. The trial was conducted to validate the response of cultivars under water logging stress.



**Figure 3.** Aerenchyma development under water logging stress and non-stress conditions in the tolerant IMH 1527 and susceptible (SMH 1) maize hybrids. Comparing the roots dissection of stress versus non-stress, sufficient numbers of aerenchyma can be seen in IMH 527 under stress conditions. On the contrary, the susceptible maize genotypes produced a comparatively much lesser number of aerenchyma cells in water logging stress.



## Conclusion

In the current study, through multi-environments, testing of maize genotypes under water logging stress as well as non-stress conditions, the three tolerant hybrids under medium duration *viz.*, IMH 1527, CMH-08-292 and DMRH 1419 and two under early such as VMH 51 and IMH 1533 were identified and selected. The medium duration hybrids were further validated for their performance under water logging stress by planting in large plots size. The anatomical study showed a sufficient number of well-developed aerenchymatous cells in the intercellular regions of tolerant genotypes as compared to the susceptible ones. The selected hybrids can be recommended for the cultivation of water stress-prone ecologies and also can be utilized as source germplasm for further diversification of water logging stress breeding programme.

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## Electron beam effects and determination of GR50 dose in tropical field corn

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**Abstract:** The maize improvement aimed at enhancing its productivity to feed the increasing world population and is largely dependent on genetic variability present in the breeding material. In the present study, additional genetic variation was induced by a physical mutagen, electron beam (EB). The experiments were conducted for determining the best dose for a 50 per cent reduction in growth (GR 50). The seeds of maize inbred line, PML 93, treated with 10 different doses (50 Gy, 100 Gy, 150 Gy, 200 Gy, 250 Gy, 300 Gy, 400 Gy, 500 Gy, 600 Gy, and 700 Gy) of EB, were tested for seedling growth parameters using pot culture method. Data on various seedling growth parameters such as per cent germination, seedling length, root length, fresh and dry weight, vigor index I and II were recorded. Significant variation was found among the treatments and 200 Gy of the electron beam was identified as the best GR 50 dose for the inbred line PML 93. The cellulase and pectolyase enzymes were deployed for the root tip cell wall digestion and karyotyping was performed to understand the effect of mutation EB doses on chromosomes. Compared to control, the mutagens with a dose of 200 Gy, induced chromosomal breakage at one or two places. These

aberrations may lead to heritable variations that can be used for crop improvement in the future.

**Keywords:** Inbred line · Electron beam · GR 50 · Karyotype · Vigour index

### Introduction

Maize is an important cereal crop having an array of diverse uses, as food, feed, and raw material for agro-based industries. Though maize crop shows high genetic variation, genetic enhancement of specific traits needs specialized methods to create novel variation. Mutagenesis is one such option in maize breeding that enables heritable variations (Saif-u-Malook *et al.*, 2015). Several mutagens have been used for mutagenesis in crop plants. They are broadly classified as physical and chemical mutagens. Further, physical mutagens are classified as classical radiation mutagen, particle mutagen, and space radiation mutagen (Ma *et al.*, 2021). The selection of suitable mutagen and the procedure to handle mutant generations to isolate desirable mutants depends on the breeding objective and the target trait (Shrivastava *et al.*, 2021). Apart from the conventional electromagnetic radiations like X-rays and gamma rays, the electron beam is now an alternative source of energy to induce mutations in plants (Mondal *et al.*, 2017; Sao *et al.*, 2020). Unlike gamma irradiators, electron beam accelerators have prompt radiation which can be stopped by switching off the source of electrons and radiofrequency power (Mittal, 2012). However, the use of electron beams in mutation breeding was limited due to the availability of the source and particularly the problem of getting low-dose delivery units for mutation breeding applications. However, a linear

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electron accelerator-based facility presently operational at Raja Ramanna Centre for Advanced Technology (RRCAT), Indore, India, overcomes all limitations (Mondal *et al.*, 2017). The same facility was used to induce the mutation in stable maize inbred line, PML 93. It is a conventionally derived inbred line, has excellent general combining ability and high yield *per se* (3.5 t/ha). It has all the required desirable characteristics to be used as female parental lines in the hybrid breeding program. However, the kernel size of the inbred line falls into the small category, as per the maize DUS descriptor (Das *et al.*, 2006.), which becomes a limitation in economical hybrid seed production and fetching farmers' preference when PML 93 is a seed parent. The mutagenesis is expected to create variability for seed size in PML 93 while retaining its high combining ability.

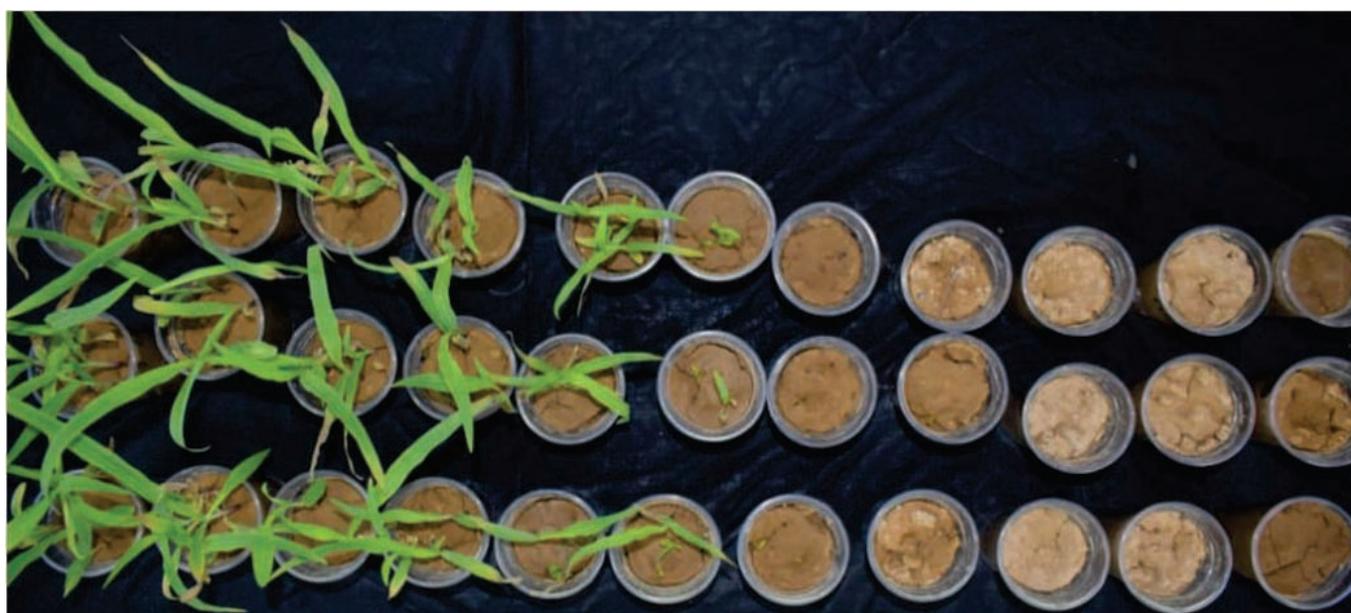
An understanding of the radio-sensitivity of the genotype is required beforehand for selecting the optimal dose for a specific genotype i.e., the dose that might produce the desired mutant (a mutant trait in low mutational load genetic background) at a frequency that can be detected in the mutagenized population (Bado *et al.*, 2016). Limited studies have documented the mutagenic potential of the electron beam in maize. It is important to have a dose that makes a trade-off between radiation-induced damage/lethality and an effective number of mutants induced by them (Ahloowalia *et al.*, 2004). Doses of mutagen that lead to 50 per cent lethality or 50 per cent

growth reduction are considered as LD50 or GR50, respectively, beyond which isolation of useful and effective mutant is difficult (Viana *et al.*, 2019). Hence, the present investigation was conducted to determine the GR 50 dose required to induce desirable mutation in maize crop using inbred line, PML 93.

## Materials and methods

### *Mutagen treatment*

The seeds of test inbred line *viz.*, PML-93 were selected from the two different lots (Lot-I and Lot-II), maintained during post rainy season, 2020 at two different locations (ICAR-Indian Agricultural Research Institute, New Delhi and ICAR-IARI, Regional Research Centre, Dharwad). Germination test of unirradiated seeds was carried out using the standard paper towel method, in which Lot-II showed 100 per cent germination. A total of 100 seeds from the lot- II were irradiated with ten different doses of electron beam *viz.*, 50 Gy, 100 Gy, 150 Gy, 200 Gy, 250 Gy, 300 Gy, 400 Gy, 500 Gy, 600 Gy, and 700 Gy at Raja Ramanna Centre for Advanced Technology, Indore, India. Unirradiated seed samples were used as the control for the comparative analysis. The control and irradiated seeds were pre-treated with Bavistin (2 g/kg) before seeding in the pots to prevent fungal infections.



Control 50Gy 100Gy 150Gy 200Gy 250Gy 300Gy 400Gy 500Gy 600Gy 700Gy

**Figure 1.** Effect of EB irradiation on the seedling growth parameters of inbred line, PML 93

### Pot experiment

A total of 30 seeds each for all ten dosages of EB were sown in the pot (6-inch diameter) containing solarized soil medium. Total three biological replications were placed in a completely randomized design under natural conditions. Pots were irrigated as and when required, with potable water. The seedling growth parameters were recorded 10 days after germination of the control seed. The growth parameters *viz.*, per cent germination (number of germinated seeds / total number of seeds)  $\times$  100), seedling length (root and shoot length), seedling fresh weight and dry weight (oven-dry weight), vigor index I [(mean root length + mean shoot length)  $\times$  per cent seed germination] and vigor index II [(mean root dry weight + mean shoot dry weight)  $\times$  per cent seed germination] were recorded on individual seedlings and the mean value of replications was used for statistical analysis using SAS 9.3 v (SSCNARS, IASRI, New Delhi).

### Karyotyping

To assess the effects of EB on chromosomes, the meristematic region of roots was studied using an enzyme-based method proposed by Snowdon *et al.* (1997) Young growing root tips of 1–2 cm length (10 days after germination) were fixed at the metaphase I stage in Carnoy's solution (cold ethanol: glacial acetic acid (3:1)). At first, the root tips were rinsed (2  $\times$  10 minutes) in double-distilled water and were incubated in citrate solution (0.01M, pH 4.5) for 15 minutes. Root tips were treated with enzyme solution containing 5:1 cellulase (Onozuka R-10 cellulase, Yakult Honsha Co. Ltd., Japan) and pectolyase (Y23 pectolyase, Seishin Pharmaceutical Ltd., Japan) prepared in citrate solution (0.01M, pH 4.5) and incubated for 1 hour 5 minutes at 37°C. Treated root tips were scrambled on glass slides in Carnoy's solution and covered with a coverslip of width about 1 cm and

visualized under a microscope (LEICA DM 750, Germany) at 100  $\times$  (100  $\times$ /1.25 oil) and images were captured through a high-resolution camera (LEICA DFC 3000 Germany) attached with it.

### Results and discussion

Analysis of variance indicated the presence of significant differences for all the studied parameters *viz.*, root length, shoot length, root dry weight, shoot dry weight, vigor index-I, and vigor index-II among different doses of EB (Table 1).

The effect of different doses of the electron beam on studied growth parameters are presented in Table 2 and Figure 2. The highest per cent seed germination was observed in the control seed (91.7 per cent) followed by seed treated with 50 Gy EB (76.7 per cent). The germination percentage was reduced to half for the seeds treated with 250 Gy of EB and it reached zero when the dosage was 500 Gy or more. Compared to control (17.8 cm), the mean root length (MRL) was found to decrease from 16.7 to 0.43 cm when the radiation dose was increased from 50 Gy to 400 Gy and further increase in the dose showed a lethal effect.

Upon exposure to different doses varying from 50 Gy to 700 Gy, the observed shoot lengths were 19.9 cm, 19.7 cm, 16.3 cm, 15.4 cm, 7.9 cm, 1.76 cm, and 1.43 cm at 50 Gy, 100 Gy, 150 Gy, 200 Gy, 250 Gy, 300 Gy, and 400 Gy, respectively. The doses higher than 400 Gy were lethal.

The mean root dry weight (MRDW) ranged from 0.02 (unirradiated) to 0.00 (300Gy). The 50 per cent or more reduction in MRDW in comparison to control was observed from the dose 150 Gy and it reached zero when the dose was raised to 300 Gy. A similar trend of decreasing mean shoot dry weight (MSDW) with increasing electron beam dosage was noticed that ranged from 0.05 g (unirradiated) to 0.00 (400 Gy). The growth reduction with respect to MSDW started at 150 Gy.

**Table 1.** Analysis of variance for growth parameters of PML 93 under different concentrations of electron beam irradiation

| Sources of variation | MSS |          |          |          |          |           |         |
|----------------------|-----|----------|----------|----------|----------|-----------|---------|
|                      | df  | RL       | SL       | RDW      | SDW      | VI- I     | VI- II  |
| Replication          | 2   | 1.62     | 0.24     | 0.00     | 0.00     | 46.54     | 0.46    |
| Treatment            | 10  | 157.19** | 253.32** | 0.0002** | 0.0014** | 3704.51** | 17.99** |
| Error                | 20  | 1.24     | 1.35     | 0.00     | 0.00     | 28.53     | 0.42    |

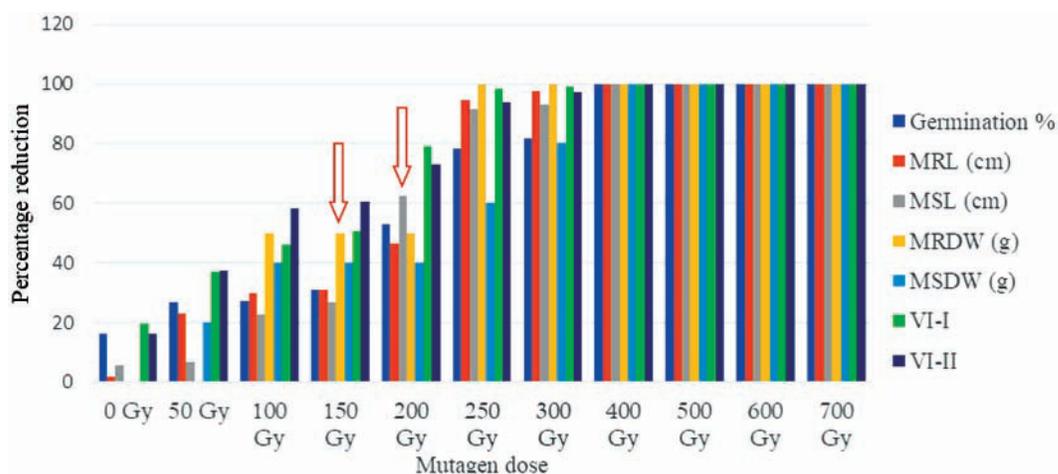
\*\* : Significant at 1% probability, Abbreviations: PT- Paper Towel, RL- Root Length, SL- Shoot Length, RDW- Root Dry Weight, SDW- Seed Dry Weight, VI- I- Vigour Index I, VI- II- Vigour Index II.

**Table 2.** Effect of Electron beam irradiation on maize seedling growth parameters

| S.No. | Treatment | Germination % | MRL (cm) | MSL (cm) | MRDW (g) | MSDW (g) | VI-I    | VI-II |
|-------|-----------|---------------|----------|----------|----------|----------|---------|-------|
| 1     | 0 Gy      | 91.7          | 17.0     | 21.1     | 0.02     | 0.05     | 3493.8  | 6.42  |
| 2     | 50 Gy     | 76.7          | 16.7     | 19.9     | 0.02     | 0.05     | 2807.2  | 5.37  |
| 3     | 100 Gy    | 67.0          | 13.1     | 19.7     | 0.02     | 0.04     | 2197.6  | 4.02  |
| 4     | 150 Gy    | 66.7          | 11.9     | 16.3     | 0.01     | 0.03     | 1880.9  | 2.67  |
| 5     | 200 Gy    | 63.3          | 11.7     | 15.4     | 0.01     | 0.03     | 1718.6  | 2.53  |
| 6     | 250 Gy    | 43.3          | 9.1      | 7.9      | 0.01     | 0.03     | 736.96  | 1.73  |
| 7     | 300 Gy    | 20.0          | 0.9      | 1.8      | 0.00     | 0.02     | 53.20   | 0.40  |
| 8     | 400 Gy    | 16.7          | 0.4      | 1.4      | 0.00     | 0.01     | 31.10   | 0.18  |
| 9     | 500 Gy    | 0.0           | 0.0      | 0.0      | 0.00     | 0.00     | 0.00    | 0.00  |
| 10    | 600 Gy    | 0.0           | 0.0      | 0.0      | 0.00     | 0.00     | 0.00    | 0.00  |
| 11    | 700 Gy    | 0.0           | 0.0      | 0.0      | 0.00     | 0.00     | 0.00    | 0.00  |
|       | Mean      | 40.5          | 7.4      | 9.5      | 0.01     | 0.02     | 1186.70 | 2.20  |
|       | SD        | 34.3          | 7.2      | 9.2      | 0.01     | 0.02     | 1320.34 | 2.49  |

Abbreviations: PT- Paper Towel, MRL- Mean Root Length, MSL- Mean Shoot Length, MRDW- Mean Root Dry Weight, MSDW- Mean Shoot Dry Weight, VI- I- Vigour Index I, VI- II- Vigour Index II.

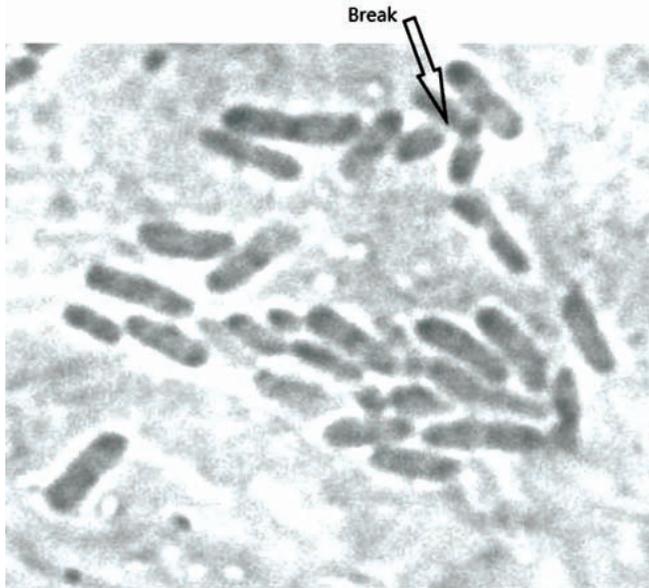
**Figure 2.** Per cent reduction in growth parameters of Electron Beam treated inbred line, PML 93



As a consequence of the decrease in mean root and shoot length compared to control, Vigor Index- I values also decreased to 3049.0, 2807.22, 2197.60, 1880.94, 1718.60, 736.96, 53.20, and 31.06 at the doses of 50 Gy, 100 Gy, 150 Gy, 200 Gy, 250 Gy, 300 Gy, and 400 Gy, respectively from the control value of 3493.77. The reduction of 50 per cent VI-I was observed at the dose 200 Gy. A similar result was observed on VI-II, where the increased dose from 50 Gy to 700 Gy resulted in the decreased VI-II from 5.37 to 0.0 compared to control (6.42). The reduction of 50 per cent VI-II was observed at dose 150 Gy.

Breeders are practicing the mutation breeding approach to induce genetic variability in crops to have an exploitable genetic variation for further crop improvement (Holme *et al.*, 2019). Generally, in plant breeding, physical and

chemical mutagenesis are the two major methods used to induce mutations in crop plants (Leitao, 2011; Mba *et al.*, 2011). But, the selection of particular mutagen depends on their efficiency and the purpose for which it is being used. Gamma irradiation is one of the frequently used physical mutagens to induce mutation in plant and animal systems and it was earlier reported that the effectiveness of EB is more than that of gamma rays (Mondal *et al.*, 2017; Gowthami, 2021). However, it is very important to standardize the dose of mutagen before its specific application to the targeted system, as the indiscriminate dose may be harmful. Hence, the present study was carried out to fix the appropriate dose of the EB mutagen to generate sufficient variations in a maize inbred line, PML 93, with minimum impact on the genome to sustain the negative effect of mutagen (Álvarez-Holguín *et al.*, 2019).



**Figure 3.** Chromosomal aberrations by the electron beam at 200 Gy in PML 93

In the ANOVA of different doses of EB, the mean sum of squares due to germination percentage, survival, seedling length (root and shoot), dry weight, wet weight, vigor index, I and II (Table 1) were highly significant indicating the induction of significant variation in growth parameter. It further implied that the tested EB doses can create genetic variation *vis-a-vis* its survivability in the target genotype (Kurtar *et al.*, 2017, Kumar *et al.*, 2018).

Further analysis of individual growth parameters for deciding GR 50 dose indicated that irradiation adversely affected both seed germination and the elongation of the root and shoot, similar to that noticed in other crops, during early seedling growth (Marcu *et al.*, 2013). Compared to the control treatment, germination, mean root length, and mean shoot length were reduced approximately to half at 250 Gy. The reduction of seedling growth after irradiation has been earlier explained based on auxin destruction, changes in ascorbic acid content and physiological injury, and biochemical disturbances in rice seedlings (Yusuf and Nair, 1974). The use of reduction in the germination percentage or root elongation and shoot elongation as an index to establish the relationship between the irradiation dose and early seed growth has its own limitations. Since, the process of seed germination and shoot elongation can be affected by various factors and their interactions, including temperature, water availability, oxygen, light, substrate, maturity of seed, and physiological age of seed (Smýkal *et al.*, 2014). Therefore, the vigor index is considered to be a more reliable parameter for estimating the 50 per cent growth reduction

dose in plants (Kumar *et al.*, 2018). Seedling vigor is a measure of the extent of damage that accumulates as viability declines, and the damage accumulates in seeds until the seeds are unable to germinate and eventually die (Copeland and McDonald, 2012; Zhao *et al.*, 2016). Vigor Index-I and II showed a reduction trend as the concentration of the EB was increased from 0 Gy to 400 Gy and further increase in mutagen dose from 500–700 Gy became lethal. Compared to the control treatment, GR 50 with respect to Vigor Index-I and II were recorded at doses 200 Gy and 150 Gy, respectively. This was depicted in the growth of the seedling also (Figure 1).

Therefore, taking into consideration of per cent germination, MRL and MSL, MRDL and MSDL, in PML 93, it can be concluded that GR 50 dose required for inducing useful mutations in this inbred line is 200 Gy. But, the 50 per cent reduction of V-I and V-II compared to the control appeared at 150 Gy (Figure 2). Hence, further karyotype analysis was conducted to decide the effective GR 50 dose. The root samples from seedlings treated with different doses as described in the material and methods section were studied for this purpose. The karyotype analysis revealed that chromosomal aberrations started at 200 Gy as minor breakage in the chromosomes and progressively heavier aberrations at doses higher than 200 Gy (Figure 3). Hence, dose 200 Gy was considered as GR 50 dose for maize genotype in general and the inbred line like PML 93.

## Conclusion

Standardization of dose for obtaining effective mutation is the basic requirement of a mutation breeding program. In the present investigation, among the 10 doses of EB employed in the study, 200 Gy was revealed as a GR 50 dose in the inbred line, PML 93. This dose can be utilized to obtain potentially useful mutations in the inbred line, PML 93.

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## Identification of stable genotypes across the ecologies using GGE analysis

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**Abstract:** Maize is an important cereal crop after wheat and rice. It is grown on a wide range of environments across the world. Genotype × environment interaction affects the relationship between phenotype and genotype in breeding programmes, particularly for quantitative traits. So, multi environment trials (MET) are used for identification of stable genotypes across the environments. To assess the differential performance of the 18 maize hybrids, an experiment was conducted under three different moisture regimes *viz.*, irrigated, rainfed and drought condition. Amongst tested hybrids ZH 161361, ZH 161311 and ZH 161083 were the most superior hybrids with grain yield 9.274, 7.845 and 7.774 t/ha across the environments. Four hybrids, *viz.*, ZH 161114, ZH 161276, ZH 161137 and ZH 161042 recorded > 70 per cent yield reduction under drought condition in comparison to optimum input level. The top performing genotypes, *viz.*, ZH 161038, ZH 161361 and ZH 161083 also performed poorly under rainfed and drought conditions. The yield reduction of these genotypes varied from 33.99 to 48.01 per cent under rainfed conditions, whereas the same was 46.2 to 70.7 per cent under drought conditions. Among the top performing hybrids ZH 161083 was the most stable

hybrid across the locations, whereas ZH 161361 and ZH 161038 were less stable hybrids. ZH 161361 was the best performing genotype both under optimum and drought condition with grain yield 17.239 and 9.274 t/ha, whereas ZH 161083 out performed under rainfed condition than rest of the hybrids.

**Keywords:** Drought · Genotype × environment interaction · GGE · Grain yield · Rainfed

### Introduction

Maize constitutes about 9 per cent of the total volume of cereals produced and is the third most important crop after rice and wheat. Maize cultivation is increasing day by day because of its high productivity, relative ease of cultivation, processing, storage and transportation. Superior maize hybrids need to be developed to meet the rising demand of maize. The development of new high yielding maize hybrid is a complex process consisting of evaluation of newly developed hybrids over the years and locations. It is highly impractical to meet all the demand of a crop during the growing period and provide all the inputs at optimum level. Most often, the crop experience limitation in different input levels and subjected to number of stresses during the period in farmer's field. The temperature rise due to the global climate change is expected to increase the risk of drought stress to the growing crops across the world. The global maize production decreased around 40 per cent due to the drought stress (Daryanto *et al.*, 2016). Haseeb *et al.* (2020) reported the minimum effects of drought were at control and 90 per cent irrigation water, whereas the adverse drought effects were observed under the

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treatment of drought at 20 per cent and 40 per cent irrigation water. Yield of maize crop is highly dependent to the amount of rainfall received, distribution, and duration, particularly in absence of assured irrigation. Further, most of the maize growing areas in India are rainfed without any assured irrigation. Identification of hybrids with high grain yield adapted to diverse environments and least affected under stress conditions is one of the most important objectives in maize breeding programs. The alternate strategy is to identify environment specific superior cultivars. Selection of genotypes purely based on yield may be misleading in presence of  $G \times E$  interaction (Kang *et al.*, 1991), as the ranking of genotypes changes due to the cross over interaction. Thus, multi-environment yield trials are essential for estimation of genotype by environment ( $G \times E$ ) interaction and identification of superior stable genotypes (Yan *et al.*, 2000). Though different methods, *viz.*, analysis of variance (ANOVA), regression analysis, principal component analysis (PCA), cluster analysis are available to detect the  $G \times E$  interaction, two models, *viz.*, AMMI (Additive Main Effects and Multiplicative Interaction) and GGE (Genotype and Genotype by Environment interaction) developed by Gauch and Zobel (1996) and Yan *et al.* (2000), respectively attracted much attention of plant breeders particularly for easy visualization and interpretation. AMMI analysis interprets the effect of the genotype (G) and environment (E) as additive effects and the  $G \times E$  interaction as a multiplicative component, whereas GGE biplot uses both genotype (G) and GE interaction effects to extract the principal components (PCs) which are plotted to form different biplots, *viz.*, mean versus stability, which won where, discriminative versus representative etc. (Yan *et al.*, 2001). GGE biplot is an effective method based on principal component analysis (PCA) to fully explore MET data. The objective of this study was to evaluate stability and adaptability of experimental maize hybrids using GGE biplot and identify the outperforming genotypes under specific environments.

## Materials and methods

### *Genetic material*

The experimental material consists of 18 hybrids include ZH 161038, ZH 161042, ZH 161050, ZH 161053, ZH 161063, ZH 161066, ZH 161077, ZH 161083, ZH 161093, ZH 161114, ZH 161129, ZH 161137, ZH 161276, ZH 161289, ZH 161311, ZH 161361, ZH 161409 and ZH 161434. These hybrids were supplied by CIMMYT under the CRMA project (Table 3).

### *Field trials*

The 18 maize hybrids were evaluated at three different locations, *viz.*, Ludhiana, Godhra and Varanasi. During *kharif* season of 2017, the material was evaluated under irrigated and rainfed conditions at Ludhiana and Godhra, respectively whereas, the same was evaluated under drought conditions at Varanasi during winter 2017. All India Coordinated Research Programme (AICRP) on maize has identified five agro-ecological zones in the country. The selected three locations belong to three different zones, *viz.*, Ludhiana represents North Western Plain Zone (NWPZ), Varanasi represents North Eastern Plain Zone (NEPZ) and Godhra represents Central West Zone (CWZ). Irrigation was withheld for one week before onset of flowering to create the water stress conditions at Varanasi. The latitude, longitude, altitude, soil type, rainfall, relative humidity and temperature of these test locations are presented in Table 1. The trials were arranged in a randomized complete block design with two replications. The row to row distance was 60 cm and 30 cm distance was maintained between the plants. The standard agronomic practices were followed according to the local agro-ecological conditions. The grain yield in tonnes/hectare (t/ha) was estimated at 15 per cent moisture for each of the entry in each location.

**Table 1.** Climatic and geographical information of the testing sites

| S.No. | Locations | Geographic Position |            | Altitude | Soil Type            | Rainfall | Humidity | Temperature |
|-------|-----------|---------------------|------------|----------|----------------------|----------|----------|-------------|
|       |           | Latitude            | Longitude  |          |                      |          |          |             |
| 1.    | Ludhiana  | 30.91°N             | 75.85°E    | 262 m    | Sandy loam to clayey | 876 mm   | 51%      | 19–36°C     |
| 2.    | Godhra    | 22.77°N             | 73.62°E    | 73m      | Loam sandy           | 530.3 mm | 35%      | 24–40°C     |
| 3.    | Varanasi  | 25°19'08"N          | 83°00'46"E | 80.71m   | Alluvial             | 1110 mm  | 57%      | 22–46°C     |

### Statistical analysis

The analysis of variance (ANOVA) was performed in SPSS 16.0 software. Analysis of variance was calculated using the model:

$$Y_{ij} = m + G_i + E_j + GE_{ij}$$

Where,  $Y_{ij}$  is the corresponding variable of the  $i$ -th genotype in  $j$ -th environment (location),  $\mu$  is the total mean,  $G_i$  is the main effect of  $i$ -th genotype,  $E_j$  is the main effect of  $j$ -th environment,  $GE_{ij}$  is the effect of  $G \times E$  interaction. Different biplots, *viz.*, discriminative versus representative plot, mean versus stability plot and which won where biplot were derived using the GEA-R software (Pacheco *et al.*, 2015).

### Results and discussion

#### Variation for grain yield

The combined ANOVA revealed significant variation for genotype, environment and their interaction ( $G \times E$ ) effect for grain yield (Table 2). The most promising hybrids were ZH 161038, ZH 161361, ZH 161083, ZH 161276, ZH 161289, ZH 161042, ZH 161311, ZH 161050, ZH 161409, ZH 161114, ZH 161053, ZH 161434 and ZH 161093 with grain yield >10.0 t/ha under optimum condition. Out of these 13 entries, two hybrids, *viz.*, ZH 161038 and ZH 161053 revealed superior performance under rainfed condition also, with grain yield 11.429 and 10.048 t/ha, respectively. Though most of the hybrids recorded

**Table 2.** Analysis of variance for yield at three environments

| Source of variation | DF    | SS       | MS       | F      | Probability |
|---------------------|-------|----------|----------|--------|-------------|
| Genotype (G)        | 17.00 | 44334.51 | 2607.91  | 34.11  | 0.00        |
| Replication (R)     | 1.00  | 210.90   | 210.90   | 2.76   | 0.10        |
| Environment (E)     | 2.00  | 82659.57 | 41329.78 | 540.59 | 0.00        |
| $G \times E$        | 34.00 | 32068.74 | 943.20   | 12.34  | 0.00        |
| Error               | 53.00 | 4052.03  | 76.45    |        |             |
| CD                  | 20.36 |          |          |        |             |
| CV (%)              | 10.73 |          |          |        |             |

**Table 3.** Mean grain yield of maize hybrids at different locations

| S.No. | Genotype  | Grain yield (t/ha) |              |              | Mean yield (t/ha) |
|-------|-----------|--------------------|--------------|--------------|-------------------|
|       |           | Optimum (E1)       | Rainfed (E2) | Drought (E3) |                   |
| 1.    | ZH 161038 | 17.314             | 11.429       | 5.071        | 11.271            |
| 2.    | ZH 161042 | 13.469             | 8.762        | 3.917        | 8.716             |
| 3.    | ZH 161050 | 12.843             | 3.929        | 5.976        | 7.583             |
| 4.    | ZH 161053 | 11.004             | 10.048       | 7.631        | 9.561             |
| 5.    | ZH 161063 | 9.79               | 7.738        | 7.417        | 8.315             |
| 6.    | ZH 161066 | 5.049              | 6.19         | 2.726        | 4.655             |
| 7.    | ZH 161077 | 6.952              | 5.714        | 5.107        | 5.924             |
| 8.    | ZH 161083 | 16.945             | 8.81         | 7.774        | 11.176            |
| 9.    | ZH 161093 | 10.415             | 7.857        | 5.786        | 8.019             |
| 10.   | ZH 161114 | 11.513             | 7.262        | 2.00         | 6.925             |
| 11.   | ZH 161129 | 9.788              | 2.619        | 7.298        | 6.568             |
| 12.   | ZH 161137 | 7.95               | 4.762        | 1.893        | 4.868             |
| 13.   | ZH 161276 | 15.638             | 7.976        | 3.50         | 9.038             |
| 14.   | ZH 161289 | 15.562             | 8.624        | 6.679        | 10.288            |
| 15.   | ZH 161311 | 12.964             | 5.409        | 7.845        | 8.739             |
| 16.   | ZH 161361 | 17.239             | 9.286        | 9.274        | 11.933            |
| 17.   | ZH 161409 | 11.989             | 5.976        | 4.036        | 7.334             |
| 18.   | ZH 161434 | 10.589             | 8.69         | 5.143        | 8.141             |

reduced grain yield under drought condition, the top three entries were ZH 161361, ZH 161311 and ZH 161083 with grain yield 9.274, 7.845 and 7.774 t/ha (Table 3).

*Performance of genotypes under different ecologies for flowering and anthesis silking interval (ASI)*

Observations on 50 per cent pollen shedding, silking and anthesis silking interval were recorded for all genotypes during the time of flowering. Significant differences in anthesis silking interval were observed under optimum, rainfed and drought conditions. ASI is highly correlated with grain yield as the adverse conditions i.e. drought causes a delay in silk emergence thus reduction in yield occurs. With the drought stress, ASI was severely affected due to non-availability of water during flowering period. There was not any visible differences in ASI reported under optimum and drought conditions (Table 4). However, under drought conditions, larger and prolonged anthesis silking interval (>8 days) was observed for the genotypes ZH 161137, ZH 161042, ZH 161066 and ZH 161409. Genotypes ZH 161050, ZH 161077 and ZH 161311 have shorter ASI.

*Yield reduction under rainfed and drought condition*

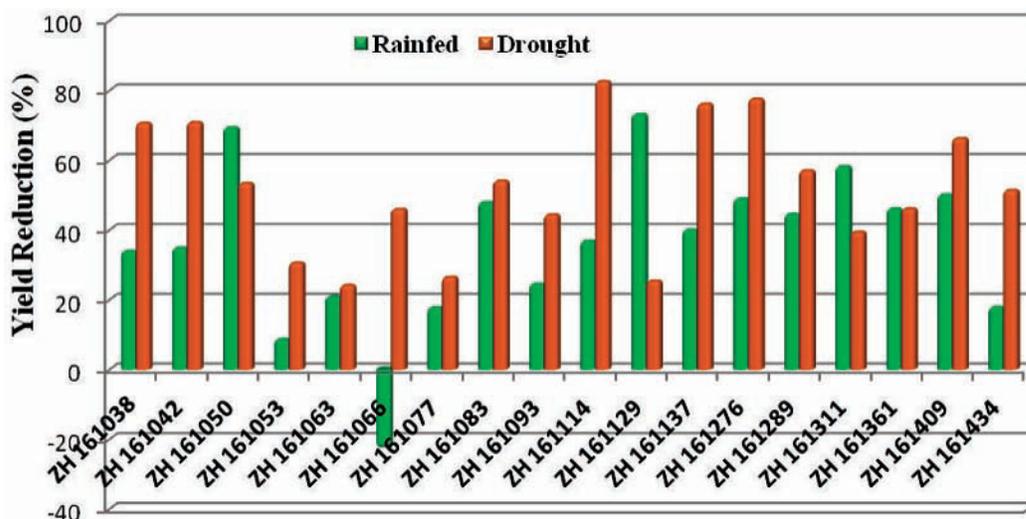
Performance of cultivars are expected to reduce under rainfed and drought condition in comparison to optimum input level, however variable response is expected for different genotypes. The most affected genotype under rainfed condition was ZH 161129 which recorded 73.24 per cent yield reduction followed by ZH 161050 (69.41 per cent) and ZH 161311 (58.28 per cent) (Figure 1). One hybrid i.e. 161066 recorded higher yield under rainfed condition (6.19 t/ha) than optimum condition (5.049 t/ha). Four hybrids, viz., ZH 161114, ZH 161276, ZH 161137 and ZH 161042 recorded >70 per cent yield reduction under drought condition in comparison when the crop was grown under optimum input level (Figure 1). The least affected hybrids under rainfed condition were ZH 161053, ZH 161077 and ZH 161434, whereas the hybrids, viz., ZH 161063, ZH 161129 and ZH 161077 were least affected under drought. The yield of ZH 161077 was most consistent across the locations, however it recorded average yield in all the three locations (optimum: 6.952 t/ha, rainfed: 5.714 t/ha and drought: 5.107 t/ha). The top performing genotypes, viz., ZH 161038, ZH 161361 and

**Table 4.** Days to anthesis and silking of the genotypes under different ecologies

| S.No. | Genotype  | Optimum (E1) |    |     | Rainfed (E2) |    |     | Drought (E3) |     |     |
|-------|-----------|--------------|----|-----|--------------|----|-----|--------------|-----|-----|
|       |           | DA           | DS | ASI | DA           | DS | ASI | DA           | DS  | ASI |
| 1.    | ZH 161038 | 55           | 57 | 2   | 55           | 57 | 3   | 102          | 107 | 5   |
| 2.    | ZH 161042 | 54           | 59 | 5   | 58           | 60 | 2   | 99           | 104 | 8   |
| 3.    | ZH 161050 | 59           | 62 | 3   | 56           | 59 | 3   | 107          | 111 | 4   |
| 4.    | ZH 161053 | 54           | 56 | 2   | 54           | 55 | 1   | 101          | 107 | 6   |
| 5.    | ZH 161063 | 53           | 56 | 3   | 55           | 58 | 3   | 97           | 102 | 5   |
| 6.    | ZH 161066 | 59           | 61 | 2   | 57           | 59 | 2   | 100          | 108 | 8   |
| 7.    | ZH 161077 | 59           | 61 | 2   | 57           | 59 | 2   | 104          | 108 | 4   |
| 8.    | ZH 161083 | 54           | 57 | 3   | 56           | 58 | 2   | 100          | 107 | 7   |
| 9.    | ZH 161093 | 53           | 55 | 2   | 55           | 57 | 2   | 102          | 108 | 6   |
| 10.   | ZH 161114 | 59           | 61 | 2   | 58           | 61 | 3   | 104          | 111 | 7   |
| 11.   | ZH 161129 | 53           | 55 | 2   | 56           | 59 | 3   | 102          | 107 | 5   |
| 12.   | ZH 161137 | 58           | 60 | 2   | 55           | 58 | 3   | 99           | 108 | 9   |
| 13.   | ZH 161276 | 56           | 58 | 2   | 58           | 60 | 2   | 105          | 110 | 5   |
| 14.   | ZH 161289 | 57           | 61 | 4   | 56           | 58 | 2   | 103          | 110 | 7   |
| 15.   | ZH 161311 | 54           | 57 | 3   | 56           | 58 | 2   | 104          | 108 | 4   |
| 16.   | ZH 161361 | 54           | 58 | 4   | 58           | 60 | 2   | 103          | 109 | 6   |
| 17.   | ZH 161409 | 54           | 58 | 4   | 58           | 61 | 3   | 103          | 111 | 8   |
| 18.   | ZH 161434 | 54           | 56 | 2   | 54           | 56 | 2   | 101          | 108 | 7   |

DA: Days to anthesis; DS: Days to silking; ASI: Anthesis-silking interval

**Figure 1.** Reduction in yield of maize hybrids under rainfed and drought condition compared to optimal condition



ZH 161083 also performed poorly under rainfed and drought condition, and reduction in yield in these genotypes varied from 33.99 to 48.01 per cent under rainfed and 46.20 to 70.71 per cent under drought. High yielding genotypes under well-watered conditions showed less amount of yield reduction under drought compared to less yielding genotypes in earlier findings also (Duvick, 1984; Castleberry *et al.*, 1984; Frederick *et al.*, 1989). However, in the present study, the genotypes could not reveal such specific trend and the high yielding hybrids also recorded yield loss even up to 70.71 per cent. Similar results were reported by Kamara *et al.* (2003), where the high yielding genotypes under optimum condition recorded about 70 per cent yield reduction in water-deficit conditions.

#### Identification of stable hybrids across the locations

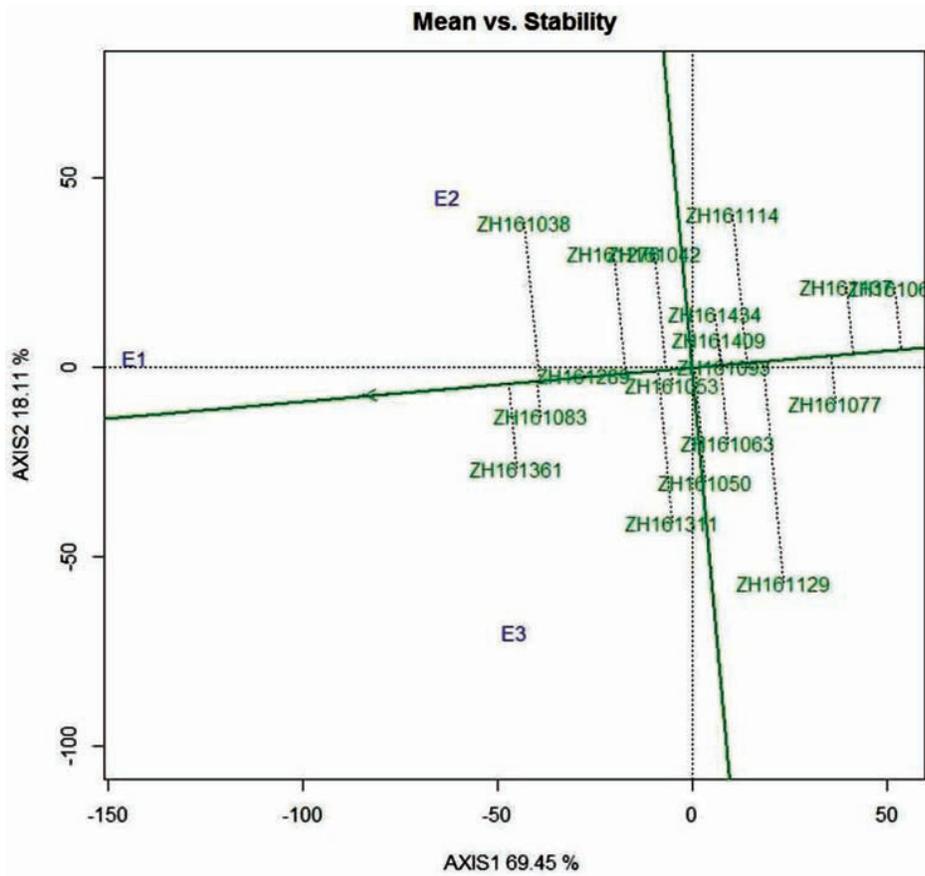
$G \times E$  interactions possess paramount importance to breeders to develop location specific and generally adapted cultivars (Kang *et al.*, 1994). The mean versus stability biplot can measure the influence of environments on a specific trait and identify the most promising genotypes across or specific location. The PC1 and PC2 explained 69.45 per cent and 18.11 per cent variation, respectively of total  $G + GE$  variation, together PC1 and PC2 could explain 87.56 per cent of variation (Figure 2). The direction of the arrow shown on the axis of the Average Environment Coordination (AEC) abscissa indicates the higher mean performance of the genotypes. Hence, the high yielding genotypes can be identified from the biplot by seeing the placement of genotypes with respect to the

direction of arrow. The AEC ordinate marks the contributions to  $G \times E$  interaction and represents the stability of genotypes across the locations. The genotypes near to origin are more stable compared to the outermost genotypes. An ideal genotype should possess the highest mean performance and absolute stability. Among the top performing hybrids, ZH 161083 was the most stable hybrid across the locations, whereas ZH 161361 and ZH 161038 were less stable hybrids. Further, ZH 161289 and ZH 161053 also revealed more stability with moderate mean (10.288 and 9.561 t/ha, respectively). Shiri and Bahrapour (2015) also used GGE biplot and reported the better performing maize hybrids across the environments. It is important to develop higher yielding hybrids for drought stress and rainfed condition which can equally perform under optimum moisture conditions. Maize hybrids of such calibre will reduce the risk of farming communities living in drought-prone areas and also contribute to increase the national maize productivity.

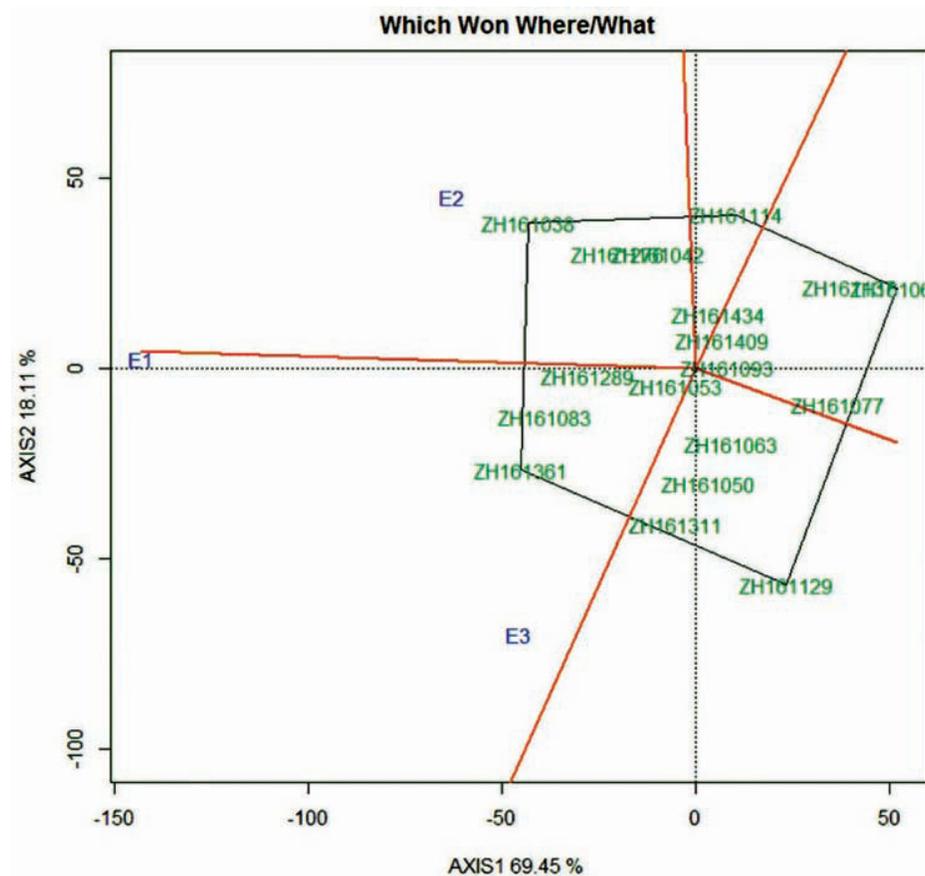
#### Which won where biplot

The “which-won-where” view of the GGE biplot helps to easily identify the best performing genotype(s) in each of the environments (Yan *et al.*, 2002). The perpendicular lines initiating from the origin, divide the polygon into different sectors (Figure 3). The vertex genotype is the best performing genotype in the environments falling in the same sector. ZH 161066, ZH 161129, ZH 161361, ZH 161038 and ZH 161114 were the vertex genotypes of the polygon (Figure 3). Mafouasson *et al.* (2018) also identified stable genotypes across the ecologies using GGE

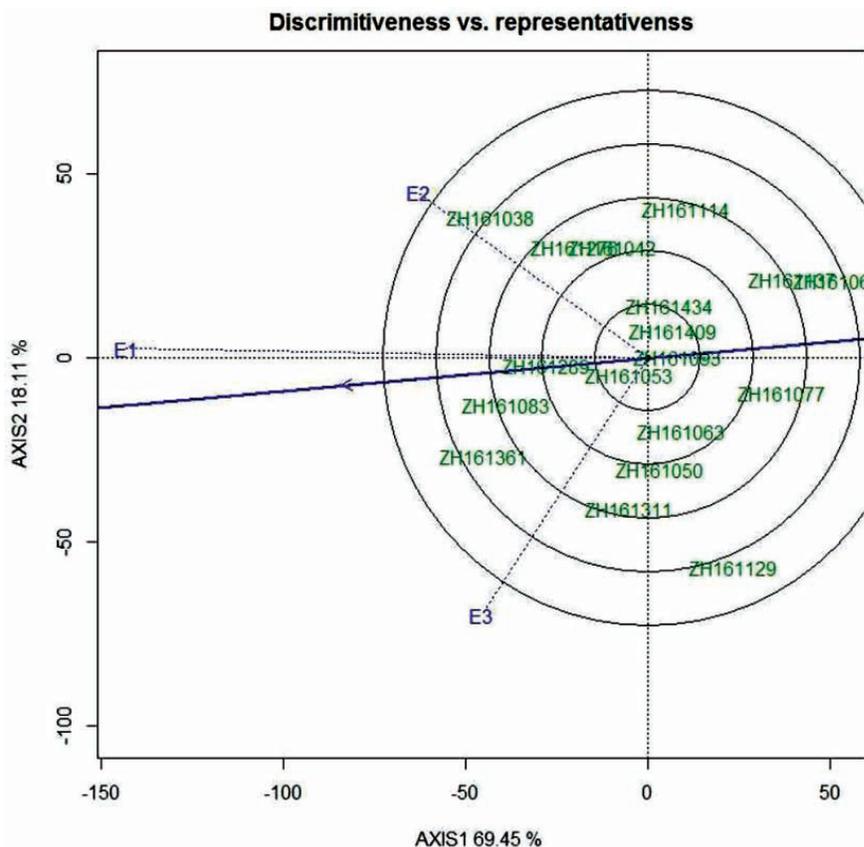
**Figure 2.** Average environment coordination views of the GGE-biplot for the mean performance and stability of genotypes



**Figure 3.** Polygon views of the GGE biplot for the genotype by environment 2-way data



**Figure 4.** Discrimitiveness vs. representativens of test environments



biplots. ZH 161361 was the best performing genotype both under optimum and drought condition with grain yield 17.239 and 9.274 t/ha, whereas ZH 161083 out performed under rainfed condition than rest of the hybrids.

#### *Discriminating power versus representativeness of test environments*

The power of an environment to differentiate among genotypes is known as discriminative power, whereas representativeness is how efficiently it can represent the mega environment. Superiority of an environment for discriminative and representative power is indicated by longer vector length of the environment and smaller angle with the average-environment axis. E1 (optimum) revealed the longest vector length as well as the smallest angle with average-environment axis. Hence, E1 is the ideal test environment among the three, as full expression of genotypes is expected under optimum condition only which is oppressed under any stress condition (Figure 4).

#### **Conclusion**

The present study could identify stable hybrids across the

different environments, viz., irrigated, rainfed and drought and at the same time genotype suitable for particular environment. However, the identified hybrids need to be tested in more number of replicated environments to validate the findings.

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## Growth and physiological indices changes in maize with differential residue and nitrogen management under conservation agriculture

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**Abstract:** A field experiment was conducted during *kharif* season 2015 for exploring influence of differential residue and nitrogen management practices on growth parameters and physiological indices of maize under conservation agriculture on a sandy loam soil at New Delhi. The treatments consisted of two cropping systems: maize-mustard-mungbean (MMuMb) and maize-wheat-mungbean (MWMB); two residue management practices of with (WR) and without residue (WoR) and four N management practices of control, prilled urea (PU), sulphur coated urea (SCU) and neem coated urea (NCU) arranged in split-split plot design and replicated thrice. The results of the study indicated that growth parameters, *viz.*, plant height, dry matter accumulation (DMA), leaf area and leaf area index (LAI) and physiological indices *viz.*, crop growth rate (CGR), relative growth rate (RGR) and net assimilation rate (NAR) in maize were increased significantly with residue retention and coated urea application. The yield attributes of maize were also enhanced by the application of WR, MWMB and NCU. After water stress significant decrease in canopy temperature was recorded with WR (-3.39°C) compared to WOR (-1.84°C). The higher dry matter accumulation (218 g/plant) was recorded under MWMB compared to

MMuMb (184 g/plant) cropping system. The highest and least leaf area and LAI in maize plants were recorded under NCU application and absolute control, respectively. SCU and PU remained at par with each other with respect to both leaf area and LAI. The highest SPAD values were observed with the NCU at 30 and 90 DAS, which were at par with SCU at 30 DAS and PU and SCU at 90 DAS. Thus, the basal application of NCU and residue retention in MWMB system was could be opted for enchaining growth parameters and physiological indices in maize under conservation agriculture.

**Keywords:** Coated urea · Conservation agriculture · Growth parameters · Maize · Residue

### Introduction

Maize, an important crop for food, feed and nutritional security in India grown in diverse agro-environments and seasons on an area of about 9.57 m ha with productivity of 3.01 t/ha and total production of 28.77 million tons (DACNET, 2021). India ranks 4<sup>th</sup> in maize area in the world but has the productivity less than half of the world's average. The bulk of the maize production in India is used as poultry feed (47 per cent). Of the rest, 14 per cent is used in the starch industry, 13 per cent is used as livestock feed, 13 per cent for food purposes, 7 per cent as processed food and 6 per cent for export and other purposes (IIMR, 2021). There is a tremendous need to increase the acreage and productivity of this crop in near future to meet rising feed, food and industrial demands, especially in view of the very fast growth in livestock and poultry sectors in India (Kar *et al.*, 2004 and 2005).

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Growth and yield of any crop under a particular environment are largely governed by soil moisture, nutrients, radiation interception and the efficiency of conversion of intercepted radiation and partitioning of dry matter to grain (Kar *et al.*, 2005; Figuerola and Berliner, 2006; Kar *et al.*, 2013).

In pre-green revolution era, coarse cereals including maize were the principal crops of rainy season in northern India. But with the introduction of high yielding varieties and expansion of irrigation facilities, rice has become prominent rainy season while wheat as winter crop after green revolution. Hardly less than one tonne organic manure per ha is being added to the soil, which leads to fast decline in organic matter content of soils. The ground water depletion due to over exploitation for planting of rice, as it is done in north-western Indo-Gangetic plains (IGP) two months before onset of monsoon and that has dangerously lowered the ground water level. Rice-wheat cropping system not only threatening sustainability of agro-ecosystem of IGP but also becoming less profitable due to increasing input cost; particularly conventional tillage practices and overuse of nitrogenous fertilizers through conventional method. Recently, conservation agriculture (CA) practices for crop production comprising of minimum soil mechanical manipulation, permanent soil cover and profitable crop rotation found to be useful in reduction in cost of crop production in addition to giving ecological services for lower carbon consumption/emission and improvement in soil health. The area under CA is increasing due to shortage of labour and escalating input prices in South Asian region and practiced on 180 m ha area worldwide (Kassam *et al.*, 2019). However, in India CA is practiced only on 1.5 m ha during 2015 (Kassam *et al.*, 2019). Adoption of CA in South Asia has skewed distribution, mainly concentrated in Indo-Gangetic Plains (IGP) in India, Pakistan, Nepal and Bangladesh (Somasundaram *et al.*, 2020). Broadcasting of nitrogenous fertilizer over retained crop residue in CA may be an inefficient method of N application, due to immobilization of inorganic N, in association with the microbial breakdown of wider C: N cereal residue (Rice and Smith, 1984; Thuy *et al.*, 2008; Xu *et al.*, 2010) and greater ammonia volatilization (Janssen, 1996; Patra *et al.*, 2004), compared to application on bare soil. Despite more favourable results of CA in research, farmers are not adopting it at their field because of several reasons and one of them is improper nutrient especially N management

practices. The five-split application of N in maize with recommended dose of nitrogen in ratio of 10:30:30:20:10 as basal, 4-leaf emergence, 8-leaf emergence, tassel emergence and early grain filling stages, respectively, enhances the grain yield as compared to three split application (Singh, 2010). But, at the same time split application entails more labour cost as compared to one time application and there is already a labour shortage in Indian agriculture. Moreover, the residue retention on the soil surface under CA becomes hurdle of split-urea application and lowers the NUE as part of it either volatilized or immobilized due to fraction of applied fertilizer rest on the residue and consumed by the microbes. For enhancing profitability in maize system through CA there is need to enhance fertilize N-use efficiency through use of slow release fertilizer which will also act as problem solving for labour shortage in agriculture. Hence, proper management practices requires for enhancing NUE and reducing environmental foot print in CA system. So, the review suggests that there is need of proper N management practices for accelerating adoption of CA. Keeping above-mentioned points in view, one time application of sulphur- and neem-coated urea has been compared with split application of prilled urea under CA in maize in our study.

## Materials and methods

The experiment was conducted during *kharif* season 2015 in '9B' block of the experimental farm of the ICAR-Indian Agricultural Research Institute, New Delhi. The field had an even topography and good drainage system. The study was a part of the long-term trial started in *Kharif* 2012. Before this, field was under maize-wheat rotation since 2007. The treatments consisted of two cropping systems: maize-mustard-mungbean (MMuMb) and maize-wheat-mungbean (MWMB); two residue management practices of with (WR) and without residue (WoR) and four N management practices consisting absolute control, prilled urea (PU), sulphur-coated urea (SCU) and neem-coated urea (NCU) arranged in split-split plot design and replicated thrice. The soil of the experimental site was sandy loam in texture with neutral pH having low N, medium P and high K availability. The growth parameters including plant height (at harvest), leaf area index and dry matter accumulation (at 90 DAS) were recorded in maize

with standard procedure. The physiological indices *viz.*, crop growth rate (g/plant/day), relative growth rate (mg/g/day) and net assimilation rate (g/cm<sup>2</sup> leaf area/day) were also computed using standard formulae.

The data recorded for different parameters were analyzed with the help of analysis of variance (ANOVA) technique (Gomez and Gomez, 1984) for split-split-plot design using SAS 9.1 software (SAS Institute, Cary, NC). The least significant difference test was used to decipher the effect of treatments at 5 per cent level of significance ( $P=0.05$ ). Critical difference (CD) values for different pair-wise comparison among the treatment effect were computed.

## Results and discussion

### Growth parameters

There was no significant effect of cropping system on plant height of maize at harvest. However, residue application significantly increased plant height at harvest

in zero-till maize compared to WoR (Table 1). The significantly taller maize plant was observed with neem-coated urea (NCU) application, which was on par with sulphur-coated urea (SCU) and prilled urea (PU). Significantly higher dry matter accumulation (218 g/plant) was recorded under MWMB compared to MMuMb (184 g/plant) cropping system. However, there was no significant effect of residue retention on DMA at 90 DAS. Among N management practices, the highest DMA was recorded with the application of NCU. Significant interaction effect of nitrogen with residue and system was also found for DMA at 90 DAS (Table 1).

The leaf area and leaf area index (LAI) of the maize were significantly enhanced by MWMB over MMuMb cropping system. Similarly, residue retention affected the leaf area and LAI significantly compared to residue removal. The highest leaf area and LAI were recorded under NCU application and SCU and PU remained at par with each other (Table 1).

The lower mineral N availability reported in our study in 0–30 cm soil depth could be the important reasons for

**Table 1.** Effect of cropping system, residue and nitrogen management on growth parameters in zero-till *Kharif* maize.

| Treatment                      | Plant height (cm) | DMA (g/plant) | Leaf area (cm <sup>2</sup> /plant) | LAI    |
|--------------------------------|-------------------|---------------|------------------------------------|--------|
| <i>Cropping system</i>         |                   |               |                                    |        |
| Maize-mustard-mungbean (MMuMb) | 206.4             | 184b          | 4094b                              | 3.06b  |
| Maize-wheat-mungbean (MWMB)    | 210.5             | 218a          | 4911a                              | 3.66a  |
| LSD ( $p=0.05$ )               | NS                | 21.1          | 79.5                               | 0.0637 |
| <i>Residue management</i>      |                   |               |                                    |        |
| Permanent beds - residue (WoR) | 205.4b            | 202           | 4125b                              | 3.08b  |
| Permanent beds + residue (WR)  | 211.5a            | 201           | 4880a                              | 3.64a  |
| LSD ( $p=0.05$ )               | 4.30              | NS            | 72.8                               | 0.053  |
| <i>Nitrogen management</i>     |                   |               |                                    |        |
| Absolute control               | 169.9b            | 122c          | 2874c                              | 2.14c  |
| N by prilled urea (PU)         | 222.0a            | 222b          | 4749b                              | 3.54b  |
| N by sulphur-coated urea (SCU) | 219.9a            | 219b          | 4944b                              | 3.69b  |
| N by neem-coated urea (NCU)    | 221.9a            | 240a          | 5443a                              | 4.06a  |
| LSD ( $p=0.05$ )               | 7.85              | 8.4           | 273.6                              | 0.2037 |
| <i>p values</i>                |                   |               |                                    |        |
| <i>System</i>                  | 0.1338            | <.0001        | <.0001                             | <.0001 |
| <i>Residue</i>                 | 0.0337            | 0.7965        | <.0001                             | <.0001 |
| <i>System × Residue</i>        | 0.2228            | 0.2048        | <.0671                             | <.0001 |
| <i>System × Nitrogen</i>       | 0.7800            | <.0001        | 0.0038                             | 0.0037 |
| <i>Residue × Nitrogen</i>      | 0.2798            | <.0001        | <.0001                             | <.0001 |
| <i>Nitrogen</i>                | <.0001            | <.0001        | <.0001                             | <.0001 |

Note: Means followed by different letters in each column are statistically different at  $LSD_{0.05}$ .

lower growth parameters under MMuMb system. Moreover, the enhancement in organic carbon and soil microbial activities recorded in this system might also have contributed for higher growth of maize in MWMB system. Some of the earlier studies also found cropping system effects on the crop growth parameters in maize (Thierfelder *et al.*, 2015; Parihar *et al.*, 2016). The residue retention helps in lowering down the drought stress effect in maize as evident from the very high (-3.39°C) canopy temperature depletion recorded in our study which might be the important reason behind the enhancement of growth parameters of maize in WR compared to WoR. In addition to this, the residue retention over a period of time resulted in positive changes in soil properties and hence improvement in crop growth was occurred. The residue retention led to enhancement in crop growth might be attributed to enhanced soil moisture (Erenstein and Laxmi, 2008; Choudhary *et al.*, 2019), lower weed population and improvement in soil health. The similar effects of residue retention's enhancement in crop growth were also reported

by many workers in varied ecologies (Tolk *et al.*, 1999; Campbell *et al.*, 2000). In contrast to this, in temperate regions negative effect of residue retention on crop growth was noticed probably due to high initial soil organic carbon and slow residue degradation and immobilization of applied nutrients (Rice and Smith, 1984; Thuy *et al.*, 2008). However, such effects were less pronounced instead reversed in tropical and sub-tropical agro-ecologies. The application of N enhanced the growth parameters in maize significantly over control and these were significantly higher in neem or sulphur-coated urea over prilled urea application. The slow and continuous supply of N with coated fertilizer (Cong *et al.*, 2010; Shivay and Prasad, 2014) to the crop lead to better photosynthesis which increased the root and shoot growth in the crop (Zhao *et al.*, 2013). The similar effects of coated fertilizer on the crop growth of maize also reported by many workers (Sharma and Prasad, 1996; Tanwar, 2014).

**Table 2.** Effect of cropping system, residue and nitrogen management on physiological indices in zero-till *kharif* maize

| Treatment                      | Crop growth rate<br>(g/plant/day) |              |              | Relative growth rate<br>(mg/g/day) |              |              | Net assimilation rate<br>(g/cm <sup>2</sup> leaf area/day) |              |
|--------------------------------|-----------------------------------|--------------|--------------|------------------------------------|--------------|--------------|--|--------------|
|                                | 0-30<br>DAS                       | 30-60<br>DAS | 60-90<br>DAS | 0-30<br>DAS                        | 30-60<br>DAS | 60-90<br>DAS | 30-60<br>DAS   | 60-90<br>DAS |
| <i>Cropping system</i>         |                                   |              |              |                                    |              |              |  |              |
| Maize-mustard-mungbean (MMuMb) | 0.85b                             | 1.31         | 3.97b        | 46.3b                              | 13.7         | 15.1         | 1.401  | 0.088        |
| Maize-wheat-mungbean (MWMB)    | 0.92a                             | 1.53         | 4.82a        | 47.6a                              | 14           | 15.7         | 1.801  | 0.033        |
| LSD ( <i>p</i> =0.05)          | 0.0346                            | NS           | 0.605        | 0.51                               | NS           | NS           | NS   | NS           |
| <i>Residue management</i>      |                                   |              |              |                                    |              |              |  |              |
| Permanent beds - residue (WoR) | 0.84b                             | 1.4          | 4.45         | 46.2b                              | 14.2         | 15.7         | 1.310b   | 0.032b       |
| Permanent beds + residue (WR)  | 0.93a                             | 1.44         | 4.35         | 47.7a                              | 13.5         | 15           | 1.893a   | 0.089a       |
| LSD ( <i>p</i> =0.05)          | 0.0351                            | NS           | NS           | 0.62                               | NS           | NS           | 0.484  | 0.043        |
| <i>Nitrogen management</i>     |                                   |              |              |                                    |              |              |  |              |
| Absolute control               | 0.57d                             | 0.93c        | 2.58c        | 41.0c                              | 14.1         | 14.5b        | 1.802a   | 0.073a       |
| N by prilled urea (PU)         | 0.88c                             | 1.46b        | 5.09a        | 47.3b                              | 14.1         | 16.7a        | 2.006a   | 0.104a       |
| N by sulphur coated urea (SCU) | 1.01b                             | 1.61ba       | 4.67b        | 49.4a                              | 13.8         | 14.8b        | 1.267b   | 0.031b       |
| N by neem coated urea (NCU)    | 1.08a                             | 1.68a        | 5.25a        | 50.2a                              | 13.5         | 15.5ba       | 1.329b   | 0.034b       |
| LSD ( <i>p</i> =0.05)          | 0.0609                            | 0.21         | 0.324        | 1.2                                | NS           | 1.4          | 0.404  | 0.0384       |
| <i>p values</i>                |                                   |              |              |                                    |              |              |  |              |
| <i>System</i>                  | 0.003                             | 0.0053       | <.0001       | 0.0011                             | 0.5494       | 0.2546       | 0.008  | 0.0004       |
| <i>Residue</i>                 | 0.0004                            | 0.6109       | 0.3846       | 0.0002                             | 0.2497       | 0.1726       | 0.0003   | 0.0002       |
| <i>System × Residue</i>        | 0.2826                            | 0.5488       | 0.2039       | 0.0922                             | 0.2443       | 0.518        | 0.0111   | <.0001       |
| <i>System × Nitrogen</i>       | 0.3681                            | 0.0309       | 0.003        | 0.1421                             | 0.0884       | 0.3558       | 0.2298   | 0.0005       |
| <i>Residue × Nitrogen</i>      | 0.0069                            | 0.2576       | <.0001       | 0.0125                             | 0.0972       | 0.0033       | 0.0254   | 0.0254       |
| <i>Nitrogen</i>                | <.0001                            | <.0001       | <.0001       | <.0001                             | 0.9048       | 0.0182       | 0.0018   | 0.0014       |

Note: Means followed by different letters in each column are statistically different at LSD<sub>0.05</sub>.

### Physiological indices

The data showed that MWMB had significant effect on CGR during 0–30 and 60–90 DAS compared to MMuMb cropping system (Table 2). Residue retention also had significant effect on CGR only in first 30 days (Table 2). Among nitrogen management practices, the highest CGR was recorded with the application of NCU which was on par with the PU and SCU during 60–90 DAS and 30–60 DAS, respectively. The data showed that RGR recorded highest at younger stage (0–30 DAS) then decline gradually in all the treatments. Significantly higher RGR was recorded with MWMB cropping system and residue retention at 0–30 DAS only while NCU application at two growth stages where it was on par with SCU at 0–30 DAS and PU at 60–90 DAS (Table 2). The data on NAR of maize at various growth stages showed that there was no significant effect of cropping systems on NAR while residue retention significantly enhanced the NAR at both the stages but the increase was more at 60 DAS. Among

the nitrogen treatments, significantly highest NAR was recorded with the application of PU, which was at par with control treatment (Table 2).

The MWMB system recorded higher CGR and RGR at initial crop growth stages (0–30 DAS) which could be attributed to enhanced leaf area and dry matter accumulation due to better soil nutrient supply observed in this system compared to MMuMb. Similar effects on CGR and RGR along with NAR of maize was recorded by WR compared to WoR, which thereby boost crop health under residue retention over period of time and better crop nutrition compared to WOR.

The application of N by NCU or SCU lead to enhancement in CGR, and RGR at most of the crop growth stages but the NAR of maize at various crop growth stages was higher in control. The higher NAR in the control could be attributed to lower dry weight and leaf area of the plant at initial growth stages observed in our study, which caused more net assimilation of photosynthates with per unit of leaf area in maize.

**Table 3.** Effect of cropping system, residue and nitrogen management on physiological stages in zero-till *kharif* maize

| Treatment                          | Days to tasseling | Days to silking | Days to maturity | Reproductive period (days) |
|------------------------------------|-------------------|-----------------|------------------|----------------------------|
| <i>Cropping system</i>             |                   |                 |                  |                            |
| Maize-mustard-mungbean (MMuMb)     | 55.3              | 62.8            | 101.1            | 45.9                       |
| Maize-wheat-mungbean (MWMB)        | 54.0              | 62.1            | 102.7            | 48.7                       |
| LSD ( $p=0.05$ )                   | NS                | NS              | NS               | NS                         |
| <i>Residue management</i>          |                   |                 |                  |                            |
| Permanent beds - residue (WoR)     | 54.3              | 62.2            | 101.7            | 47.4                       |
| Permanent beds + residue (WR)      | 54.9              | 62.8            | 102.1            | 47.2                       |
| LSD ( $p=0.05$ )                   | NS                | NS              | NS               | NS                         |
| <i>Nitrogen management</i>         |                   |                 |                  |                            |
| Absolute control                   | 61.5a             | 70.0a           | 102.3            | 40.8b                      |
| N by prilled urea (PU)             | 51.9b             | 59.6b           | 101.3            | 49.4a                      |
| N by sulphur coated urea (SCU)     | 52.4b             | 60.7b           | 101.8            | 49.3a                      |
| N by neem coated urea (NCU)        | 52.6b             | 59.6b           | 102.2            | 49.6a                      |
| LSD ( $p=0.05$ )                   | 2.01              | 2.82            | NS               | 2.40                       |
| <i>p values</i>                    |                   |                 |                  |                            |
| <i>System</i>                      | 0.0728            | 0.4448          | 0.0632           | 0.0021                     |
| <i>Residue</i>                     | 0.4391            | 0.5513          | 0.3451           | 0.8413                     |
| <i>System × Residue</i>            | 0.5910            | 0.7978          | 0.5976           | 0.4856                     |
| <i>System × Nitrogen</i>           | 0.8204            | 0.6408          | 0.2379           | 0.5288                     |
| <i>Residue × Nitrogen</i>          | 0.8551            | 0.3701          | 0.3642           | 0.6779                     |
| <i>Nitrogen</i>                    | <.0001            | <.0001          | 0.2896           | <.0001                     |
| <i>System × Residue × Nitrogen</i> | 0.5299            | 0.7752          | 0.4016           | 0.5774                     |

Note: Means followed by different letters in each column are statistically different at  $LSD_{0.05}$

Moreover, the application of N fertilization resulted in timely tasseling and silking in maize compared to control where it got delayed by 9 to 10 days. However, the maturity was arrived on same time, which resulted in decreasing of reproductive period of crop by almost 9 days. This could be attributed to better growth parameters of leaf area of the crop with N fertilization compared to control, which helped in achieving crop developmental stages on time.

#### Physiological stages

The data showed that cropping system and residue retention had no significant effect on days to 50 per cent tasseling, 50 per cent silking, maturity and reproductive period. However, days to 50 per cent tasseling and silking was recorded significantly higher in control plots (61.5 and 70 days) compared to lowest PU (51.9 days and 59.9 days) which were statistically at par with SCU and NCU (Table 3). Days to maturity were found to be non-significant with

nitrogen management practices. Reproductive period recorded significantly higher with the application of NCU, which was on par with the application of PU and SCU. No interaction effect was recorded among the cropping system, residue retention and nitrogen management practices for physiological stages in maize (Table 3).

#### NDVI values

Normalized differential vegetation index (NDVI) value shows direct correlation with plant growth and development and is one of the indicators for plant health. Cropping system and residue retention had no significant effect on NDVI value at any crop growth stage but significantly higher NDVI was recorded in NCU at 30 DAS, which was statistically at par with PU and SCU (Table 4). At 60 and 90 DAS, PU recorded significantly higher NDVI which was on par with the SCU and NCU but significantly superior over control.

**Table 4.** Effect of cropping system, residue and nitrogen management on NDVI, SPAD and canopy temperature depression (CTD) in fourth zero-till *kharif* maize

| Treatment                             | NDVI   |        |        | SPAD values |        |        | CTD (°C) |
|---------------------------------------|--------|--------|--------|-------------|--------|--------|----------|
|                                       | 30 DAS | 60 DAS | 90 DAS | 30 DAS      | 60 DAS | 90 DAS | 60 DAS   |
| <i>Cropping system (System)</i>       |        |        |        |             |        |        |          |
| Maize-mustard-mungbean (MMuMb)        | 0.509  | 0.608  | 0.485  | 45.4        | 36.3   | 43.9   | -2.45    |
| Maize-wheat-mungbean (MWMb)           | 0.525  | 0.612  | 0.479  | 45.7        | 38.3   | 44.0   | -2.78    |
| LSD ( $p=0.05$ )                      | NS     | NS     | NS     | NS          | NS     | NS     | NS       |
| <i>Residue management (Residue)</i>   |        |        |        |             |        |        |          |
| Permanent beds - residue (WoR)        | 0.524  | 0.603  | 0.473  | 45.5        | 36.9   | 43.6   | -1.84a   |
| Permanent beds + residue (WR)         | 0.510  | 0.617  | 0.491  | 45.6        | 37.7   | 44.3   | -3.39b   |
| LSD ( $p=0.05$ )                      | NS     | NS     | NS     | NS          | NS     | NS     | 1.041    |
| <i>Nitrogen management (Nitrogen)</i> |        |        |        |             |        |        |          |
| Absolute control                      | 0.404b | 0.505b | 0.422b | 30.2c       | 30.9c  | 34.4b  | -2.59    |
| N by prilled urea (PU)                | 0.552a | 0.658a | 0.512a | 48.3b       | 41.8a  | 46.9a  | -2.83    |
| N by sulphur coated urea (SCU)        | 0.546a | 0.639a | 0.486a | 50.9ab      | 37.5b  | 46.1a  | -2.43    |
| N by neem coated urea (NCU)           | 0.566a | 0.637a | 0.509a | 52.9a       | 38.9ab | 48.4a  | -2.61    |
| LSD ( $p=0.05$ )                      | 0.051  | 0.0497 | 0.0365 | 3.2026      | 3.2811 | 4.0628 | NS       |
| <b><i>p values</i></b>                |        |        |        |             |        |        |          |
| <i>System</i>                         | 0.9607 | 0.8088 | 0.5989 | 0.7610      | 0.1018 | 0.9528 | 0.3114   |
| <i>Residue</i>                        | 0.6178 | 0.4136 | 0.1745 | 0.9550      | 0.4461 | 0.6363 | <.0001   |
| <i>System × Residue</i>               | 0.7945 | 0.3143 | 0.1952 | 0.0632      | 0.5087 | 0.9057 | 0.7661   |
| <i>System × Nitrogen</i>              | 0.7940 | 0.9918 | 0.1651 | 0.2316      | 0.2185 | 0.1298 | 0.1346   |
| <i>Residue × Nitrogen</i>             | 0.4035 | 0.3960 | 0.0940 | 0.5619      | 0.5001 | 0.1419 | 0.4247   |
| <i>Nitrogen</i>                       | 0.7516 | <.0001 | <.0001 | <.0001      | <.0001 | <.0001 | 0.8493   |

Note: Means followed by different letters in each column are statistically different at  $LSD_{0.05}$ .

### SPAD values

SPAD (Soil Plant Analysis Development)/chlorophyll meter value also show direct correlation with plant growth and development and indicate plant health. The highest SPAD values were observed with the NCU at 30 and 90 DAS, which were at par with SCU at 30 DAS and PU and SCU at 90 DAS (Table 4). However, PU recorded significantly higher value at 60 DAS and was at par with NCU but the least SPAD values were in the control. The enhanced availability of N in soil lead to increased N uptake and hence the application of residue and NCU and other fertilization lead to increased values of NDVI and SPAD values as these values directly correlate with the N in leaves. The similar finding of better nutrition lead enhanced NDVI and SPAD values were also reported by Mohanty *et al.* (2015).

### Canopy temperature depression (CTD)

Data on CTD were recorded at 60 DAS in maize after a dry spell of 12 days (Table 4). There was no significant effect of cropping system and nitrogen management practice on CTD, but residue retention had the significant effect on CTD compared to residue removal. More reduction in canopy temperature was recorded WR (-3.39°C) compared to WoR (-1.84°C).

### Conclusion

It was concluded that the basal application of NCU and residue retention in MWMb system was found significantly superior for enhancing the growth parameters and physiological indices in maize under conservation agriculture.

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## Precision nutrient management influences growth, yield attributes and yield of *kharif* maize (*Zea mays* L.)

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**Abstract:** An investigation was conducted during *kharif* season of 2016 for exploring the influence of precision nutrient management on the growth and yield of maize in sandy loam soil at research farm of ICAR–IARI, New Delhi. The results of the study indicated that growth parameters (plant height, dry matter accumulation and leaf area index) and yield attributes (cobs/ha, cob length, grains/row and grains/cob) in maize were improved significantly with precision nutrient management practices. The Soil-Test Crop Response (STCR)-based precision nutrient management practice increased grain yield of maize to the tune of 2.9-122.7 per cent over control and other precision nutrient management practices. The grain yield of maize in STCR-based nutrition was on par with green seeker-based nutrition in 33 per cent basal N+GS guided at knee high and tasseling (T<sub>3</sub>), 60 per cent basal N + GS guided at tasseling (T<sub>8</sub>) and 70 per cent basal N + GS guided at tasseling (T<sub>9</sub>). This was followed by 30–35 per cent basal N + 30–35 per cent N at 25 days after sowing (DAS)+GS guided at the tasseling stage. Thus, it was concluded that application of 33 per cent basal N + GS guided N or 60–70 per cent basal N + GS guided N at tasseling or 30–35 per cent basal N + 30–35 per cent N at knee high + GS guided at tasseling stage could be a

better choice for enhancing growth, yield attributes and yield in maize production.

**Keywords:** Maize · Precision nitrogen management · Green Seeker · Yield attributes

### Introduction

Maize (*Zea mays* L.) also known as “Queen of cereals”, is the world’s third most important cereal after wheat and rice and is grown in different agro-ecological regions. In addition to staple food for human beings and quality feed for animals, maize serves as a basic raw material to thousands of industrial products *viz.*, starch, oil, protein, alcoholic beverages, food sweeteners, pharmaceutical, cosmetic, film, textile, gum, package and paper industries. In India, it is mainly grown in the rainy (*kharif*) season, which contributes about 70 per cent of the total maize production of the country. As a fodder and grain crop, it is extensively grown in Uttar Pradesh, Rajasthan, Madhya Pradesh, Bihar and Karnataka and now it is gaining popularity as maize, for crop diversification perspective, is being considered as a suitable alternative to rice in the unsustainable rice-wheat cropping system belt of the Indo Gangetic plains. Besides abiotic and biotic stresses, poor soil fertility (including micronutrient deficiencies) and low nutrient use efficiency are among the most important factors limiting maize productivity and yield stability in India (Yadav *et al.*, 2016).

Nitrogen is one of the most important nutrients required in crop production. It is subjected to losses in different forms like ammonia, nitrate, nitrous oxide through different processes such as denitrification,

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volatilization and leaching losses which cause environmental pollution like eutrophication of water bodies, increase in nitrate content in groundwater etc. Nitrous oxide has 298 times more global warming potential than carbon dioxide and its emissions are due to poor nitrogen management in the intensive crop production system (Prasad and Shivay, 2019). The potential for enrichment of ground and surface waters with nitrates also increases with excessive N fertilizer applications causing eutrophication of aquatic ecosystem and methemoglobinemia in infants (blue baby syndrome) (Jat *et al.*, 2016). However, insufficient N availability to crop results in low yields and significantly reduced profits compared to a properly fertilized crop. Efficient nutrient management programmes supply plant nutrients in adequate quantities to sustain maximum crop productivity and profitability while minimizing the environmental impacts of nutrient use (Jat *et al.*, 2013). Ensuring optimum nutrient availability through effective nutrient management practices requires knowledge of the interactions between the soil, plant and environment. The use of some tools for in-season N management like Soil Plant Analysis Development (SPAD)/chlorophyll meter, Green Seeker, Site-Specific Nutrient Management (SSNM) through Soil-Test Crop Response (STCR) or Nutrient Expert helps in fulfilling the crop nutrient requirement with less environmental footprints apart from higher crop productivity and profitability of maize-based production systems in India (Kumar *et al.*, 2014; Kumar *et al.*, 2015; Pooniya *et al.*, 2015; Jat *et al.*, 2016 and Mohanty *et al.*, 2016). Therefore, the adoption of precision nutrient management in maize is expected to pay a rich dividend in India for enhancing crop yield and soil health in future. However, such tools of optical sensor-based N management have been evaluated and standardized in rice (Bijay-Singh *et al.*, 2015) and wheat (Bijay-Singh *et al.*, 2011) but negligible information is available in maize. Keeping this in view, the available tools like SPAD, SSNM and STCR have been tested with newly developed NDVI algorithms for maize in our study for precision nutrient management.

## Materials and methods

A field experiment was conducted at the research farm of ICAR–Indian Agricultural Research Institute, New Delhi, India during *kharif* season of 2016. The different

treatments of nutrient application at different growth stages of maize crop *viz.*, Control (T<sub>1</sub>), RDF (1/3+1/3+/1/3 N splitting at basal, knee-high and tasseling) (T<sub>2</sub>), STCR (1/3 + 1/3 + /1/3 N splitting at basal, knee-high and tasseling) (T<sub>3</sub>), Nutrient expert (1/3 + 1/3 + 1/3 N splitting at basal, knee-high and tasseling) (T<sub>4</sub>), 33 per cent basal N + Green Seeker based N at knee-high & tasseling stage (T<sub>5</sub>), 60 per cent basal N + Green Seeker based N at knee-high (T<sub>6</sub>), 70 per cent basal N + Green Seeker based N at knee-high (T<sub>7</sub>), 60 per cent basal N + Green Seeker based N at tasseling stage (T<sub>8</sub>), 70 per cent basal N + Green Seeker based N at tasseling stage (T<sub>9</sub>), 30 per cent Basal N + 30 per cent at 25 DAS + Green Seeker based N at tasseling stage (T<sub>10</sub>) and 35 per cent Basal N + 35 per cent at 25 DAS + Green Seeker based N at tasseling stage (T<sub>11</sub>). The experiment was laid out in a randomized complete block design (RCBD) with three replications comprising the 11 test treatments with a nitrogen-rich strip. The soil was sandy loam low in organic carbon and available nitrogen and phosphorus and medium in available potassium with a slightly alkaline pH of 7.6.

The growth parameters including, plant height, leaf area index (LAI), dry matter accumulation, SPAD and Normalized difference vegetation index (NDVI) were recorded in maize with standard procedure. The yield attributes were also recorded at harvest of the maize. The plant barrenness percentage in each experimental unit was calculated by using the formula given below:

$$\text{Bareness (\%)} = \frac{\text{Plants / plot} - \text{Cobs / plot}}{\text{Plants / plot}} \times 100$$

After harvesting, all the cobs from each plot were sun-dried and after removing the cob sheath, shelling was done using a hand maize sheller. The moisture percentage in the grain was adjusted to 15 per cent to obtain grain yield.

The data recorded for different parameters were analyzed with the help of the analysis of variance (ANOVA) technique for randomized complete block design using SAS 9.3 software (SAS Institute, Cary, NC). The least significant difference test was used to decipher the effect of treatments at a 5 per cent level of significance ( $P=0.05$ ). The Least significant difference (LSD) values for different pair-wise comparisons amongst the treatment effect were computed. Correlation analysis was performed using ProcCORR in the SAS 9.3 software.

## Results and discussion

### Growth attributes

The highest plant height was recorded with T<sub>3</sub> (STCR) at all growth stages at 30, 60 and 90 DAS (70.44, 191.7 and 197.3 cm) and the lowest with control (Table 1). However, the highest leaf area was recorded under T<sub>8</sub>, which was at par with STCR and the other GS based nitrogen management in T<sub>7</sub> to T<sub>11</sub>. The highest leaf area index (LAI) was recorded at 60 DAS across treatments that might be because at the later stage of the maize crop (90 DAS) the leaves dry up while the number of leaves at the initial stage (30 DAS) are lower. The highest dry matter accumulation (DMA) was also recorded at 90 DAS across all treatments, which could be attributed to the maximum biomass accumulated at the later stage of the maize crop as the leaves dry up but in general, leaves do not detach from the plant. Hence, more DMA was recorded at later growth stages compared to initial growth stages in the crop in contrast to leaf area, which was highest at 60 DAS. Amongst nitrogen management practices, the highest DMA was recorded with T<sub>3</sub> and the lowest was recorded with control at all stages. As the soils of most of the maize growing areas in India are inherently

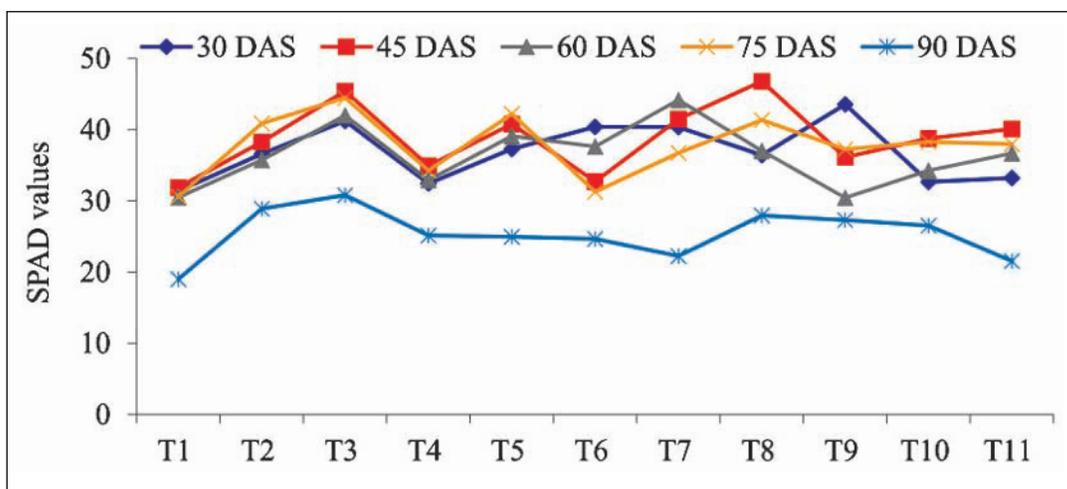
low in soil organic matter and nitrogen is the major limiting plant nutrient, with nitrogen (N) availability being routinely supplemented through the application of fertilizers (Prasad, 2011). As N is the key element in the chlorophyll of the plant and its concentration/supply to the plant have a direct correlation with the photosynthetic area of the plant. The timely supply of N adjusted to crop demand might lead to optimum N nutrition to the crop, which might have enhanced photosynthetic areas in precision nutrient management (PNM) practices and enhanced plant height and DMA in the maize crop. The continuous supply of N to the crop leads to better photosynthesis, which increases the roots and shoots growth in the crop. Efficient nutrient management programmes supply plant nutrients in adequate quantities to sustain maximum crop productivity and profitability while minimizing the environmental impacts of nutrient use (Jat *et al.*, 2013).

The better nutrition made more functional photosynthesis areas that in turn increased the crop growth rate (CGR) in all these treatments. We observed less deviation in the SPAD reading over the period in comparison to normalized difference vegetation index (NDVI), which shows that NDVI is better than SPAD values in maize (Figure 1). At a later stage, NDVI decreases drastically which is not the case with SPAD values as this measure

**Table 1.** Effect of precision nitrogen management practices on the plant population, plant height, leaf area index, dry matter accumulation and NDVI of *kharif* maize

| Treatment       | Plant height (cm)    | LAI                | Dry matter accumulation (g/plant) | Normalized difference vegetation index (NDVI) |                     |                     |
|-----------------|----------------------|--------------------|-----------------------------------|---|---------------------|---------------------|
|                 | 90 DAS               | 90 DAS             | 90 DAS                            | 30 DAS  | 60 DAS              | 90 DAS              |
| T <sub>1</sub>  | 113.7 <sup>d</sup>   | 1.89 <sup>c</sup>  | 91.9 <sup>d</sup>                 | 0.51 <sup>d</sup>                             | 0.52 <sup>d</sup>   | 0.24 <sup>d</sup>   |
| T <sub>2</sub>  | 185.9 <sup>ab</sup>  | 2.57 <sup>ab</sup> | 165.7 <sup>b</sup>                | 0.54 <sup>cd</sup>                            | 0.65 <sup>bc</sup>  | 0.27 <sup>bcd</sup> |
| T <sub>3</sub>  | 197.3 <sup>a</sup>   | 2.60 <sup>ab</sup> | 201.6 <sup>a</sup>                | 0.62 <sup>a</sup>                             | 0.72 <sup>a</sup>   | 0.37 <sup>a</sup>   |
| T <sub>4</sub>  | 179.6 <sup>abc</sup> | 2.41 <sup>ab</sup> | 132.5 <sup>c</sup>                | 0.59 <sup>bc</sup>                            | 0.62 <sup>c</sup>   | 0.27 <sup>bcd</sup> |
| T <sub>5</sub>  | 168.7 <sup>bc</sup>  | 2.76 <sup>a</sup>  | 169.5 <sup>b</sup>                | 0.60 <sup>bc</sup>                            | 0.66 <sup>abc</sup> | 0.26 <sup>cd</sup>  |
| T <sub>6</sub>  | 166.6 <sup>c</sup>   | 2.45 <sup>ab</sup> | 161.9 <sup>b</sup>                | 0.50 <sup>d</sup>                             | 0.62 <sup>c</sup>   | 0.27 <sup>bcd</sup> |
| T <sub>7</sub>  | 191.7 <sup>a</sup>   | 2.41 <sup>ab</sup> | 169.4 <sup>b</sup>                | 0.55 <sup>bcd</sup>                           | 0.68 <sup>ab</sup>  | 0.27 <sup>bcd</sup> |
| T <sub>8</sub>  | 197.2 <sup>a</sup>   | 2.70 <sup>a</sup>  | 194.8 <sup>a</sup>                | 0.61 <sup>ab</sup>                            | 0.68 <sup>ab</sup>  | 0.30 <sup>b</sup>   |
| T <sub>9</sub>  | 196.0 <sup>a</sup>   | 2.61 <sup>a</sup>  | 193.0 <sup>a</sup>                | 0.60 <sup>bc</sup>                            | 0.67 <sup>abc</sup> | 0.29 <sup>bc</sup>  |
| T <sub>10</sub> | 179.3 <sup>abc</sup> | 2.13 <sup>bc</sup> | 173.6 <sup>b</sup>                | 0.54 <sup>cd</sup>                            | 0.67 <sup>abc</sup> | 0.28 <sup>bc</sup>  |
| T <sub>11</sub> | 184.0 <sup>abc</sup> | 2.49 <sup>ab</sup> | 175.1 <sup>b</sup>                | 0.54 <sup>cd</sup>                            | 0.66 <sup>bc</sup>  | 0.26 <sup>bcd</sup> |
| p-Value         | <.0001               | 0.0469             | <.0001                            | 0.0017  | 0.0002              | <.0001              |

*Note:* T<sub>1</sub>: Control, T<sub>2</sub>: RDF, T<sub>3</sub>: STCR, T<sub>4</sub>: NE-SSNM, T<sub>5</sub>: 33% RDN + GS-KH + GS-T, T<sub>6</sub>: 60% RDN + GS-KH, T<sub>7</sub>: 70% RDN + GS-KH, T<sub>8</sub>: 60% RDN + GS-T, T<sub>9</sub>: 70% RDN + GS-T, T<sub>10</sub>: 30% RDN + 30% RDN at 25 DAS + GS-T and T<sub>11</sub>: 35% RDN + 35% RDN at 25 DAS + GS-T. [Means followed by a similar lowercase letter within a column are not significantly different according to least significant difference test (P=0.05)].



**Figure 1.** Effect of precision nitrogen management practices on SPAD values of *kharif* maize at different crop stages

Note: T<sub>1</sub>: Control, T<sub>2</sub>: RDF, T<sub>3</sub>: STCR, T<sub>4</sub>: NE-SSNM, T<sub>5</sub>: 33% RDN + GS-KH + GS-T, T<sub>6</sub>: 60% RDN + GS-KH, T<sub>7</sub>: 70% RDN + GS-KH, T<sub>8</sub>: 60% RDN + GS-T, T<sub>9</sub>: 70% RDN + GS-T, T<sub>10</sub>: 30% RDN + 30% RDN at 25 DAS + GS-T and T<sub>11</sub>: 35% RDN + 35% RDN at 25 DAS + GS-T.

the health of the leaves not the overall vigour of the crop. Hence, SPAD did not take into account overall crop health. A positive and significant correlation between the NDVI values at 30, 60 DAS and all the growth parameters were observed in *kharif* maize, which indicates that the NDVI could be used as the best predictor for maize health (Table 1). The enhanced availability of N in the soil leads to increased N uptake that in turn leads to increased values of NDVI and SPAD as these values directly correlate with the N in leaves. Singh *et al.* (2013) found that the greenness of wheat leaves at the maximum tillering stage was found to be a function of N applied at planting. Similar findings of better nutrition (STCR/GS) leading to enhanced NDVI and SPAD values in wheat have been reported by Mohanty *et al.* (2016). Precision nutrition (NE-SSNM) linked enhancement in maize growth rate also reported by Ghosh (2015) and Kumar *et al.* (2014) in the same agro-ecologies.

#### Crop growth rate and relative growth rate

The highest crop growth rate (CGR) was recorded at 0–30 and 60–90 DAS in STCR, while at 30–60 DAS it was with T<sub>5</sub> (33 per cent basal + Green Seeker guided) (Table 2). However, the relative growth rate (RGR) at 0–30 DAS was highest in STCR but at 60–90 DAS was with GS guided application (T<sub>8</sub> to T<sub>11</sub>). In all the cases, the CGR and RGR were lowest in control treatments where no nitrogen was applied (Table 2).

#### Yield attributes in maize

Significantly, higher number of cobs were recorded in all nitrogen applied treatments over control, while the highest plant barrenness (11.55 per cent) was registered in control plots (Table 3). The cob girth was statistically similar in all the treatments but the cob length increased significantly with different precision nitrogen management practices and was highest in STCR that was statistically at par with RDF and GS guided T<sub>7</sub>, T<sub>8</sub> and T<sub>9</sub> treatments. The enhanced nutrient availability, which gave higher crop growth parameters and translated into producing more cobs. This subsequently enhanced cobs/ha along with enhancement in cob length but no differences were observed for cob girth and grain rows/cob. As cob girth and grain rows/cob are more genetically driven parameters of the crop that requires higher management practices to get significantly influenced (Saif-ul-malook *et al.*, 2014). The enhancement in crop growth parameters could be attributed to increased leaf area, plant height and dry matter accumulation in maize, which gave significantly higher cob parameters due to enhanced photosynthetic areas for more resource utilization.

Enhancement in these cob parameters linked with better nutrition/NE-SSNM in maize as reported by Kalpana and Krishnarajan (2002); Kumar *et al.* (2014) and Ghosh (2015). The highest grains/row and grains/cob were registered with GS guided T<sub>9</sub> which was statistically similar to T<sub>3</sub> and other PNM except for T<sub>11</sub> and T<sub>6</sub> in the

**Table 2.** Effect of precision nitrogen management practices on crop growth rate and relative growth rate of *kharif* maize

| Treatment       | Crop growth rate (g/plant/day) |                      |                    | Relative growth rate (mg/g/day) |           |                     |
|-----------------|--------------------------------|----------------------|--------------------|---------------------------------|-----------|---------------------|
|                 | 0–30 DAS                       | 30–60 DAS            | 60–90 DAS          | 0–30 DAS                        | 30–60 DAS | 60–90 DAS           |
| T <sub>1</sub>  | 0.71 <sup>e</sup>              | 0.82 <sup>d</sup>    | 1.53 <sup>d</sup>  | 44.32 <sup>e</sup>              | 9.47      | 10.04 <sup>c</sup>  |
| T <sub>2</sub>  | 0.92 <sup>cd</sup>             | 1.17 <sup>abc</sup>  | 3.43 <sup>b</sup>  | 47.91 <sup>d</sup>              | 11.9      | 14.14 <sup>ab</sup> |
| T <sub>3</sub>  | 1.18 <sup>a</sup>              | 1.30 <sup>ab</sup>   | 4.24 <sup>a</sup>  | 51.59 <sup>a</sup>              | 10.74     | 14.48 <sup>ab</sup> |
| T <sub>4</sub>  | 0.94 <sup>cd</sup>             | 0.87 <sup>cd</sup>   | 2.61 <sup>c</sup>  | 48.34 <sup>cd</sup>             | 11.05     | 12.91 <sup>ab</sup> |
| T <sub>5</sub>  | 0.91 <sup>d</sup>              | 1.36 <sup>a</sup>    | 3.38 <sup>b</sup>  | 47.84 <sup>d</sup>              | 13.21     | 13.23 <sup>ab</sup> |
| T <sub>6</sub>  | 1.01 <sup>bcd</sup>            | 1.16 <sup>abcd</sup> | 3.22 <sup>bc</sup> | 49.24 <sup>bcd</sup>            | 11.11     | 13.27 <sup>ab</sup> |
| T <sub>7</sub>  | 1.11 <sup>ab</sup>             | 1.31 <sup>ab</sup>   | 3.23 <sup>bc</sup> | 50.75 <sup>ab</sup>             | 11.26     | 12.29 <sup>bc</sup> |
| T <sub>8</sub>  | 1.01 <sup>bcd</sup>            | 1.27 <sup>ab</sup>   | 4.21 <sup>a</sup>  | 49.41 <sup>bcd</sup>            | 11.75     | 15.14 <sup>a</sup>  |
| T <sub>9</sub>  | 1.06 <sup>abc</sup>            | 1.25 <sup>ab</sup>   | 4.13 <sup>a</sup>  | 50.00 <sup>abc</sup>            | 11.29     | 14.88 <sup>a</sup>  |
| T <sub>10</sub> | 0.94 <sup>cd</sup>             | 1.07 <sup>abcd</sup> | 3.78 <sup>ab</sup> | 48.28 <sup>cd</sup>             | 11.04     | 15.30 <sup>a</sup>  |
| T <sub>11</sub> | 1.01 <sup>bcd</sup>            | 1.00 <sup>bcd</sup>  | 3.83 <sup>ab</sup> | 49.39 <sup>bcd</sup>            | 9.93      | 15.45 <sup>a</sup>  |
| <i>p</i> value  | 0.0002                         | 0.045                | <.0001             | <.0001                          | 0.5352    | 0.0088              |

Note: T<sub>1</sub>: Control, T<sub>2</sub>: RDF, T<sub>3</sub>: STCR, T<sub>4</sub>: NE-SSNM, T<sub>5</sub>: 33% RDN + GS-KH + GS-T, T<sub>6</sub>: 60% RDN + GS-KH, T<sub>7</sub>: 70% RDN + GS-KH, T<sub>8</sub>: 60% RDN + GS-T, T<sub>9</sub>: 70% RDN + GS-T, T<sub>10</sub>: 30% RDN + 30% RDN at 25 DAS + GS-T and T<sub>11</sub>: 35% RDN + 35% RDN at 25 DAS + GS-T. [Means followed by a similar lowercase letter within a column are not significantly different according to the least significant difference test ( $p=0.05$ )].

case of grains/cob and with all other treatments except T<sub>1</sub>, T<sub>6</sub>, T<sub>8</sub> and T<sub>11</sub> for grains/row. The increase in growth and cob parameters led to a better source-sink relationship, which might enhance the grains/cob and grain weight/cob. A similar finding of enhancement in grain attributes in ZT maize was also reported in earlier studies (Kumar *et al.*, 2014; Ghosh, 2015).

#### Grain yield

The grain yield of maize increased significantly with the application of STCR based nutrition that was on par with T<sub>2</sub>, T<sub>8</sub> and T<sub>9</sub> (Table 3). This shows the GS guided N application develops a better source-sink relationship as this synchronises the N supply with crop demand. The increase with STCR was 122.7, 6.3 and 14.3, 13.8, 31.2, 15.0, 4.7, 2.9, 12.0 and 14.1 per cent higher over the T<sub>1</sub>, T<sub>2</sub> and T<sub>4</sub> to T<sub>11</sub>, respectively. However, in comparison to control, the yield in all fertilization treatments was almost more than double in most of the cases that warrant the necessity of efficient nitrogen management in maize. The progressive growth and yield attributes in maize led to a better source-sink relationship in maize that in turn gave higher grain yield of maize due to enhanced mineral N at various crop growth stages found in this study (data not presented). The yield attributes improvement led to increased yield in particular treatment further supported

by a significant positive correlation found in our study (Table 4). Moreover, the crop yield is a function of the yield attributes and their efficiency in particular treatment. The blanket recommendations based on the fixed-time application of fertilizer N doses at specified growth stages do not consider the dynamic soil nutrient supply and crop nutrient requirements and lead to the untimely and inefficient application of fertilizer nutrients (Biradar *et al.*, 2012). Therefore, crop need-based fertilizer management in maize can help in improving recovery efficiency and reducing losses of nutrients.

The significant and positive correlation among various crop growth parameters in our study confirms that the photosynthetic area enhancement has a direct relation with other growth parameters. The enhancement in leaf area leads to more photosynthesis that in turn increased the DMA, CGR and RGR under STCR and GS based N application treatments over control and other practices (Table 4). Similar findings of the PNM linked enhancement in crop growth parameters of maize were reported by Kumar *et al.* (2014) and Ghosh (2015). The significant correlation between the growth parameters and cob parameters observed in this study supports our hypothesis of growth lead to enhanced cob parameters. The barrenness in maize significantly reduced due to precision N fertilization as this is the main nutrient responsible for crop growth by way of its direct involvement in

**Table 3.** Effect of precision nitrogen management practices on yield attributes and yield of *kharif* maize

| Treatment       | Cobs ('000/ha)     | Barrenness (%)      | Cob length (cm)      | Cob girth (cm) | Grains per row       | Grains per cob       | 1000-grains weight (g) | Grain yield (t/ha)  |
|-----------------|--------------------|---------------------|----------------------|----------------|----------------------|----------------------|------------------------|---------------------|
| T <sub>1</sub>  | 58.97 <sup>b</sup> | 11.55 <sup>a</sup>  | 12.67 <sup>d</sup>   | 11.80          | 21.11 <sup>d</sup>   | 267.4 <sup>d</sup>   | 221.7                  | 2.51 <sup>e</sup>   |
| T <sub>2</sub>  | 71.53 <sup>a</sup> | 1.50 <sup>ef</sup>  | 15.73 <sup>abc</sup> | 12.90          | 27.22 <sup>a</sup>   | 355.6 <sup>abc</sup> | 249.3                  | 5.26 <sup>abc</sup> |
| T <sub>3</sub>  | 71.08 <sup>a</sup> | 0.69 <sup>f</sup>   | 17.64 <sup>a</sup>   | 12.76          | 26.89 <sup>ab</sup>  | 367.0 <sup>a</sup>   | 243.3                  | 5.59 <sup>a</sup>   |
| T <sub>4</sub>  | 72.60 <sup>a</sup> | 2.36 <sup>cde</sup> | 13.03 <sup>d</sup>   | 12.01          | 25.89 <sup>abc</sup> | 339.1 <sup>abc</sup> | 231.7                  | 4.89 <sup>c</sup>   |
| T <sub>5</sub>  | 71.19 <sup>a</sup> | 2.04 <sup>de</sup>  | 15.53 <sup>bc</sup>  | 12.92          | 25.78 <sup>abc</sup> | 346.8 <sup>abc</sup> | 234.7                  | 4.91 <sup>c</sup>   |
| T <sub>6</sub>  | 70.14 <sup>a</sup> | 2.95 <sup>cd</sup>  | 12.79 <sup>d</sup>   | 12.08          | 23.68 <sup>bed</sup> | 325.2 <sup>bc</sup>  | 242.0                  | 4.26 <sup>d</sup>   |
| T <sub>7</sub>  | 70.51 <sup>a</sup> | 3.31 <sup>bc</sup>  | 16.40 <sup>abc</sup> | 13.20          | 27.11 <sup>a</sup>   | 356.1 <sup>abc</sup> | 243.0                  | 4.86 <sup>c</sup>   |
| T <sub>8</sub>  | 71.22 <sup>a</sup> | 2.37 <sup>cde</sup> | 17.00 <sup>ab</sup>  | 12.78          | 25.73 <sup>abc</sup> | 360.3 <sup>ab</sup>  | 234.7                  | 5.34 <sup>abc</sup> |
| T <sub>9</sub>  | 69.81 <sup>a</sup> | 4.27 <sup>b</sup>   | 16.69 <sup>ab</sup>  | 12.96          | 27.33 <sup>a</sup>   | 379.6 <sup>a</sup>   | 232.0                  | 5.43 <sup>ab</sup>  |
| T <sub>10</sub> | 71.88 <sup>a</sup> | 2.82 <sup>cd</sup>  | 14.53 <sup>cd</sup>  | 12.32          | 27.00 <sup>ab</sup>  | 360.0 <sup>ab</sup>  | 238.3                  | 4.99 <sup>bc</sup>  |
| T <sub>11</sub> | 72.63 <sup>a</sup> | 2.12 <sup>de</sup>  | 15.46 <sup>bc</sup>  | 12.44          | 23.11 <sup>cd</sup>  | 318.7 <sup>c</sup>   | 253.3                  | 4.90 <sup>c</sup>   |
| p-Value         | <.0001             | <.0001              | 0.0001               | 0.1308         | 0.0117               | 0.0012               | 0.0762                 | <.0001              |

Note: T<sub>1</sub>: Control, T<sub>2</sub>: RDF, T<sub>3</sub>: STCR, T<sub>4</sub>: NE-SSNM, T<sub>5</sub>: 33% RDN + GS-KH + GS-T, T<sub>6</sub>: 60% RDN + GS-KH, T<sub>7</sub>: 70% RDN + GS-KH, T<sub>8</sub>: 60% RDN + GS-T, T<sub>9</sub>: 70% RDN + GS-T, T<sub>10</sub>: 30% RDN + 30% RDN at 25 DAS + GS-T and T<sub>11</sub>: 35% RDN + 35% RDN at 25 DAS + GS-T. [Means followed by a similar lowercase letter within a column are not significantly different according to the least significant difference test ( $P=0.05$ )].

**Table 4.** Correlation matrix among the different yield attributes and yield of *kharif* maize

| Parameter          | Cobs/ha  | Cob length | Cob girth | Grain per row | Grains per cob | Grain weight per cob | Grain yield | 1000-grains weight |
|--------------------|----------|------------|-----------|---------------|----------------|----------------------|-------------|--------------------|
| Cobs/ha            | -        |            |           |               |                |                      |             |                    |
| Cob length         | 0.267    |            |           |               |                |                      |             |                    |
| Cob girth          | 0.302    | 0.698***   |           |               |                |                      |             |                    |
| Grain/row          | 0.421*   | 0.463**    | 0.468**   |               |                |                      |             |                    |
| Grains/cob         | 0.514**  | 0.550***   | 0.623***  | 0.899***      |                |                      |             |                    |
| Grain weight/cob   | 0.557*** | 0.532**    | 0.671***  | 0.797***      | 0.860***       |                      |             |                    |
| Grain yield        | 0.788*** | 0.522**    | 0.573***  | 0.688***      | 0.809***       | 0.859***             |             |                    |
| 1000-grains weight | 0.439*   | 0.272      | 0.417*    | 0.308         | 0.298          | 0.740***             | 0.570***    |                    |
| Harvest index      | 0.690*** | 0.224      | 0.360*    | 0.560***      | 0.684***       | 0.705***             | 0.769***    | 0.463**            |

The \*, \*\* or \*\*\* indicates significance at 5, 1 and 0.1% probability.

photosynthesis as a constituent of chlorophyll and its significant positive interactions with other nutrients utilization. Moreover, the hybrid maize responses to N fertilization also enhanced and under no fertilization the barrenness could be much higher and sometimes in low fertility soil, it could not reproduce. Thus, it was concluded that application of 33 per cent basal N + GS guided N or 60–70 per cent basal N + GS guided N at tasseling or 30–35% basal N + 30–35 per cent N at knee high + GS guided at tasseling stage could be a better choice for enhancing growth, yield attributes and yield in maize production.

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## Potassium fertilization effects on growth and yield of hybrid maize under semi-arid conditions of Kandahar province of Afghanistan

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**Abstract:** A field experiment was carried out at the main block of Tarnak Farm of Afghanistan National Agricultural Sciences and Technology University (ANASTU) Kandahar, Afghanistan during the summer season of 2015, to determine the effects of different potassium fertilizer levels on growth and yields parameters of hybrid maize. The experiment consisted eight potash fertilizer levels viz. 0, 30, 60, 90, 120, 150, 180 and 210 kg K<sub>2</sub>O/ha in a randomized complete block design (RCBD) and replicated thrice. Results showed that application of 60 kg K<sub>2</sub>O/ha significantly improved plant height, leaf area index and gains/cob over control and statistically on par with other higher K levels. The highest dry matter accumulation, grains/row and 1000-grains weight were recorded with the application of 210 kg K<sub>2</sub>O/ha, which was statistically at par with all other treatments except control. Cob length and grain yield were significantly highest with 180/210 kg K<sub>2</sub>O/ha which was on par with nutrient level of up to 90 kg K<sub>2</sub>O/ha. The yield obtained with application of 90 kg K<sub>2</sub>O/ha was on par with 60 kg K<sub>2</sub>O/ha. Therefore, it is concluded that application of 60 kg K<sub>2</sub>O/ha to be recommended for enhancing productivity of hybrid maize under semi-arid condition of Kandahar province of Afghanistan.

**Keywords:** Dry matter accumulation · Leaf area index · Yield attributes · Yield · *Zea Mays*

### Introduction

In Afghanistan, maize is grown on 0.142 million hectares area. The total production is 0.312 million tonnes with an average productivity of 2.2 tonnes/ha which is less than half of the global productivity. Out of this, 0.055 m tonnes is used as human food and 0.2 m tonnes for animal feed. Afghanistan imports large quantities of maize from neighboring countries, as domestic production is not sufficient for 30 million populations. Currently, the main constraints to maize production for small-scale farmers in Afghanistan are the lack of improved adapted varieties, and shortage of high-quality fertilizer (Jilani *et al.*, 2013). Due to the poor inherent soil fertility in Afghanistan, the use of fertilizer is quite common. After nitrogen and phosphorus, potassium (K) is the nutrient most likely to limit plant growth. It is needed for energy metabolism, starch synthesis, photosynthesis, nitrogen fixation, and sugar degradation and it is not currently considered a threat to water quality (Anonymous, 2022); however, K fertility management is important because plants with optimum K levels are more resistant to environmental stresses, including drought. Application of potassium @ 375 kg/ha increased grain weight/cob, 1,000-grains weight, grain yield and biological yields of different maize hybrids (Ali *et al.*, 2004). Maize yield parameters like cob weight, cob length, cob girth and grain yield increased steadily with increasing potassium doses and reached optimum values at 155.8 and 144.8 kg K/ha (Kenyanya *et al.*, 2014). Therefore, it is necessary that suitable amount of potassic fertilizer should be applied to enhance

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crop productivity in a cost-effective manner. Keeping these aspects in view an experiment was undertaken to study the effect of potassic fertilizer levels on growth and yield of hybrid maize.

## Materials and methods

The experiment was conducted in the main block of Tarnak Farm of Afghanistan National Agricultural Sciences and Technology University (ANASTU) Kandahar, Afghanistan during the summer season of 2015. The rainfall received during crop growing period from May to August was 9.6 mm. The soil of the experimental site was sandy clay loam with 7.4 pH and was low in nitrogen, medium in available phosphorus and high in available potassium. The experiment was laid out in randomized block design with three replications. The potassium treatments were 0, 30, 60, 90, 120, 150, 180 and 210 kg K<sub>2</sub>O/ha, applied as basal dose through muriate of potash. Nitrogen and phosphorus were applied equally to all plots as per recommended dose of 150 and 60 kg/ha, respectively. Maize cultivar CS-200 was sown using seed rate 25 kg/ha at 70 cm × 20 cm spacing. Due to subtropical and arid climate of experimental area, irrigation were given based on soil moisture condition, the crop was irrigated nine times during growing season. First irrigation was done at 13 days after sowing (DAS) and subsequently, eight irrigation were applied as and when required at different plant growth stages, until the crop attained its physiological maturity. Growth parameters were recorded from three randomly selected plants from

each plot using a standard procedure. The data were collected at 30, 60, 90 DAS and at harvest (115 DAS). The data for yield parameters such as grains/cob, grains/row, cob girth, cob diameter, cob length and 1000-grains weight were recorded from six randomly sampled plants in each experimental unit. The data recorded for different parameters were analyzed statistically by the method of analysis of variance (ANOVA) in complete randomized block design by using SAS 9.3 software. The standard error of means was calculated in all the cases. Wherever the 'F' test was found significant at 5 per cent level of significance correspondence least significant differences (LSD) values were calculated to compare the significance of results. The correlation analysis was performed using the MS excel.

## Results and discussion

### *Growth attributes*

The growth parameters viz., plant height, dry matter accumulation (DMA) and leaf area index (LAI) at various crop growth stages responded to potassium application in maize up to 60 kg K<sub>2</sub>O/ha (Table 1). The application of 30 kg K<sub>2</sub>O/ha resulted in the increase of maize plant height by 29.3, 21.8, 16.4 and 12.3 per cent over control at 30, 60, 90 DAS and at harvest, respectively. The similar increase was also reported in leaf area index. The application of 30 kg K<sub>2</sub>O/ha increased dry matter accumulation by 30.0, 11.7, 32.8 and 26.9 per cent at 30, 60, 90 DAS and at harvest, respectively which was at par

**Table 1.** Effect of potassium levels on plant height, leaf area index and dry matter accumulation of maize at different growth stages

| K levels<br>(kg K <sub>2</sub> O/ha) | Plant height (cm) |        |        |            | Leaf area index |        |        | Dry matter accumulation (g/plant) |        |        |            |
|--------------------------------------|-------------------|--------|--------|------------|-----------------|--------|--------|-----------------------------------|--------|--------|------------|
|                                      | 30 DAS*           | 60 DAS | 90 DAS | At harvest | 30 DAS          | 60 DAS | 90 DAS | 30 DAS                            | 60 DAS | 90 DAS | At harvest |
| 0                                    | 16.7              | 75.3   | 145.5  | 154.9      | 0.32            | 3.05   | 3.46   | 2.0                               | 91.3   | 124.4  | 179.5      |
| 30                                   | 21.6              | 91.7   | 169.4  | 174.0      | 0.42            | 3.18   | 3.72   | 2.6                               | 102.0  | 165.2  | 227.7      |
| 60                                   | 25.6              | 102.4  | 187.7  | 188.2      | 0.45            | 3.70   | 3.88   | 2.8                               | 109.8  | 173.7  | 237.9      |
| 90                                   | 26.8              | 103.9  | 188.3  | 188.8      | 0.44            | 3.77   | 3.95   | 2.9                               | 110.7  | 175.0  | 239.4      |
| 120                                  | 27.0              | 104.1  | 189.0  | 189.7      | 0.44            | 3.78   | 4.01   | 3.0                               | 112.8  | 177.8  | 242.7      |
| 150                                  | 27.0              | 104.8  | 190.1  | 190.8      | 0.44            | 3.85   | 4.06   | 3.2                               | 114.2  | 179.9  | 245.0      |
| 180                                  | 28.0              | 106.3  | 191.0  | 191.2      | 0.47            | 3.90   | 4.08   | 3.3                               | 115.1  | 185.8  | 252.2      |
| 210                                  | 28.3              | 106.7  | 191.7  | 192.3      | 0.47            | 3.93   | 4.11   | 3.3                               | 115.4  | 187.2  | 253.9      |
| SEm(±)                               | 1.61              | 5.19   | 7.11   | 5.88       | 0.022           | 0.145  | 0.114  | 0.11                              | 4.44   | 11.8   | 11.76      |
| LSD (P=0.05)                         | 4.88              | 15.75  | 21.58  | 17.82      | 0.068           | 0.439  | 0.347  | 0.35                              | 13.46  | 35.7   | 35.66      |

DAS\*= Days after sowing

up to 210 kg K<sub>2</sub>O/ha over control. The similar findings of increased plant height and leaf area and leaf area index (LAI) in maize were reported by many workers in South Asia (Tabri and Akil, 2010; Ahmad *et al.*, 2015). Mastoi *et al.* (2013) also found that potassium application at 60 kg/ha by integrating organic and inorganic K fertilizers increased plant height (2 to 15 per cent), and fresh biomass (13 to 60 per cent) in Pakistan. Similarly, the fertilizer level of 30 K<sub>2</sub>O kg/ha resulted in significant increase in growth characters i.e. plant height, leaf area index and dry matter production at different growth stages in Kashmir, Budgam in India (Khuspe *et al.*, 2016). It might be due to potassium @ 60 kg/ha as basal application improved the root system in hybrid maize (Gul *et al.*, 2015). Moreover, the potassium application improves utilization of water, its application accelerates photosynthesis process, water uptake through roots and reduces the per cent of senescent stalks, lodging and increased crushing strength and rind thickness (Bukhsh *et al.*, 2012).

As a result, the application of potassium improves leaf area index, dry matter accumulation and other allometric parameters. The enhanced growth due to potassium application was due to several roles of K in crop plants that might help in increasing plant height and leaf area index over control. The important reasons might be increased root growth (Valadabadi and Farahani, 2009), improved stalk strength (Sala *et al.*, 2014), increased photosynthesis (Hussain *et al.*, 2015) and balancing the internal nutrient status specially Na: K ratios in saline soils (Wang *et al.*, 2013), and significant improvement in most of the physiological processes (Wang *et al.*, 2013).

#### *Yields attributes and yield of maize*

The yield attributes and yield of maize increased significantly with potassium application in our study (Table 2). Significantly higher cob length in maize was recorded by the application of potassium over control. The highest cob length of maize (20 cm) was recorded by the application of 210 kg K<sub>2</sub>O/ha and was at par with higher levels of potassium application beyond 90 kg K<sub>2</sub>O/ha (18.6 cm) in our study. The cob length increased due to K nutrition (175 kg/ha) which corroborates with our findings as reported earlier by Ijaz *et al.* (2014). The enhancement in the growth parameters resulted in the generation of the better sink which might have increased the sink strength and hence improved the cob length significantly. The positive correlation estimated in our study for cob length with growth parameters further affirmed our assumption (Figure 1 and 2). Thus, the better source is a key for the best sink in maize.

Significantly higher number of grains/rows (35.5) was recorded by the application of potassium 210 kg K<sub>2</sub>O/ha. The application of 30 kg K<sub>2</sub>O/ha resulted increased in grains/row by 15.5 per cent and 1,000-grains weight by 11.7 per cent over control which was at par with all K levels in our study. The highest number of grains/cobs (463) was recorded with the application of 210 kg K<sub>2</sub>O/ha, however it was not significant with higher doses beyond 60 kg K<sub>2</sub>O/ha. The yield attributes of maize viz., cob length, gains/cob, increased with application of 90 and 60 kg K<sub>2</sub>O/ha, respectively over no K application. The yield obtained in 90 kg K<sub>2</sub>O/ha was on par with 60 kg K<sub>2</sub>O/ha. The enhancement in the growth parameter which

**Table 2.** Effect of potassium levels on yield attributes and yield of maize

| K levels (kg K <sub>2</sub> O/ha) | Grains/row | Grains/cob | Cob length (cm) | 1,000-grains weight (g) | Grain yield (t/ha) |
|-----------------------------------|------------|------------|-----------------|-------------------------|--------------------|
| Control                           | 28.3       | 377        | 15.8            | 213                     | 4.3                |
| 0                                 | 32.7       | 410        | 17.3            | 238                     | 5.7                |
| 30                                | 33.0       | 438        | 17.6            | 255                     | 7.5                |
| 60                                | 33.2       | 442        | 18.6            | 258                     | 7.8                |
| 90                                | 35.0       | 443        | 18.9            | 260                     | 8.0                |
| 120                               | 35.2       | 452        | 19.0            | 262                     | 8.4                |
| 150                               | 35.4       | 457        | 19.7            | 262                     | 8.6                |
| 180                               | 35.5       | 463        | 20.0            | 263                     | 8.6                |
| SEm±                              | 1.43       | 15.4       | 0.52            | 10.16                   | 0.32               |
| LSD (P=0.05)                      | 4.33       | 46.6       | 1.58            | 30.80                   | 0.96               |

acted as a source for better photosynthesis and accumulation of more sugars for generation of the better sink and ultimately enhanced the yield attributing characters in maize. However, more genetic controlled attributed like cob girth, cobs/plant and grain rows/cob were not

significantly influenced by K fertilization (data not reported). This shows that the parameters like cob length, grains/cob, grains/row and 1,000-grains weight can be increased in maize by K fertilization. The better source and good sink with the continued supply of nutrients and

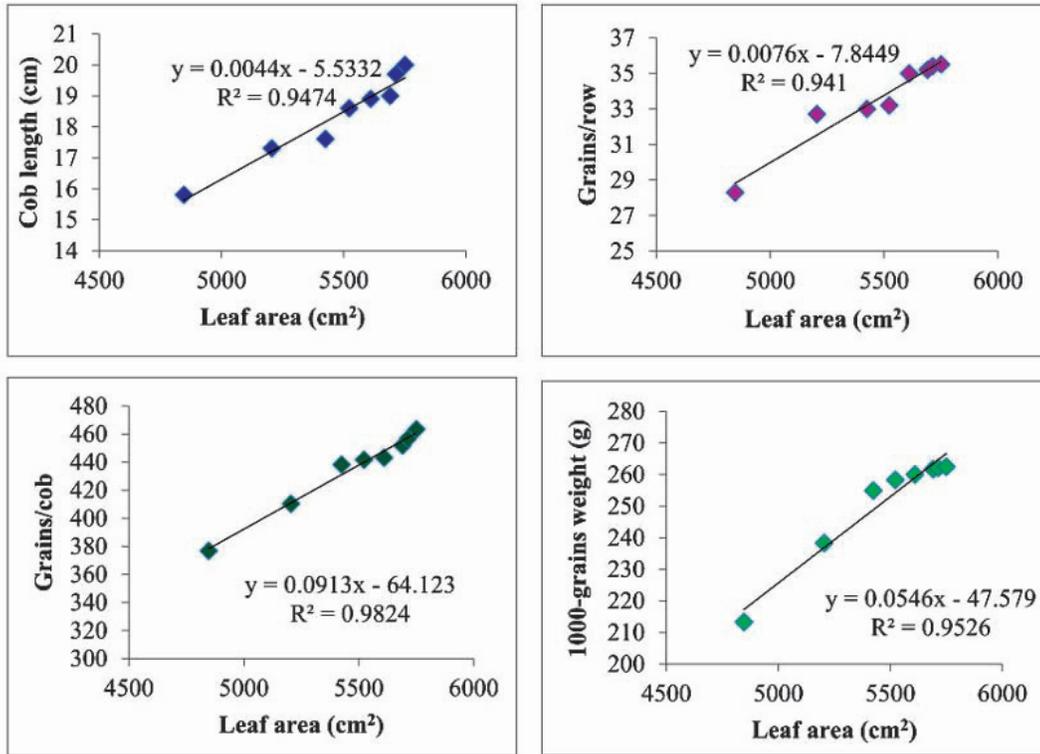


Figure 1. Correlation among the various yields attributes with the leaf area at 90 DAS in maize

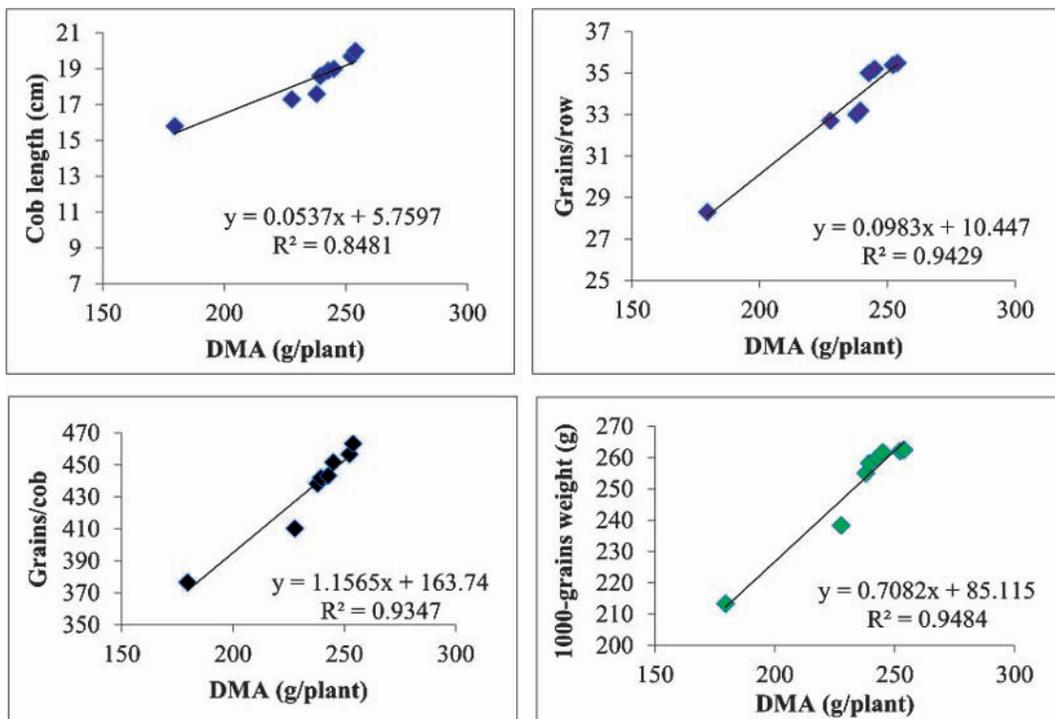


Figure 2. Correlation among the various yields attributes with the dry matter accumulation (DMA) at harvest in maize

photosynthetic might have helped in the production of bolder grains and thus, enhanced the grain weight/cob as well as 1,000-grains weight in our study. However, Tabri and Akil (2010) reported that K application had no significant effect on 1,000-seed weight and weight of cobs.

Significantly higher grain yield (8.6 t/ha) were recorded with the application of 180 and 210 kg K<sub>2</sub>O/ha over control (4.3 t/ha). The application of lower doses of potassium @ 30 and 60 kg K<sub>2</sub>O/ha resulted in increased grain yield (t/ha) to the tune of 32.6 and 74.6 per cent compared to control, respectively. The enhanced growth attributes like dry matter, plant height and leaf area increased the yield attributes of maize in our study. The strong significant positive correlation among these parameters confirms that these are linked and enhancement in growth led to increased yield attributes in maize in Kandahar conditions (Figure 1 and 2).

Application of potassium with little dose of 30 kg K<sub>2</sub>O/ha could enhance the dry matter accumulation, grains/rows and 1000-grains weight while 60 kg K<sub>2</sub>O/ha enhances plant height, leaf area index and gains/cob of maize and further increase in level up to 90 kg K<sub>2</sub>O/ha increased key parameters of cob length and grain yield. The application of potassium up to 60 kg K<sub>2</sub>O/ha significantly enhanced most of the growth parameters and yield attributes of maize, while application of 90 kg K<sub>2</sub>O/ha resulted in significantly higher grain yield of maize under semi-arid conditions of Kandahar province of Afghanistan. It is concluded that application of 60 kg K<sub>2</sub>O/ha could enhance key yield attributes and yield of hybrid maize under semi-arid condition of Kandahar province of Afghanistan.

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## Effects of sowing date on the growth and yield of maize cultivars (*Zea mays* L.) in spring season

Narender Singh · M. C. Kamboj · Preeti Sharma

**Abstract:** A field experiment was conducted to study the effect of date of sowing on grain yield of spring maize at CCS Haryana Agricultural University, Regional Research station, Karnal during the year 2015, 2016 and 2017. Seven maize hybrids (HQPM 1, HQPM 4, HQPM 5, HM 9, HM 10, HM 11 and HM 12) were tested with combination of five sowing dates (last week of January, first week of February, second week of February, third week of February and fourth week of February). The delayed planting led to decreased days to 50 per cent silking and maturity in maize. The highest grain yield, returns over variable cost and B: C ratio were obtained when the crop sown in the first week of February followed by the last week of January, second week of February, third week of February and fourth week of February. The crop duration reduced to 114.3 from 126.4 days when crop planted in fourth week of February compared to planting in last week of January. The hybrid HM 10 gave the highest grain yield, returns over variable cost and B: C ratio followed by HM 11, HQPM 4 and HQPM 1. Thus, growing of HM 10 and planting in First week of February is the best option for higher yield and profit from spring maize in Haryana.

**Keywords:** Grain yield · Sowing date · Spring maize

### Introduction

Maize is one of the most important cereal crops next to wheat and rice in the world. Globally, it is known as queen of cereals because it has the highest genetic yield potential among the cereals. Worldwide, it was cultivated on 197.0 million hectares and recorded the production of 1,148 million tonnes of grains with an average grain yield of 5.82 t/ha (Anonymous, 2020a). In India, maize is grown on 9.9 million hectares with 30.0 million tonnes of production and average grain yield of 3.03 t/ha (Anonymous 2020b). Presently, India ranks 4th, 7th and 6th with respect to area, production and consumption of maize, respectively at the world level. Rice-wheat is major cropping system in Haryana, resistance in *Phalaris minor* in wheat crop and deteriorating of soil health are the major problems in paddy-wheat cropping system. Crop rotation/diversification is must to way out these serious problems. The spring maize is sown in the months of February and March after harvesting of potato and is harvested in June. The sudden rise in temperature in the month of February during the past few years has adversely affected wheat grain filling; this problem can be solved by replacing late sown wheat with the spring maize in Haryana. In Haryana area of maize in *kharif* season is about 6,200 ha and production is about 17,000 tonnes and with average productivity of 2.74 t/ha (Statistical abstract of Haryana 2019–20), but area in spring maize is increasing every year and is grown on around 10,000–12,000 hectares with productivity of nearly 4.5 t/ha. During spring season, the maize has very high yield potential and it may yield up to 10.0 t/ha. In Haryana, short duration paddy-potato-spring maize is a profitable cropping system as compared to conventional rice-wheat system and it may be useful in

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crop diversification by replacing rice-wheat system and overcoming the above said problems.

## Materials and methods

A field experiment on the effect of date of sowing on grain yield of spring maize was conducted at Regional Research Station, Karnal during the year 2015, 2016 and 2017 to work out the performance of seven maize hybrids (HQPM 1, HQPM 4, HQPM 5, HM 9, HM 10, HM 11 and HM 12) with five sowing dates (last week of January, first week of February, second week of February, third week of February and fourth week of February). The experiment was laid out in split-plot design with three replications. The soil of experimental site was clay loam in texture, alkaline (pH 8.3) with electrical conductivity of 0.4 dS/m, low in organic carbon (0.28 per cent) and available phosphorus (P) (7 kg/ha) and medium in available potash (K) (193 kg/ha). The grain yield (t/ha), days to 50 per cent tasseling, days to 50 per cent silking days to maturity and economics as returns over variable cost and B: C ratio were recorded in this experimental studies.

## Results and discussion

On three years mean basis, the highest grain yield was obtained when the crop was sown during first week of February (8.64 t/ha) followed by last week of January (8.27 t/ha), second week of February (8.09 t/ha), third week of February (7.34 t/ha) and the lowest in fourth week of February (6.60 t/ha). Dahmardeh (2012) observed that sowing dates affect the gain yield of spring maize, similar trend was observed for B: C ratio. The highest B:C ratio (1.9) was found when the crop was sown during first week of February followed by last week of January (1.8), second week of February (1.8), third week of February (1.6) and the lowest fourth week of February (1.5) (Table 2). Year-wise data also shows similar trend (Table 1). The hybrid HM 10 (8.29 t/ha) gave the highest grain yield followed by HM 11 (8.10 t/ha), HQPM 4 (7.98 t/ha) and HQPM 1 (7.85 t/ha). In other studies early sowing gave the highest yield of maize (Sherstha *et al.*, 2016). Similarly, Khanal *et al.*, (2019) observed that February 1 sown had higher yield than 2<sup>nd</sup> date of sowing i.e. February 12 as well as 3<sup>rd</sup> date of sowing i.e. February 23, so earlier planting in spring maize was the best than delayed sowing. The least grain yield was recorded in

**Table 1.** Effect of different sowing dates and hybrids on grain yield on spring maize during 2015-2017

| Treatment               | Grain yield (t/ha) |      |      | Mean |
|-------------------------|--------------------|------|------|------|
|                         | 2015               | 2016 | 2017 |      |
| <i>Sowing dates</i>     |                    |      |      |      |
| Last week of January    | 8.20               | 8.40 | 8.20 | 8.27 |
| First week of February  | 8.51               | 8.80 | 8.60 | 8.64 |
| Second week of February | 8.02               | 8.22 | 8.02 | 8.09 |
| Third week of February  | 7.41               | 7.61 | 7.00 | 7.34 |
| Fourth week of February | 7.01               | 7.20 | 5.60 | 6.60 |
| LSD (P=0.05)            | 0.26               | 0.25 | 0.40 | 0.30 |
| <i>Hybrids</i>          |                    |      |      |      |
| HQPM 1                  | 7.90               | 8.10 | 7.56 | 7.85 |
| HQPM 4                  | 8.01               | 8.21 | 7.71 | 7.98 |
| HQPM 5                  | 7.61               | 7.81 | 7.31 | 7.58 |
| HM 9                    | 7.62               | 7.82 | 7.32 | 7.59 |
| HM 10                   | 8.33               | 8.53 | 8.01 | 8.29 |
| HM 11                   | 8.15               | 8.33 | 7.83 | 8.10 |
| HM 12                   | 7.30               | 7.50 | 7.10 | 7.30 |
| LSD (P=0.05)            | 0.39               | 0.40 | 0.40 | 0.35 |

hybrid HM 12 (Table 2). Days to 50 per cent tasseling, 50 per cent silking and maturity were decreased with delayed sowing of spring maize crop. The crop duration reduced to 114.3 from 126.4 days when crop planted in fourth week of February compared to planting in last week of January. Among the hybrids tested days taken to 50 per cent tasseling was the highest in HQPM 5 (73.1) as compared to rest of the hybrids. Among hybrids tested, days to 50 per cent silking were delayed in HQPM 5 (76.4) as compared to all other hybrids. Among hybrids tested the maximum days to maturity were observed in HQPM 5 (123.0) as compare to rest of the hybrids (Table 2). Shrestha *et al.* (2018) observed that plant cultivar response differently with different planting date. Optimum planting date plays important role in optimum growth and thus helps in achieving higher yield for that crop season. Delayed planting greatly affects the growth, development and productivity of maize plants and lesser duration of crop leads to lower yield as duration is normally directly linked to higher crop productivity. It brings changes in weather parameters such as temperature, solar radiation, humidity during crop season that are responsible for changes in morphology, plant physiology and molecular level of plants. Thus, planting date has prime importance for crop production due to its variation in weather.

**Table 2.** Effect of different sowing dates and hybrids on grain yield, days to 50% tasseling, days to 50% silking, days to maturity and economics of different hybrids (mean of 2015-2017)

| Treatment               | Grain yield (t/ha) | Days to 50% tasseling | Days to 50% silking | Days to maturity (Nos.) | Return over variable cost (Rs./ha) | B:C |
|-------------------------|--------------------|-----------------------|---------------------|-------------------------|------------------------------------|-----|
| <i>Sowing dates</i>     |                    |                       |                     |                         |                                    |     |
| Last week of January    | 8.27               | 83.0                  | 86.0                | 126.4                   | 51,031                             | 1.8 |
| First week of February  | 8.64               | 79.5                  | 83.0                | 123.3                   | 56,269                             | 1.9 |
| Second week of February | 8.09               | 71.0                  | 72.3                | 120.2                   | 48,763                             | 1.8 |
| Third week of February  | 7.34               | 65.0                  | 67.4                | 117.2                   | 38,779                             | 1.6 |
| Fourth week of February | 6.60               | 58.1                  | 61.2                | 114.3                   | 30,028                             | 1.5 |
| LSD (P=0.05)            | 0.30               | 0.3                   | 0.3                 | 0.6                     | –                                  | –   |
| <i>Hybrids</i>          |                    |                       |                     |                         |                                    |     |
| HQPM 1                  | 78.5               | 72.3                  | 75.1                | 121.2                   | 45,383                             | 1.7 |
| HQPM 4                  | 79.8               | 72.4                  | 75.3                | 121.0                   | 47,320                             | 1.8 |
| HQPM 5                  | 75.8               | 73.1                  | 76.4                | 123.0                   | 42,013                             | 1.7 |
| HM 9                    | 75.9               | 70.1                  | 73.2                | 118.5                   | 42,065                             | 1.7 |
| HM 10                   | 82.9               | 72.0                  | 75.0                | 120.0                   | 51,600                             | 1.8 |
| HM 11                   | 81.0               | 72.4                  | 75.4                | 121.0                   | 49,150                             | 1.8 |
| HM 12                   | 73.0               | 68.3                  | 71.3                | 118.2                   | 37,288                             | 1.6 |
| LSD (P=0.05)            | 4.0                | 0.4                   | 0.4                 | 0.4                     | –                                  | –   |

## Conclusion

The highest grain yield (8.64 t/ha), returns over variable cost and B: C ratio was recorded in spring maize crop when it was sown during first week of February followed by last week of January (8.27 t/ha) and second week of February (8.09 t/ha) while these were the highest with HM 11 hybrid.

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