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# Maize Journal

(An International Journal of Maize Research and Related Industries)

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**Maize Technologists Association of India**
Pusa Campus, New Delhi 110 012

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1. To bring together all professionals from public and private organizations involved in maize research, extension, production, processing, value addition, seed production/ marketing, energy, socio-economic and environmental issues.
2. To facilitate linkages among maize stake holders to disseminate up-to-date and relevant technology/information needed for end through organizing conferences/symposia/ seminars/ meetings, *etc.*
3. To publish a multidisciplinary scientific journal of international standards exclusively devoted to the maize research named "Maize Journal"

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**(An International Journal of Maize and Related Industries)**

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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iii. **Crop Protection**
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REVIEW PAPER

# Banded leaf and sheath blight: threat to maize

**Ramandeep Kaur[1] · Ashmita[1] · Gurwinder Kaur[1] · Priti Sharma[1] · Yogesh Vikal[1] · Ramesh Kumar[2]**

**Abstract:** Banded leaf and sheath blight (BLSB) is a destructive disease of maize and it leads to 100 per cent yield losses. The causal organism of BLSB is *Rhizoctonia solani* and the organism grows under favourable humidity (88–90 per cent) and temperature (15–35$^{o}$C) conditions. The management practices used to control the disease spread are inadequate at this moment due to its broad host range. Therefore, the identification and exploitation of genetic resistance is an economical and sustainable option to control this disease. However, limited genetic variability poses a major challenge for resistance breeding against BLSB in maize. A few resistant sources have been identified and used for the identification of QTLs and candidate genes responsible for BLSB resistance. Here, we summarized the management practices, genetic resources, and different molecular approaches used to identify the QTLs and key candidate genes associated with BLSB resistance in maize.

**Keywords:** Maize · BLSB · Rhizoctonia solani · Resistance · Susceptible

## Introduction

Maize *(Zea mays* L.), is one of the most important crop among cereals and it belongs to the family *Poaceae*. It is also known as the 'Queen of Cereals' due to its high yield potential (Choudhri, 2019). It hailed from Mexico and Central America. Worldwide production of maize in 2019-20 was 1147.7 million metric tonnes (FAOSTAT, 2020). The nutritional importance of this crop in millions of people around the world is extensively recognized. It consists of roughly 72 per cent starch, 10 per cent protein, and 4 per cent fat (Ranum *et al*., 2014). Each part of maize has its industrial importance, be it the grain or the leaves, stalk, tassel, or cob, all are useful for making various products (Kumar *et al*., 2012; Singh *et al*., 2021). It is also famous by the name corn that is largely adaptive all over the world and is cultivated in tropical, subtropical, and temperate regions. It is among the most flexible and emerging cash crop having immense adaptabilities under varied climatic conditions (Kaul *et al*., 2017).

The reduction in the grain yield in maize is due to its susceptibility to various biotic and abiotic stresses. Losses due to the disease may vary over the year and location. There are more than 112 diseases that affect maize production throughout the world. Out of which, 65 diseases were reported in many places in India. Leaf spots, leaf blights, downy mildew, stalk rots, seed rots, and banded leaf and sheath blight are major diseases of maize crop (Saxena, 2002).

Among these, Banded leaf and sheath blight (BLSB) is one of the crucial diseases in maize also known as ear rot, banded sheath rot, sheath blight, banded sclerotial disease, and horizontal banded blight. The fungus responsible for causing BLSB is *Rhizoctonia solani*. It was reported for the first time in 1927 by Bertus in Sri Lanka and was called as 'sclerotial disease' (Sharma *et al*., 2002). In India, the incidence of this disease was first reported by Ullstrup in the *Tarai* region of UP (Now Uttarakhand). Among the states, Himachal Pradesh, Uttar Pradesh, Haryana, Punjab, Madhya Pradesh, Rajasthan, West Bengal, Meghalaya, Bihar, and Jharkhand experience the regular occurrence of this disease (Chaudhary *et al*., 2016).

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### *Economic losses*

The disease was earlier reported as a minor disease on maize (Payak and Renfro, 1966). The importance of the disease was only realized in the early 1970s when an epidemic occurred in warm and humid foothill areas in the Mandi district of Himachal Pradesh (Sharad Shroff, 2016). Lal *et al*. (1980) have estimated a loss in grain yield in ten cultivars ranging from 23.9 to 31.9 per cent, whereas Singh and Sharma (1976) estimated 10–40 per cent in other cultivars. Lal *et al.* (1985) had suggested that grain yield loss can go up to an extent of 90 per cent.

The BLSB disease results in premature death, stalk breakage, and ear rot. About 11–40 per cent losses were reported in 10 different varieties of maize and losses in grain yield were reported up to 97 per cent (Butchaih, 1977) while in another study a yield loss of up to 100 per cent was reported (Sudjono, 1995). In the USA, yearly disease loss estimates for corn production ranges from 2–15 per cent. The expected average economic loss due to corn diseases in the USA, Ontario and Canada, was the US $76.51 per acre from 2012 to 2015, with the total corn production being nearly 54 billion bushels. In Asia, loss estimates were around 12 per cent. Depending on the severity, grain yield loss varies between 11 and 40 per cent though, it can increase up to 100 per cent when the ear rot phase predominates and where the conditions are favourable for the pathogen. This alarming situation is further worsened by the lack of adequate level of resistance in maize germplasm.

### *Pathogen*

The pathogen is soilborne as it survives in the soil and on diseased crop debris in the form of sclerotia or mycelium (Figure 1). Sclerotia can survive for several years in the soil. The fungi proliferate by irrigation and by the motion of affected soil and debris. The disease starts from the first and second leaf sheath, moves towards the ears, and leads to ear rot. Near the ear, the mycelium of light brown colour and small round black coloured sclerotia were perceived (Singh and Shahi, 2012).

### *Disease cycle and climatic conditions*

The pathogen requires optimum temperature and humidity to grow and spread from diseased plant to healthy plant

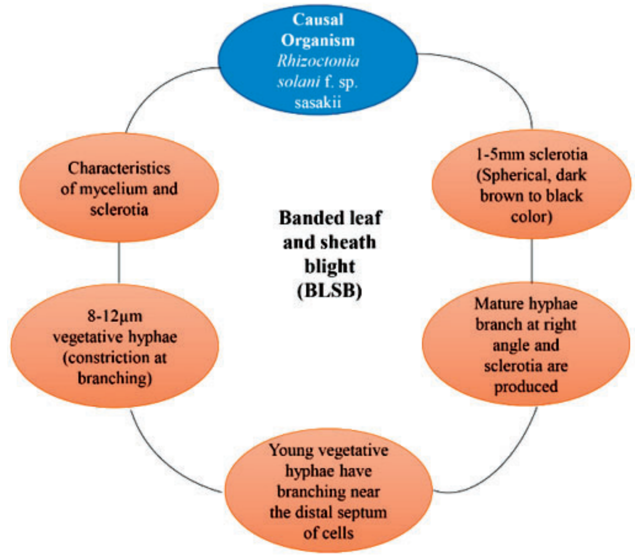


**Figure 1.** Characteristics of BLSB Disease

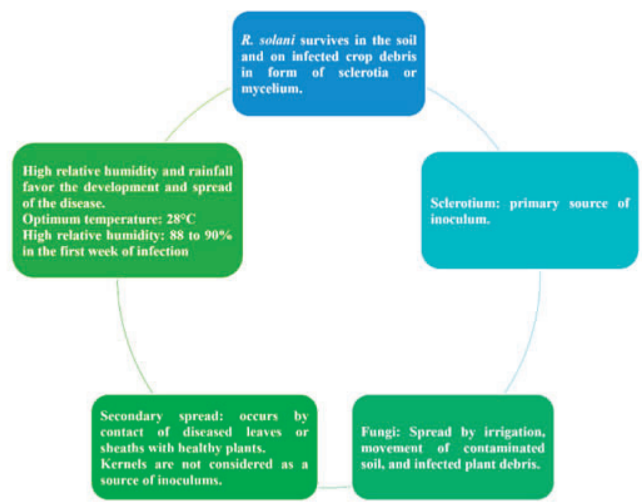


**Figure 2.** Disease cycle and climatic conditions

by a different mode of migration as explained in Figure 2. The favourable temperature for this disease to occur is 28°C and it also requires a relatively high humidity of about 88–90 per cent (Sharad Shroff, 2016). Grain yield losses can reach up to 100 per cent if the ear rot phase predominates. The pathogen propagates further when favourable conditions prevail (Madhavi *et al.*, 2011). The disease completely destroys plants as it affects photosynthetic rate initially and then yield at later stages. The symptoms are discussed in Figure 3.

### *Need to identify resistant sources*

The development of resistant hybrids with the help of classical plant breeding is further worsened due to the

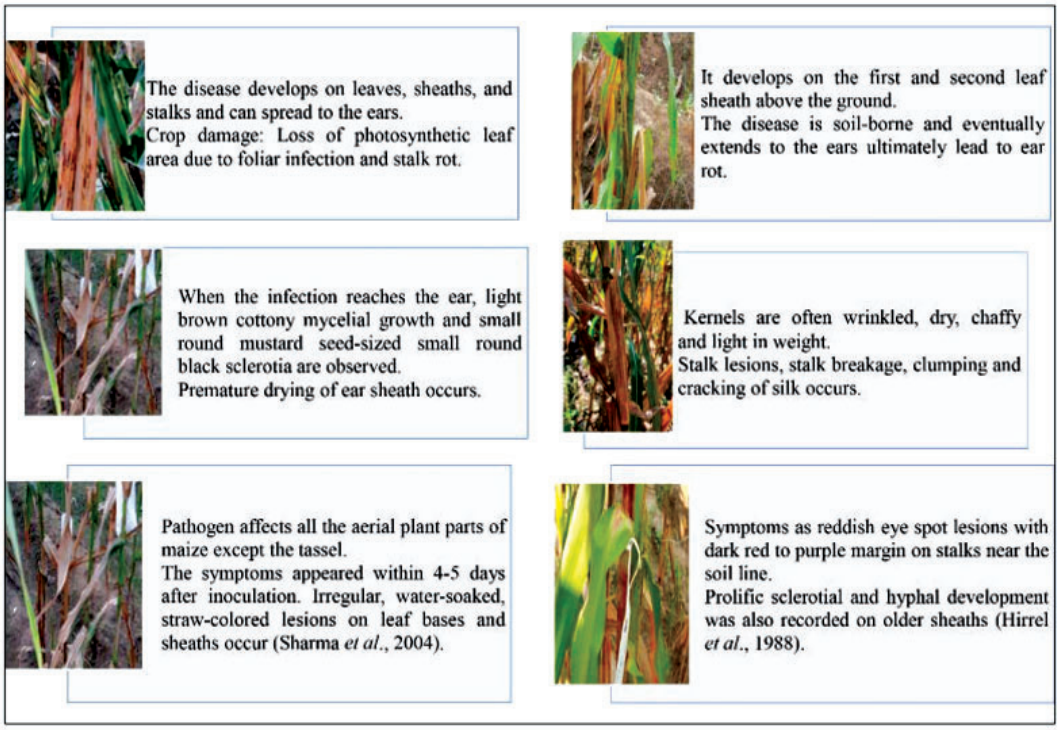


**Figure 3.** Symptoms of Banded leaf and sheath blight

limited availability of genetic resistance (Pan and Rush, 1997; Han *et al*., 2002). The wild grass named teosinte *(Z. mays* spp. *Parviglumis*) is postulated as an ancestor of maize. Teosinte can also cross-breed with modern maize varieties and as a result, fertile maize-teosinte hybrids can be formed that can reproduce naturally (Singh *et al*., 2017). It has been reported that wild species are very significant sources of genetic variability and these can be exploited in breeding programs. In teosinte, the genetic variability for different important agronomic traits for disease and insect resistance has been identified (Pasztor and Borsos, 1990; Srinivasan and Brewbaker, 1999). It is a valuable source of novel and diverse alleles and thus resistant to various fungal diseases. Various studies have shown that teosinte lines are resistant sources to *Ustilago maydis*, *Gray leaf spot*, and Southern leaf blight caused by *C. heterostrophus* (Chavan *et al*., 2014; Lennon *et al*., 2017). It consists of different loci resistant to northern corn leaf blight, corn leaf spot, *H. mayids*, corn smut diseases (Sahoo *et al.*, 2021), whereas different teosinte derived maize population has been screened and found to be resistant to *Fusarium*, downy mildew, and BLSB resistance (Maazou *et al*., 2017; Adhikari., 2019). Teosinte accession has been screened at PAU, Ludhiana for BLSB by artificial inoculation method and it was reported that teosinte accessions were moderately susceptible to BLSB (Garg *et al*., 2019).

*Approaches*

Different approaches have been used for the management of the disease like the use of toxins specific to fungi, validamycin, topsin, topsin-M, mancozeb, SAAF and carbendazim in different varying concentrations, using bioagents, *Trichoderma harzianum, T. viride, T. virens* and *Pseudomonas fluorescens* (Akhtar *et al*., 2010).

*Chemical control*

Chemical control involves two fungicides, Rhizolex and Thiophenate-M. These fungicides proved effective in preventing damage to the crop and also the yield increase was almost equal (Sharma and Rai, 1999). Seed and soil treatment with carbendazim decreased the disease severity index but caused an elevated increase in plant height and weight of the plant (both fresh and dry weight). Moreover, the seeds treated with *P. fluorescens* gave better results over others in all the parameters (Rani *et al*., 2013). Propiconazole was found to be effective when initial stages after planting but foliar sprays of Carbendazim was found to be ineffective against the disease (Saxena, 2002). Bavistin, Rhizolex, and Thiophenate M, these chemicals showed high control of BLSB under field conditions (Sharma *et al*., 2002).

*Mechanical controls*

Mechanical controls by removing the two lower leaves with leaf sheath (Madhavi *et al.*, 2018). But to date, no method had been of much use to control this disease.

*Biocontrol*

Biocontrol for controlling the disease involves the treatment of maize seeds with peat-based *Pseudomonas fluorescens* formulation. PF-1 and PF-6 isolates showed a great effect in forbidding the effect of a pathogen in causing the disease. Foliar sprays for controlling the disease can also be used (Sivakumar *et al.*, 2000). *Bacillus subtilis* BNt8 as a biocontrol agent against the disease was used and other different fungi species like the *Glomus* sp, *Acaulospora mellea*, *Trichoderma* sp and *Gliocladium* sp can also be used to reduce the attack of the disease-causing fungi (Djaenuddin *et al.*, 2017). The use of water agar before inoculating it with any of the biocontrol moiety is important to receive an increased control over the pathogen. The leaf sheath inoculation method also helps to reduce the disease growing on the plant (Pascual *et al.*, 2000).

Conventional control measures, through cultural practices or by the use of fungicides, are not enough to control the disease and are not an economically viable option. The development of resistant variety has been slowed down due to the unavailability of disease-resistant sources.

*Genetic resistance to BLSB*

Association mapping strategy has been used for finding the genes that are responsible for causing disease and identified loci are also connected to drought tolerance (Lin *et al*., 2013). About four QTLs for BLSB resistance have been located on chromosomes 6, 7 and 10, respectively (Pin *et al*., 2009). Different studies have been done to identify resistance sources to the disease. Around 20 genotypes from the CIMMYT-Asian Regional Maize Program (CIMMYT-ARMP), seven CM (Coordinated Maize) lines which were developed by various public sector institutions in India, and two LM (Ludhiana Maize) lines were screened. Different sources of resistance were found in different states of India (Garg *et al*., 2011). About 200 maize lines were screened for 10 of the major diseases occurring in the plant to find out the resistance sources at 9 different regions of the country. A total of these 18 lines showed resistance to BLSB. These lines can now be used for the mapping of genes resistant to particular diseases in maize plants (Hooda *et al*., 2012). About 11 QTLs were identified in $F_{2:4}$ and 8 QTLs in $F_{2:3}$ mapping population developed from nine inbred lines were identified as associated resistance to BLSB (Zhao *et al*., 2006; Garg *et al*., 2009). Various QTLs were identified for different morphological traits in the teosinte-derived maize population for BLSB resistance (Table 1) (Adhikari *et al*., 2021). GWAS approach was used to identify a novel allele ZmFBL41 (F-box protein) which is involved in maize resistance to *R. solani* by regulating lignin coding enzyme (Li *et al*., 2019).

*Use of omics*

Proteomic analysis reveals two biochemical markers β-1-3-glucanase and peroxidase for screening and resistance breeding against disease (Shamim *et al*., 2020). RNAseq analysis was performed to study the transcriptional changes in the B73 maize line in response to *R. Solani* and it was concluded that about 388 DEG (Differentially Expressed Genes) were involved in response to *R. solani* and these were considered as core immune genes in maize (Cao *et al*., 2021).

*Future prospective*

As many studies identified QTLs for BLSB resistance but still there is a lack and need to identify more loci for BLSB resistance such that resistant hybrids can be developed or identification make possible to transfer QTLs to susceptible lines through Marker Assisted Selection. Secondly, a pyramiding of QTLs can also be done to develop resistant lines. There is a need to collect more germplasm from CIMMYT and USDA to screen against BLSB. In addition, further research is required in the field of proteomics and transcriptomics as these will help to identify the regions conferring resistance to BLSB. More extensive work needed addressing the genetic inheritance will provide exciting novel insights. Refined genetic approaches and system analysis will be instrumental to get deeper insights into the mechanisms of BLSB resistance in maize.

**Table 1.** QTLs associated with different morphological traits for banded leaf and sheath blight resistance

| Population Type | Trait | Single marker analysis/ QTLS/ Chromosome No. | Marker interval | Reference |
|---|---|---|---|---|
| $F_{2:4}$ Elite inbreds R15 and 478 | BLSB | *qBLSB-1* | umc1245-dupssr12 | Zhao *et al*. (2006) |
| | | *qBLSB-2a* | umc1250-bnlg 1721 | |
| | | *qBLSB-2b* | bnlg1662-bnlg 1940 | |
| | | *qBLSB-2c* | bnlg1036-umc2150 | |
| | | *qBLSB-3* | phi19322-umc1010 | |
| | | *qBLSB-4* | umc2281-umc1662 | |
| | | *qBLSB-5* | phi10918-umc2164 | |
| | | *qBLSB-6a* | bnlg1600-umc1818 | |
| | | *qBLSB-6b* | bnlg1006-umc1723 | |
| | | *qBLSB-6c* | umc1859-bnlg1759 | |
| | | *qBLSB-10* | mmc0501-phi054 | |
| Teosinte and inbred line DI-103$BC_1F_5$ | Ear length, kernel rows per ear, & kernels per row | phi10918-linked QTL | - | Adhikari *et al*. (2021) |
| | Test weight, | umc1500-linked QTL | - | |
| | Ears per plant | umc2000-linked QTL | - | |
| | Plant height | Phi420701 QTL | - | |
| $F_{2:3}$ developed from 9 inbred lines (CA00106, CA00310, CM104, CM105, CM300, CM139, CM140, LM5 and LM6) | BLSB resistance | Chromosome 4 | (bnlg252-bnlg1621) | Garg *et al.* (2008) |
| | | Chromosome 8 | (umc2146-umc1172) | |
| | | Chromosome 9 | (phi108411-umc2346) | |
| | | Chromosome 2 | (umc2363-umc1622) | |
| | | Chromosome 3 | (umc2101-umc1892) | |
| | | Chromosome 6 | (umc1127) | |
| | | Chromosome 7 | (umc1066-bnlg1792 | |
| | | Chromosome 10 | (bnlg1518-bnlg1526) | |
| CML429 × DM9 $F_{2:3}$ | BLSB resistance, Disease incidence | (2 QTLs) on Chromosome 6 | bnlg107 and umc1796 | Chen *et al.* (2009) |
| | | (2 QTLs) on Chromosomes 7&10 | bnlg1161 and phi059 | |
| R 15 (resistant) × Ye 478 (susceptible) w $F_{2:4}$ | BLSB resistance | BLSB2-1 | umc1285-nc003 | Lin *et al.* (2008) |
| | | BLSB2-2 | umc2150-bnlg172 | |
| | | BLSB2-3 | bnlg1662-bnlg1606 | |
| | | BLSB2-4 | bnlg1606-bnlg1940 | |
| | | BLSB3-2 | umc1659-umc1052 | |
| | | BLSB6-1 | bnlg1538-umc181 | |
| | | BLSB6-2 | umc1818-umc1083 | |
| | | BLSB9-1 | umc1231-umc2343 | |
| | | BLSB7-1 | bnlg2132-umc1016 | |
| | | BLSB3-1 | bnlg1523-bnlg1447 | |
| | | BLSB7-2 | umc1125-umc1154 | |
| | | BLSB3-2 | umc1659-umc1052 | |
| | | BLSB4-1 | bnlg2162-umc1051 | |
| | | BLSB4-2 | umc2281-umc1662 | |
| | | BLSB4-3 | bnlg1621-umc1299 | |
| | | BLSB10-1 | phi118-umc1319 | |
| | | BLSB6-3 | umc1723-umc1014 | |
| | | BLSB10-1 | phi118-umc1319 | |
| | | BLSB10-2 | mmc0501-phi054 | |

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REVIEW PAPER

# Maize - an alternative cereal for crop diversification in changing climate

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**Abstract:** In India, the cereal consumption and hence, production patterns have changed with more rice and wheat and less coarse cereals such as millet, maize and sorghum, resulting in consistent decline in groundwater availability especially in the country's northern states, raising concerns for cereal production and self-sufficiency. *Kharif* maize, with less water requirement than paddy, has been recognized as a candidate crop in crop diversification plan to break predominant wheat- paddy status quo. Moreover, maize - wheat cropping system lead to about 10 per cent higher productivity of wheat than paddy-wheat crop rotation. But poor competitiveness of maize v/s paddy, price instability, yield gaps and instability of performance, less mechanized, rice-wheat oriented infrastructure, market and processing are major issues for a large scale cultivation of maize. Maize yield enhancement, which is a major driver for its economic viability and farmer's acceptance, need to be addressed on priority dovetail with mechanisation, nutritional enrichment and infrastructure development especially for specialty corns. Maize, being a multi-purpose crop and its strong emergence as industrial crop ensures its stable and perpetual demand. In India, as our major cereal consumption needs are met from domestic production, the country has an excellent opportunity for strengthening crops like maize as an alternate cereal with less water requirement, reduced environmental burden of agriculture and more nutrition.

**Keywords:** Alternate cereal · Accelerated breeding · Maize · Genetic enhancement · Productivity

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## Introduction

India is on the brink of a severe water crisis due to heavy-water extraction for irrigated agriculture and a weakening monsoon. Green revolution, though, led to self-sufficiency in cereal production but fuelled largely by depletion of freshwater resources for irrigation, nutrient pollution from injudicious fertilizer application and rising greenhouse gas emissions.

As vegetarian Indian diets generally derive a large fraction of nutrients from cereals, these mounting population and environmental concerns necessitate decisive assessment of the rice-wheat status quo of the Indian food system and need for alternate pathways toward healthier food baskets with less environmental burden (De Fries *et al*., 2015). While emphasizing the considerable urgency for improved compatibility between food security and environmental stewardship in India, Davis *et al*. (2018) mentioned that a national analysis of the potential nutritional and water use benefits of alternative cereals (viz., maize, millets, and sorghum) is still lacking for India. Endorsement of maize, a lower blue water footprint crop (maize has 49 mm $H_2O$ per year cumulative blue water requirement, whereas rice has 307 mm $H_2O$ per year), as a candidate to replace paddy during *Kharif* season will contribute in a profound way as three-fourth of Indian maize production is in the *Kharif* season which depends mainly on the south west monsoon (Kayatz *et al*., 2019). India's public distribution system is the best platform to begin/realise the transition to alternate cereals.

The impacts of climate volatility on cropping systems/ patterns and their productivity, and on severity and alteration of biotic and abiotic stresses, have also emerged as serious challenges for sustainable agriculture. The spread of desert locust, *Schistocerca gregaria*, in western parts of India in winter 2019-20 is the recent example of the threats due to climate volatility. There are many other examples of changes in incidence and severity of pests. The deadly polyphagous pest fall armyworm (FAW), *Spodoptera frugiperda*, with a strong preference for maize (*Zea mays* L.), has invaded India in May 2018. Since its first report from Karnataka, it has spread to almost all the states (except Himalayan region). Pink stem borer, *Sesamia inferens*, a key insect pest of paddy was earlier restricted to the peninsular India. Its incidence was reported in other crops also. In Punjab, it used to appear in paddy but not in wheat till recently. The predominance of paddy-wheat crop rotation may have hastened the incidence of some pests along with their climate change-aided biological and ecological adaptation to the conditions in new areas.

In this scenario, we need to shift our agriculture towards more resilience and sustainability to ensure national food and nutritional security and farmers' livelihood. Maize is an important alternative to paddy for crop diversification particularly in Northern India in the wake of depleting water resources, reduction in on farm diversity and environmental pollution due to paddy straw burning. Moreover, maize-wheat cropping system leads to 10–12 per cent higher productivity of wheat than paddy-wheat crop rotation. But poor competitiveness of maize crop in comparison to paddy in terms of low productivity, price instability, wide gaps in potential and farmer's field yields, less mechanization, rice-wheat oriented infrastructure, marketing and processing are major issues for a large scale cultivation of maize.

We, here, made efforts to highlight the researchable aspects to be strengthened to make maize as a commercially viable alternative cereal in prevailing Indian scenario.

*Maize – a crop of diversified utility*

Maize is queen of cereals, surpasses all other cereals and food crops in its ability to adapt to diverse agro-ecological niches and being cultivated from 58$^o$N to 55$^o$S latitude. In India, it is traditionally a *Kharif* season crop, but it is now cultivated in *Rabi* and spring seasons too. Being a $C_4$ plant, it has a competitive edge over $C_3$ cereals and has inherent potential to cope up upcoming challenges of drought, high temperature, and carbon dioxide limitation conditions. The grain feed industry is growing at a Cumulative Annual Growth Rate (CAGR) of 6–7 per cent globally and in India at 9 per cent with poultry, cattle, piggery and aqua feed sectors emerging as major growth drivers (Rakshit *et al.*, 2021). The Indian animal feed market was worth INR 400.5 billion in 2018. The market is further projected to reach INR 898.5 billion by 2024, growing at a CAGR of 14.3 per cent during 2019–2024 (India-animal-feed-market Report-20-21).

With the largest global livestock population, the dairy sector is playing a critical role in providing livelihood opportunities to millions of people, largely women, in rural sector of India. Since 2006, India has witnessed a major shift from vegetarian diets toward diets containing larger amounts of meat. Factors like urbanization, greater exposure to newer cultures, and increasing disposable incomes could be possible reasons for the shift.

Poultry is a highly vertically integrated industry in India and matches the efficiency levels of many western countries (Opportunities in poultry sector in India, 2020). Feed accounts for about 60 per cent of the maize consumption in India. The major driver of maize demand is poultry feed which accounts for 47 per cent (as the main source of energy in poultry feeds) while other livestock feed accounts for 13 per cent of total maize consumption (Rakshit *et al.*, 2021). Piggery is also becoming an important and providing employment opportunities to rural farmers with supplementary income to improve their living standards. The availability of feed plays an important role in transforming subsistence pig farming to a market-oriented meat business. As a fish feed, maize enables generative fish gain maturity very fast.

Maize also serves as an important source of green fodder and stover fodder amongst the non-legumes as its plant biomass is a good source of protein (11.4 per cent) and total digestible nutrients (66.2 per cent - which are highest among non-legume fodders). The proteins and total digestible nutrients, respectively, in pearl millet are 8.8 per cent and 58.2 per cent, sorghum 9.0 per cent and 55.0 per cent, Napier *bajra* 8.7 per cent and 59.3 per cent and guinea grass 10.8 per cent and 62.4 per cent. Moreover, maize fodder is soft and easy to cut, does not require much labour and has low machinery cost for processing.

Maize is also preferred for silage making over other fodders due to its softness, higher starch content (about 18 per cent) and no anti-nutritional components. Green fodder and silage which is cut at silking to milking stage is more nutritious, less fibrous and more digestible. Green maize is rich in protein and possesses sufficient quantities of soluble sugars required for proper ensiling. Cows fed on maize silage produced more milk and consumed more silage dry matter in than those fed sorghum silage (Lance *et al.*, 1964). Maize silage is used extensively for lactating dairy cows that require high-energy feed for maximum milk production (Marsalis *et al.*, 2010).

*Development of dual purpose maize*

For optimum milk production, around 40 kg of green fodder is required to feed per animal per day. However, a huge deficit exits between the demand and supply of green fodder in India. The specific research efforts to develop maize for dual purpose (grain + stover) and fodder (green maize harvested at dough stage) have not been rigorous. Given the prevalent fodder shortage in India, the quality stover from superior dual-purpose maize varieties can replace sorghum stover with substantial decrease in the feeding cost (Erenstein *et al.*, 2011). Therefore, there is an imperative need to develop maize varieties which can be used as green fodder and for silage making by harvesting at milking stage or as dual purpose for both grain and quality dry stover at harvest. In addition to serve as food, dual-purpose maize can meet both the poultry industry demand for grain and the demand for good quality stover to feed cattle. Focusing on maize breeding for fodder is very important for the promotion of dairy industry which is essentially required for diversification of agriculture.

PAU bred J1006, fodder purpose variety, and released in 1989 is a ruling variety even today along with an introduction, African Tall. In an evaluation by the National *Dairy Development Board* (*NDDB*) of PAU-bred hybrids, PMH-1 was reported as a very good for silage making.

*Maize grain productivity enhancement*

Undoubtedly, we have made significant progress in enhancing productivity which rose from 1 to 3 t/ha during 1965-66 to 2019-20 at the national level. To further enhance productivity, however, some traits need to be given greater emphasis. Development of longer maturity duration hybrids than those presently available, and incorporation of temperate germplasm from US and other regions having high genetic potential and rapid grain filling, need to be emphasized (Sandhu *et al.*, 2019). With combine harvesting, lodging resistant has gained importance for which strong root system, thick stem, low-medium ear placement and stay green traits are needed. The long duration hybrids should also have rapid dry down of ears/ grains. Development of hybrids well adapted to high plant density (HPD) is another key area to break the yield plateau (Dhillon *et al.*, 2019). Historical perspective of maize breeding and quantum jump in maize productivity in last centenary in tropical countries has well demonstrated that yield potential per plant has not increased over the years. With high plant density (HPD), productivity per unit area was increased. Tropical maize is inherently distinguished with tall stem, numerous long dropping leaves, bushy tassel, rapid post-silking leaf senescence and prone to lodging (Sandhu and Dhillon, 2021). There are a few reports of evaluation of hybrids under HPD (Dhillon *et al.*, 1978) but no systematic efforts have been made to develop the plant type through incorporating key traits- plant height, leaf angle, anthesis to silking interval (ASI), barrenness, kernel number and weight, tassel branching along with resistance to lodging in tropical maize. The selections for these traits over years have led to success of HPD in temperate maize. Therefore, there is need to re-design maize for amenability to HPD stress (Sandhu and Dhillon, 2021).

Temperature and moisture extremes, unseasonal rainfall, droughts, floods, cloud bursts, cyclonic storms etc. are becoming more frequent and consequently, the impacts of climate volatility on severity and alteration of biotic and abiotic stresses, have emerged as serious challenges for the scientific community (Dhillon *et al.*, 2019). Water logging (WL) is an emerging stress resulting from large and unpredictable fluctuations in weather patterns. The contingent flooding by excessive rainfall and inadequate drainage constraints the maize production in Asian tropics and other parts of the world (Singh *et al.*, 2017) and over 18 per cent of the total maize cultivating area in South and Southeast Asia alone are frequently affected by water logging problems. Maize is susceptible to water logging stress especially during germination and at the flowering stage when excessive rains lead to pollen wash and hampers seed set (Dhillon *et al.*, 2019). So, there is an urgent need for deeper insights into the mechanisms of crop adaptation to water logging. QTLs for adventitious root formation (Mano *et al.*, 2005); for leaf injury (Mano *et al.*, 2006); for aerenchyma formation under non-flooding conditions (Mano *et al.*, 2007); for root dry weight, root length, plant height, shoot dry weight, total dry weight and WL tolerance coefficient (Qiu *et al.*, 2007); for aerenchyma formation in non-flooding conditions (Mano *et al.*, 2008); for effective brace root tier number and total brace root tier number (Ku *et al.*, 2012); for seedling height, shoot dry weight, shoot fresh weight, root length, root dry weight and root fresh weight (Zhang *et al.*, 2013); for chlorophyll content, brace roots, root lodging and yield (Zaidi *et al.*, 2015) and QTLs for shoot fresh weight, root fresh weight, root length, root dry weight, shoot dry weight and seedling (Osman *et al.*, 2017) have been reported which have potential to improve WL tolerance in maize. The molecular markers available and strategies to utilize have been reviewed by Sandhu *et al.* (2021). The identified QTLs should be fine mapped and these tightly linked DNA markers can be further used for transferring WL tolerance traits from tolerant inbred to economically important inbred lines having desirable agronomical backgrounds through marker-assisted backcrossing (MABC) or development of WL tolerant superior line through marker-assisted recurrent selection (MARS). Most of the QTLs were identified for root biomass and adventitious root whereas other important root parameters like root surface area, root density, root volume has not been accounted for by researchers due to its labour-intensive phenotyping. The invent of high throughput phenotyping can add flavour to evaluation methods, which may enable the scientist to have a detailed understanding of WL tolerance especially for root traits.

Bridging yield gaps through agronomic interventions to realize potential yield and promotion of drip irrigation to harness the potential of spring maize are other major areas seeking attention. These should get priority as these efforts will have immediate impact. National maize productivity is 3.05 t/ha whereas experimental yields of improved hybrids hover around 6.5–7.5 t/ha. To make maize a commercially viable crop, this wide gap needs to be plugged in through large scale front-line demonstrations of the high yielding varieties, along with the improved production and protection technologies particularly during *Kharif* season. Some of the factors which also need to be emphasized are use of good quality seed, precision sowing, weed control, and taking care of the crop facing biotic/abiotic stress.

*Redesigning maize as a nutri-cereal*

Now, there is clarion call to re-orient our research strategies to nutritional security of masses. With its high content of carbohydrates, fats, proteins, some of the important vitamins and minerals, maize acquired a well-deserved reputation as a ‘poor man’s nutri-cereal (Prasanna *et al.*, 2020). Normal maize protein possesses low nutritional significance to humans because of very limited amounts of major amino acids, such as lysine (1.6–2.6 per cent) and tryptophan (0.2–0.6 per cent) (Chandran *et al.*, 2019), which is less than half of the recommended dose specified for human nutrition. Development of high yielding, high lysine and tryptophan maize, commonly known as Quality Protein Maize (QPM) and biofortified maize enriched in beta-carotene (provitamin A) holds significant promise. The bioavailability of Zn in maize grains is only 20 per cent in the human gut (Andersson *et al.*, 2017). The major impediment of low bioavailability of Zn has been the presence of phytic acid/phytate that constitutes nearly 75–80 per cent of the total phosphorus in maize grains (Raboy, 2001). Phytate being negatively charged has a strong tendency to chelate positively charged metal ions, such as Zn, thereby resulting in highly insoluble salts with poor bioavailability of the nutrient (Zhou and Erdman, 1995). Hence, bringing down the phytate in maize could be an important strategy for Zn biofortification. Further, the high carotene content of yellow grained maize is considered very useful in imparting yellow colour to egg yolk so is important in poultry industry. Poultry diets based on maize and soybean [*Glycine max* (L.) Merr.] require methionine supplementation because both grains have inadequate methionine concentrations. Feed supplementation with commercially produced methionine is used to alleviate these deficiencies but also increases poultry production costs (Waldroup *et al.*, 1981). Genetic approaches to increase the level of methionine in maize grain will provide a more nutritionally balanced feed source.

*Specialty maize*

There is a good possibility of diversifying maize cultivation by promoting specialty maize types, namely baby corn, pop corn, sweet corn and waxy corn. Baby corn is a very delicious, nutritious vegetable and has high export potential to further boost to its cultivation. Sweet corn is being utilised in hotels and big restaurants. A significant quantum of green biomass is available from specialty corn cultivation, which can efficiently be used as animal fodder. These are high value crops and therefore helpful in increasing farm income. In baby corn, prolificacy and male sterility are important traits. In pop corn, popping ratio is 20–25:1 which needs to be upgraded. Waxy maize having nearly 100 per cent amylopectin compared 75 per cent amylopectin and 25 per cent amylase in normal maize kernels, is gaining popularity as instant energy source. There is a significant export market for waxy maize in Europe and Asia.

Major constraint in the commercial cultivation of specialty maize types, like that in biofortified, is their poor agronomic performance as compared to normal corn. Thus, research efforts need to be strengthened for which resources need to be generated may be through Research-Industry interface. In addition, infrastructure facilities like small scale processing units, cold chains, storage facilities and strong linkage of farmers and industry are need to be addressed.

*New breeding approaches coupled with accelerated line development*

Breeding methods namely modified S1 recurrent selection, recurrent selfed plant mass selection and comprehensive recurrent selection have higher genetic gain per year compared to population improvement methods available earlier. Advances in next-generation sequencing technologies have taken the implementation of SNPs for genetic analysis to a new level. The use of marker assisted selection (MAS) in plant breeding programs has been accelerated since the advent of the genomics era. Rapid progress has been made in the developed world in rapid development of inbred line through doubled haploid (DH) technology using haploid inducer stocks. This approach shortens the time span required for the line development by 3-4 generations. This should be integrated as systematic and regular activity in commercial breeding program. The efforts on the identification of desirable genes and QTLs governing biotic and abiotic stresses and their funnelling into maize gene pool must be expanded. *Maize* stem borer, *Chilo partellus*, is already a serious pest and now FAW, *Spodoptera frugiperda*, with a strong preference for maize, has emerged as a very serious one. The increased incidence of banded leaf and sheath blight caused by *Rhizoctonia solani*, also needs an urgent attention. Heat and moisture stress tolerance are also key components for spring crop. Transfer of identified QTLS for these stresses in spring maize parental lines through MAS will expedite the breeding program.

The availability of new molecular tools has potential to accelerate introgression breeding and have enhanced interest in the identification and exploitation of desirable genes in landraces and wild progenitors. Teosintes are a valuable source for broadening and enriching the maize gene pool. The close genetic relationship between the two subspecies, namely maize and annual teosinte (*Zea mays* sp. *mexicana*) has greatly stimulated interest in this regard. *Zea mays* sp. *mexicana* has excellent potential to produce high biomass and can expand the fodder production potential of maize. Research is going on at PAU to use the genes for resistance to leaf and sheath blight in this subspecies (Garg *et al*., 2019). Other teosintes, namely *Zea perennis* and *Zea diploperennis* showed resistance to biotic stresses and *Zea luxurians* and *Zea nicaraguensis* are adapted to frequent rainfalls and possess unique flooding resistance traits. The annual teosinte, *Zea mays* sp. *parviglumis*, the closest ancestor of modern maize, has good tillering and re-growth ability and hence, can serve as an excellent source to fodder maize breeders to develop multi-tiller high biomass yielding cultivars (Choudhary *et al*., 2020).

## Conclusion

To realise the demand driven potential of maize as 'the future cereal crop', a dedicated effort by researchers and holistic support by policy makers is an inevitability. Under Punjab conditions, with existing crop productivity of near 4 t/ha, there is a loss of about Rs. 17,500 Rs/ha in economic returns to maize growers in comparison to paddy. Such profit gaps has to be plugged in for initial promotion of large scale maize cultivation. It is necessary to have coupling of strategies and interventions around technological innovations, policy support, marketing linkages, supporting infrastructure and strong public private partnership for economic viability of maize crop. Improvement in the maize value chain across its various stages will be extremely crucial for making Indian maize competitive in international market both in terms of quality and prices.

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RESEARCH PAPER

# Post-emergence herbicides enhance yield and profitability in *Kharif* maize (*Zea mays* L.)

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**Abstract:** A field research on 'Evaluation of post-emergence herbicides in maize (*Zea mays* L.)' was carried out at the research farm of ICAR-Indian Institute of Maize Research, Pusa Campus, IARI, New Delhi, India during *Kharif* season of 2017. It was found that the early post emergence tank-mix application of the pre-emergence (PE) and post-emergence (POE) herbicides and their sequential application increased crop growth parameters *viz.*, plant population, plant height, dry matter accumulation (DMA), leaf area index (LAI) crop growth rate (CGR) and relative growth rate (RGR), yield attributes (cobs/ha, cob length, grains/row and grains/cob) in *Kharif* maize. However, no significant effects of different weed management practices were recorded on cob girth and grain rows/cob of the crop in our study. The application of the pre-emergence (PE) and post-emergence (POE) herbicides increased maize grain yield to the tune of 9.2 to 14.6 and 17.5 to 20 per cent over recommended practice (atrazine *fb* hand weeding), respectively. The increase in net returns over weedy check ranged from Rs. 15.12 $\times 10^3$ ($T_5$) to 56.19 $\times 10^3$ ($T_{10}$)/ha and gave 3-5 times more net returns in different herbicides applied treatments. The range of the added cost in weed management with post-emergence herbicide was Rs 5.02 $\times 10^3$ to 6.55 $\times 10^3$/ha over weedy check. Significantly the highest net returns (Rs. 56.19 $\times 10^3$/ha), additional net returns (Rs. 53.09 $\times 10^3$/ha) and benefit-cost (BC) ratio (1.59) in *Kharif* maize obtained by application of 75% recommended dose of atrazine as pre-emergence followed by topramezone (25.2 g/ha) at 25 DAS which was at par with the application of 75% atrazine as pre-emergence followed by at tembotrione (120 g/ha) 25 days after sowing (DAS) and with early post-emergence tank-mix application of these combinations.

**Keywords:** Crop growth · Crop yield · Maize · Net returns · Post-emergence herbicide

## Introduction

Maize is the third most important crop of India after rice and wheat that occupied 9.6 million ha area with average productivity of 3.0 tonnes/ha compared to the world average of 5.8 tonnes/ha. In India, maize is primarily cultivated during the *Kharif* season where weed is the most important yield-limiting factor. Maize is infested by a wide range of weed flora, viz. *Panicum spp.*, *Echinochloa colona*, *Cyperus rotundus*, *Commelina benghalensis and Trianthema portulacastrum* dominate during early stages of the crop growth whereas *Dactyloctenium aegyptium* toward the tasseling and maturity of the crop (Saini and Angiras, 1998). Even with a light infestation of weeds under ideal situation the weeds should be controlled throughout the crop-growing season for getting higher yield. However, the most critical period for crop weed competition are first six weeks after planting of crop because of initial slow growth and wider row spacing of maize, coupled with congenial weather conditions allow luxuriant weed growth which may reduce yield by 28–100 per cent (Pandey *et al.*, 1999; Dass *et al.*, 2012). During this critical period, weeding is

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essentially required by chemical or non-chemical means. Weeding by hands (labour) and mechanical means are expensive and many a times timely operations are not possible due to continuous rains in monsoon season. Research and development in herbicide technology has opened up new possibilities for chemical weed management practices.

There are good pre-emergence herbicidal options like, atrazine and pendimethalin are available in maize (Singh *et al*., 2015) but the availability of post-emergence herbicides in need of the maize cultivation in south and central Asia in view of escalating labour prices. Topramezone and tembotrione are the selective, post-emergence herbicides that have been recently introduced for use in maize. These HPPD inhibiting herbicides are most effective in newly developing tissues that emerge bleached, because of failure to properly assemble photosynthetic units and thus they control weeds (Schonhammer *et al.*, 2006). The herbicide like tembotrione (Singh *et al*., 2012) and halosulfuron along with 2,4-D are most common post-emergence herbicide used in maize worldwide (Kumar *et al*., 2015). These provide an easy option for herbicidal-based weed management in maize especially in later seasons as the critical period of crop-weed completion extends up to 50 days whereas pre-emergence herbicides are effective up to 25–30 days. Thus, for enhancing yield and profitability in maize cultivation of India as well to reduce drudgery in crop cultivation identification of suitable post-emergence herbicide will be boon for the farmers. Thus, a study was conducted to explore the possibilities of the good option of herbicidal-based weed management in maize to improve crop growth and yield while simultaneously controlling weeds.

## Materials and methods

A field experiment was conducted during the *Kharif* season of 2017 at ICAR-Indian Institute of Maize Research, Pusa Campus, New Delhi. The experimental farm used in the present study was under maize-wheat cropping system for last five years. The study site (New Delhi) is situated at 28$^o$ 40’N latitude, 77$^o$ 11’E longitude and at an altitude 228.6 m above mean sea level. The climate of the area is semi-arid, characterized by hot summer and severe cold winter. The season that received 565 mm rainfall and mean maximum and minimum temperature of 33.8$^o$C and 23.3$^o$C, respectively. However, as effective rainfall calculated by CROPWAT-FAO was only 394 mm and long dry spell and hence two irrigations were applied for successful crop cycle. The soil of experimental field was sandy loam in texture and alkaline in nature (pH 7.6), EC (dS/m at 25°C) was 0.38, low in organic carbon (0.42 per cent) and available nitrogen (236.8±23.5 kg/ha), and medium in available phosphorus (14.8±1.5 kg/ha) and potash (232.4±15.2 kg/ha). The experiment was laid out in a randomized complete block design with three replications and twelve treatments. The treatments tested were $T_1$: Weedy check, $T_2$: Weed free check, $T_3$: Atrazine 1000 g/ha (PE) *fb* hand weeding at 25 DAS, $T_4$: Topramezone 30 g/ha at 25 DAS, $T_5$: Halosulfuron 75 g/ha at 25 DAS , $T_6$: Tembotrione 150 g/ha at 25 days after sowing (DAS), $T_7$: Topramezone 25.2 g/ha + Atrazine 750 g/ha at 15 DAS , $T_8$: Halosulfuron 67.5 g/ha + Atrazine 750 g/ha at 15 DAS, $T_9$: Tembotrione 120 g/ha + Atrazine 750 g/ha at 15 DAS, $T_{10}$: Atrazine 750 g/ha (PE) *fb* Topramezone 25.2 g/ha at 25 DAS, $T_{11}$: Atrazine 750 g/ha (PE) *fb* Halosulfuron 67.5 g/ha at 25 DAS, $T_{12}$: Atrazine 750 g/ha (PE) *fb* Tembotrione 120 g/ha at 25 DAS. The single cross hybrid maize cv. PMH 1 seeds were dibbled on the ridges spaced at 0.70 meters at 0.20 meters spacing using 20 kg seed/ha. The net plot was 12.6 m$^2$. The herbicide doses were calculated as per the treatments and applied as aqueous spray @ 400 litres/ha water using knapsack sprayer fitted with a flat-fan nozzle. Hand weeding was done to maintain the weed free plot at 15 DAS, 30 DAS and 50 DAS with the help of *khurpi*. A hand weeding was also done in $T_3$ (as standard check) at 25 DAS. All the nutrients and plant protection practices were followed as per the recommendation. The crop growth observations of plant population, plant height, leaf area, CGR, RGR, and DMA at different stages of maize crop were taken following standard protocols to know about periodically effect of weeds on the performance of maize crop growth. The crop yield attributes and yields of crop were recorded using standard methods in all experimental plots. The economics for the cost of cultivation, gross returns, net returns, benefit cost ratio and net returns/rupee invested were worked out on the basis of prevailing market rates of the inputs and minimum wages of the labour announced as per the government in the region. Analysis of variance (ANOVA) was done to determine treatment effects by using SAS 9.3. The least significant difference (LSD) test was used as a post hoc mean separation test ($P < 0.05$).

## Results and discussion

*Crop growth performance*

The initial plant stands in maize (after thinning and gap filling operation) was statistically similar in all the treatments but the final plant stand (at harvest) was statistically superior in all the weed management treatments compared to weedy check. At the harvest, the maximum number of plants was observed with $T_2$ (Weed free check) that were statistically similar with $T_3$ (atrazine 1000 g/ha (PE) *fb* hand weeding at 25 DAS) and $T_7$ (Topramezone 25.2 g/ha + atrazine 750 g/ha at 15 DAS) treatments. The reasons for lower plant population in these treatments could be initially the maize and weeds during *Kharif* season grows faster as most of the *Kharif* season weeds are ephemeral in nature and completes life cycle in the very short span of time.

The significantly smallest plants observed in control plots (weedy check) at all the growth stages compared to weed management treatments. At 30 DAS, the tallest plant was rerecorded with $T_2$ (weed free check) which was statistically at par with $T_3$ (standard check) and combine/sequential application of the herbicides with $T_9$ and $T_{12}$. At 60 and 90 DAS and the highest plant height was recorded with standard check that was statistically similar with $T_2$ (weed free check) and tank-mix application of post-emergence herbicide *viz*., tembotrione (120 g/ha) or topramezone (25.2 g/ha) at 15 DAS or as sequential application at 25 DAS with 75% recommended dose of the atrazine in $T_7$, $T_9$, $T_{10}$ and $T_{12}$. Our findings of enhancement in the growth parameters of maize with lesser weed competition corroborates with Kumar *et al.* (2013). However, Soltani *et al.* (2007) and Thomas *et al*. (2010) reported that plant height did not influence with doses of topramezone which indicates the specificity of this herbicide to maize having no phytotoxic effect. This could probably be due to broad-spectrum weed control which resulted in efficient utilization of resources and more biomass accumulation. Almost similar finding were reported by Subramanyam *et al.* (2007).

The highest LAI (Table 2) was recorded under weed free check treatment, which was at par with $T_{10}$ and the other weed management treatments ($T_3$, $T_7$ and $T_{12}$). Alike plant height, significantly lower LAI was recorded in $T_1$ (weedy check treatment) at all growth stages. However, at 90 DAS, the LAI was statistically similar in all the treatments, which could be due to lesser weed population

**Table 1.** Effect of post-emergence herbicides application on the plant population and plant height of *Kharif* maize

| Treatment | Plant population (× $10^3$ plants/ha) | | Plant height (cm) | | |
|---|---|---|---|---|---|
| | Initial | At harvest | 30 DAS | 60 DAS | 90 DAS |
| $T_1$ | 71.40 | $54.1^{f}$ | $44.56^{g}$ | $131.44^{f}$ | $141.96^{f}$ |
| $T_2$ | 71.51 | $70.5^{a}$ | $78.33^{a}$ | $172.00^{ab}$ | $185.76^{ab}$ |
| $T_3$ | 71.45 | $69.2^{ab}$ | $69.33^{abc}$ | $173.56^{a}$ | $187.44^{a}$ |
| $T_4$ | 71.53 | $59.93^{d}$ | $54.78^{ef}$ | $159.22^{bcde}$ | $171.96^{bcde}$ |
| $T_5$ | 71.19 | $58.19^{e}$ | $57.33^{def}$ | $149.89^{e}$ | $161.88^{e}$ |
| $T_6$ | 71.22 | $60.56^{d}$ | $51.90^{fg}$ | $168.78^{abc}$ | $182.28^{abc}$ |
| $T_7$ | 71.52 | $69.19^{ab}$ | $66.46^{bcd}$ | $161.78^{abcde}$ | $174.72^{abcde}$ |
| $T_8$ | 71.49 | $67.92^{bc}$ | $60.22^{cdef}$ | $155.67^{cde}$ | $168.12^{cde}$ |
| $T_9$ | 71.11 | $68.98^{b}$ | $75.94^{ab}$ | $162.89^{abcde}$ | $175.92^{abcde}$ |
| $T_{10}$ | 71.28 | $68.88^{b}$ | $66.67^{bcd}$ | $167.56^{abcd}$ | $180.96^{abcd}$ |
| $T_{11}$ | 71.59 | $67.26^{c}$ | $63.56^{cde}$ | $154.11^{de}$ | $166.44^{de}$ |
| $T_{12}$ | 71.37 | $68.81^{b}$ | $77.56^{a}$ | $173.89^{a}$ | $187.80^{a}$ |
| p-Value | 0.537 | <.0001 | <.0001 | <.0001 | <.0001 |
| LSD (P=0.05) | NS | 1.45 | 9.79 | 13.79 | 14.89 |

***Note**:* $T_1$: Weedy check, $T_2$: Weed free check, $T_3$: Atrazine 1000 g/ha (PE) *fb* hand weeding at 25 DAS, $T_4$: Topramezone 30 g/ha at 25 DAS, $T_5$: Halosulfuron 75 g/ha at 25 DAS , $T_6$: Tembotrione 150 g/ha at 25 DAS, $T_7$: Topramezone 25.2 g/ha + atrazine 750 g/ha at 15 DAS , $T_8$: Halosulfuron 67.5 g/ha + atrazine 750 g/ha at 15DAS, $T_9$: Tembotrione 120 g/ha + atrazine 750 g/ha at 15 DAS, $T_{10}$: Atrazine 750 g/ha (PE) *fb* topramezone 25.2 g/ha at 25 DAS, $T_{11}$: Atrazine 750 g/ha (PE) *fb* halosulfuron 67.5 g/ha at 25 DAS, $T_{12}$: Atrazine 750 g/ha (PE) *fb* tembotrione 120 g/ha at 25 DAS. Means followed by a similar lower case letter within a column are not significantly different according to least significant difference test (P=0.05).

and biomass accumulation in these treatments at various crop growth stages. The dry matter accumulation (Table 2 & Figure 1) was recorded with $T_{10,}$ $T_{11}$ and $T_{12}$, which was on par with $T_2$ and $T_8$ at 30 DAS. However, at 60 and 90 DAS it was similar in all weed management treatments, it might be due to more resource availability in absence of the intense crop-weed competition the higher crop growth and dry matter accumulation was recorded in the better weed control plots (Subramanyam, 1998).

The highest CGR and RGR (Table 3) at 0-30 DAS was recorded in herbicides application treatments with $T_{10}$, which was statistically on par with $T_8$, $T_{11}$ and $T_{12}$ and it was at par with all weed management treatments at 30-60 and 60-90 DAS. At 30-60 DAS the highest RGR was in $T_{12}$ (atrazine *fb* tembotrione). At 60-90 DAS, RGR was statistically similar. In all the cases, the CGR and RGR were lowest in control treatments where no application of herbicides was done. It indicates that the weed management is very essential for the higher growth rate of maize and can cause significant reduction in biomass accumulation in maize if weed management is ignored in its production. The similar findings of the enhanced growth with lesser weed completion were also observed by Sinha *et al.* (2001).

**Table 2.** Effect of post-emergence herbicides application on leaf area index and dry matter accumulation of *Kharif* maize

| Treatment | Leaf area index (LAI) | | | Dry matter accumulation (g/plant) | | |
|---|---|---|---|---|---|---|
| | 30 DAS | 60 DAS | 90 DAS | 30 DAS | 60 DAS | 90 DAS |
| $T_1$ | 1.65[g] | 3.10[f] | 2.23 | 20.00[e] | 39.33[b] | 90.07[b] |
| $T_2$ | 2.80[a] | 5.35[a] | 3.04 | 39.67[ab] | 98.33[a] | 225.18[a] |
| $T_3$ | 2.46[bc] | 4.84[abc] | 2.37 | 37.67[abc] | 91.00[a] | 208.39[a] |
| $T_4$ | 2.06[ef] | 3.79[e] | 2.45 | 31.67[bcd] | 94.67[a] | 216.79[a] |
| $T_5$ | 1.93[fg] | 3.83[e] | 2.47 | 24.67[de] | 89.67[a] | 205.34[a] |
| $T_6$ | 2.20[cdef] | 3.95[de] | 2.53 | 30.67[cd] | 96.00[a] | 219.84[a] |
| $T_7$ | 2.42[bcd] | 4.97[abc] | 2.68 | 36.67[abc] | 95.00[a] | 217.55[a] |
| $T_8$ | 2.12[def] | 4.45[bcde] | 2.88 | 40.67[a] | 90.67[a] | 207.63[a] |
| $T_9$ | 2.28[cde] | 4.52[bcd] | 2.25 | 35.67[abc] | 88.33[a] | 202.28[a] |
| $T_{10}$ | 2.37[cde] | 5.03[ab] | 2.85 | 41.33[a] | 95.67[a] | 219.08[a] |
| $T_{11}$ | 2.35[cde] | 4.36[cde] | 2.67 | 40.67[a] | 96.67[a] | 221.37[a] |
| $T_{12}$ | 2.73[ab] | 4.97[abc] | 2.90 | 41.00[a] | 97.00[a] | 222.13[a] |
| p-Value | <.0001 | <.0001 | 0.1236 | 0.0001 | <.0001 | <.0001 |
| LSD (P=0.05) | 0.325 | 0.661 | NS | 8.17 | 17.18 | 39.35 |

***Note:*** $T_1$: Weedy check, $T_2$: Weed free check, $T_3$: Atrazine 1000 g/ha (PE) *fb* hand weeding at 25 DAS, $T_4$: Topramezone 30 g/ha at 25 DAS, $T_5$: Halosulfuron 75 g/ha at 25 DAS , $T_6$: Tembotrione 150 g/ha at 25 DAS, $T_7$: Topramezone 25.2 g/ha + atrazine 750 g/ha at 15 DAS , $T_8$: Halosulfuron 67.5 g/ha + atrazine 750 g/ha at 15DAS, $T_9$: Tembotrione 120 g/ha + atrazine 750 g/ha at 15 DAS, $T_{10}$: Atrazine 750 g/ha (PE) *fb* topramezone 25.2 g/ha at 25 DAS, $T_{11}$: Atrazine 750 g/ha (PE) *fb* halosulfuron 67.5 g/ha at 25 DAS, $T_{12}$: Atrazine 750 g/ha (PE) *fb* tembotrione 120 g/ha at 25 DAS. Means followed by a similar lowercase letter within a column are not significantly different according to least significant difference test (P=0.05).

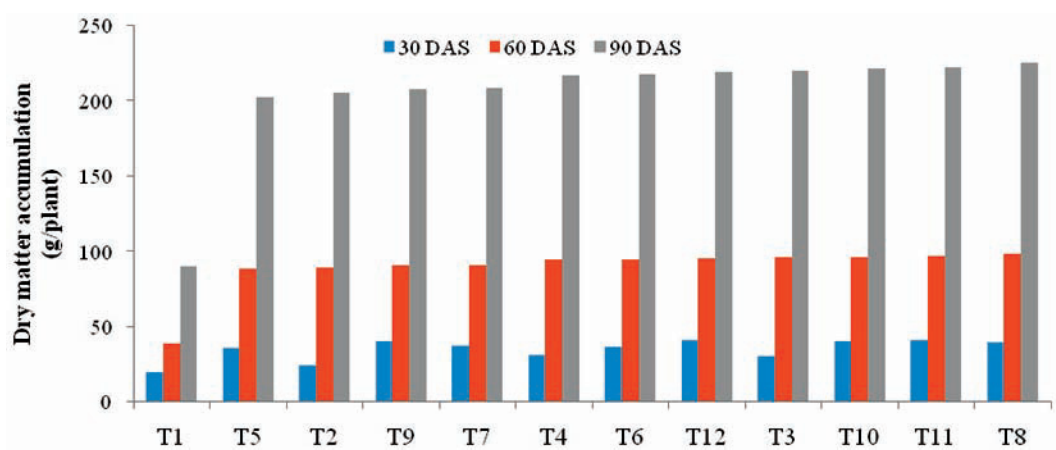



**Figure 1.** Effect of post-emergence herbicide application on dry matter accumulation in *Kharif* maize at various crop stages.

**Table 3.** Effect of post-emergence herbicides application 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 |
| $T_1$ | 0.67[e] | 0.64[b] | 1.69[b] | 43.32[e] | 53.08[b] | 65.07[b] |
| $T_2$ | 1.32[ab] | 1.96[a] | 4.23[a] | 53.20[ab] | 66.41[a] | 78.40[a] |
| $T_3$ | 1.26[abc] | 1.78[a] | 3.91[a] | 52.33[abc] | 65.14[a] | 77.14[a] |
| $T_4$ | 1.06[bcd] | 2.10[a] | 4.07[a] | 49.96[bc] | 65.86[a] | 77.85[a] |
| $T_5$ | 0.82[de] | 2.17[a] | 3.86[a] | 46.10[de] | 65.06[a] | 77.05[a] |
| $T_6$ | 1.02[cd] | 2.18[a] | 4.13[a] | 49.47[cd] | 66.04[a] | 78.03[a] |
| $T_7$ | 1.22[abc] | 1.94[a] | 4.09[a] | 51.99[abc] | 65.89[a] | 77.88[a] |
| $T_8$ | 1.36[a] | 1.67[a] | 3.90[a] | 53.63[a] | 65.15[a] | 77.14[a] |
| $T_9$ | 1.19[abc] | 1.76[a] | 3.80[a] | 51.64[abc] | 64.74[a] | 76.74[a] |
| $T_{10}$ | 1.38[a] | 1.81[a] | 4.11[a] | 53.85[a] | 65.93[a] | 77.93[a] |
| $T_{11}$ | 1.36[a] | 1.87[a] | 4.16[a] | 53.56[a] | 66.13[a] | 78.13[a] |
| $T_{12}$ | 1.37[a] | 1.87[a] | 4.17[a] | 53.73[a] | 66.15[a] | 78.15[a] |
| p-Value | 0.0001 | 0.0021 | <.0001 | <.0001 | <.0001 | <.0001 |
| LSD (P=0.05) | 0.27 | 0.58 | 0.74 | 3.53 | 2.82 | 2.82 |

***Note:*** $T_1$: Weedy check, $T_2$: Weed free check, $T_3$: Atrazine 1000 g/ha (PE) *fb* hand weeding at 25 DAS, $T_4$: Topramezone 30 g/ha at 25 DAS, $T_5$: Halosulfuron 75 g/ha at 25 DAS , $T_6$: Tembotrione 150 g/ha at 25 DAS, $T_7$: Topramezone 25.2 g/ha + atrazine 750 g/ha at 15 DAS ,$T_8$: Halosulfuron 67.5 g/ha + atrazine 750 g/ha at 15DAS, $T_9$: Tembotrione 120 g/ha + atrazine 750 g/ha at 15 DAS, $T_{10}$: Atrazine 750 g/ha (PE) *fb* topramezone 25.2 g/ha at 25 DAS, $T_{11}$: Atrazine 750 g/ha (PE) *fb* halosulfuron 67.5 g/ha at 25 DAS, $T_{12}$: Atrazine 750 g/ha (PE) *fb* tembotrione 120 g/ha at 25 DAS. Means followed by a similar lower case letter within a column are not significantly different according to least significant difference test (P=0.05).

*Yield attributes*

The data on yield attributes are presented in Table 4. The significantly higher cobs were recorded in $T_2$ while the highest bareness (39.0 per cent) was registered in control (weedy check). However, in other treatments of weed management, the bareness ranged from 2.03 to 6.41 per cent only. The cobs/ha in standard checks i.e. atrazine 1000 g/ha (PE) *fb* hand weeding at 25 DAS were statistically similar with application of post-emergence herbicide treatments ($T_7$, $T_9$, $T_{10}$ and $T_{12}$). Similarly, the increase in the cob length in the range of 30.7 to 42.3 per cent in all weed management treatments over weedy check might be due to higher LAI and the dry matter accumulation in absence of crop-weed also reported by Walia *et al*. (2007). The grain rows/cob was found non-significant but weed management practices influenced grains per row and grains per cob in maize. The cob girth could be more genetically linked and thus was not changed with the various management practices could be the probable reason for the statistically similar in weedy check and other treatments (Sanodiya *et al*., 2013). The highest grains/row and grains/cob were registered with $T_2$ which was statistically similar to $T_{12}$ and other post-emergence herbicide application treatments. Significantly the highest 1000-grains weight in maize was recorded with $T_{12}$. The more weed competition might also lead to decreased seed size and thus decreased 1000-grain weight while it increased the kernel weight in more resource available plots with the lesser crop-weed competition. The similar observation reported by Gopinath and Kundu (2008).

*Crop yield*

The cob yield of maize increased significantly with the weed-free check ($T_2$), which was on par with $T_7$, $T_9$, $T_{10}$ and $T_{12}$ in the range of 33.2 ($T_5$) to 135.2 ($T_{10}$) per cent over weedy check. Similarly, significantly the highest stover yield with application of atrazine (PE) *fb* topramezone (POE) at 25 DAS ($T_{10}$), which was on par with $T_2$, $T_7$, $T_9$, $T_{11}$ and $T_{12}$. Increased in the range from 16.2 ($T_5$) to 62.1 ($T_{10}$) per cent over weedy check. The grain yield as an economic produce in maize increased significantly with weed free check ($T_2$), which was on par with $T_{10}$ and $T_{12}$. The increase in the grain and biological yield in *Kharif* maize over weedy check were 63 to 208 and 22.1 ($T_5$) to 87.2 ($T_{10}$) per cent, respectively (Table 5 & Figure 2), which were almost more than

**Table 4.** Effect of post-emergence herbicides application on yield attributes of *Kharif* maize

| Treatment | Cobs ('000/ha) | Bareness (%) | Cob length (cm) | Cob girth (cm) | Grain rows per cob | Grains per row | Grains per cob | 1000–grains weight (g) |
|---|---|---|---|---|---|---|---|---|
| $T_1$ | 32.96$^{e}$ | 39.04$^{a}$ | 14.83$^{b}$ | 13.47 | 13.56 | 26.00$^{f}$ | 352.00$^{e}$ | 226.67$^{d}$ |
| $T_2$ | 69.94$^{a}$ | 0.76$^{e}$ | 18.73$^{a}$ | 13.60 | 14.22 | 35.89$^{a}$ | 510.30$^{a}$ | 278.33$^{b}$ |
| $T_3$ | 67.52$^{b}$ | 2.41$^{cde}$ | 20.81$^{a}$ | 14.63 | 14.22 | 34.11$^{abcde}$ | 485.33$^{abc}$ | 280.00$^{b}$ |
| $T_4$ | 56.33$^{d}$ | 5.96$^{b}$ | 20.24$^{a}$ | 14.48 | 14.11 | 32.00$^{de}$ | 451.48$^{cd}$ | 266.67$^{bc}$ |
| $T_5$ | 54.89$^{d}$ | 5.66$^{b}$ | 20.07$^{a}$ | 14.09 | 14.00 | 31.78$^{e}$ | 444.89$^{d}$ | 260.00$^{c}$ |
| $T_6$ | 56.67$^{d}$ | 6.41$^{b}$ | 19.93$^{a}$ | 14.38 | 14.00 | 32.78$^{cde}$ | 458.89$^{cd}$ | 270.00$^{bc}$ |
| $T_7$ | 67.41$^{b}$ | 2.58$^{cde}$ | 20.64$^{a}$ | 14.04 | 14.00 | 34.22$^{abcd}$ | 479.11$^{abcd}$ | 266.67$^{bc}$ |
| $T_8$ | 64.30$^{c}$ | 5.34$^{b}$ | 20.33$^{a}$ | 14.42 | 13.56 | 33.00$^{bcde}$ | 447.41$^{d}$ | 263.33$^{bc}$ |
| $T_9$ | 65.67$^{bc}$ | 4.78$^{bc}$ | 21.10$^{a}$ | 14.31 | 13.78 | 34.67$^{abc}$ | 477.04$^{abcd}$ | 280.00$^{b}$ |
| $T_{10}$ | 67.48$^{b}$ | 2.03$^{de}$ | 20.57$^{a}$ | 14.31 | 14.00 | 34.00$^{abcde}$ | 475.26$^{bcd}$ | 270.00$^{bc}$ |
| $T_{11}$ | 64.67$^{c}$ | 3.82$^{bcd}$ | 20.00$^{a}$ | 14.36 | 14.00 | 32.78$^{cde}$ | 458.89$^{cd}$ | 276.67$^{bc}$ |
| $T_{12}$ | 67.19$^{b}$ | 2.36$^{cde}$ | 19.38$^{a}$ | 14.38 | 14.22 | 35.22$^{ab}$ | 500.78$^{ab}$ | 298.33$^{a}$ |
| p-Value | <.0001 | <.0001 | 0.0021 | 0.2892 | 0.6211 | <.0001 | <.0001 | <.0001 |
| LSD(P=0.05) | 0.97 | 2.68 | 2.39 | NS | NS | 2.43 | 34.98 | 18.27 |

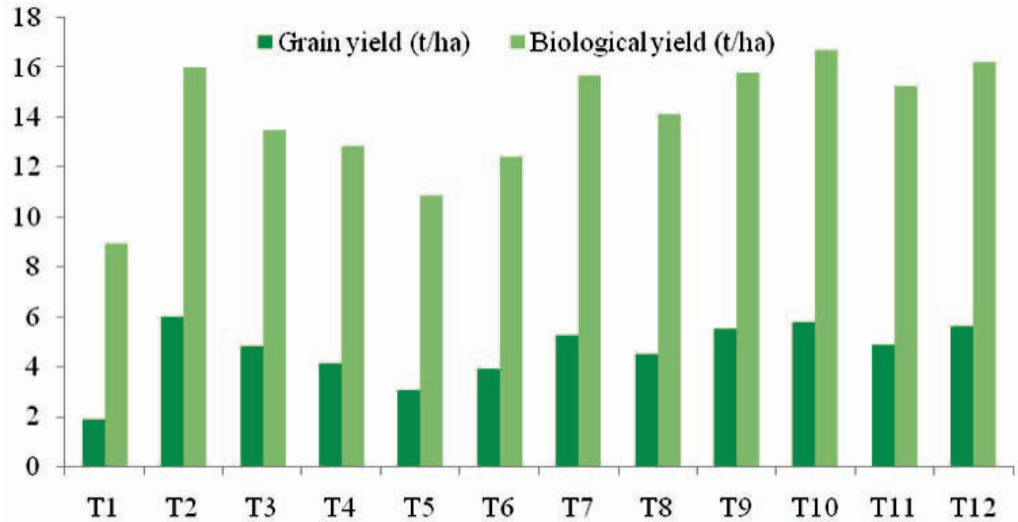
***Note**:* $T_1$: Weedy check, $T_2$: Weed free check, $T_3$: Atrazine 1000 g/ha (PE) *fb* hand weeding at 25 DAS, $T_4$: Topramezone 30 g/ha at 25 DAS, $T_5$: Halosulfuron 75 g/ha at 25 DAS , $T_6$: Tembotrione 150 g/ha at 25 DAS, $T_7$: Topramezone 25.2 g/ha + atrazine 750 g/ha at 15 DAS ,$T_8$: Halosulfuron 67.5 g/ha + atrazine 750 g/ha at 15DAS, $T_9$: Tembotrione 120 g/ha + atrazine 750 g/ha at 15 DAS, $T_{10}$: Atrazine 750 g/ha (PE) *fb* topramezone 25.2 g/ha at 25 DAS, $T_{11}$: Atrazine 750 g/ha (PE) *fb* halosulfuron 67.5 g/ha at 25 DAS, $T_{12}$: Atrazine 750 g/ha (PE) *fb* tembotrione 120 g/ha at 25 DAS. Means followed by a similar lower case letter within a column are not significantly different according to least significant difference test (P=0.05).

**Table 5.** Effect of post-emergence herbicides application on yields, shelling and harvest index of *Kharif* maize

| Treatment | Cob yield (t/ha) | Stover yield (t/ha) | Grain yield (t/ha) | Shelling (%) | Harvest index (%) |
|---|---|---|---|---|---|
| $T_1$ | 3.07$^{h}$ | 5.86$^{f}$ | 1.87$^{h}$ | 61.04$^{c}$ | 20.94$^{f}$ |
| $T_2$ | 7.43$^{a}$ | 8.59$^{abc}$ | 6.01$^{a}$ | 80.68$^{a}$ | 37.45$^{a}$ |
| $T_3$ | 6.06$^{cde}$ | 7.43$^{de}$ | 4.80$^{cde}$ | 79.22$^{ab}$ | 35.66$^{ab}$ |
| $T_4$ | 5.35$^{ef}$ | 7.51$^{cde}$ | 4.12$^{ef}$ | 77.21$^{ab}$ | 31.98$^{d}$ |
| $T_5$ | 4.09$^{g}$ | 6.81$^{ef}$ | 3.04$^{g}$ | 74.49$^{b}$ | 27.84$^{e}$ |
| $T_6$ | 5.02$^{fg}$ | 7.40$^{de}$ | 3.93$^{f}$ | 78.48$^{ab}$ | 31.64$^{d}$ |
| $T_7$ | 6.66$^{abcd}$ | 9.04$^{ab}$ | 5.24$^{bc}$ | 78.76$^{ab}$ | 33.39$^{cd}$ |
| $T_8$ | 5.86$^{def}$ | 8.29$^{bcd}$ | 4.49$^{def}$ | 77.11$^{ab}$ | 31.65$^{d}$ |
| $T_9$ | 6.90$^{abc}$ | 8.91$^{ab}$ | 5.50$^{abc}$ | 79.71$^{ab}$ | 34.82$^{bc}$ |
| $T_{10}$ | 7.22$^{ab}$ | 9.50$^{a}$ | 5.76$^{ab}$ | 79.96$^{a}$ | 34.45$^{bc}$ |
| $T_{11}$ | 6.41$^{bcd}$ | 8.88$^{ab}$ | 4.87$^{cd}$ | 76.15$^{ab}$ | 31.90$^{d}$ |
| $T_{12}$ | 7.00$^{abc}$ | 9.23$^{ab}$ | 5.64$^{ab}$ | 80.49$^{a}$ | 34.72$^{bc}$ |
| p-Value | <.0001 | <.0001 | <.0001 | <.0001 | <.0001 |
| LSD (P=0.05) | 0.97 | 1.09 | 0.73 | 5.35 | 2.15 |

***Note**:* $T_1$: Weedy check, $T_2$: Weed free check, $T_3$: Atrazine 1000 g/ha (PE) *fb* hand weeding at 25 DAS, $T_4$: Topramezone 30 g/ha at 25 DAS, $T_5$: Halosulfuron 75 g/ha at 25 DAS , $T_6$: Tembotrione 150 g/ha at 25 DAS, $T_7$: Topramezone 25.2 g/ha + atrazine 750 g/ha at 15 DAS ,$T_8$: Halosulfuron 67.5 g/ha + atrazine 750 g/ha at 15DAS, $T_9$: Tembotrione 120 g/ha + atrazine 750 g/ha at 15 DAS, $T_{10}$: Atrazine 750 g/ha (PE) *fb* topramezone 25.2 g/ha at 25 DAS, $T_{11}$: Atrazine 750 g/ha (PE) *fb* halosulfuron 67.5 g/ha at 25 DAS, $T_{12}$: Atrazine 750 g/ha (PE) *fb* tembotrione 120 g/ha at 25 DAS. Means followed by a similar lower case letter within a column are not significantly different according to least significant difference test (P=0.05).

**Figure 2.** Effect of post-emergence herbicides application on grain and biological yields of *Kharif* maize




double than weedy check. The enhancement in the growth attributes might have lead to increased cob and stover yield in these treatments in our study. Kumar *et al*. (2013) reported yield enhancement in atrazine + halosulfuron @ 1.0 kg *a.i.*/ha + 90 g *a.i.*/ha as (POE) application in zero-till maize. The signifcantly lowest shelling percentage and harvest index of maize were recorded with weedy check and the highest were recorded with $T_2$, which was at par with all other weed management practices except $T_5$ which shows that the better weed management has the potential for a better source-sink relationship also reported by Sanodiya *et al.* (2013).

*Economics*

The application of post-emergence herbicides gave the highest net returns of Rs. 56.19 $\times 10^3$/ha ($T_{10}$) which was at par with $T_{12}$. However, the lowest net returns were

**Table 6.** Effect of post-emergence herbicides application on economics of *Kharif* maize production

| Treatment | Gross returns ($\times 10^3$ Rs./ha) | Net returns ($\times 10^3$ Rs./ha) | BC ratio | Added cost ('000 Rs/ha) | Additional return over a weedy check ('000 Rs./ha) | Additional return over a weedy check (Rs./Rs. invested) |
|---|---|---|---|---|---|---|
| $T_1$ | 32.50$^{h}$ | 3.09$^{g}$ | 0.11$^{g}$ | - | - | - |
| $T_2$ | 94.17$^{a}$ | 53.52$^{ab}$ | 1.32$^{abc}$ | 11.23 | 50.42$^{ab}$ | 4.49$^{d}$ |
| $T_3$ | 75.79$^{cde}$ | 41.03$^{cd}$ | 1.18$^{bcd}$ | 5.35 | 37.93$^{cd}$ | 7.09$^{bc}$ |
| $T_4$ | 66.28$^{ef}$ | 31.61$^{de}$ | 0.91$^{de}$ | 5.26 | 28.51$^{de}$ | 5.42$^{cd}$ |
| $T_5$ | 50.07$^{g}$ | 15.12$^{f}$ | 0.43$^{f}$ | 5.54 | 12.02$^{f}$ | 2.17$^{e}$ |
| $T_6$ | 63.46$^{f}$ | 28.58$^{e}$ | 0.82$^{e}$ | 5.47 | 25.48$^{e}$ | 4.66$^{d}$ |
| $T_7$ | 83.76$^{abc}$ | 49.32$^{abc}$ | 1.43$^{ab}$ | 5.02 | 46.22$^{abc}$ | 9.21$^{a}$ |
| $T_8$ | 72.26$^{def}$ | 37.32$^{de}$ | 1.07$^{cde}$ | 5.53 | 34.22$^{de}$ | 6.19$^{cd}$ |
| $T_9$ | 87.24$^{ab}$ | 52.81$^{ab}$ | 1.53$^{a}$ | 5.02 | 49.71$^{ab}$ | 9.90$^{a}$ |
| $T_{10}$ | 91.64$^{a}$ | 56.19$^{a}$ | 1.59$^{a}$ | 6.04 | 53.09$^{a}$ | 8.79$^{ab}$ |
| $T_{11}$ | 78.34$^{bcd}$ | 42.38$^{bcd}$ | 1.18$^{bcd}$ | 6.55 | 39.28$^{bcd}$ | 6.00$^{cd}$ |
| $T_{12}$ | 89.56$^{a}$ | 54.11$^{a}$ | 1.53$^{a}$ | 6.04 | 51.01$^{a}$ | 8.45$^{ab}$ |
| p-Value | <.0001 | <.0001 | <.0001 | – | <.0001 | <.0001 |
| LSD (P=0.05) | 11.15 | 11.15 | 0.31 | – | 11.72 | 1.99 |

***Note:*** $T_1$: Weedy check, $T_2$: Weed free check, $T_3$: Atrazine 1000 g/ha (PE) *fb* hand weeding at 25 DAS, $T_4$: Topramezone 30 g/ha at 25 DAS, $T_5$: Halosulfuron 75 g/ha at 25 DAS , $T_6$: Tembotrione 150 g/ha at 25 DAS, $T_7$: Topramezone 25.2 g/ha + atrazine 750 g/ha at 15 DAS ,$T_8$: Halosulfuron 67.5 g/ha + atrazine 750 g/ha at 15DAS, $T_9$: Tembotrione 120 g/ha + atrazine 750 g/ha at 15 DAS, $T_{10}$: Atrazine 750 g/ha (PE) *fb* topramezone 25.2 g/ha at 25 DAS, $T_{11}$: Atrazine 750 g/ha (PE) *fb* halosulfuron 67.5 g/ha at 25 DAS, $T_{12}$: Atrazine 750 g/ha (PE) *fb* tembotrione 120 g/ha at 25 DAS. Means followed by a similar lower case letter within a column are not significantly different according to least significant difference test (P=0.05).

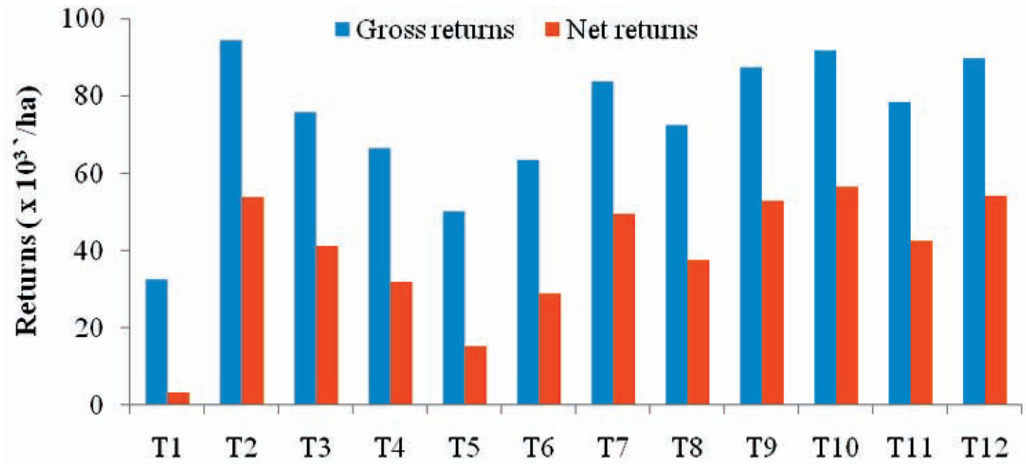



**Figure 3.** Effect of post-emergence herbicides application on gross and net returns of *Kharif* maize

recorded in control (Rs. 3.09 $\times 10^3$/ha). The mean increase in net returns ranged from Rs. 15.12 ($T_5$) to 56.19 ($T_{10}$) $\times 10^3$/ha across the herbicides applied treatments, which gave three to five times more net returns compared to weedy check. The significantly highest BC ratio was observed in the $T_{10}$ that was at par with $T_2$, $T_7$, $T_9$ and $T_{12}$. The added cost and additional return showed in Table 6 & Figure 3. The uniform cost of Rs. 29,410/ha was incurred in maize cultivation. The range of the added cost in weed management was Rs. 5.02 to 6.55 $\times 10^3$/ha. The significantly highest additional net returns were found in the PE atrazine *fb* POE application of topramezone and tembotrione with $T_{10}$ and $T_{12}$. The additional returns Rs./Rs. invested was found significantly highest with $T_9$ which was at par with $T_7$. The increase in the net returns was due to increase in grain and stover yields of maize in these treatments as compared to weedy check which leads to enhanced returns in maize cultivation as the range of the added cost in weed management was Rs. 5.02 to 6.55 $\times 10^3$/ha only. Singh *et al*. (2012) also reported enhancement in net returns from maize with the use of the tembotrione. The economic viability of the post-emergence herbicide application became more relevant in the scenario of the escalating farm labour prices.

## Conclusion

The application of post-emergence herbicide *viz*., tembotrione (120 g/ha) / topramezone (25.2 g/ha) as tank-mix as early post-emergence at 15 DAS or as sequential application at 25 DAS with 75% recommended dose of the atrazine (750 g/ha) gave significantly enhanced crop growth, yield and net returns in *Kharif* maize. Thus, these post-emergence herbicides could be adopted for higher productivity and profitability in *Kharif* maize production.

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RESEARCH PAPER

# Evaluating the performance of new stabilized urea fertilizer (AGROTAIN incorporated urea) in maize-wheat system

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**Abstract:** The nitrogen use efficiency of available urea fertilizer is low and it ranged from 30-50% in major food crops due to high volatilization and leaching losses. Nitrification and urease inhibitors and neem coating are the suggested means to reduce the nitrogen losses, thereby increasing crop nitrogen use efficiency (NUE). To improve the NUE, a study was conducted during 2020-21 to evaluate the performance of neem coated urea (NCU) (nitrification inhibitor) and Agrotain incorporated urea (AIU) (urease inhibitor) in maize-wheat system. The experiment comprised of control (No nitrogen), 100% NCU, 100% AIU, 80% AIU and 60% AIU in a randomized complete block design with four replications. The SPAD and NDVI values at different crop growth stages in maize-wheat system found at par with 100% AIU and 100% NCU. As compared to 100% NCU, grain yield of maize and wheat increased by 6.0% and 4.9% with 100% AIU, respectively. However, the system yield (wheat equivalents) was 5.6 and 1.9% higher with 100 and 80% AIU, respectively in comparison to 100% NCU. The 100% and 80% AIU recorded the higher net returns in maize by 7.7% and 3.0% as compared to 100% NCU, respectively. The system net returns were 6.2% higher with 100% AIU over 100% NCU. The agronomic efficiency of N ($AE_N$) and partial factor productivity of N ($PFP_N$) was decreased with the increased doses of N. Higher $AE_N$ was observed with 100% AIU as compared to 100% NCU. Compared to NCU, $AE_N$ and $PEP_N$ of the system increased 30% and 27.4% under 80% AIU. Our study revealed that new stabilized urea fertilizer (AIU) saved 20% (30 kg N ha$^{-1}$) of NCU at similar or higher yields and N-use efficiency in maize-wheat system of north-west India. The AIU urea with neem was more efficient than neem alone and may play a great role in reducing the N-losses. thereby improving productivity while reducing the environmental footprints. AIU with neem could allow farmers to achieve equal to or higher yields to neem alone while using 20% less nitrogen fertilizer.

**Keywords:** Agrotain incorporated urea · Maize-wheat system · Net returns · Nitrogen use efficiency · System yield

## Introduction

Urea and nitrogen based fertilizers are commonly used to supply nitrogen (N) to agricultural crops (Heffer and Michel, 2016). Urea is one of the most extensively used N fertilizers due to its high solubility and N concentration and lower cost of production (Behera *et al.,* 2013; Li *et al.,* 2015), it accounts for over 55% of all N fertilizers (IFA, 2017). According to the reports of FAI (2019-2020), the consumption of urea in India grew moderately at a growth rate of ~1.3% from 31.9 MT (2016) to 33.6 MT (2020). However, domestic urea production remained unchanged at 24.5 MT in 2020 compared to 24.1 MT in 2019 and ~27% of the urea demand was met through imports in 2020. The efficiency of the use of nitrogen from urea based fertilizer is low in many instances ~30-50% due to complex transformation of N in soil which led to loss of N (Afshar *et al.,* 2018; Panday *et al.,* 2020). Approximately 60% of global N fertilizer is majorly used

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for producing cereals: rice, wheat and maize**.** On average, the recovery rate of the urea N fertilizer was 34% for wheat (Galindo *et al.,* 2021), 46% for rice (Ladha *et al.,* 2005) and 33% for maize (Sindelar *et al.,* 2015).

However, urea can be an inefficient source of N in soil because it can be lost through multiple pathways such as volatilization, nitrate leaching and nitrous oxide emissions (Zaman and Blennerhassett, 2010). Urea undergoes rapid hydrolysis by urease enzyme releasing carbon dioxide and ammonia, which might be lost from soil to the atmosphere through volatilization (Dawar *et al.,* 2011; Soares *et al.,* 2012). The hydrolysis of urea increased the soil pH and favours ammonia volatilization; these nitrogen losses can be higher than 35% of the applied N (Cai *et al.,* 2002; Soares *et al.,* 2012). A main focus for ensuring the sustainability of crop productivity is to increase N use efficiency by optimized N management practices that reduce the risk of $NO_3$ leaching and $NH_3$ emission. Use of urease and nitrification inhibitors to urea fertilizer is one approach to improve N use efficiency of fertilizer. Common urease and nitrification inhibitors are N-(n-butyl) thiophosphoric triamide (NBPT) and neem coated urea (NCU), respectively. Nitrification inhibitors slow down the conversion of $NH_4^+$ to $NO_3^-$ by deactivating ammonia oxidation by ammonium-oxidizing bacteria, leading to a reduction in the emission of $N_2O$ from soils (Ruser and Schulz, 2015).

Ammonia volatilization is a major source for N losses and the main cause of low N use efficiency of urea-based fertilizers (Li *et al.,* 2008). However, the efficiency of urea based fertilizers can be improved with use of urease inhibitors to delay hydrolysis, prolonged urease diffusion interval and allow the urea to move to deeper layers of soil along with rainfall (Rawluk *et al.,* 2001). There are several compounds that have been developed to delay urea hydrolysis and behave as urease inhibitors, but only NBPT (N-(n-butyl) thiophosphoric triamide) traded as "Agrotain" has been used outside of India (North America, South America, Europe, Australia and Southern Africa) for more than 20 years. The application of NBPT has been reported to reduce urease activity in soil and thus reducing $NH_3$ emission (Sanz-Cobena *et al.,* 2012; Sanz-Cobena *et al.,* 2014). When NBPT is applied to soil it gets converted into its oxygen analogue NBPTO (N-(n-butyl) phosphoric triamide) which strongly competes with the urease enzyme for active sites on urea and slows down the hydrolysis of urea (Sigurdarson *et al.,* 2018). NBPT can block urea hydrolysis for 7 to 10 days after its application and results in smaller increase of soil pH around the urea granule reduces the ammonia volatilization losses (Fu *et al.,* 2020). Recent literatures have revealed that urea coated with NBPT has reduced the volatilization losses from the soil surface under various soil and environmental conditions (Pan *et al.,* 2016; Silva *et al.,* 2017; Cantarella *et al.,* 2018).

The objectives of this study was to evaluate the performance of agrotain incorporated urea (AIU) on nitrogen use efficiencies (agronomic efficiency and partial factor productivity), crop yields, net returns yield attributes, chlorophyll content and NDVI, in maize-wheat system. We hypothesized that application of Agrotain incorporated urea (AIU) would increase crop yield, thereby decreasing nitrogen losses, maintaining leaf chlorophyll content, and maximizing above-ground plant attributes.

## Materials and methods

*Site description and experiment details*

A field study was conducted during 2020-2021 with maize (*Kharif* season) and wheat (*Rabi* season) crops at ICAR (Indian Council of Agricultural Research)-CSSRI (Central Soil Salinity Research Institute) research farm Karnal (29°708625N, 76°7956765E and at an elevation of 243 m above msl), India. This region has a sub-tropical climate with wet summers and dry winters, with annual precipitation 670 mm of which 75–80% occurs in June - September (monsoon season). The soil texture was silty loam (0-15 cm layer) with medium in organic carbon (0.69%), low in available N (157 kg ha$^{-1}$), low in P (18.5 kg ha$^{-1}$), K (210 kg ha$^{-1}$) and neutral pH (7.45). Different soil chemical properties analysed prior to treatment application are given in Table 1. The experiment was conducted in plot size of 20 m$^2$ (5 m × 4 m) in a randomized complete block design with four replications. The five treatments were; T1- untreated control (control), T2- 100% (150 kg N ha$^{-1}$) neem coated urea (NCU), T3- 100% (150 kg N ha$^{-1}$) agrotain incorporated urea (AIU), T4- 80% (120 kg N ha$^{-1}$) agrotain incorporated urea (AIU), T5- 60% (90 kg N ha$^{-1}$) agrotain incorporated urea (AIU). The source of the AIU was Koch Agronomic Services (Wichita, Kansas, USA) and all the AIU was treated with neem after granulation.

**Table 1**. Initial soil properties of the experimental site

| Soil parameters | Soil depth (cm) | | |
|---|---|---|---|
| | 0–15 | 15–30 | 30–45 |
| Soil pH | 7.45 | 7.61 | 7.61 |
| EC (dS $m^{-1}$) | 0.70 | 0.65 | 0.67 |
| SOC (%) | 0.69 | 0.40 | 0.24 |
| Available N (kg $ha^{-1}$) | 157 | 133 | 108 |
| Available P (kg $ha^{-1}$) | 18.5 | 14.1 | 10.0 |
| Available K (kg $ha^{-1}$) | 210 | 210 | 237 |

*Crop management*

Before sowing, field was levelled at zero level and ploughed well before implementation of different treatments as per protocols. Cultivar CP 838 of maize was sown on 7th June and row to row spacing of 67.5 cm was maintained. The seed rate of 20 kg $ha^{-1}$ was adopted for optimum plant population. In *Rabi season*, wheat cultivar HD-2967 was sown on 5th November with multi crop planter. Inter row distance of 22.5 cm was maintained at a seed rate of 100 kg $ha^{-1}$. Irrigation was applied at all the critical crop growth stages of both the crops. Recommended plant protection measures were implemented for keeping crop free from weeds, insects, pests and diseases. Both maize and what crop was harvested at physiological maturity.

*Fertilizer management*

Maize and wheat were fertilized with recommended dose of 150 kg N+ 60 kg P+ 60 kg K per hectare. During the experiment, full dose of P (60 kg $ha^{-1}$) and K (60 kg $ha^{-1}$) were applied as basal at the time of sowing in the form of single super phosphate and muriate of potash, respectively, while N fertilizer was split into two equal doses, half applied at 21 days after sowing (DAS) and remaining half at 50 DAS. To determine the agronomic efficiency of applied N, partial factor productivity of N ($PFP_N$) was calculated using Eq. 1 & 2 and expressed in kg grain per kg of N applied.

Agronomic efficiency of N ($AE_N$) = $(Y-Y_0) / F$ ...(1)

Partial factor productivity of N ($PFP_N$) = $Y / F$ ...(2)

Where

Y= yield under nutrient applied plot, $Y_0$ = yield under control plot (with not nutrient applied), F = amount of N-nutrient applied

*Crop observations*

For physiological studies, Minolta 502 SPAD chlorophyll meter was used to determine the leaf chlorophyll content at different intervals of time. The normalized difference vegetation index (NDVI) was measured using a portable N tech "Greenseeker" NDVI meter at different intervals of time. Measurements were taken horizontally across the plot from the middle rows in the plot to avoid border effects. NDVI and SPAD readings were taken from 11:00 h to 14:00 h under stable and clear sky to prevent data fluctuations. Plant height, no. of grains per cob/spikelet, 1000-grain weight, spike length, and grain yield were recorded for each plot. The yield attributes of maize and wheat crop were determined during the crop growth period as per the standard procedures.

Maize and wheat were harvested and threshed manually. At maturity, the grain and straw yields of both maize and wheat were determined on a total area of 14 $m^2$. Grain yield was expressed as Mg $ha^{-1}$ at 14% and 12% grain moisture content for maize, and wheat, respectively. To compare the productivity of maize and wheat crop and total system productivity of both the crops, the grain yield of maize was converted into wheat equivalent yield (WEY) (Mg $ha^{-1}$) and calculated using the Eq. (3).

WEY (Mg $ha^{-1}$) = [maize yield (Mg $ha^{-1}$) * MSP of respective crop (INR Mg $ha^{-1}$)]

/ MSP of wheat (INR Mg $ha^{-1}$)] ...(3)

Where, MSP is minimum support price of Govt. of India, INR is Indian Rupee

*Economics*

The data on crop management practices like tillage, irrigations, seed, pesticides, fertilizer, labour use, etc. and their costs under each treatment were recorded using a standard data format. Variable costs of each treatment were summed up to calculate the total cost of production. Gross returns were calculated on the prevailing market prices of the produce (grain and straw). Net returns were calculated by deducting the total cost of production from the gross returns. The gross returns, net returns and BC ratio were statistically analysed using analysis of variance (ANOVA) at 5% level of significance. The total net return of maize-wheat system was calculated by subtracting total cost of production from gross returns of system.

*Statistical analysis*

The data for different parameters were analysed by ANOVA (analysis of variance) using SAS 9.1 software (SAS Institute, Cary, NC). Fisher's least significant difference (LSD) test was used for comparing differences between two means at the 5% probability level.

## Results and Discussion

*Crop growth attributes*

SPAD and NDVI values changed with the crop stage, highest value at 60 days after sowing (DAS) (Figure 1 & 2). At the respective crop stages, there was no much difference with respect to the varied N-fertilizer doses in both maize and wheat crop. But it was significant with the control (No N-fertilizer) for SPAD and NDVI values. The treatment of 100% AIU resulted in higher SPAD and NDVI numerical values than 100% NCU (30, 44, 50, 60 and 72 DAS). At 44 and 50 DAS, SPAD values showed increment of 3% and 3.5% with 100% AIU over 100% NCU treatment. At 60 DAS, 100% NCU (40.38 and 0.76) showed the highest value followed by 100% AIU (39.68 and 0.74). SPAD and NDVI value were observed higher by 3.0-56.8% than control at different intervals of time. A positive correlation exists between N and leaf chlorophyll content with addition of urease inhibitor (Buscaglia and Varco, 2002; Kawakami *et al.,* 2012) due to increased nitrogen uptake as N is a chief structural component of chlorophyll in plants (Makino and Osmond, 1991).

*Crop yield attributes*

Yield attributes data (Table 2) indicated a significant treatment effect for no. of grains per cob and 100-grains (seed index) of maize crop. Results showed no

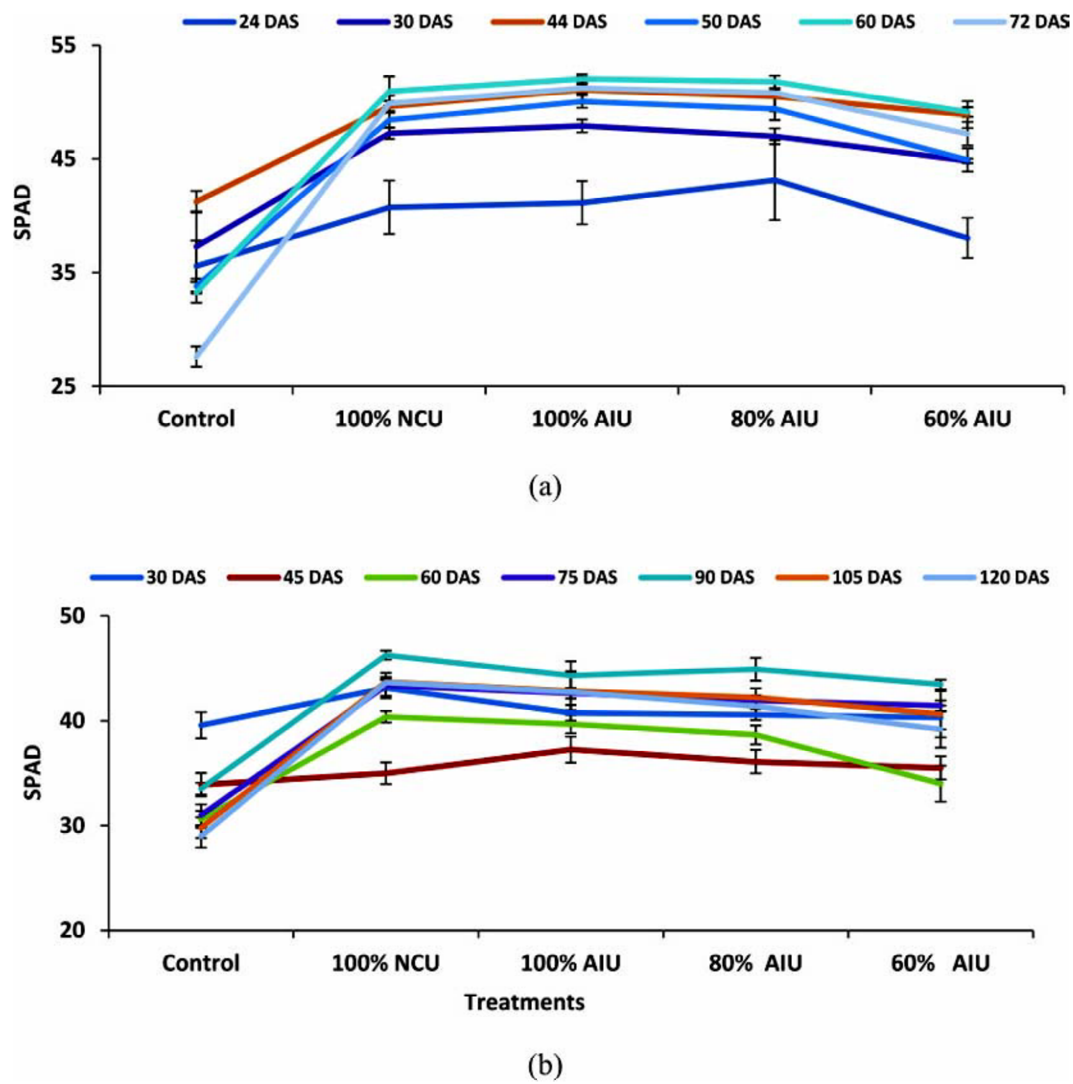


**Figure 1.** SPAD values of (a) maize (b) wheat under different N-doses (error bars indicate SE of the mean)

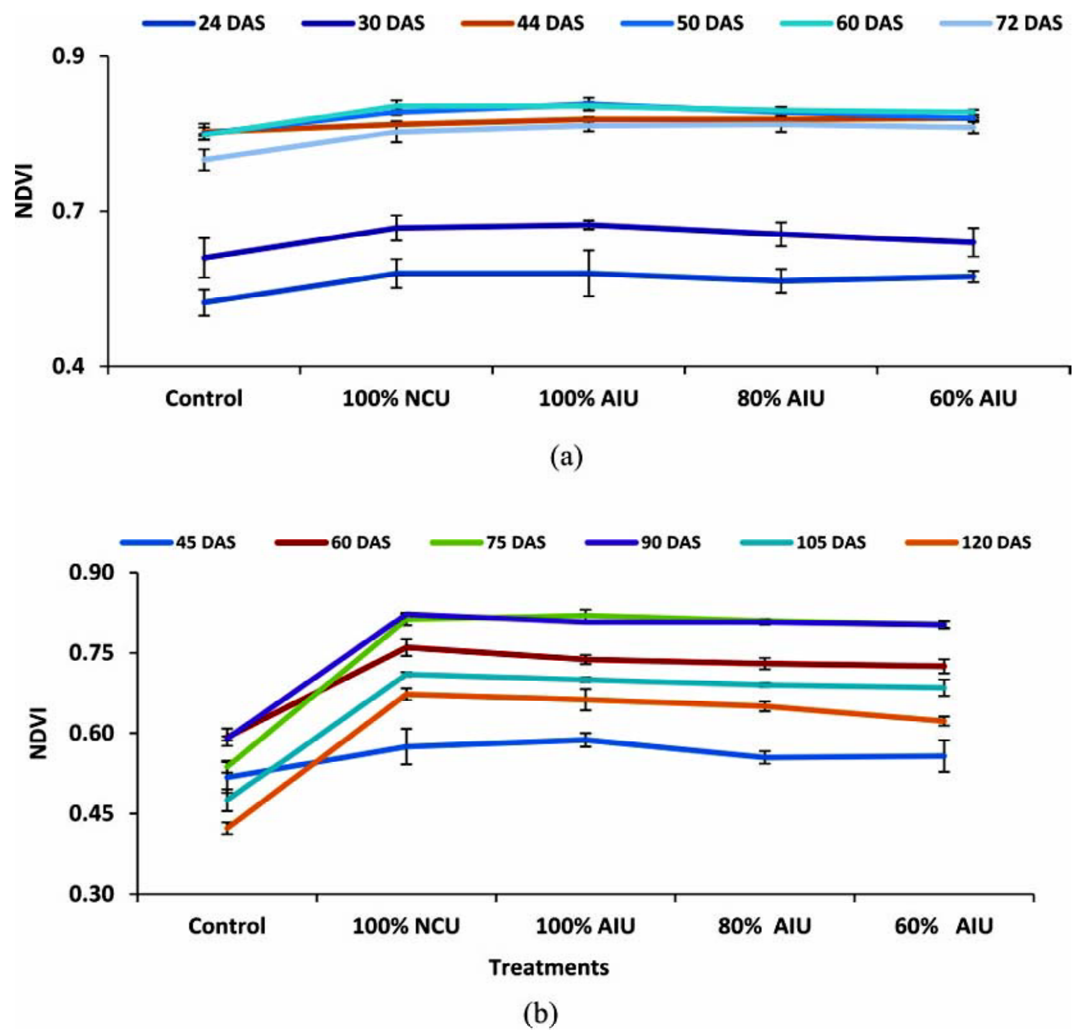


**Figure 2.** NDVI values of (a) maize (b) wheat under different N doses (error bars indicate SE of the mean)

**Table 2.** Effect of different N- doses on yield attributes of maize and wheat

| Treatment | Maize | | Wheat | | |
|---|---|---|---|---|---|
| | No. of grains per cob | 100- grains weight | Spike length(cm) | No. of grains per spike | 1000- grains weight |
| Control | 507.60b* | 27.64c | 13.00b | 29.33c | 38.16a |
| 100% NCU | 607.05a | 31.84ab | 15.80a | 59.35ab | 39.19a |
| 100% AIU | 627.70a | 33.95a | 15.90a | 63.23a | 41.36a |
| 80% AIU | 624.60a | 33.12a | 15.78a | 61.90a | 39.28a |
| 60% AIU | 581.80ab | 30.71b | 15.49a | 55.85b | 39.00a |

*Means followed by a similar lowercase letters within a column are not significantly different at 0.05 level of probability using LSD test

differences in no. of grains per cob between 100% NCU (607.1), 100% AIU (627.7), 80% AIU (624.6), and 60% AIU (581.8) however all the N fertilized treatments was significantly higher over control (507.6). Meanwhile, the grains weight of 100 seeds was improved by 6.6% and 4% with 100% AIU and 80% AIU as compared to 100% NCU, respectively.

In wheat, spike length was recorded similar under all N fertilized treatments but significantly decreased under control. The 80% AIU was on par with 100% AIU and 100% NCU with respect to the no. of grains per spike. Compared to 100% NCU, the no. of grains per spike with 100% AIU and 80% AIU were increased by 6.5% and 4.3%, respectively. The 1000-grains weight of wheat under different treatments was found similar but with 100% AIU it was 5.5% higher over 100% NCU. These results are consistent with studies of Dawar *et al.* (2011); Curitiba *et al.* (2013) and Khan *et al.* (2014) as they reported the improvement in yield parameters with application of NBPT incorporated urea as compared to urea.

*Crop and system productivity*

The Result showed that grain yield of maize with 80% AIU (9.14 Mg ha$^{-1}$) was on par with 100% AIU (9.49 Mg ha$^{-1}$) and 100% NCU (8.95 Mg ha$^{-1}$). The lowest yield was recorded with control treatment and it was followed by 60% AIU (Table 3). The maize yield was increased by 6% with 100% AIU as compared to 100% NCU. Stover yield was also observed similar with all N fertilized treatments. The highest stover yield of 13.21 Mg ha$^{-1}$ was recorded with 100% AIU which was 5.8% higher than 100% NCU (12.49 Mg ha$^{-1}$). The application of urea along with urease inhibitor generally exhibited higher yield and N use efficiency by inhibiting N losses through regulating transformation of inorganic N in soil (Abalos *et al.,* 2014). Previous studies on urease inhibitor (NBPT) also concluded that application of the urea along with NBPT improve maize yields as compared to conventional urea (Venterea *et al.,* 2011; Qiao *et al.,* 2015; Zhao *et al.,* 2017). Similarly, Mohd Zuki *et al.,* (2020) also reported that NBPT-coated urea at normal rate and 20% lower rate improved the maize yield by 42% and 48% in comparison to urea. Wheat grain yield with 100% AIU (6 Mg ha$^{-1}$) was found at par with 100% NCU (5.72 Mg ha$^{-1}$). Straw yield under different N fertilized treatments varied from 7.21 Mg ha$^{-1}$ to 8.03 Mg ha$^{-1}$ (Table 3). Meanwhile, application of 100% AIU increased the grain and straw yield by 4.9% and 4.4% as compared to 100% NCU. The outcomes showed a strong agreement with Silva *et al.,* (2017); as their meta-analysis results directed that urease inhibitor treated urea can increase crop yield potential up to 5.3-7.5% for major crops. Similar results were found by McClallen (2014), where the addition of urease inhibitor (NBPT) significantly increased wheat yield production. In literature, the field trials showed increased yield of different crops species following the use of new urease inhibitor (Zaman *et al.,* 2009; Khan *et al.,* 2015).

Highest system yield (WEY) of 14.99 Mg ha$^{-1}$ was recorded with 100% AIU and the lowest with control (7.46 Mg ha$^{-1}$) (Table 5). System yield exhibited a statistical difference only between the control and 60% AIU treatments ($P < 0.05$). The system yield under 100% NCU, 100% AIU and 80% AIU were recorded at par, which varied from 14.19 Mg ha$^{-1}$ to 14.99 Mg ha$^{-1}$. The 100% AIU increased the system yield by 5.6% compared 100% NCU in maize-wheat system. Correspondingly, meta-analysis study of Linquist *et al.* (2013) revealed that when enhanced efficiency nitrogen fertilizers were analysed, urease inhibitor (NBPT) and neem product suggest increased yield and N uptake of rice systems under different soil conditions.

*Crop and system net returns*

The seasonal cost of production, gross return, net return and BC ratio for maize and wheat crop are presented in Table 4. The total production costs of maize varied from 35,720 to 43,370 INR ha$^{-1}$ under different treatments. In maize, 100% AIU resulted in highest gross (1, 77,472 INR ha$^{-1}$) and net return (1, 34,104 INR ha$^{-1}$) which was 6.1% and 7.7% higher over the 100% NCU. This was due to lower cost of production and higher net incomes from 100% AIU. Results indicated highest B:C ratio was recorded with 100% AIU (3.1) and found at par with 100% NCU and 80% AIU treatment.

In wheat, 100% AIU recorded highest cost of production, gross and net return than other treatments; however, it was at par with 100% NCU and 80% AIU in terms of gross and net returns. The gross and net income under 100% AIU was higher by 4,456 INR ha$^{-1}$ (3.2%)

**Table 3.** Effect of different N- doses on grain yield, stover or straw yield, agronomic efficiency of N ($AE_N$) and partial factor productivity of N ($PFP_N$) of maize–wheat system

| Treatment | Maize | | | | Wheat | | | |
|---|---|---|---|---|---|---|---|---|
| | Grain yield (Mg ha$^{-1}$) | Stover yield (Mg ha$^{-1}$) | $AE_N$ (kg grain kg N$^{-1}$) | $PFP_N$ (kg grain kg N$^{-1}$) | Grain yield (Mg ha$^{-1}$) | Straw yield (Mg ha$^{-1}$) | $AE_N$ (kg grain kg N$^{-1}$) | $PFP_N$ (kg grain kg N$^{-1}$) |
| Control | 4.9c* | 8.53b | - | - | 2.81c | 3.94c | - | - |
| 100% NCU | 8.95a | 12.49a | 26.93c | 59.63c | 5.72ab | 7.69a | 20.48d | 38.13d |
| 100% AIU | 9.49a | 13.21a | 30.53b | 63.27c | 6.00a | 8.03a | 22.31c | 40.00c |
| 80% AIU | 9.14a | 12.92a | 35.25a | 76.17b | 5.81ab | 7.84a | 26.32b | 48.42b |
| 60% AIU | 7.95b | 11.05ab | 33.75a | 88.31a | 5.43ab | 7.21b | 30.85a | 60.33a |

*Means followed by a similar lowercase letters within a column are not significantly different at 0.05 level of probability using LSD test

**Table 4.** Economics of maize-wheat cultivation under different N fertilizer treatments

| Treatment | Maize | | | | Wheat | | | |
|---|---|---|---|---|---|---|---|---|
| | TCP (INR $ha^{-1}$) | GR (INR $ha^{-1}$) | NR (INR $ha^{-1}$) | BCR | TCP (INR $ha^{-1}$) | GR (INR $ha^{-1}$) | NR (INR $ha^{-1}$) | BCR |
| Control | 35720d* | 91795d | 56075d | 1.6c | 30454e | 67349c | 36895c | 1.2c |
| 100% NCU | 42714b | 167248b | 124534b | 2.9a | 36506b | 137981a | 101475ab | 2.8ab |
| 100% AIU | 43370a | 177472a | 134104a | 3.1a | 36506a | 142437a | 105930a | 2.9a |
| 80% AIU | 42511b | 170803ab | 128292ab | 3.0a | 36075c | 137838a | 101762ab | 2.8ab |
| 60% AIU | 40658c | 148591c | 107933c | 2.7b | 35648d | 128813b | 93165b | 2.6b |

*Means followed by a similar lowercase letters within a column are not significantly different at 0.05 level of probability using LSD test
**TCP-total cost of production, GR-gross return, NR-net return, BCR- benefit-cost ratio

and 4,455 INR $ha^{-1}$ (4.3%) compared to 100% NCU (Table 4). The BC ratio was 3.6% higher with 100% AIU (2.9) compared to conventional urea (2.8). Higher crop yields along with lower production costs in 100% AIU resulted in higher profitability compared with commonly adopted fertilizer sources. Our results lined up with Allende-Montalbán *et al.* (2021), they recorded increase in net economic benefit with application of urease inhibitor (NBPT) as compared with equal amount of urea in maize-wheat rotation. However, Zheng *et al.,* (2021) observed the economic benefit with nitrification inhibitor and urease inhibitor as compared to urea. The economic analysis of system revealed the highest net returns with 100% AIU (2, 40,033 INR $ha^{-1}$) and it was higher by 6.2% compared to 100% NCU (Table 4). Similar results have been reported by Liu *et al.,* (2022), reported that use of urea with inhibitors improved the net economic returns by 24.7% on average compared with the farmers' N practice in the wheat-maize rotation.

*N-use efficiencies ($AE_N$ and $PFP_N$)*

Results showed that different nitrogen treatments influenced the N-use efficiencies i.e. agronomic efficiency of N ($AE_N$) and partial factor productivity of N ($PFP_N$) of maize (Table 3). The highest $AE_N$ was recorded with 80% AIU (35.25 kg grain kg $N^{-1}$) and the lowest with 100% NCU (26.93 kg grain kg $N^{-1}$). $AE_N$ under 100% AIU was increased by 15.4% in contrast to 100% NCU. The $PFP_N$ was recorded higher with 60% AIU (88.31 kg grain kg $N^{-1}$) and it was followed by 80% AIU (76.17 kg grain kg $N^{-1}$) in maize. Application of 100% AIU and 80% AIU had higher $PFP_N$ than 100% NCU with an increment of 6.1% and 27.7%, respectively. Urease inhibitor has capability to delay the hydrolysis of urea and improved the use efficiencies of nitrogen by crop. Numerous researchers have done meta-analysis on urease inhibitor (NBPT) and showed positive effect of urea with NBPT in reducing the N losses and increasing nitrogen use efficiency under different crops (Abaldos *et al.,* 2014; Silva *et al.,* 2017; Cantarella *et al.,* 2018). Silva *et al.* (2011) also reported that the agronomic efficiency of AIU at lower N rate (96 kg N $ha^{-1}$) was higher and had the potential to perform better in increasing the NUE than higher N rates (120 kg N $ha^{-1}$).

The $AE_N$ and $PFP_N$ exhibited increased trend with decreased N fertilization in which higher values under 60% AIU than other N- fertilized treatments in wheat crop. With

**Table 5.** System yield (wheat equivalents), net returns, agronomic efficiency and partial factor productivity of N ($AE_N$ and $PFP_N$) of maize-wheat system

| Treatment | System yield (Mg $ha^{-1}$) | System net returns (INR $ha^{-1}$) | $AE_N$ (kg grain kg $N^{-1}$) | $PFP_N$ (kg grain kg $N^{-1}$) |
|---|---|---|---|---|
| Control | 7.46c* | 92969d | - | - |
| 100% NCU | 14.19a | 226009b | 22.43a | 47.31c |
| 100% AIU | 14.99a | 240033a | 25.09a | 49.95c |
| 80% AIU | 14.46a | 230054ab | 29.19a | 60.27b |
| 60% AIU | 12.95b | 201098c | 30.55a | 71.97a |

*Means followed by a similar lowercase letters within a column are not significantly different at 0.05 level of probability using LSD test

100% AIU, the $AE_N$ and $PEP_N$ was increased by 8.9% and 4.9% as compared to 100% NCU, respectively. However, our results indicating a benefit of incorporating agrotain to urea fertilizer on agronomic use efficiency of N are consistent with the study of Mozaffari *et al.,* (2007) and Li *et al.,* (2015). Furthermore, Kawakami *et al.,* (2012) reported that use of AIU in silt loam soil increased N uptake and N use efficiency by 17% and 41%, respectively.

The $PFP_{N-}$ of system ranged from 47.32 to 71.97 kg grain kg $N^{-1}$. The $AE_N$ of maize-wheat system with 60% AIU was increased by 36.2%, 21.8% and 4.7% as compared to 100% NCU, 100% AIU and 80% AIU, respectively (Table 5). The increase in $AE_N$ was not significant at $P<0.05$. The highest $PEP_N$ was recorded with 60% AIU (71.97 kg grain kg $N^{-1}$) followed by 80% AIU (60.27 kg grain kg $N^{-1}$). As compared to 100% NCU, increase of 5.5% and 27% was observed with 100% AIU and 80% AIU, respectively.

## Conclusions

This study provides a better understanding on the effect of urease inhibitors like Agrotain (NBPT), which improves the nitrogen use efficiency in maize-what system under western IGP. In the maize-wheat system, 80% AIU with neem was on par with 100% AIU and 100% NCU with respect to growth attributes, yield attributes, yields (grain and straw/stover) and net returns. It is concluded that with, similar yield and net returns can be obtained in maize-wheat system. With the use of AIU, 20% nitrogen can be saved because of higher N-use efficiencies in maize-wheat system. This would help in developing new methodology or strategies for more efficient utilization of input resources in soil and enhancement of sustainable crop production.

## Acknowledgement

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RESEARCH PAPER

# Response of tropical maize inbred lines to high plant density stress in terms of grain yield and disease attributes

**Surinder Sandhu · Harleen Kaur · Aditi Sarkar · Ashutosh Kushwah**

**Abstract:** The significantly lower maize productivity in tropical countries in comparison on temperate zones has always been an area of high concern. The increase in the number of plants per unit area, through the adoption of high plant density (HPD), has been extensively used in temperate maize but this strategy has not been well exploited in tropical maize. The proposed study is to identify HPD adept donor inbreds to generate commercial hybrids to achieve high productivity in tropical/ subtropical maize. A set of 20 inbred lines, selected from diverse origin of tropical maize germplasm pool, was screened under normal plant density (NPD) and high plant density to assess their tolerance to crowding stress in terms of grain yield and prevalent diseases during *Kharif* season. The present study indicated that maize inbred lines PML 387, PML 503 and PML 892 with good seed yield potential and disease resistance may serve as potential donors to generate crowding stress tolerant hybrids for commercial hybrid breeding programme. With systematic evaluation of HPD amenable parents and hence, generated hybrids, there is a feasibility to enhance the maize productivity on commercial scale with new agronomic intervention of altered plant to plant spacing.

**Keywords:** Banded leaf and sheath blight · Grain yield · High plant density · Maize · Maydis leaf blight

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## Introduction

Maize has the highest potential for grain yield among all other crops of the grass family. Though the introduction of single-cross hybrids led to an improvement in maize yields significantly, genetic gains are still low particularly under marginal environments in the developing world (Chakradhar *et al.*, 2017). Globally, the USA is the highest producer of maize followed by China, Brazil, the European Union, Ukraine, Argentina, India and Mexico (Anonymous, 2019). In the USA, since the 1930s, the drastic increase in maize grain yield per unit land area has been attributed to an increase in plant density per unit area. Many studies have suggested that yield potential has not been changed in terms of yield per plant rather stress tolerance in plants has been increased for obtaining high yield potential under a wider range of environmental conditions (Tokatlidis and Koutroubas, 2004). Increasing plant density includes lodging resistance, reduced angle of leaf, reduced anthesis to silking interval and smaller architecture of tassel (Leivar *et al.*, 2012). Lower height of plant and ear and a strong stem can reduce lodging and is favourable for high plant density. Upright leaf allows the plant to intercept light more efficiently in its entire canopy thus increasing the photosynthetic efficiency. The small and less leafy plant reduces the level of competition over the other plants (Sangoi and Salvador, 1998). Sass and Loeffel (1959) had implied that the primary cause of barrenness is the lengthening of anthesis to the silking interval. Therefore, shorter anthesis-silking interval is one of major attributes for proper seed setting.

High-plant density has its limitations as well. Planting at high density increases interplant competition for resources and negatively affects final yield. This might be due to apical dominance, barrenness, reduced number of ears per plant and reduced kernels set per ear (Sangoi and Salvador, 1998). Earlier studies in this domain indicated that crowding stress reduced the ability of plants to use soil N prominently during the post-silking period (Yan *et al*., 2017). The increased incidence of biotic stresses has also been indicated (Yu *et al.*, 2017). They reported that high plant density corresponds to reduced distance between individual maize plants and adversely affect the ventilation and light conditions of maize canopies, which facilitate the transmission of spores of pathogens. In addition, the ability of maize roots to absorb nutrients is reduced under high plant density; limited nutrient uptake decreases the soluble sugar content and physiological activity of stalk, thereby reducing the resistance of stalk to pathogens (Anderson and White, 1994; Xue *et al.*, 2016). In short, regardless of whether maize varieties are resistant or susceptible to disease, high plant density increases the mortality and degree of stem rot and sheath blight (Thomison *et al.*, 2011). Despite all the constraints, it is expected that high density apposite inbred lines would generate high-density responsive hybrids. Al- Naggar and Atta (2017) emphasized that hybrids developed under low plant population do not perform well under higher plant population. The present study was aimed at the identification of efficient inbred lines that harbour the ability to tolerate crowding stress in terms of grain yield and prevalent leaf blights of *Kharif* season.

## Materials and methods

A set of diverse 20 inbred lines (Table 1), were planted in randomized block design with three replications in two environments: normal plant density (NPD) comprising 33,333 plants/acre (at a recommended row to row and plant to plant spacing of 60 cm and 20 cm, respectively) and high-plant density (HPD) comprising 43,333 plants/ acre (at row to row and plant to plant spacing of 60 cm and 15 cm, respectively). The experiment was carried out at Field Experimental Area, Department of Plant Breeding and Genetics, PAU, Ludhiana across two years during *Kharif* 2019 and 2020. The lines were screened against maydis leaf blight (MLB) and banded leaf and sheath blight (BLSB) under natural conditions. Standard disease rating scale (1-9) was followed for recording the severity of maydis leaf blight and banded leaf and sheath blight under two planting regimes and the genotypes screened were grouped into four categories *viz.*, resistant, moderately

**Table 1.** List of test maize inbreds with pedigree used for assessing yield performance under different planting regimes

| S.No. | Entry | Pedigree |
|---|---|---|
| 1 | LM 6 | MS Pool $C_2$ $IC_2$-5-1-2-1-1-2-1-1---f |
| 2 | LM 13 | JCY 3-7-1-1-1---f |
| 3 | LM 14 | CA 00 310-1-1-1 |
| 4 | LM 21 | LM21-6-3-1-2-1--f |
| 5 | LM 25 | SE 563 |
| 6 | PML 41 | (Tux $C_2$ $IC_2$-5...x Pop24)-10-2-1-1-4-1-1-1-1-1-1-1-1-1-1 |
| 7 | PML 243 | HS-2785-5-1-(2)-4-#-1- --f |
| 8 | PML 333 | CM 122-1-1-1 |
| 9 | PML 368 | LM 13 Selection N/G -4-1-1-1-1 |
| 10 | PML 387 | [{Tux 162xLM 5-6-1....x LM 5.Pop 24-4-4..}x LM 13]BC 2-1- 3-#-1 |
| 11 | PML 503 | (SE 563 x LM 13)-1-1-1-2-4--f |
| 12 | PML 892 | 900 MG-x-x-1-2-3-1---f |
| 13 | PML 920 | NSX082012-x-1-1-2-4-2--f |
| 14 | PML 1012 | LM6-$BC_3$ waxy S 5-182-3-3--1-1--f |
| 15 | PML 1224 | WNC DMR 10R YFWS 1206 |
| 16 | PML 1225 | WNC-DMR 10R YFWS 8337-1-1-2-11-f |
| 17 | PML 1229 | WNC DMR 10R YFWS 8344-2-2-1-1--f |
| 18 | PML 1230 | SW 93-D-313-23 Pop 49-2-1-1-3-3 f |
| 19 | PML 1231 | SMH 3900-#-1-2-3-2-1----f |
| 20 | CML 451 | ((NPH-28-1/G25)/NPH-28-1)-1-2-1-1-3-1-B |

resistant, moderately susceptible and susceptible based on their disease reaction (Hooda *et al.*, 2018). Yield per plot (Y/plot; in kilogram) was recorded in terms of ear weight per plot (also called field weight per plot) immediately after crop harvest. Ear weight per plot was converted into grain yield after accounting for grain moisture content (to be recorded in the field) and shelling percentage. Statistical analysis of phenotypic data was conducted using SAS software (SAS Institute 2011).

## Results and discussion

*Response in maize inbreds to grain yield under different planting regimes*

The characterization of maize inbred lines for grain yield over years under different planting regimes is depicted in Table 2. In 2019, mean grain yield was 6.70 and 7.49 under NPD and HPD, respectively whereas in 2020, the mean grain yield was 6.64 under NPD and enhanced to 8.03 under HPD, leading to per cent average increase of 11.63 per cent in 2019 and 20.95 per cent in 2020. The overall yield performance across years and planting regimes have identified five top performing maize inbreds *viz.*, PML 1224, PML 1012, PML 1225, PML 387 and PML 1231 (> 7.8 q/acre grain yield) which can be utilized as parents with high seed potential. The grain yield of these identified parental lines was > 8.5 quintal per acre under HPD with the highest grain yield of 9.15 q/acre in PML 1224. The mean per cent yield increment (YD %) across years ranged from 6.13 (LM 6) to 29.22 (PML 503) as depicted in Table 2. Five inbred lines (PML 368, PML 503, PML 892, PML 1231 and LM 25) exhibited more than 20 per cent increase in yield under HPD when

**Table 2.** Profiling of maize inbred lines for mean grain yield under normal plant density (NPD) and high plant density (HPD) during *Kharif* 2019 and 2020 and per cent increase in yield (YD %) under HPD

| Inbred | Grain yield (q/acre) | | | | |
|---|---|---|---|---|---|
| | *Kharif* 2019 | | *Kharif* 2020 | | YD % under HPD across years |
| | NPD | HPD | NPD | HPD | |
| LM 6 | 5.8 | 6.2 | 5.6 | 5.9 | 6.13 |
| LM 13 | 6 | 6.6 | 6.8 | 8.1 | 14.56 |
| LM 14 | 7.5 | 7.5 | 7 | 7.9 | 6.43 |
| LM 21 | 5.9 | 6.7 | 5.8 | 7 | 17.12 |
| **LM 25** | **6.4** | **7.5** | **6.1** | **7.7** | **21.71** |
| PML 41 | 7 | 7.9 | 6.9 | 8.3 | 16.57 |
| PML 243 | 6.4 | 6.6 | 6.9 | 8 | 9.53 |
| PML 333 | 6 | 6.9 | 5.9 | 6.9 | 15.97 |
| **PML 368** | **6.2** | **7.7** | **6.8** | **8.6** | **25.33** |
| PML 387 | 7.7 | 8.2 | 6.8 | 8.7 | 17.22 |
| **PML 503** | **6.2** | **8** | **6.8** | **8.8** | **29.22** |
| **PML 892** | **7.2** | **8.3** | **6.8** | **8.6** | **20.87** |
| PML 920 | 5.8 | 6.2 | 6.8 | 8.3 | 14.48 |
| PML 1012 | 8 | 9.3 | 7.4 | 8.6 | 16.23 |
| PML 1224 | 8.9 | 9.2 | 7 | 9.1 | 16.69 |
| PML 1225 | 7.6 | 8.9 | 6.9 | 8.3 | 18.70 |
| PML 1229 | 5.9 | 6.3 | 6.2 | 7.3 | 12.26 |
| PML 1230 | 6 | 6.4 | 5.9 | 6.8 | 10.96 |
| **PML 1231** | **6.8** | **8.1** | **7.1** | **9.2** | **24.35** |
| CML 451 | 6.8 | 7.2 | 7.2 | 8.4 | 11.27 |
| Mean | 6.70 | 7.49 | 6.64 | 8.03 | |
| SE | 0.45 | 0.66 | 0.50 | 0.78 | |
| CD (1%) | 1.22 | 1.78 | 1.35 | 2.11 | |
| CD (5%) | 0.91 | 1.33 | 1.01 | 1.57 | |
| CV (%) | 8.21 | 10.72 | 9.21 | 11.86 | |

compared to NPD. Figure 1 depicted the average per cent increase in yield across years under HPD. A wide variability in YD % indicated the wide scope of selection of inbred lines to develop hybrids amenable to HPD. Lower plant density delay canopy closure and decrease light interception, which leads to higher yield per plant but lower yield per unit area (Andrade *et al*., 1999) while, high plant density enhances interplant competition for water, nutrients and assimilates (Edmeades *et al*., 2000). High plant density also increases the anthesis-silking interval (ASI) and stimulates barrenness (Sangoi, 2001), which leads to reduced kernel number per unit area, the main maize-yield component.

As the increase in number of plants per unit area imposes crowding stress on inbreds, grain yield based stress tolerance index (STI) has been worked out. The Figure 2 depicted the variability in response of test inbreds to STI ranging 0.78 to 1.64. Amongst the top l5 lines which showed more than 15 YD %, 13 lines were having STI more than 1.10, inferring STI served as a good indicator to characterize lines for crowding stress tolerance. Inbred lines viz., PML 387, PML 892, PML 1012, PML 1224, PML 1225 and PML 1231 were showing maximum value of STI, thereby exhibiting good level of plant density tolerance as compared to other test inbred lines.

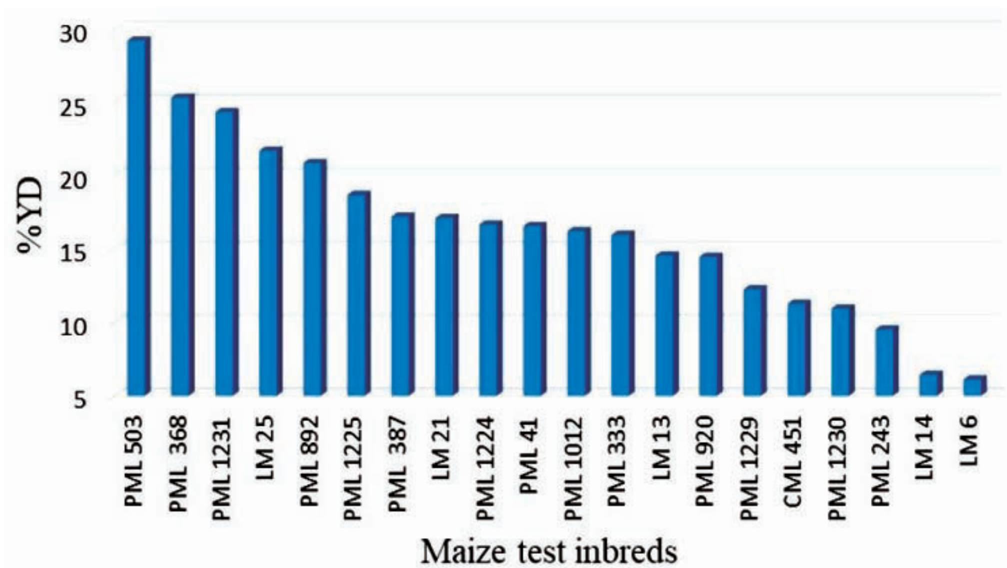



**Figure 1.** Percent yield increase (%YD) under high plant density

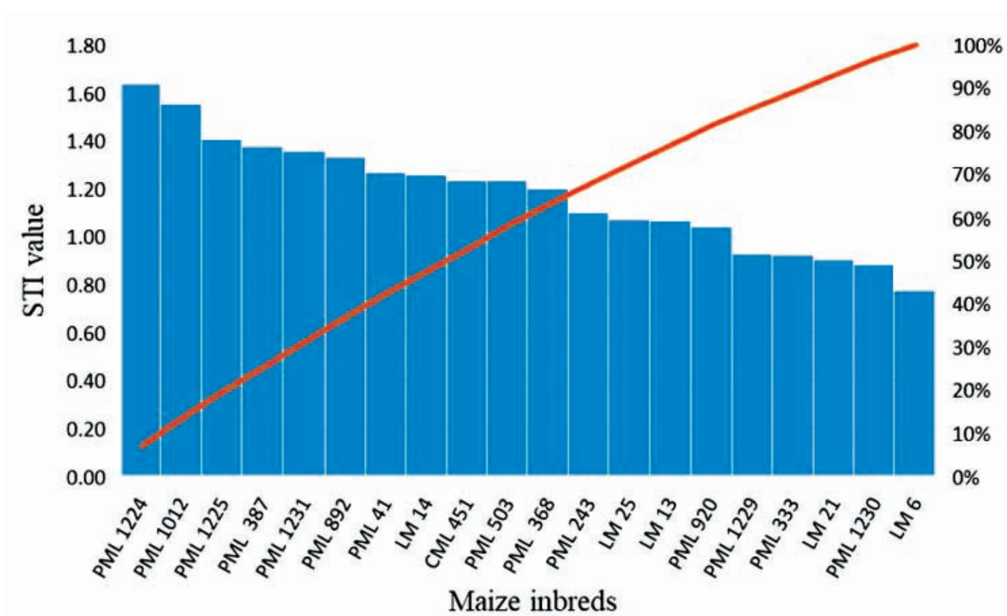



**Figure 2.** Profiling of maize inbreds for grain yield stress tolerance index (STI)

*Response in maize inbreds to disease attributes under different planting regimes*

*Maydis leaf blight*

Data in Table 3 showed that under NPD, 14 maize lines with disease rating scale ranging from 2.5-3.0 showed resistant reaction and 5 lines with 3.5-4.0 disease rating scale gave moderately resistant reaction to maydis leaf blight. Only one line PML 1224 having 5.5 severity scale was found moderately susceptible. On comparing MLB severity in two planting regimes, maize lines showed comparatively higher disease incidence in HPD than under NPD. Under HPD, 7 and 10 genotypes gave resistant and moderately resistant reaction, respectively (Figure 3). However, two genotypes-PML 333 and PML 1232 gave moderately susceptible and one genotype- PML 1224 showed susceptible reaction. Out of these 20 lines, 4 lines- LM 21, LM 13, LM 14 and PML 41 did not show any increase in MLB severity under HPD condition. Four inbreds viz. LM 25, PML 368, PML 387 and PML 243 gave ≤ 20 per cent increase in MLB severity under HPD condition.

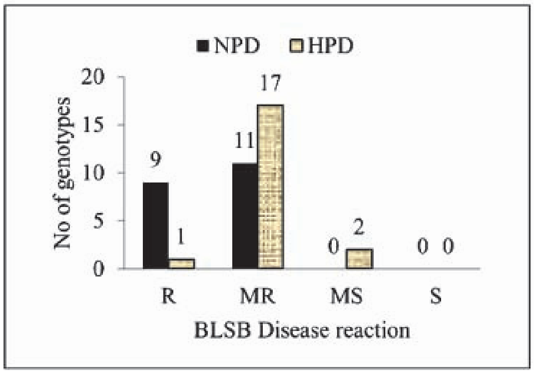
*Banded leaf and sheath blight*

Data in Table 3 showed that under HPD, only one line – PML 1229 gave resistant reaction; however, under NPD, nine lines gave resistant reaction to BLSB of maize. Large proportion fall in moderately resistant group with 17 and 11 lines under HPD and NPD conditions, respectively. Two lines- PML 1224 and CML 451 gave moderately susceptible reaction with severity scale of 5.2 under HPD conditions (Figure 3). There was increase in severity of BLSB in all maize lines when planted under high plant density conditions. Six lines viz. LM 6, PML 1224, PML 503, LM 14, PML 892 and PML 1229 gave ≤ 20 per cent severity increase in BLSB severity under HPD conditions (Table 3). Maize crop is subjected to extensive yield losses due to biotic stresses at different growth stages during monsoon season. To compensate the yield loss, high plant density is a good strategy to meet the current and future food necessities of high population and their rising dietary needs. Though previous studies have shown that densification may lead to increase in disease incidence.

**Table 3.** Per cent increase in disease severity of maydis leaf blight and banded leaf and sheath blight in maize inbreds under normal (NPD) and high plant density (HPD) during *Kharif* season

| S.No. | Entry | Maydis leaf blight severity | | | Banded leaf and sheath blight severity | | |
|---|---|---|---|---|---|---|---|
| | | NPD (1–9)* | HPD (1–9) | Percent increase | NPD (1–9) | HPD (1–9) | Percent increase |
| 1 | LM 6 | 2.5 | 3.5 | 40.0 | 3.0 | 3.5 | 16.7 |
| 2 | LM 13 | 2.5 | 2.5 | 0.0 | 3.0 | 4.0 | 33.3 |
| 3 | LM 14 | 2.5 | 2.5 | 0.0 | 4.5 | 5.0 | 11.1 |
| 4 | LM 21 | 2.5 | 2.5 | 0.0 | 3.2 | 4.5 | 40.6 |
| 5 | LM 25 | 3.0 | 3.5 | 16.7 | 3.5 | 4.5 | 28.6 |
| 6 | PML 41 | 3.5 | 3.5 | 0.0 | 3.5 | 4.5 | 28.6 |
| 7 | PML 243 | 2.5 | 3.0 | 20.0 | 3.5 | 4.5 | 28.6 |
| 8 | PML 333 | 4.0 | 6.0 | 50.0 | 2.5 | 4.0 | 60.0 |
| 9 | PML 368 | 3.0 | 3.5 | 16.7 | 3.5 | 4.5 | 28.6 |
| 10 | PML 387 | 2.5 | 3.0 | 20.0 | 3.0 | 4.0 | 33.3 |
| 11 | PML 503 | 2.0 | 3.0 | 50.0 | 3.0 | 3.5 | 16.7 |
| 12 | PML 892 | 2.5 | 3.5 | 40.0 | 3.0 | 3.5 | 16.7 |
| 13 | PML 920 | 4.0 | 5.0 | 25.0 | 3.3 | 4.5 | 38.5 |
| 14 | PML 1012 | 3.5 | 4.5 | 28.6 | 2.5 | 4.0 | 60.0 |
| 15 | PML 1224 | 5.5 | 7.5 | 36.4 | 4.5 | 5.2 | 15.6 |
| 16 | PML 1225 | 3.0 | 4.0 | 33.3 | 3.0 | 5.0 | 66.7 |
| 17 | PML 1229 | 2.5 | 4.5 | 80.0 | 2.5 | 3.0 | 20.0 |
| 18 | PML 1230 | 2.5 | 2.5 | 0.0 | 3.5 | 4.5 | 28.6 |
| 19 | PML 1231 | 4.0 | 5.5 | 37.5 | 3.5 | 4.5 | 28.6 |
| 20 | CML 451 | 3.0 | 4.5 | 50.0 | 3.5 | 5.2 | 48.6 |

*1–3.0: Resistant; 3.1–5.0: Moderately resistant; 5.1–7.0: Moderately susceptible; 7.1–9.0: Susceptible



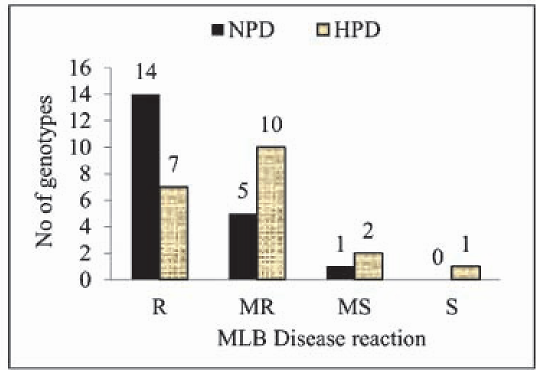




**Figure 3.** Disease reaction of maize inbreds to (a) banded leaf and sheath blight and (b) maydis leaf blight under normal plant density (NPD) and high plant density (HPD) conditions during *kharif* season

In our study, most of the maize inbreds tested have shown comparatively higher disease under HPD than NPD regime. Further, preliminary results reported three lines viz. PML 387, PML 503 and PML 892 giving moderately resistant reaction to maydis leaf blight and banded leaf and sheath blight under HPD but need further confirmation under artificial screening too. These parental lines with multiple resistance can be well utilized to generate desired resistant commercial hybrids.

Hence, this study indicated that prior selection of parental lines and their evaluation under HPD is a prerequisite for the development of hybrids for HPD. On the basis of seed yield performance and response against MLB and BLSB, maize inbreds PML 387, PML 503 and PML 892 were suggested as potential donors to generate HPD amenable hybrids. However, the significant increase in productivity of inbreds needs to be validated with multi-site screening and nutrient management for commercial level utilization. PML 1229 exhibiting resistant reaction for BLSB under HPD may serve as good genetic resource after confirmation through artificial screening.

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RESEARCH PAPER

# Evaluation of canopy temperature depression and relative water content as indicators for drought tolerance in maize (*Zea mays* L.) inbreds

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**Abstract:** Drought tolerance is a complex trait involving morphological and biochemical mechanisms. Physiological changes due to moisture stress, which reflect an adaptive mechanism in genotype, are worth measuring for relative assessment of differences. Considering this, the present study was conducted to determine the effect of water stress on canopy temperature depression (CTD) and relative water content (RWC) to evaluate drought tolerance of maize inbreeds. Thirty maize inbreeds were evaluated in greenhouse experiment based on completely randomized design with three replications at two irrigation levels (well-watered and low moisture-stressed). Inbreeds CML-470, DMR-N6, CML-415 and LM-12 had cooler canopies under drought conditions. The difference between CTD under irrigated and drought conditions was small initially (1.20) but increased progressively at 2nd week of stress imposition (1.70) and was highest at 3rd week of stress imposition (2.70) in all maize genotypes. High CTD can be used as a selection criterion to improve tolerance to drought and heat. RWC under drought conditions recorded the highest value in DMR-N6 followed by L-9 and V-335. The rate of RWC in plant with high resistance against drought was higher than others. In other words, plant having higher yields under drought stress should have high RWC. Relative water content can be said to be a good parameter suitable to screening drought-tolerant maize genotypes.

**Keywords:** Canopy temperature depression · Drought tolerance · Maize inbreeds · Relative water content

## Introduction

Maize (*Zea mays* L.) is a common staple food in many countries, especially those in the tropics and subtropics. In terms of acreage and production, maize is the third most popular food grain crop in the world. It grows successfully in low-land, tropical, sub-tropical, and temperate climates around the world as a versatile crop (Elias, 1995). Maize is recognized as the "Queen of Cereals" around the world because it has the highest genetic yield potential of all cereals. Maize production is hampered by a number of abiotic stresses causes in various places of the world. Drought is one of the most prominent factors restricting maize output and productivity among them (Araus *et al*., 2002). It is the most significant constraint to maize production potential realization.

Drought stress has a negative impact on maize output around the world (Feller *et al*., 2014; Wu *et al*., 2017). The drought tolerance in maize is a viable solution to the problem of yield loss induced by drought stress. The drought tolerant lines are being developed by breeders and agronomists, and their properties are being studied. As a result, elucidating the processes of maize response to drought stress and adaptability in order to fulfill rising food demands is critical. Drought stress causes plants to exhibit a variety of morphological, physiological, and biochemical

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responses, including a deep root system, small leaves, and a range of drought-resistance mechanisms, including reduced water loss, and increased its uptake, reduced transpirational losses, and sustained high leaf tissue water potential, as well as higher chlorophyll (Chl) contents and photosynthetic efficiency (Farooq *et al*., 2009, 2012; Liu *et al*., 2015). On the one hand, drought-tolerant lines must improve their roots' ability to absorb water from the soil. The root system of maize can minimize its investigation of the surface soil and grow deeper soil by reducing the number and increasing the volume of each cortical cell, allowing maize to obtain more water from soil (Chimungu *et al*., 2014). Drought tolerance, on the other hand, can be improved by reducing leaf density and managing stomata opening.

Lower leaf or canopy temperatures imply increased potential to absorb soil moisture or sustain a better water status for the plant. The degree to which transpiration cools leaves under a severe environmental load, leading to greater stomatal conductance and net photosynthesis, is shown by canopy temperature in relation to air temperature (Araus *et al*., 2008). Because of the large natural variation in crops and its link with yield under both stress and non-stress circumstances, canopy temperature depression has emerged as a viable substitute (Sofi *et al*., 2019). Relative water content (RWC) properly reflects cell volume and reveals the balance between absorbed and consumed water through transpiration, making it a useful indicator of plant water status in terms of the physiological consequences of cellular water shortage. For carbon fixation, maize uses the NADP-malic enzyme-dependent C4-dicarboxylic acid pathway. The C4 route, on the other hand, offers no special ability to endure low leaf water potential, which was previously utilized as a reliable predictor of drought-tolerant triticale and maize genotypes (Grzesiak *et al*., 2006).

## Materials and methods

The present study was conducted at the green house facility of the Division of Genetics and Plant Breeding, Faculty of Agriculture Wadura, SKUAST-K Sopore. Thirty inbreds of maize *viz.*, L-1, L-2, L-9, L-18, L-6, L-10, L-8, HKI- 101, CML-129, HKi-1015-W8, CML-470, L-72, CML-488, CML-167, LM-14, DMR-N6, CML-135, CML-415, LM-12, CML-139, CML-425, CML-286, CML-474, V-338, V-5, V-412, V-351, V-405, V-400, V-335 were evaluated in well-watered and water-deficit conditions.

Canopy temperature was measured using a hand-held infrared thermometer (Fluke 68 Max, Fluke Corporation USA) at five inches from leaf surface inclined at 45 degrees in three stages *viz.*, two-, three- and four-week intervals, after imposition of stress between 10 am-2 pm. Three readings per replications were recorded and averaged out. Canopy temperature depression was calculated as difference of air temperature and canopy temperature.

$$CTD = AT - CT$$

Where,
CTD = Canopy temperature depression
AT = Air temperature
CT = Canopy temperature

Relative water content (RWC) estimates the current hydration of leaf tissue to its maximum potential hydration of each genotype was measured at two weeks after stress imposition from top most fully expanded leaves. Relative water content was measured at noon (between 11.00-1.00 noon). Four leaves were taken from each replication from both water stressed and well-watered treatments and were weighed immediately to obtain their fresh weight. Then the leaves were submerged in distilled water in beakers till saturation for 6 h, after which the turgid weight was measured. Afterwards the leaves were subjected to drying at 60°C for 72 h to determine their dry weights. From these data sets *viz.*, FW, TW and DW, RWC was calculated as follows and expressed as percentage (Barr and Weatherley, 1962).

$$RWC\ (\%) = \frac{FW - DW}{TW - DW} \times 100$$

Where,
RWC = Relative water content
FW = Fresh weight
DW = Dry weight
TW = Turgour weight

## Results

Thirty maize inbred lines were evaluated to study the effect on CTD under drought and irrigated conditions in green house conditions and the comparative performance of genotypes for CTD under drought and irrigated conditions is presented in Table 1. Observations were taken at two, three and four weeks after stress (WAS).

**Table 1.** Canopy temperature depression across three growth stages under irrigated and drought conditions in maize genotypes

| Inbreds | Irrigated | | | Sub Mean | Drought | | | Sub Mean | Factor Mean | | Mean across irrigated conditions | Mean across drought conditions | Change under drought |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | CTD2 | CTD3 | CTD4 | | CTD2 | CTD3 | CTD4 | | Inbreds | Stages | Im | Dm | Im-Dm |
| L-1 | 3.60 | 2.50 | 1.70 | 2.60 | 2.90 | 0.30 | -1.20 | 0.67 | 1.63 | 2.90 | 2.60 | 0.67 | 1.93 |
| L-2 | 2.60 | 2.00 | 1.50 | 2.00 | 2.30 | 0.80 | -1.50 | 0.53 | 1.27 | 1.60 | 2.00 | 0.53 | 1.47 |
| L-9 | 3.20 | 3.00 | 2.20 | 2.80 | 2.10 | 0.70 | -1.90 | 0.30 | 1.55 | -0.35 | 2.80 | 0.30 | 2.50 |
| L-18 | 2.60 | 1.50 | 1.30 | 1.80 | 1.70 | 1.10 | -1.20 | 0.53 | 1.16 | | 1.80 | 0.53 | 1.27 |
| L-6 | 4.00 | 3.20 | 2.30 | 3.20 | 2.70 | -0.40 | -2.30 | 0.00 | 1.58 | | 3.20 | 0.00 | 3.20 |
| L-10 | 2.70 | 2.10 | 0.30 | 1.70 | 1.60 | 0.70 | -1.90 | 0.13 | 0.91 | | 1.70 | 0.13 | 1.57 |
| L-8 | 4.20 | 3.00 | 1.70 | 3.00 | 2.90 | 0.60 | -1.40 | 0.70 | 1.83 | | 3.00 | 0.70 | 2.30 |
| HKI-101 | 4.02 | 3.60 | 1.50 | 3.00 | 2.70 | 0.90 | -2.20 | 0.47 | 1.74 | | 3.00 | 0.47 | 2.53 |
| CML-129 | 3.30 | 2.20 | 0.60 | 2.00 | 1.90 | 1.20 | -1.60 | 0.50 | 1.26 | | 2.00 | 0.50 | 1.50 |
| HKI-1015-W8 | 2.70 | 1.90 | 0.70 | 1.80 | 1.60 | 0.60 | -2.30 | -0.03 | 0.87 | | 1.80 | -0.03 | 1.83 |
| CML-470 | 4.40 | 3.70 | 0.80 | 3.00 | 3.60 | 1.70 | -1.30 | 1.33 | 2.15 | | 3.00 | 1.33 | 1.67 |
| L-72 | 3.80 | 2.40 | 0.40 | 2.20 | 2.50 | 0.70 | -1.80 | 0.47 | 1.33 | | 2.20 | 0.47 | 1.73 |
| CML-488 | 4.40 | 3.60 | 1.20 | 3.10 | 2.40 | 1.60 | -2.00 | 0.67 | 1.87 | | 3.10 | 0.67 | 2.43 |
| CML-167 | 3.30 | 2.60 | 1.20 | 2.40 | 1.30 | 0.70 | -2.60 | -0.20 | 1.08 | | 2.40 | -0.20 | 2.60 |
| LM-14 | 2.70 | 2.30 | 1.60 | 2.20 | 1.80 | 0.50 | -0.60 | 0.57 | 1.38 | | 2.20 | 0.57 | 1.63 |
| DMR-N6 | 3.60 | 2.50 | 1.20 | 2.40 | 2.30 | 1.50 | -0.80 | 1.00 | 1.71 | | 2.40 | 1.00 | 1.40 |
| CML-135 | 2.70 | 2.00 | 0.50 | 1.70 | 1.70 | 0.70 | -2.10 | 0.10 | 0.91 | | 1.70 | 0.10 | 1.60 |
| CML-415 | 4.80 | 2.60 | 0.30 | 2.60 | 2.70 | 1.00 | -1.40 | 0.77 | 1.67 | | 2.60 | 0.77 | 1.83 |
| LM-12 | 3.20 | 2.30 | 0.70 | 2.10 | 2.00 | 0.50 | -0.20 | 0.77 | 1.42 | | 2.10 | 0.77 | 1.33 |
| CML-139 | 4.50 | 3.50 | 1.20 | 3.10 | 2.60 | -1.30 | -1.70 | -0.13 | 1.47 | | 3.10 | -0.13 | 3.23 |
| CML-425 | 2.30 | 1.40 | 0.50 | 1.40 | 1.50 | 0.70 | -2.60 | -0.13 | 0.63 | | 1.40 | -0.13 | 1.53 |
| CML-286 | 4.80 | 2.80 | 0.80 | 2.80 | 2.60 | 0.50 | -1.20 | 0.63 | 1.71 | | 2.80 | 0.63 | 2.17 |
| CML-474 | 3.20 | 2.40 | 0.50 | 2.00 | 2.30 | 0.30 | -3.00 | -0.13 | 0.94 | | 2.00 | -0.13 | 2.13 |
| V-338 | 3.20 | 2.20 | 0.30 | 1.90 | 2.40 | 0.60 | -2.40 | 0.20 | 1.05 | | 1.90 | 0.20 | 1.70 |
| CML-451 | 3.70 | 2.90 | 1.00 | 2.50 | 2.90 | 0.20 | -1.70 | 0.47 | 1.49 | | 2.50 | 0.47 | 2.03 |
| V-412 | 2.40 | 1.30 | 0.60 | 1.40 | 1.60 | 0.60 | -1.30 | 0.30 | 0.86 | | 1.40 | 0.30 | 1.10 |
| V-351 | 4.00 | 2.80 | 1.00 | 2.60 | 2.80 | 0.60 | -2.40 | 0.33 | 1.46 | | 2.60 | 0.33 | 2.27 |
| V-405 | 3.80 | 2.10 | 0.70 | 2.20 | 2.10 | 0.40 | -0.80 | 0.57 | 1.38 | | 2.20 | 0.57 | 1.63 |
| V-400 | 3.70 | 2.50 | 0.50 | 2.20 | 2.50 | 0.70 | -2.50 | 0.23 | 1.22 | | 2.20 | 0.23 | 1.97 |
| V-335 | 2.80 | 1.50 | 0.40 | 1.60 | 1.60 | 0.50 | -2.00 | 0.03 | 0.80 | | 1.60 | 0.03 | 1.57 |
| Mean | **3.50** | **2.50** | **1.00** | **2.30** | **2.30** | **0.64** | **-1.70** | **0.39** | | | **2.30** | **0.39** | **1.92** |

**CD ($p<0.05$)** Regimes = 0.02; Stages = 0.03; Genotypes = 0.10; Regimes*Stages =0.04; Regimes*Genotypes = 0.15; Stage*Genotypes = 0.18; Regimes*Stages*Genotypes = 0.26

CTD at S1 under irrigated conditions had mean value of 3.50 with highest value recorded in CML-415 and CML-286 (4.80) followed by CML-139 (4.50) and CML-470 and CML-488 (4.40) and was lowest in CML-425 (2.30) while as under drought it had mean value of 2.30 with highest value recorded in CML-470 (3.60) followed by L-1, L-8, CML-451 (2.90) and V-351 (2.80) and was lowest in CML-167 (1.30).

CTD at S2 under irrigated conditions had mean value of 2.50 with highest value recorded in CML-470 (3.70) followed by HKI-101, CML-488 (3.60) and CML-139 (3.50) and was lowest in V-412 (1.30) while as under drought conditions it had mean value of 0.64 with highest value recorded in CML-470 (1.70) followed by CML-488 (1.60) and DMR-N6 (1.50) and was lowest in CML-139 (-1.30).

**Table 2.** ANOVA for CTD in maize inbreeds lines

| Source of variation | DF | CTD |
|---|---|---|
| Genotype | 29 | 2.377 |
| Water regime | 1 | 71.983 |
| Stages | 2 | 117.373 |
| Genotype × Regime | 29 | 0.911 |
| Genotype × Stage | 58 | 0.931 |
| Regime × Stage | 2 | 77.378 |
| Genotype × Regime × Stage | 58 | 0.638 |
| Error | 360 | 0.265 |

Significant at 5% level

CTD at S3 under irrigated conditions had mean value of 1.0 with highest value recorded in L-6 (2.3) followed by L-9 (2.2) and L-1, L-8 (1.7) and was lowest in L-10, CML-415, V-338 (0.3) while as under drought conditions it had mean value of -1.7 with highest value recorded in LM-12 (-0.2) followed by LM-14 (-0.6) and DMR-N6, V-405 (-0.8) and was lowest in CML-167, CML-425 (-2.6).

Analysis of variance for canopy temperature depression across three growth stages under irrigated and drought conditions is presented in Table 2 which shows that mean square due to genotypes was significant for CTD. Thirty maize inbreeds genotypes were evaluated to study the effect on relative water content under irrigated and drought conditions in green house and the comparative performance of genotypes for RWC under irrigated and drought conditions is presented in Table 3. Readings were taken at two weeks after stress (WAS).

Under irrigated conditions, RWC had a mean value of 60.85 with highest value in V-412 (85.7) followed by L-1 (78.5) and CML-135, V-351 (78.4) and lowest in V-405 (25.1) followed by DMR-N6 (42). Under drought conditions it had a mean value of 48.31 with highest value in DMR-N6 (78.2) followed by L-9 (77.7) and V-335 (67.7) and lowest in HKI-1015-W8 (22.8) followed by L-8 and CML-129 (30.2). Per cent reduction under drought was highest in CML-129 (36.6) followed by V-400 (34.6) and V-351 (34.1) and lowest in V-405 (-36.4) followed by DMR-N6 (-36.2).

Analysis of variance for relative water content under irrigated and drought conditions is presented in Table 4 which shows that mean square due to genotypes, water regime and interaction genotype × water regime was significant.

**Table 3.** Mean performance of maize inbreeds for relative water content

| Inbreeds | Relative water content (%) | | |
|---|---|---|---|
| | Irrigated | Drought | Percent change |
| L-1 | 78.50 | 54.50 | 24.00 |
| L-2 | 72.70 | 38.70 | 34.00 |
| L-9 | 71.90 | 77.70 | -5.80 |
| L-18 | 60.50 | 49.40 | 11.10 |
| L-6 | 57.20 | 50.00 | 7.20 |
| L-10 | 51.30 | 33.30 | 18.00 |
| L-8 | 54.80 | 30.20 | 24.60 |
| HKI-101 | 62.10 | 35.40 | 26.70 |
| CML-129 | 66.80 | 30.20 | 36.60 |
| HKI-1015-W8 | 52.90 | 22.80 | 30.10 |
| CML-470 | 61.50 | 50.40 | 11.10 |
| L-72 | 60.00 | 70.60 | -10.60 |
| CML-488 | 68.30 | 38.70 | 29.60 |
| CML-167 | 43.70 | 65.20 | -21.50 |
| LM-14 | 75.60 | 43.50 | 32.10 |
| DMR-N6 | 42.00 | 78.20 | -36.20 |
| CML-135 | 78.40 | 46.60 | 31.80 |
| CML-415 | 62.50 | 42.30 | 20.20 |
| LM-12 | 69.80 | 46.60 | 23.20 |
| CML-139 | 48.30 | 65.50 | -17.20 |
| CML-425 | 61.50 | 51.50 | 10.00 |
| CML-286 | 48.90 | 31.00 | 17.90 |
| CML-474 | 55.50 | 40.40 | 15.10 |
| V-338 | 51.90 | 48.00 | 3.90 |
| CML-451 | 55.50 | 39.30 | 16.20 |
| V-412 | 85.70 | 600 | 25.70 |
| V-351 | 78.40 | 44.30 | 34.10 |
| V-405 | 25.10 | 61.50 | -36.40 |
| V-400 | 70.40 | 35.80 | 34.60 |
| V-335 | 53.80 | 67.70 | -13.90 |
| Mean | 60.85 | 48.31 | 12.54 |

**CD (P< 0.05)** Genotypes = 1.830
Water regime = 0.473
Genotype × Water regime = 2.588

**Significant at 1% level and * Significant at 5% level

**Table 4.** Analysis of variance of maize inbreds for relative water content

| Source of variation | DF | RWC (%) |
|---|---|---|
| Genotype | 1 | 484.413** |
| Water regime | 29 | 7,077.425** |
| Genotype × Water regime | 29 | 659.201** |
| Error | 120 | 2.557 |

**Significant at 1% level and * Significant at 5% level

## Discussion

The current investigation found a significant variation in the CTD of irrigated and stressed maize inbreeds during three weeks. In all maize genotypes, the difference between CTD under irrigated and drought conditions was initially moderate (1.20), but rose gradually during the 2nd week of stress imposition (1.70) and was maximum at the 3rd week of stress imposition (2.70). As CTD represents an entire, integrated physiological response to drought and high temperature, so it can be used to monitor plant water status (Idso, 1982). CTD can be utilized as a selection criterion to improve drought and heat resistance. Talebi (2011) and Guendouz *et al*. 2012 obtained similar results in durum wheat, as did Kumar *et al*. (2016) in chickpea, and Dar *et al*. (2018) in maize.

As a result of deeper roots, cooler canopy temperatures have been linked to enhanced plant availability to water (Lopes and Reynolds, 2010). These researchers discovered that genotypes with cooler canopy temperatures yielded 30 per cent more and had a 40 per cent increase in root dry weight at 60-120 cm. CTD is used in practical applications such as plant response to environmental stress (Howell *et al*., 1986), irrigation scheduling (Wanjura *et al*., 1995; Evett *et al*., 1996), cultivar comparison for water use (Hatfield *et al*., 1987; Pinter *et al*., 1990) and heat (Reynolds *et al*., 1998) and drought tolerance (Reynolds *et al*., 1998) (Royo *et al*., 2002). Drought stressed plants displayed higher canopy temperatures than well-watered plants (Siddique *et al*., 2000). High CTD has been used as a selection criterion to improve tolerance to drought and heat (Amani *et al*., 1996; Ayeneh *et al*., 2002). The genotypes with negative values of CTD suggest that these genotypes are very sensitive to water stress. CTD is used a tool for predicting performance (Reynolds *et al*., 1998; Ayeneh *et al*., 2002). So, in the present evaluation inbreeds like CML-470, DMR-N6, CML-415 and LM-12 depicting low canopy temperature can be considered to be performing better under drought stresses.

V-412 had the highest RWC while V-405 had the lowest in irrigated conditions. Under drought conditions, however, DMR-N6 had the highest RWC value, whereas HKI-1015-W8 had the lowest. Under drought, CML-129 had the highest per cent decline in RWC, whereas V-405 had the lowest. RWC levels are higher in drought-tolerant plants than in others. In other words, plants with high RWC should have higher yields under drought stress. Drought-tolerant maize cultivars can be identified using relative water content as screening criteria. RWC was higher under irrigated conditions than drought, according to Velicevici *et al*. (2012). Leaf RWC is one of the most accurate growth/biochemical markers for determining the severity of stress (Alizade, 2002). The rate of RWC in drought-resistant plants is higher than in other plants. In other words, plants with high RWC should have higher yields under drought stress). According to Ludlow (1990), RWC is an alternative measure of plant water status that reflects metabolic activity in tissues and lethal leaf water status. Chen and colleagues (1990) determined that under water stress, water potential and relative water content dropped, and that relative water content in leaves reduced significantly in drought susceptible varieties compared to resistant varieties. According to Guang-hua Yin *et al*. (2012), relative water content varied significantly depending on soil water content and maize variety, but not with interactions.

RWC values that are higher under well-watered and water-stressed conditions imply that plant tissues can absorb and hold more water than tissues with low RWC values (Rosales *et al*., 2012). As a result, relative water content might be considered a useful measure for identifying drought-tolerant maize genotypes. Drought tolerance may be associated with genotypes that have a high RWC in this study.

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RESEARCH PAPER

# Studies on response of maize genotypes against PEG–6000 for various effects under drought stress

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**Abstract:** Drought is one of the most serious problems posing a grave threat to cereal production, including maize. The present study was conducted in the laboratory of the division of Genetics and Plant Breeding, Faculty of Agriculture, Wadura. Ten genotypes of maize were evaluated comprising of released composites in white and yellow group along with derived lines from crosses of GM-6, a drought tolerant composite. The present investigation was performed under artificially imposed drought stress by PEG-6000 at four levels of drought treatment {distilled water (0), 10, 15 and 20 per cent}. Results indicated significant differences among the genotypes and drought stress levels. Further, the results indicated that significant decrease was observed in primary root length, root biomass, seminal root number and lateral root number. Based on the results, genotypes KDM-111 followed by SMC-3 and DT-1 exhibited lesser reduction in total root length and KG-2 followed by SMC-3 and SMC-4 in root weight, GM-6 followed by KDM-111 and KG-2 in number of seminals and GM-6 followed by KDM-111 and C-15 in number of laterals. Hence, there was a progressive decline in all the parameters across all genotypes with an increase in PEG concentration from 0–20 per cent level, respectively.

**Keywords:** Drought · Genotype · Maize · PEG · Root

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## Introduction

Water stress is one among the major limiting factors in rainfed agriculture resulting in reduced crop growth and productivity (Ghassemi-Golezani *et al.*, 2009). Water is the basic requirement for all the growth stages of plant and it is also limited in some areas such as arid and semi-arid region of the world. Water is required by the plant for its growth and development. Without water the plant goes under drought condition and severally affects its growth stages and ultimately yield of crops is reduced. Maize (*Zea mays* L.) is one of the most versatile cereal crops having wider growth adaptability under varied agro-climatic conditions and also has the highest yield potential among the cereals. It is one of the important cereal crops in India after rice and wheat, grown in all the states. Also, it is a staple food and commercial crop which is sensitive to drought. The most important stage of crop is seed germination in the presence of water so, growing seeds under drought stress conditions, effects the growth of plant (Albuquerque *et al.*, 2003). The polyethylene glycol (PEG) has high molecular weight and is important for controlling germination of seed by creating osmotic stress in plants. The PEG inhibits seed germination by creating stress in plants (Sidari *et al.*, 2008). Several physiological characteristics have been reported as being reliable indicators for the selection of germplasm possessing drought tolerance. These characteristics include seed germination and seedling growth in nutrient solutions with low osmotic potential (Richards *et al.*, 1978; Blum *et al.*, 1981; Ashraf *et al.*, 1992). The success of these approaches requires evidence that the drought tolerance of cultivars tested under laboratory reflects this character under field conditions (Sammons *et al.*, 1978).

The aim of this study was to investigate the effects of osmotic stress generated by PEG-6000 on germination stage and water stress at seedling growth of maize genotypes. The primary objective of the present study was to screen out the most tolerant and most sensitive maize genotypes under drought stress.

## Materials and methods

The present study was conducted during 2017 at the green house faculty of the Division of Genetics and Plant Breeding, Faculty of Agriculture, Wadura, SKUAST-K Sopore. Ten genotypes of maize comprising of released composites in white and yellow group along with derived lines from crosses of GM-6 a drought tolerant composite were evaluated in the present study *viz.*, composite-8 (C-8), C-15, Kishan Ganga-2 (KG-2), Gujarat Makka-6 (GM-6), SMC-3, SMC-4, DT-1, DT-2, KDM- 111 and Mahi Dhawal, respectively. In case of experiment with polyethylene glycol, PEG-6000 (HIMEDIA) was used in four concentrations *viz.*, control (0%), 10, 15 and 20 per cent. Four seeds for each genotype were surface sterilized with 0.5 per cent NaOCl for one minute, rinsed thoroughly with distilled water and were put in petri plates containing moist filter paper with different concentrations of PEG and allowed to germinate in a germinator at 25$^{o}$C and 75 per cent humidity in darkness. Radicle length, root biomass, seminal root number and lateral root number was measured after seven days. The design used was completely randomized design with four replications.

## Results

*In vitro response of maize genotypes to PEG mediated screening*

The genotypes used in the present study were evaluated for response to PEG-6000 given in Table 1 at different levels (0, 10, 15 and 20%) and the results are given below under appropriate headings:

*1. Length of radical (cm)*

The data recorded under controlled conditions for length of radical under different levels of PEG-6000 are as under:

*0% level:* Under 0%, the length of radical had a mean value of 14.56 with the highest value recorded in DT-1 (17.50) followed by SMC-4 (17.50) and C-8 (16.75) and was lowest in KDM-111 (9.50) followed by KG-2 (12.75).

*10% level:* Under 10%, the length of radical had a mean value of 8.15 with highest value recorded in DT-1 (11.00) followed by GM-6 (9.50) and DT-2 (9.50) and the lowest was in SMC-3 (4.25) followed by KG-2 (7.00).

*15% level:* Under 15%, the length of radical had a mean value of 5.5 with highest value recorded in GM-6 (7.50) followed by SMC-4 (6.50) and DT-1 (6.00) and the lowest was in SMC-3 (3.50) followed by Mahi Dhawal (4.50).

*20% level:* Under 20%, the length of radical had a mean value of 3.97 with highest value recorded in DT-2 (5.00) followed by KG-2 (5.00) and GM-6 (4.50) and the lowest was in KDM-111 (2.75) followed by SMC-3 (3.00).

**Table 1.** Mean response of maize genotypes to PEG mediated screening

| Genotype | Total root length (cm) | | | | Root weight (g) | | | | Number of seminals | | | | Number of laterals | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | 0% | 10% | 15% | 20% | 0% | 10% | 15% | 20% | 0% | 10% | 15% | 20% | 0% | 10% | 15% | 20% |
| C 8 | 16.75 | 8.50 | 5.50 | 3.50 | 0.51 | 0.46 | 0.48 | 0.23 | 5.00 | 4.00 | 3.00 | 2.50 | 25.50 | 20.50 | 18.00 | 3.50 |
| C 15 | 15.25 | 9.00 | 5.50 | 3.75 | 0.52 | 0.53 | 0.44 | 0.33 | 6.50 | 5.50 | 3.50 | 3.00 | 27.50 | 22.50 | 9.50 | 2.00 |
| SMC 3 | 15.50 | 4.25 | 3.50 | 3.00 | 0.33 | 0.27 | 0.20 | 0.14 | 4.50 | 3.50 | 2.00 | 1.50 | 17.50 | 14.00 | 11.00 | 9.00 |
| SMC 4 | 17.50 | 7.75 | 6.50 | 4.50 | 0.56 | 0.50 | 0.47 | 0.22 | 6.00 | 4.50 | 2.50 | 1.50 | 17.50 | 13.00 | 12.00 | 7.50 |
| KG 2 | 12.75 | 7.00 | 5.50 | 5.00 | 0.49 | 0.22 | 0.20 | 0.11 | 3.50 | 2.500 | 1.50 | 1.00 | 22.00 | 20.00 | 12.50 | 4.50 |
| DT 1 | 17.50 | 11.00 | 6.00 | 3.50 | 0.67 | 0.63 | 0.59 | 0.39 | 9.50 | 7.500 | 6.00 | 4.50 | 34.00 | 29.50 | 22.50 | 5.50 |
| DT 2 | 13.50 | 9.50 | 5.50 | 5.00 | 0.63 | 0.48 | 0.54 | 0.36 | 8.50 | 6.500 | 4.50 | 4.00 | 32.00 | 20.50 | 26.500 | 6.50 |
| KDM 111 | 9.50 | 7.50 | 5.00 | 2.75 | 0.61 | 0.50 | 0.37 | 0.29 | 4.50 | 2.500 | 1.50 | 1.00 | 31.00 | 27.00 | 25.000 | 1.00 |
| GM 6 | 13.85 | 9.50 | 7.50 | 4.50 | 0.32 | 0.27 | 0.23 | 0.18 | 6.50 | 4.500 | 3.50 | 1.00 | 25.00 | 19.00 | 7.500 | 1.00 |
| Mahi Dhawal | 13.50 | 7.50 | 4.50 | 4.20 | 0.62 | 0.35 | 0.29 | 0.18 | 5.50 | 4.000 | 4.00 | 3.50 | 10.50 | 7.50 | 6.500 | 5.50 |
| Mean | 14.56 | 8.15 | 5.5 | 3.97 | 0.52 | 0.42 | 0.38 | 0.24 | 6.00 | 4.50 | 3.20 | 2.35 | 24.25 | 19.35 | 15.1 | 4.6 |
| CD ($p \leq 0.05$) | Genotype = 1.07<br>PEG Levels =0.68 | | | | Genotype = 0.04<br>PEG Levels = 0.03 | | | | Genotype = 0.74<br>PEG Levels = 0.47 | | | | Genotype = 2.14<br>PEG Levels = 1.35 | | | |

*2. Root biomass (g)*

The data recorded under controlled conditions for root biomass under different levels of PEG-6000 are as under:

*0% level:* Under 0%, the root biomass had a mean value of 0.52 with highest value recorded in DT-1 (0.67) followed by DT-2 (0.63) and Mahi Dhawal (0.62) and the lowest was in GM-6 (0.32) followed by SMC-3 (0.33).

*10% level:* Under 10%, the root biomass had a mean value of 0.42 with highest value recorded in DT-1 (0.63) followed by C-15 (0.53) and SMC-4 (0.50) and the lowest was in KG-2 (0.22) followed by GM-6 (0.27).

*15% level:* Under 15%, the root biomass had a mean value of 0.38 with highest value recorded in DT-1 (0.59) followed by DT-2 (0.54) and C-8 (0.48) and the lowest was in SMC-3 (0.20) followed by KG-2 (0.20).

*20% level:* Under 20%, the root biomass had a mean value of 0.24 with highest value recorded in DT-1 (0.39) followed by DT-2 (0.36) and C-15 (0.33) and the lowest was in SMC-3 (0.14) followed by GM-6 (0.18).

*3. Number of seminals*

The data recorded under controlled conditions for number of seminals under different levels of PEG-6000 are as under:

*0% level:* Under 0%, the number of seminals had the highest value recorded in DT-1 (9.50) followed by DT-2 (8.50) and C-15 (6.50) and the lowest was in KG-2 (3.50) followed by SMC-3 (4.50), with a mean value of 6.00.

*10% level:* Under 10%, the number of seminals had a mean value of 4.50 with highest value recorded in DT-1 (7.50) followed by DT-2 (6.50) and C-15 (5.50) and the lowest was in KG-2 (2.50) followed by KDM-111 (2.50).

*15% level:* Under 15%, the number of seminals had a mean value of 3.20 with the highest value recorded in DT-1 (6.00) followed by DT-2 (4.50) and Mahi Dhawal (4.00) and the lowest was in KG-2 (1.50) followed by KDM-111 (1.50).

*20% level:* Under 20%, the number of seminals had the highest value recorded in DT-1 (4.50) followed by DT-2 (4.00) and Mahi Dhawal (3.50) and was lowest in KDM-111 (1.00) followed by GM-6 (1.00), having a mean value of 2.35.

*4. Number of laterals*

The data recorded under controlled conditions for number

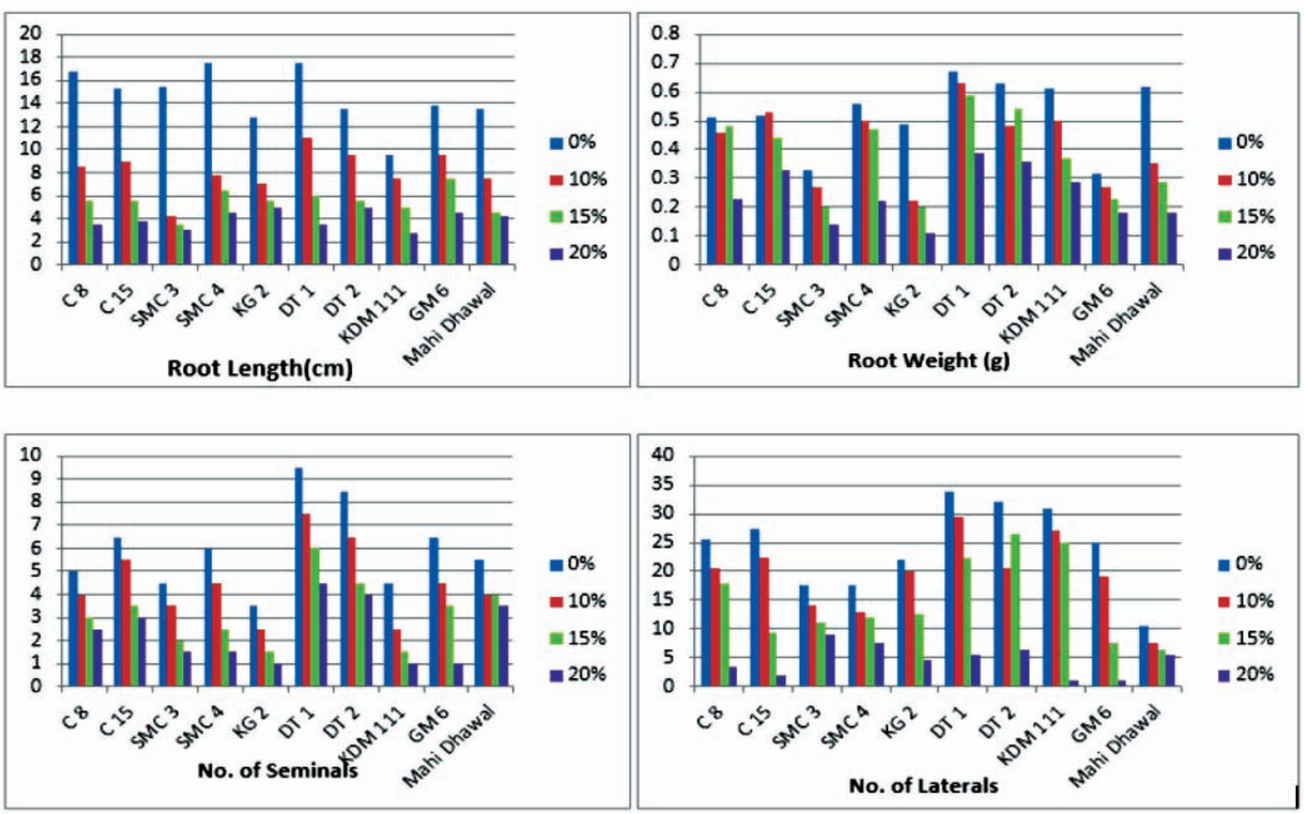
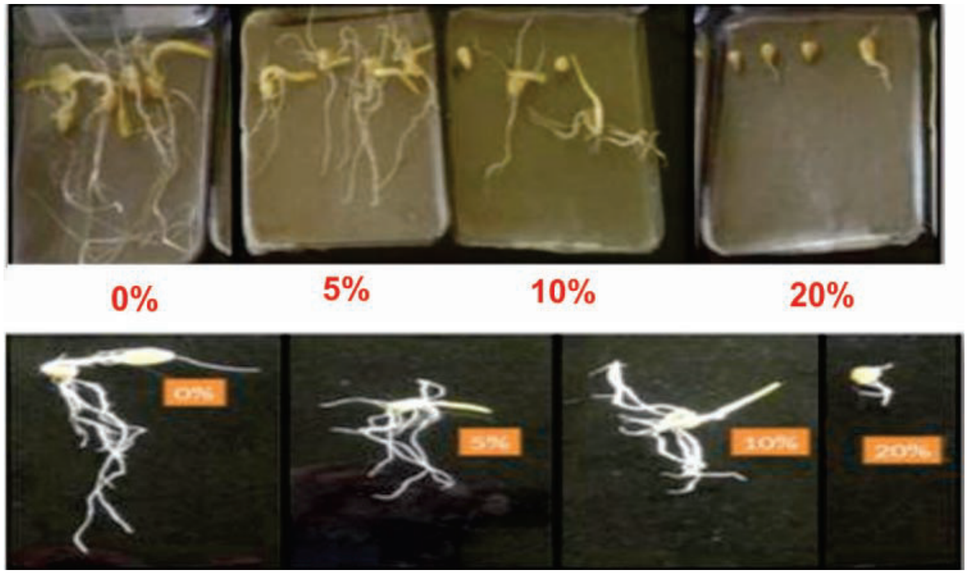



**Figure 1.** Response of maize genotypes to PEG-6000 mediated screening

**Figure 2.** PEG-6000 response of DT-1 genotype under different concentrations




of laterals under different levels of PEG-6000 are as under:

*0% level:* Under 0%, the number of laterals had a mean value of 26.45 with the highest value recorded in C-8 (37.50) followed by C-15 (37.50) and DT-1(34.00) and the lowest was in SMC-3 (17.50) followed by SMC-4 (17.50).

*10% level:* Under 10%, the number of laterals had a mean value of 21.85 with the highest value recorded in C-8 (35.50) followed by C-15 (32.50) and DT-1 (29.50) and the lowest was in Mahi Dhawal (7.50) followed by SMC-4 (13.00).

*15% level:* Under 15%, the number of laterals had a mean value of 18.5 with the highest value recorded in C-8 (31.00) followed by C-15 (30.50) and DT-2 (26.50) and the lowest was in Mahi Dhawal (6.50) followed by GM-6 (7.50).

*20% level:* Under 20%, the number of laterals had a mean value of 9.50 with the highest value recorded in C-8 (23.50) followed by C-15 (21.00) and DT-2 (10.50) and was lowest in KDM-111 (1.00) followed by GM-6 (1.00).

*Analysis of variance for root traits scored under laboratory screening*

Analysis of variance for various root traits scored under laboratory conditions is presented in Table 2 which showed that mean square due to genotypes was significant for all the traits.

## Discussion

In the present study, polyethylene glycol (PEG) compounds have been used to stimulate osmotic stress effects in petri dish (*in vitro*) for plants to maintain uniform water potential throughout the experimental period. There was a progressive decline in all the parameters across all genotypes with increase in PEG concentration from 0–20 per cent level and the parameters, length of radical, root biomass, number of seminals and number of laterals had the highest value under control and had significant differences under water stress. Under 0%, length of radical had the highest value in DT-1 and lowest in KDM-111. Under 10 per cent, it had the highest value in DT-1 and the lowest in SMC-3. Under 15 per cent, it

**Table 2.** Analysis of variance for root traits under PEG-6000 mediated screening

| Source of variation | df | Total root length (cm) | Root weight (g) | Number of seminals | Number of laterals |
|---|---|---|---|---|---|
| Genotype | 9 | 9.361** | 0.105** | 17.735** | 515.200** |
| Replication | 3 | 422.593 | 0.271 | 50.746 | 1002.617 |
| Genotype × replication | 27 | 5.982 | 0.007 | 0.653 | 43.330 |
| Error | 40 | 1.125 | 0.002 | 0.538 | 4.475 |

**Significant at 0.05% level

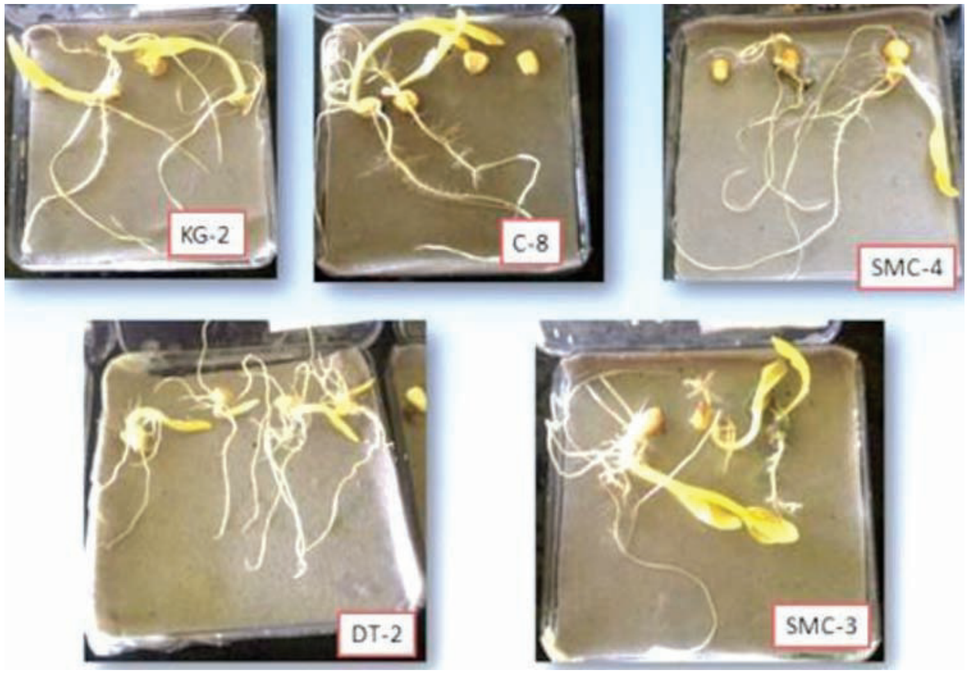



**Figure 3.** PEG-6000 response of maize genotypes under various concentrations

had the highest value in GM-6 and lowest in SMC-3. Under 20 per cent, the highest value was recorded in DT-2 and lowest in KDM-111. However, root biomass under 0, 10 and 15 per cent had the highest value in DT-1 and the lowest in GM-6, KG-2 and SMC-3, while root biomass had the highest value in DT-2 and the lowest in SMC-3 under 20 per cent level of PEG-6000. Similarly, number of seminals under 0, 10, 15 and 20 per cent had the highest value in DT-1 and lowest in KG-2 and under various PEG concentrations (0, 10, 15 and 20 per cent) number of laterals had the highest value recorded in C-8 and lowest in SMC-3, Mahi Dhawal and KDM-111. Results of the current study were in agreement with other experiments in different plants including (Kalefetoglu *et al*., 2009) in chickpea, (Almansouri *et al.,* 2001; Soltani *et al*., 2006) in wheat.

The polyethylene glycol (PEG)-induced inhibition of germination has been attributed to osmotic stress (Dodd and Donovan, 1999; Sidari *et al.*, 2008). The solutions of high molecular weight polyethylene glycol are often used to control water potential in seed germination studies (Hardegree and Emmerich, 1990). Water stress acts by decreasing the percentage and rate of germination and seedling growth (Delachiave and De Pinho, 2003). Water stress not only affects seed germination but also increases mean germination time in crop plants (Willenborb *et al.*, 2004). The adverse effect of water shortage on germination and seedling growth has been well reported in different crops such as corn (Farsiani and Ghobadi, 2009; Khayatnezhad *et al.,* 2010; Mostafavi *et al.*, 2011), wheat (Jajarmi, 2009), sorghum (Gill *et al.*, 2002), sunflower (Mohammed *et al.*, 2002). This agreed with the results of Khayatnezhad *et al.* (2010) and Mostafavi *et al.*, (2011) in corn, Jajarmi (2009) in wheat and Mostafavi (2011) in safflower. According to Ayaz *et al.* (2000) decrease of seed germination under stress conditions was due to the occurrence of some metabolic disorders. It seems that decrease of germination percentage and rate is related to reduction in water absorption into the seeds at imbibitions and seed turgescence stages (Hadas, 1977). Water deficit affects the germination of seed and the growth of seedlings negatively as germination is one of the most important traits in early stage of growth in most plants. The adverse effect of water shortage on germination and seedling growth has been well reported in different crops (Mostafavi *et al.*, 2011). An artificially created water-stress environment is used to provide the opportunity in selecting superior genotypes out of a large population. Therefore, it is necessary to identify genotypes that are tolerant to drought at the primary growth stage.

## Conclusion

Keeping in view the above stated research finding it can be concluded that genotypes *viz.*, KDM-111 followed by SMC-3 and DT-1 exhibited lesser reduction in total root length and KG-2 followed by SMC-3 and SMC-4 in root weight, GM-6 followed by KDM-111 and KG-2 in number of seminals and GM-6 followed by KDM-111 and C-15 in number of laterals. There was a progressive decline in all the parameters across all genotypes with increase in PEG concentration from 0–20 per cent level. The genotypes had significant differences under water deficit stress and hence, those genotypes having potential genetic sources to maintain the higher growth under stress conditions are considered to be drought tolerant.

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RESEARCH PAPER

# Molecular variability in *Curvularia lunata* isolates causing *Curvularia* leaf spot of maize in Karnataka

**S. I. Harlapur · Ramangouda Hadimani · R. M. Kachapur · S. R. Salakinkop · S. C. Talekar · Krishna Iliger**

**Abstract:** Molecular variability of *Curvularia lunata* pathogen causing *Curvularia* leaf spot of maize was carried out by using ITS primers and subsequently subjected to sequence analyses. Results revealed that, phylogenetic analysis grouped *Curvularia lunata*, isolates into two main clusters with 0.005 similarity coefficient. The similarity coefficient ranged from 0.001 to 0.005 indicating good level of similarity. UPGMA cluster analysis based on genetic distance coefficients clearly separated all the isolates. Out of five isolates Bailhongal, Badami, Davangere and Dharwad isolates were grouped under cluster I, whereas Mundagod isolate formed separate cluster II. In cluster I, Bailhongal and Dharwad isolates were showed almost one per cent of divergence. In cluster II only one isolate Mundagod was present with nearly 5.5 per cent divergence compared to cluster I, this indicated there was 96.5 per cent of similarity among the isolates of *Curvularia lunata*.

**Keywords:** *Curvularia* leaf spot · Primers · Ecology · Maize

## Introduction

Maize (*Zea mays* L.) is the world’s leading crop with wider adaptability across agro-ecological zones with versatile uses. It is the main raw material for industrial products like glucose, sorbitol, dextrose and oil. Besides it is used as livestock, poultry and animal feed, and for manufacturing of starch and starch based products. It can contribute to diet diversification and improve nutrition in human beings through exploitation of quality protein maize. In India, maize is the important food crop after rice and wheat cultivated in an area 9.86 million hectare with an annual production of 30.16 million tonnes and an average productivity of 3.06 t/ha (FAOSTAT, 2020). The maximum area and production of maize is in Uttar Pradesh and the highest productivity is in Andhra Pradesh (4637 kg/ha) followed by Tamil Nadu (4,134 kg/ha). In Karnataka, maize is cultivated in an area of 1.40 million hectare with a production of 3.96 million tonnes and an average productivity of 2.9 t/ha which is almost same as the national productivity.

Maize is subjected to as many as 112 diseases on global basis. In India, there is a record available for 35 diseases. The annual loss due to maize diseases in India was estimated to the tune of 13.2 to 39.5 per cent (Payak and Sharma, 1985). *Curvularia* leaf spot of maize caused by *Curvularia andropogonist* which was a minor disease earlier has emerged as an important disease all over the maize growing area in India (Choudhary *et al.*, 2011). In Karnataka, the disease was reported for the first time from Dharwad district during 2008 (Anonymous, 2008) and further the occurrence of the disease and causal organism as *C. lunata* Wakker (Boedikun) on the basis of morphological characters was reported by Harlapur *et al.* (2012 and 2014).

The symptoms of the disease start as minute, chlorotic, pinhead sized translucent spots on the leaf surface. Subsequently the spots increase in size and necrosis start from the centre. The disease severity of *Curvularia* leaf spot of maize in Karnataka ranged from 18.5 to 61.5 per cent with an estimated 22.6 per cent loss

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in grain yield (Harlapur *et al*., 2012). In India though *C. lunata* has a narrow host range infecting maize and paddy, it extracts significant morphological as well as pathogenic variability. Although some of the earlier work had thrown light on its morphological characterization and pathogenic variability, but that does not provide a clear-cut situation in the variability existing in the pathogen. Thus, the present study was undertaken to study the molecular variability of *C. lunata* along with morphological variability.

## Materials and method

Molecular variability of the pathogen was studied using ITS primers and subsequently subjected to sequence analysis. The detailed methodology followed for preparation of formulation is as below.

*Curvularia lunata* isolates were collected from 15 locations of major maize growing areas of northern Karnataka (Table 1) and were assessed for molecular variability. The DNA was isolated from all the isolates of *C. lunata*, by following standard CTAB method with minor modification (Cuervo-Parra *et al.,* 2011). For DNA isolation, 2-3 of fungal mat grown potato dextrose broth was taken and homogenized using pestle and mortar in liquid nitrogen. To the above solution 1 ml of lysis buffer was added. The suspension in pestle and mortar was extracted with equal volume of phenol: chloroform: isoamyl alcohol (1:1 W/V) in centrifugation tube, centrifuged at 10,000 rpm for 15–20 minutes at $4^0$C. Supernatant was taken in fresh centrifuge tube and 2.5 µl RNase and 2.5µl protienase-k was added and incubated at room temperature for 30 minutes. Chilled isopropanol of about $1/3^{rd}$ volume (300-400 µl) was added and centrifuged @ 10,000 rpm for 15minutes at $4^o$C. The 500 µl buffer was added and centrifuged at 10,000 rpm for 5 min at $4^o$C. DNA, pellet was washed with 70 per cent ethanol; air dried and responded in 500 µl of $T_{10}E_1$. This DNA obtained was further quantified by agarose gel electrophoresis.

*Polymerase chain reaction (PCR)*

The ribosomal DNA (rDNA) unit contains genetic and non-genetic or spacer region. Each repeat unit consists of copy of 18S, 5.83S and 28S like rDNA and its spacer like internal transcribed spacer (ITS). The rDNA has been employed to analyze evolutionary events because it is highly conserved, whereas ITS rDNA is more variable hence, it was used for investigation. Forward ITS-1 (5'-TCCGTAGGTGAACCTGCG-3') and reverse primers ITS-4 (5'-TCCTCCGCTTATTGATATGC-3') were used.

*Master mix for PCR*

Amplification reaction mixture was prepared in 0.2 ml thin-walled PCR tubes containing following components. The total volume of each reaction mixture was 20 µl.

| Reaction mixture | Quantity |
|---|---|
| Template DNA (25 ng/µl) | 1.00 µl |
| Primer (5PM/µl) | F-1.00 µl<br>R-1.00 µl |
| dNTPs mix (2.5 mM each) | 1.00 µl |
| 10 x assay buffer with 15 mM $MgCl_2$ | 2.00 µl |
| *Taq* DNA polymerase (6.0U µl-1) | 0.50 µl |
| Sterile distilled water | 13.50 µl |
| Total | 20.00 µl |

Except template the master mix was distributed to PCR tubes (19 µl tube) and later 1 µl of template DNA from the respective isolates was added making the final volume of 20 µl. The amplification was performed in the thermal cycler by following method suggested by Cuervo-Parra *et al.* (2011).

**Table 1.** The details of the sites (GPS position) from where the isolates have been collected

| S.No. | Location | Name of the isolate | GPS Details | |
|---|---|---|---|---|
| | | | Latitude | Longitude |
| 1. | Kalghatagi | CI Kal | 15.18 | 74.97 |
| 2. | Navalagund | CI Nav | 15.57 | 75.37 |
| 3. | Dharwad | CI Dha | 15.46 | 75.01 |
| 4. | Davangere | CI Dav | 14.47 | 75.91 |
| 5. | Kerimattihalli | CI Ker | 14.77 | 75.39 |
| 6. | Haveri | CI Hav | 14.80 | 75.40 |
| 7. | Badami | CI Bad | 15.82 | 75.59 |
| 8. | Bagalkot | CI Bag | 16.18 | 75.69 |
| 9. | Mundagod | CI Mum | 14.97 | 75.03 |
| 10. | Nargund | CI Nar | 15.72 | 75.38 |
| 11. | Hulakote | CI Hul | 13.26 | 77.35 |
| 12. | Bailhongal | CI Bai | 15.82 | 74.86 |
| 13. | Gokak | CI Gok | 16.16 | 74.83 |
| 14. | Naganoor | CI Nag | 15.18 | 75.10 |
| 15. | Raichur | CI Rai | 16.21 | 77.34 |

**Table 2.** Blast results of nucleotide sequence of *Curvularia lunata* ITS region

| S.No. | Isolate code | Place | Accession number | NCBI BLAST Hit results | Location | Host | Max. Indent |
|---|---|---|---|---|---|---|---|
| 1 | CI Dha | Dharwad | KR633084.1 | *Curvularia lunata* strain CX-3 18S ribosomal RNA gene, partial sequence; internal transcribed spacer | China | Maize | 99 |
| 2 | IC Dav | Davangere | KR633132.1 | *Curvularia lunata* strain SD-5 18S ribosomal RNA gene, partial sequence; internal transcribed spacer | China | Maize | 99 |
| 3 | CI Bad | Badami | KU315128.1 | *Curvularia lunata* strain CSV3, 5.8S ribosomal RNA gene and internal transcribed spacer | Jammu & Kashmir | Maize | 99 |
| 4 | CI Mun | Mundagod | JX256431.1 | *Curvularia lunata* voucher MFLUCC 10-0706 internal transcribed spacer | Thailand | Maize | 99 |
| 5 | CI Bai | Bailhongal | KU215414.1 | *Curvularia lunata* strain CL-1 18S ribosomal RNA gene, partial sequence; internal transcribed spacer | China | Maize | 99 |

*Gel electrophoresis*

Agarose gel electrophoresis was performed to resolve the amplified product using 1.4 per cent agarose in 1X TBE (Tris Borate EDTA) buffer, 0.5 µl/ml of Ethidium bromide and loading buffer (0.25 per cent Bromophenol Blue in 40 per cent sucrose). Four microlitre of loading dye was added to 20 µl of PCR product and loaded to the agarose gel. Electrophoresis was carried out at 75 V for 1.5 hr. The gel was observed under UV light and documented using gel documentation unit.

*Sequencing of ITS region*

The ITS region was sequenced for five isolates of *C. lunata* belonging to different geographical regions to confirm organism and to know the variability present in them.

## Results and discussion

The full length ITS rDNA region was amplified with ITS-1 (5'-TCCGTAGGTGAACCTGCG-3') and ITS-4) 5'-TCCTCCGCTTATTGATATGC-3') primers for fifteen isolates of *C. lunata.* DNA amplicon was observed at the region 560 bp by checking the amplified products on 1.2 using the NCBI BLAST programme, these isolates were confirmed as *C. lunata*. Cuervo-Para *et al.* (2011) obtained the similar results where the DNA of three strains of *C. lunata* electrophoresis and amplicon was observed at the region of 560 bp with universal ITS fungal primers. By using the NCBI-BLAST programme confirmed the isolates as *C. lunata*. The list of isolates, accession number, per cent homology and name identified are given in Table 2.

*Phylogenetic analysis*

The ITS rDNA region sequence was used in this analysis because it has been shown to be more informative and closest phylogenetic relative in *Curvularia lunata,* isolates. In order to evaluate whether the grouping pattern obtained on the basis of the ITS sequences would be useful frame to identify and align, these isolates and were identified at the species level by morphological characters using the existing taxonomic criteria analysis and analysis of their ITS rDNA region gene sequences. In the present case, isolates of *Curvularia lunata w*ere used as an out group vice-versa to interpret the clustering of isolates as distinct or related out group of genus. Phylogenetic analysis grouped *Curvularia lunata,* isolates into two main clusters with 0.005 similarity coefficient. The similarity coefficient ranged from 0.001 to 0.005 indicating good level of similarity.

*Curvularia lunata*

The UPGMA cluster analysis-based of genetic distance coefficients clearly separated all isolates. The five isolates were grouped into two clusters. The cluster I included the isolates which belonged to Dharwad, Bailhongal, Badami and Davangere, whereas the isolates of Mundagod were grumped in to cluster II. The per cent divergence between the clusters was less. Further, the divergence within the clusters indicated that the isolates of Dharwad and Bailhongal showed only 1 per cent and the isolates of Badami and Davanagere showed 2.5 to 3.0 per cent divergence. (Figure 1). Form the amplification of ITS region (Figure 2) and sequencing data of ITS region, it is evident that the genetic diversity within *Curvularia lunata,* among the collected isolates is low. The presence of this

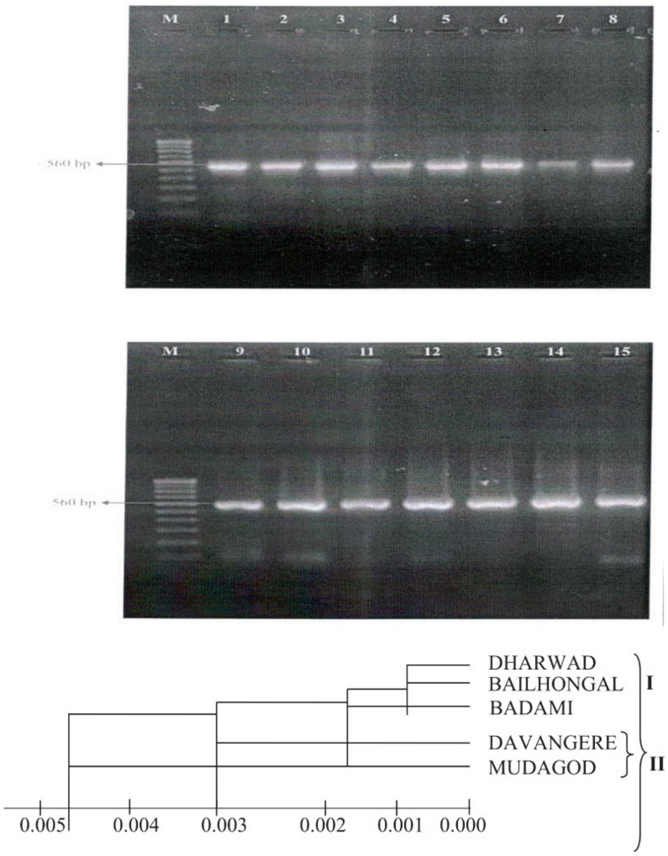
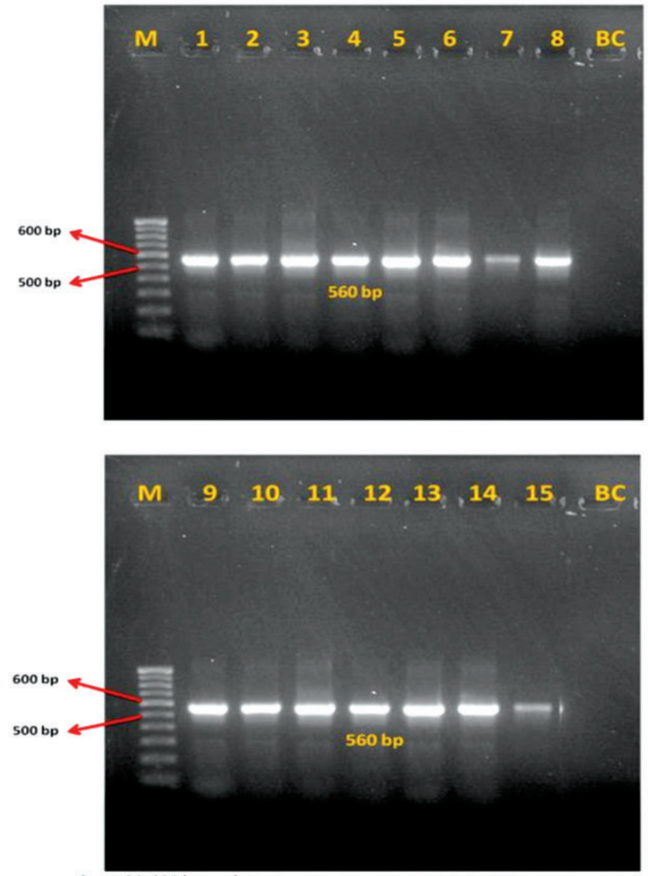



**Figure 1.** Phylogenetic relationship on ITS rDNA among the isolates of *Curuvlaria lunata* from different regions of northern Karnataka




Lane: M; 100 bp marker
BC: Buffer control, 1: Kal, 2: Nav, 3: Dhar, 4: Dav, 5: Ker, 6: Hav, 7: Bad, 8: Bag, 9: Mun, 10: Nar, 11: Hul, 12: Bai, 13: Gok, 14: Nag and 15: Rai.

**Figure 2.** PCR amplification of ITS region of

low genetic variability of the isolates in the regions helps to manage the *Curvularia* leaf spot of maize by following similar management practices in the regions.

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RESEARCH PAPER

# Estimation of crop losses and eco-friendly management of maize cyst nematode

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**Abstract:** Maize cyst nematode (*Heterodera zeae*) is the most important nematode pest of maize and reported to cause significant losses in Rajasthan due to monocropping, favourable soil and environmental conditions. Therefore, an experiment was carried out to estimate the avoidable yield losses caused by *H. zeae* on maize with the application of carbofuran at 2 kg *a.i.*/ha in naturally infested field with test nematode. Results revealed that application of chemical significantly reduced nematode population and avoided yield losses of grain and straw to the tune of 25.1–26.3 per cent and 21.9–25.2 per cent, respectively. In another trial, efficacy of neem, karanj and mahua (deoiled cakes, dry leaves and seed kernel powder) were tested as row and broadcast application @ 0.5 t/ha for the management of this nematode. Results revealed that neem seed kernel @ 0.5 t/ha when applied as row application was most effective for the management of maize cyst nematode whereas karanj seed kernel @ 0.5 t/ha applied in rows was best to enhance yield of grain (56.4–59.6 per cent) and straw (52.6–57.6 per cent) followed by neem seed kernel and karnaj cake @ 0.5 t/ha over untreated control.

**Keywords:** *Heterodera zeae* · Karnaj cake · Maize · Neem seed kernel

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## Introduction

Maize (*Zea mays* L.) being queen of cereals is cultivated under the wide range of agro-climatic conditions all over the world. In India, maize occupies third rank after rice and wheat. In India, maize is extensively grown in Rajasthan, Bihar, Gujarat, Haryana, Karnataka, Delhi, Himachal Pradesh, Madhya Pradesh, Andhra Pradesh, Tamil Nadu, Maharashtra, Punjab and Uttar Pradesh etc. In Rajasthan, it is mainly cultivated in Udaipur, Rajsamand, Banswara, Bhilwara, Chittorgarh, Dungarpur, Ajmer, Kota, Bundi and Jhalawar districts of Rajasthan.

Maize is primarily a *Kharif* season crop but presently, it is also being popularized as an important *Rabi* crop in certain parts of the country including southern Rajasthan depending upon the climate, hybrid seed availability, irrigation facilities etc. In summer, it is grown as fodder crop while in *Kharif* and *Rabi*, it is mainly grown for human consumption as it provides high quality of staple food among rural masses.

In India, productivity of maize is very low because it is mainly cultivated as rainfed crop with poor management of pest and pathogens including plant parasitic nematodes. A large number of phytonematodes have been found associated with maize crop such as species of *Heterodera*, *Pratylenchus*, *Tylenchorhynchus*, *Meloidogyne*, *Hoplolaimus*, *Helicotylenchus*, *Rotylenchulus*, *Radopholus*, *Longidorus*, *Trichodorus*, *Xiphinema*, *Belanolaimus*, *Ditylenchus* and *Aphelenchus* (Yadav and Verma, 1971; Windham, 1998; Berg *et al.*, 2001; Kumar *et al.*, 2018). These nematodes apart from causing loss interact with other disease causing agents and adversely affect the quality and quantity of maize production.

Amongst phytonematodes, maize cyst nematode (*Heterodera zeae*) is economically most important and causes severe losses to this crop. It was first discovered by Koshy *et al.* (1970) from Chhapli village of the then Udaipur district of Rajasthan. It is widely distributed in maize growing areas of Rajasthan, Bihar, Madhya Pradesh, Gujarat, Haryana, Uttar Pradesh, Maharashtra and Delhi (Koshy and Swarup, 1971; Rathore *et al.*, 2007; Meena *et al.*, 2013; Mehta *et al.*, 2016). It causes severe damage to maize (Krusberg *et al.*, 1997; Dodwadia *et al.*, 2015). Significant yield losses are caused by this nematode under favouarable conditions (Srivastava *et al.*, 2001). The severity of losses caused by *H. zeae* on maize is higher in Rajasthan, probably due to favourable soil conditions, monocropping and ignorance of management practices conditions (Meena and Singh, 1995). In view of disease severity and crop losses, attempts were made to manage it with use of pesticides (Sethi and Srivastava, 1986; Kaul and Sethi, 1988a; Baheti *et al.*, 2015). However, health hazards, residual toxicity, environmental pollution, high application cost, and poor adoption at farmers' fields are limiting factors. Though attempts were made (Mishra and Prasad, 1974; Bhatti, 1988; Mehta *et al.*, 2015; Kumhar *et al.*, 2018) to evolve economical and eco-friendly techniques for the management of plant parasitic nematode on various crops yet information regarding management of maize cyst nematode on maize is lacking. Therefore, present investigation was conceived so as to evolve eco-friendly management of maize cyst nematode.

## Materials and methods

*Estimation of avoidable losses*

An experiment was designed to estimate the avoidable yield losses caused by maize cyst nematode, *H. zeae* on maize. A field trial was carried out on farmer's field naturally infested with maize cyst nematode, *Heterodera zeae* at Bujhda near Udaipur. Before layout, soil samples were collected and processed in the laboratory to ensure good nematode population required for experimentation. It was determined 1000 and 1140 larvae/100 cc soil during I$^{st}$ Year and II$^{nd}$ year, respectively. Two treatments *viz.*, carbofuran 3G @ 2 kg *a.i.*/ha (66.67 kg commercial formulation) and untreated control were taken. Chemical was weighed for each experimental plot of 10 m$^2$ size, mixed well in 1 kg of sand for uniform distribution in each experimental plot and applied at the time of sowing. Experiment was laid out in paired plot method as suggested by LeClerg (1971) and both the treatments were replicated ten times. All intercultural operations (fertilizer application, weeding, hoeing, irrigation etc.) were done as per recommended package of practices for better growth of the crop. Fungicide and insecticide application was also applied for safeguarding the crop from pests and diseases. Crop was harvested after maturity and observations on number of cyst per plant, cyst per 100 cc soil, larvae per 100 cc soil, grain yield, and straw yield were recorded. For estimation of soil population, samples collected from the experimental plots were brought to the laboratory and thoroughly mixed. Hundred cc soil was taken in bowl, added water, stirred thoroughly and passed through 16 mesh sieve. The content obtained were then passed through 100 mesh sieve and catch of sieve was transferred in a beaker and later on a blotting paper, examined under microscope for counting of cysts. The filtrate of 100 mesh sieve was further passed through 400 mesh sieves and catch of sieve was placed over Baermann's funnel assembly. After 24 hours, the nematode suspension was drawn from the funnel in a beaker and kept for some time as such to allow the nematodes to settle down at the bottom. Nematode larvae population was counted under microscope.

After harvesting, root samples were collected from each experimental plot and brought to the laboratory. Roots were gently washed in running tap water to remove adhering soil particles. Well cleaned roots were cut into small pieces and observed thoroughly under microscope for counting of cysts population per plant. For grain yield, cobs were collected separately from each experimental plot in well aerated cotton cloth bags and kept in shade for drying. Thereafter, grains were separated from cobs with the help of maize sheller, cleaned and weighed to obtained grain yield data. For straw yield, dry plants were cut and tied with small piece of rope. Then weighed with the help of weighing balance to record straw yield. Yield data were compiled after termination of the experiment and expressed in t/ha. Avoidable losses (Treated-Untreated/Treated*100) and per cent increase in yield and decrease in nematode population over control (Treated-Untreated/Untreated*100) was determined.

*Organic amendments*

In view of disease severity and crop losses caused by tiny and tough organism, attempts were made to test various pesticides for the management of nematodes including maize cyst nematode on maize and proved effective but due to health hazards, residual toxicity, environmental pollution and high cost, their adoption at farmer's level has been limited. Therefore, present investigation was made to evolve eco-friendly management options for the maize cyst nematode, *H. zeae* on maize. With this view, a field trial was planned and conducted to test the efficacy of neem, karanj and mahua cakes, leaves powder and seed kernels for the management of maize cyst nematode, *H. zeae* on maize. These plant products were applied @ 0.5 t/ha as row and broadcast application. Carbofuran @ 2 kg *a.i.*/ha was kept and maintained as standard chemical check along with untreated check. The required quantity of cakes, leaves, seed kernels and chemicals were calculated and weighed separately for each plot (10 $m^2$). All plant products were mixed well in 1 kg sand for uniform distribution and then applied in experimental plots at the time of sowing. Soil samples were collected to estimate the initial nematode population. It was estimated 1020 and 1180 larvae/100 cc soil during Ist and IInd Year, respectively. The experiment was laid out in randomized block design with five replications. All agronomical practices were adopted throughout the cropping period. Observations *viz*., number of cyst per plant, cyst per 100 cc soil, larvae per 100 cc soil, grain and straw yield were recorded for comparison of treatments. Data were compiled and analyzed for interpretation of findings (Table 2–3).

## Results and discussion

*Assessment of avoidable losses*

Results revealed that 28.4 cyst per plant, 9.8 cyst per 100 cc soil and 1055.4 larvae per 100 cc soil were recorded in plots treated with carbofuran @ 2 kg *a.i.*/ha compared to 50.5, 17.0 and 1940.6 in untreated plots during $I^{st}$ year, respectively (Table 1). Similar trend was noticed during $II^{nd}$ year also.

Results revealed that application of carbofuran @ 2 kg *a.i.*/ha in field infested with maize cyst nematode, *H. zeae* significantly enhanced grain and straw yield over untreated control (Table 2).

During $I^{st}$ year 3.50 and 11.66 t/ha grain and straw yield was recorded in chemical treated plots while in untreated plots, it was found to be 2.58 and 8.72 t/ha, respectively. Similar trend was also noticed during $II^{nd}$ year where grain yield of 3.74 and 2.80 t/ha and straw yield of 12.60 and 9.84 t/ha in treated and untreated plots, respectively. Application of chemical avoided losses to the tune of 26.3 per cent in grain yield and 25.2 per cent in straw yield during $I^{st}$ year. During $II^{nd}$ year, it was recorded 25.1 and 21.9 per cent, respectively. Per cent increase in grain (35.7 per cent) and straw yield (33.7 per cent) over untreated control were obtained during $I^{st}$ year. During $II^{nd}$

**Table 1.** Estimation of avoidable yield losses caused by maize cyst nematode, *Heterodera zeae* on maize

| Treatment | Nematode population | | | | | | Per cent decrease over control | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Cyst/ plant | | Cyst/100 cc soil | | Larvae/100 cc soil | | Cyst/ plant | | Cyst/100 cc soil | | Larvae/100 cc soil | |
| | Ist Year | IInd Year | Ist Year | IInd Year | Ist Year | IInd Year | Ist Year | IInd Year | Ist Year | IInd Year | Ist Year | IInd Year |
| Carbofuran 2 kg a.i./ha ($T_1$) | 28.4 | 33.2 | 9.8 | 11.6 | 1055.4 | 1170.6 | 43.8 | 40.1 | 42.4 | 38.9 | 45.6 | 41.3 |
| Untreated control ($T_2$) | 50.5 | 55.4 | 17.0 | 19.0 | 1940.6 | 1995.8 | - | - | - | - | - | - |
| 't' test | Sig. | Sig. | Sig. | Sig. | Sig. | Sig. | - | - | - | - | - | - |

**Table 2.** Estimation of yield per cent avoidable loss and per cent increase over control caused by maize cyst nematode, *Heterodera zeae* on maize

| Treatment | Yield (t/ha) | | | | Per cent avoidable loss | | | | Per cent increase over control | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Grain | | Straw | | Grain | | Straw | | Grain | | Straw | |
| | Ist Year | IInd Year | Ist Year | IInd Year | Ist Year | IInd Year | Ist Year | IInd Year | Ist Year | IInd Year | Ist Year | IInd Year |
| Carbofuran 2 kg a.i./ha ($T_1$) | 3.50 | 3.74 | 11.66 | 12.6 0 | 26.3 | 25.1 | 25.2 | 21.9 | 35.7 | 33.6 | 33.7 | 28.0 |
| Untreated control ($T_2$) | 2.58 | 2.80 | 8.72 | 9.84 | – | – | – | – | – | – | – | – |
| 't' test | Sig. | Sig. | Sig. | Sig. | – | – | – | – | – | – | – | – |

*Data are the average of ten replications*

**Table 3.** Effect of botanicals as soil amendment for the management of *Heterodera zeae* on maize

| Treatment | Nematode population | | | | | | Yield (t/ha) | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Cyst/ plant | | Cyst/100 cc soil | | Larvae/100 cc soil | | Grain | | Straw | |
| | Ist Year | IInd Year | Ist Year | IInd Year | Ist Year | IInd Year | Ist Year | IInd Year | Ist Year | IInd Year |
| *Control v/s Rest* | | | | | | | | | | |
| Control | 51.4 | 53.2 | 15.6 | 17.4 | 1850.8 | 2050.6 | 2.60 | 2.75 | 8.60 | 9.70 |
| Rest | 36.7 | 39.7 | 11.2 | 13.0 | 1358.5 | 1568.9 | 3.43 | 3.55 | 11.10 | 12.20 |
| 'F' test | Sig. | Sig. | Sig. | Sig. | Sig. | Sig. | Sig. | Sig. | Sig. | Sig. |
| *Method of application* | | | | | | | | | | |
| Row (R) | 32.2 | 34.8 | 10.0 | 11.9 | 1279.7 | 1483.9 | 3.70 | 3.80 | 12.07 | 13.10 |
| Broad cast (B) | 41.2 | 44.6 | 12.4 | 14.1 | 1437.3 | 1654.0 | 3.16 | 3.29 | 10.13 | 11.29 |
| SEm ± | 0.37 | 0.36 | 0.14 | 0.17 | 16.90 | 15.00 | 0.029 | 0.029 | 0.098 | 0.099 |
| CD at 5% | 1.04 | 1.03 | 0.40 | 0.47 | 47.57 | 42.22 | 0.083 | 0.083 | 0.277 | 0.278 |
| *Between treatment* | | | | | | | | | | |
| Neem cake 0.5 t/ha ($T_1$) | 34.7 | 37.8 | 10.4 | 12.0 | 1260.2 | 1450.2 | 3.54 | 3.66 | 11.53 | 12.50 |
| Karanj cake 0.5 t/ha ($T_2$) | 36. 4 | 39.8 | 10.8 | 12.5 | 1320.4 | 1503.5 | 3.69 | 3.79 | 11.88 | 13.00 |
| Mahua cake 0.5 t/ha ($T_3$) | 39.5 | 42.7 | 12.0 | 13.9 | 1447.9 | 1685.1 | 3.26 | 3.38 | 10.51 | 11.47 |
| Neem leaf 0.5 t/ha ($T_4$) | 42.1 | 45.9 | 13.1 | 15.0 | 1591.5 | 1831.0 | 2.99 | 3.08 | 9.68 | 10.65 |
| Karanj leaf 0.5 t/ha ($T_5$) | 42.8 | 46.5 | 13.7 | 15.7 | 1661.4 | 1902.8 | 3.09 | 3.22 | 10.03 | 10.93 |
| Mahua leaf 0.5 t/ha ($T_6$) | 45.2 | 48.1 | 14.3 | 16.4 | 1725.4 | 1956.5 | 2.84 | 2.93 | 9.20 | 10.35 |
| Neem seed kernel 0.5 t/ha ($T_7$) | 30.7 | 32.6 | 9.4 | 10.7 | 1129.3 | 1322.8 | 3.75 | 3.83 | 12.03 | 13.18 |
| Karanj seed kernel 0.5 t/ha ($T_8$) | 32.7 | 35.6 | 10.0 | 11.5 | 1192.5 | 1380.1 | 3.82 | 3.98 | 12.40 | 13.65 |
| Mahua seed kernel 0.5 t/ha ($T_9$) | 36.7 | 40.1 | 11.2 | 12.9 | 1330.4 | 1562.2 | 3.42 | 3.53 | 11.13 | 12.10 |
| Carbofuran 2 kg a.i./ha ($T_{10}$) | 26.3 | 28.1 | 7.3 | 9.2 | 926.1 | 1095.2 | 3.92 | 4.09 | 12.63 | 14.13 |
| SEm ± | 0.83 | 0.81 | 0.32 | 0.37 | 37.79 | 33.54 | 0.066 | 0.066 | 0.220 | 0.221 |
| CD at 5% | 2.33 | 2.29 | 0.89 | 1.05 | 106.37 | 94.40 | 0.185 | 0.185 | 0.620 | 0.621 |
| *Two way Interaction* | | | | | | | | | | |
| Method × Treatment | | | | | | | | | | |
| Row × Neem cake 0.5 t/ha ($RT_1$) | 29.8 | 32.2 | 9.0 | 10.6 | 1160.4 | 1320.2 | 3.87 | 3.95 | 12.65 | 13.50 |
| Row × Karanj cake 0.5 t/ha ($RT_2$) | 31.0 | 33.6 | 9.4 | 11.2 | 1220.8 | 1381.4 | 4.05 | 4.09 | 13.00 | 14.05 |
| Row × Mahua cake 0.5 t/ha ($RT_3$) | 33.6 | 36.4 | 10.4 | 12.4 | 1325.0 | 1540.2 | 3.54 | 3.63 | 11.52 | 12.40 |
| Row × Neem leaf 0.5 t/ha ($RT_4$) | 38.2 | 42.0 | 12.0 | 14.0 | 1552.4 | 1760.0 | 3.16 | 3.23 | 10.35 | 11.20 |
| Row × Karanj leaf 0.5 t/ha ($RT_5$) | 38.8 | 42.6 | 12.6 | 14.8 | 1622.6 | 1864.8 | 3.30 | 3.41 | 10.80 | 11.45 |
| Row × Mahua leaf 0.5 t/ha ($RT_6$) | 42.0 | 45.0 | 13.4 | 15.8 | 1700.8 | 1932.6 | 2.88 | 2.98 | 9.55 | 10.80 |
| Row × Neem seed kernel 0.5 t/ha ($RT_7$) | 26.4 | 28.6 | 8.2 | 9.4 | 1048.2 | 1245.0 | 4.10 | 4.14 | 13.25 | 14.25 |
| Row × Karanj seed kernel 0.5 t/ha ($RT_8$) | 28.0 | 30.2 | 8.8 | 10.4 | 1085.0 | 1290.0 | 4.15 | 4.30 | 13.55 | 14.80 |
| Row × Mahua seed kernel 0.5 t/ha ($RT_9$) | 31.4 | 34.0 | 9.8 | 11.6 | 1210.2 | 1448.4 | 3.73 | 3.81 | 12.20 | 13.10 |
| Row × Carbofuran 2 kg a.i./ha ($RT_{10}$) | 22.6 | 23.8 | 6.4 | 8.6 | 872.0 | 1056.6 | 4.25 | 4.45 | 13.85 | 15.45 |
| Broadcast × Neem cake 0.5 t/ha ($BT_1$) | 39.6 | 43.4 | 11.8 | 13.4 | 1360.0 | 1580.2 | 3.20 | 3.37 | 10.40 | 11.50 |
| Broadcast × Karanj cake 0.5 t/ha ($BT_2$) | 41.8 | 46.0 | 12.2 | 13.8 | 1420.0 | 1625.6 | 3.33 | 3.48 | 10.75 | 11.95 |
| Broadcast × Mahua cake 0.5 t/ha ($BT_3$) | 45.4 | 49.0 | 13.6 | 15.4 | 1570.8 | 1830.0 | 2.98 | 3.12 | 9.50 | 10.54 |
| Broadcast × Neem leaf 0.5 t/ha ($BT_4$) | 46.0 | 49.8 | 14.2 | 16.0 | 1630.6 | 1902.0 | 2.82 | 2.92 | 9.00 | 10.10 |
| Broadcast × Karanj leaf 0.5 t/ha ($BT_5$) | 46.8 | 50.4 | 14.8 | 16.6 | 1700.2 | 1940.8 | 2.88 | 3.02 | 9.25 | 10.40 |
| Broadcast × Mahua leaf 0.5 t/ha ($BT_6$) | 48.4 | 51.2 | 15.2 | 17.0 | 1750.0 | 1980.4 | 2.80 | 2.88 | 8.85 | 9.90 |
| Broadcast × Neem seed kernel 0.5 t/ha ($BT_7$) | 35.0 | 36.6 | 10.6 | 12.0 | 1210.4 | 1400.6 | 3.40 | 3.52 | 10.80 | 12.10 |
| Broadcast × Karanj seed kernel 0.5 t/ha ($BT_8$) | 37.4 | 41.0 | 11.2 | 12.6 | 1300.0 | 1470.2 | 3.48 | 3.65 | 11.25 | 12.50 |
| Broadcast × Mahua seed kernel 0.5 t/ha ($BT_9$) | 42.0 | 46.2 | 12.6 | 14.2 | 1450.6 | 1676.0 | 3.10 | 3.24 | 10.05 | 11.10 |
| Broadcast × Carbofuran 2 kg a.i./ha ($BT_{10}$) | 30.0 | 32.4 | 8.2 | 9.8 | 980.2 | 1133.8 | 3.58 | 3.72 | 11.40 | 12.80 |
| SEm ± | 1.17 | 1.15 | 0.45 | 0.53 | 53.45 | 47.44 | 0.093 | 0.093 | 0.311 | 0.312 |
| CD at 5% | NS | 3.24 | NS | NS | NS | NS | 0.262 | 0.261 | NS | NS |

*Data are the average of five replications* year, it was 33.6 and 28.0 per cent in grain and straw yield, respectively (Table 2).

These results are in accordance with the findings of Herling (1974) who estimated yield losses up to the extent of 40 per cent on maize caused by *H. avenae* in East and West Germany. Similarly, Srivastava *et al.* (2001) reported losses to the tune of 11–26 per cent on maize by maize cyst nematode, *H. zeae* under sandy loam soils with the application of carbofuran @ 2 kg *a.i.*/ha. The results of present investigation show that losses caused by maize cyst nematode, *H. zeae* are in accordance with the previous workers.

*Nematode management*

In comparison to chemicals, some of the merits of organic amendment are building up of soil fertility, comparatively cheaper, having beneficial effect on succeeding crop, harmless to beneficial soil microbes, easy in application, non-pollutant, no extra care and precautions. Therefore, in present investigation neem, karanj, and mahua cakes, leaves and seed kernels were used @ 0.5 t/ha as organic amendment for the management of maize cyst nematode, *H. zeae* on maize. Among different organic amendments, minimum cysts per plant (30.7 and 32.6) were observed with neem seed kernel followed by karanj seed kernel (32.7 and 35.6) and neem cake (34.7 and 37.8) applied @ 0.5 t/ha during *Kharif* I$^{st}$ year and II$^{nd}$ year, respectively (Table 3). However, soil application of carbofuran at 2 kg *a.i.*/ha was significantly better over botanicals tested and produced 26.3 and 28.1 cysts per plant. Results also showed that cysts per plant were significantly reduced with row application (32.2 and 34.8) in comparison to broadcast application (41.2 and 44.6) (Table 3).

Application of neem seed kernel @ 0.5 t/ha in rows reduced cyst per plant to the tune of 48.6 and 46.2 per cent whereas, in karanj seed kernel and neem cake @ 0.5 t/ha, it was observed 45.5 and 43.2 per cent and 42.0 and 39.5 per cent, respectively lesser over untreated control (Table 4). The highest reduction in cyst per plant (56.0

**Table 4.** Per cent changes in nematode parameters and yield of maize through soil amendment with different plant products

| Treatment | Per cent decrease over control | | | | | | Yield increase over control (%) | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Cyst/plant | | Cyst/100 cc soil | | Larvae/100 cc soil | | Grain | | Straw | |
| | Ist Year | IInd Year | Ist Year | IInd Year | Ist Year | IInd Year | Ist Year | IInd Year | Ist Year | IInd Year |
| Row × Neem cake 0.5 t/ha ($RT_1$) | 42.0 | 39.5 | 42.3 | 39.1 | 37.3 | 35.6 | 48.9 | 43.6 | 47.1 | 39.2 |
| Row × Karanj cake 0.5 t/ha ($RT_2$) | 39.7 | 36.8 | 39.7 | 35.6 | 34.0 | 32.6 | 55.8 | 48.7 | 51.2 | 44.9 |
| Row × Mahua cake 0.5 t/ha ($RT_3$) | 34.6 | 31.6 | 33.3 | 28.7 | 28.4 | 24.9 | 36.2 | 32.0 | 34.0 | 27.8 |
| Row × Neem leaf 0.5 t/ha ($RT_4$) | 25.7 | 21.1 | 23.1 | 19.5 | 16.1 | 14.2 | 21.5 | 17.5 | 20.4 | 15.5 |
| Row × Karanj leaf 0.5 t/ha ($RT_5$) | 24.5 | 19.9 | 19.2 | 14.9 | 12.3 | 9.1 | 26.9 | 24.0 | 25.6 | 18.0 |
| Row × Mahua leaf 0.5 t/ha ($RT_6$) | 18.3 | 15.4 | 14.1 | 9.2 | 8.1 | 5.8 | 10.8 | 8.4 | 11.1 | 11.3 |
| Row × Neem seed kernel 0.5 t/ha ($RT_7$) | 48.6 | 46.2 | 47.4 | 46.0 | 43.4 | 39.3 | 57.7 | 50.6 | 54.1 | 46.9 |
| Row × Karanj seed kernel 0.5 t/ha ($RT_8$) | 45.5 | 43.2 | 43.6 | 40.2 | 41.4 | 37.1 | 59.6 | 56.4 | 57.6 | 52.6 |
| Row × Mahua seed kernel 0.5 t/ha ($RT_9$) | 38.9 | 36.1 | 37.2 | 33.3 | 34.6 | 29.4 | 43.5 | 38.6 | 41.9 | 35.1 |
| Row × Carbofuran 2 kg a.i./ha ($RT_{10}$) | 56.0 | 55.3 | 59.0 | 50.6 | 52.9 | 48.5 | 63.5 | 61.8 | 61.1 | 59.3 |
| Broadcast × Neem cake 0.5 t/ha ($BT_1$) | 23.0 | 18.4 | 24.4 | 23.0 | 26.5 | 22.9 | 23.1 | 22.6 | 20.9 | 18.6 |
| Broadcast × Karanj cake 0.5 t/ha ($BT_2$) | 18.7 | 13.5 | 21.8 | 20.7 | 23.3 | 20.7 | 28.1 | 26.6 | 25.0 | 23.2 |
| Broadcast × Mahua cake 0.5 t/ha ($BT_3$) | 11.7 | 7.9 | 12.8 | 11.5 | 15.1 | 10.8 | 14.6 | 13.5 | 10.5 | 8.7 |
| Broadcast × Neem leaf 0.5 t/ha ($BT_4$) | 10.5 | 6.4 | 9.0 | 8.1 | 11.9 | 7.3 | 8.5 | 6.2 | 4.7 | 4.1 |
| Broadcast × Karanj leaf 0.5 t/ha ($BT_5$) | 9.0 | 5.3 | 5.1 | 4.6 | 8.1 | 5.4 | 10.8 | 9.8 | 7.6 | 7.2 |
| Broadcast × Mahua leaf 0.5 t/ha ($BT_6$) | 5.8 | 3.8 | 2.6 | 2.3 | 5.5 | 3.4 | 7.7 | 4.7 | 2.9 | 2.1 |
| Broadcast × Neem seed kernel 0.5 t/ha ($BT_7$) | 31.9 | 31.2 | 32.1 | 31.0 | 34.6 | 31.7 | 30.8 | 28.0 | 25.6 | 24.7 |
| Broadcast × Karanj seed kernel 0.5 t/ha ($BT_8$) | 27.2 | 22.9 | 28.2 | 27.6 | 29.8 | 28.3 | 33.9 | 32.7 | 30.8 | 28.9 |
| Broadcast × Mahua seed kernel 0.5 t/ha ($BT_9$) | 18.3 | 13.2 | 19.2 | 18.4 | 21.6 | 18.3 | 19.2 | 17.8 | 16.9 | 14.4 |
| Broadcast × Carbofuran 2 kg a.i./ha ($BT_{10}$) | 41.6 | 39.1 | 47.4 | 43.7 | 47.0 | 44.7 | 37.7 | 35.3 | 32.6 | 32.0 |
| Untreated control | – | – | – | – | – | – | – | – | – | – |

and 55.3 per cent) was noticed with row application of carbofuran @ 2 kg *a.i.*/ha. Cysts per plant were observed lower in seed kernel compared to cake and leaf treatments. In the same way, neem products proved better over karanj and mahua. Similar order was noticed with regards to cyst and larvae population in soil (Table 4).

The results obtained in present investigation are also in accordance with the findings of number of earlier researchers. Mishra and Prasad (1974) also reported that effect of karanj, neem, mahua, groundnut, cotton, and linseed and sesamum cake on various nematodes attacking wheat, mungbeen and tomato. Although, all cakes were effective in reducing nematode population except cotton and linseed cake but neem cake was best among all the cakes tested. Alam and Khan (1974) reported that highest reduction in the population of stylet bearing nematodes took place in neem cake followed by cakes of mahua, mustard, groundnut and castor on spinach. Efficacy of plant products was also tested by Mojumder *et al.* (2002). They conducted an experiment on seed coating of pea with neem seed kernel, neem seed coat and neem cake (20 per cent), achook, neemark and nimbecidine (5 per cent) against *M. incognita* and *R. reniformis* and found that neem seed kernel was most effective in reducing root-knot galls, juveniles of *M. incognita* and all stages of *R. reniformis* and enhanced the plant growth and crop yield. Nangegowda *et al.* (1998) tested the efficacy of neem seed kernel, neem leaf, neem cake, nimbicidine and carbofuran against *M. incognita* in tomato nursery. They reported that all the neem products and carbofuran significantly reduced the nematode population. However, carbofuran was more effective followed by neem seed kernel and cake. Ram and Baheti (2004) tested the efficacy of neem, castor and karanj products (leaf and seed kernel) as seed treatment (10% w/w) along with soil application (0.25 t/ha) for the management of *Rotylenchulus reniformis* on cowpea (Pusa Barsati). Results revealed that plant products were effective in improving plant growth and reducing nematode reproduction over untreated check. However, neem seed kernel was found to be the most effective and superior among all the plant products. Mehta *et al.* (2015) observed maximum increase in plant growth parameters with neem (*Azadirachta indica*) leaves powder @ 4 g/plant followed by aak (*Calotropis procera*) and water hyacinth (*Eichornia crassipes*) leaves powder @ 4 g/plant as soil application over untreated control on maize infested with *H. zeae*. Similar results have also been reported by Baheti *et al.* (2015) on sweet corn, Kumar *et al.* (2018) on maize infested by various plant parasitic nematodes.

The suppression of nematodes in amended soil might be due to the effect of several combined factors. Production of volatile fatty acids, phenols, ammonia, amino acids etc. during decomposition of plant products might have caused inhibitory effect on the nematodes. The decomposed products might be either directly toxic to nematodes or the microbial metabolites produced during decomposition might have toxic effect to nematodes or enhance activity of predators and parasites attacking the nematode.

In present investigation also maize cyst nematode, *H. zeae* produced less number of cysts per plant, cysts and larvae per 100 cc soil under botanical treatments over untreated control which is in accordance with the findings of the previous workers. Comparative studies were also carried out between row and broadcast application of plant products for the management of *H. zeae* on maize. Results showed that all the plant products exhibited better response in row application over broadcast application.

Results pertaining to grain yield revealed that neem, karanj and mahua cakes, powdered leaves and seed kernels when applied @ 0.5 t/ha as soil amendment in *H. zeae* infested field, enhanced the yield. Among different botanical treatments, maximum grain yield (3.82 and 3.98 t/ha) was obtained with karanj seed kernel applied @ 0.5 t/ha followed by neem seed kernel @ 0.5 t/ha (3.75 and 3.83 t/ha) and karanj cake @ 0.5 t/ha (3.69 and 3.79 t/ha) during I$^{st}$ year and II$^{nd}$ year, respectively. Higher grain yield was recorded in case of seed kernel application compared to cake and leaf. Similarly, karanj products proved better than those of neem and mahua. However, highest numerical grain yield (3.92 and 4.09 t/ha) was recorded with the application of carbofuran @ 2 kg *a.i.*/ha over karanj seed kernel though statistically at par with each other during both the years.

Comparative studies of row and broadcast methods of application indicated that row application was significantly superior over broadcast application w.r.t. grain yield. It was obtained 3.70 and 3.80 t/ha in row while 3.16 and 3.29 t/ha in broadcast application during both the cropping seasons. Per cent increase in grain yield with the application of different botanicals over untreated was calculated and it was maximum (59.6 and 56.4 per cent) with karanj seed kernel @ 0.5 t/ha when applied in rows followed by neem seed kernel (57.7 and 50.6 per cent) and karanj cake (55.8 and 48.7 per cent) when applied @ 0.5 t/ha. However, highest increase in grain yield (63.5

and 61.8 per cent) was recorded with carbofuran @ 2 kg *a.i.*/ha when applied in rows during I[st] year and II[nd] year, respectively Similar trend was observed with respect to straw yield also. The efficacy of different botanicals has also been observed by several workers. Patel *et al.* (2000) studied the efficacy of Indian mustard, cakes of castor, neem, mahua, piludi, press-mud, poultry manure, FYM and celrich @ 1 or 2 per cent on wheat cv. Lok-1 and observed that all treatments were effective in increasing grain yield. Ravindra *et al.* (2003) evaluated the efficacy of neem and pongamia cake @ 10, 20 and 30 g/plant, applied directly to the base. Pongamia cake @ 30 g/plant recorded maximum green and cured leaf yield of tobacco. Similarly, Ram and Baheti (2006) and Khoarniya and Baheti (2020) reported the efficacy of botanicals for the management of reniform and root-knot nematode infecting pulses.

These findings support that application of botanicals as soil amendment enhanced plant growth and crop yield in nematode prone areas. This might be due to the fact that soil amendment with botanicals improve physical condition of soil, reduce population of plant parasitic nematodes, enhances the activity of beneficial soil microbes. Different plant products exert different reactions w.r.t. plant growth parameters. However, application of seed kernels led to better yield over cake and leaf because seed kernels gave better protection from nematode attack. Similarly, karanj products proved better over neem and mahua because of the fact that karanj products might be providing better nourishment to the crop or have other attributes responsible for enhancing yield.

## Conclusion

Application of carbofuran @ 2 kg *a.i.*/ha avoided grain and straw yield losses of maize to the tune of 25.1–26.3 and 21.9–25.2 per cent, respectively in field infested with *H. zeae*. Soil amendment with neem seed kernel @ 0.5 t/ha as row application was found better for the management of maize cyst nematode whereas karanj seed kernel at same dose was effective in enhancing the yield.

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RESEARCH PAPER

# Participatory maize improvement for enhancing yield and production

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**Abstract:** Most of the data on different types of farmer participation in selection of better plant ideotypes suggest that there is little to lose and much to gain by involving farmers, and more generally the users, in the process of plant breeding. Decentralized-participatory plant breeding should not be seen as "an alternative" type of plant breeding, but rather as an approach to specifically address situations such as marginal environments, or where there is a variety of different requirements (quality, crop duration, management, etc.). In the past, researchers have relied heavily on the extension department of the Ministry of Agriculture to transfer technologies to the farmers. Without intensive interaction with farmers in the development of maize varieties, there is a possibility of ignoring certain characteristics that would be of importance to farmers in their decision making process of whether to adopt a variety or not. In the very poor, rain-fed rice-growing areas of South Asia that the green revolution passed by, participatory plant breeding is now paying off with strong early adoption of farmer selected varieties that provide 40% higher yields in farmers' fields. The approach needs to be more widely tested in the heterogeneous rain-fed environments of Africa, where involving farmers, especially women farmers, in selecting varieties has shown early successes for beans, maize and rice. The cost effectiveness of the approach for wider use also needs to be evaluated.

**Keywords:** Farmers field · Maize · Participatory breeding · Varieties

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## Introduction

Participatory plant breeding (PPB) is the process by which farmers are routinely involved in a plant breeding programme with opportunities to make right decisions throughout. It involves scientists, farmers, and others, such as consumers, extensionists, vendors, industry, and rural cooperatives in plant breeding research. The primary aim of this activity is to enable tribal and farm families to initiate PPB along with scientists. In the recent years there has been an increasing interest towards participatory research in general, and towards participatory plant breeding in particular. It is termed "participatory" because many actors, and especially the users, can have a research role in all major stages of the breeding and selection process. PPB is an extension of Participatory Varietal Selection (PVS).

Participatory breeding will help to convert on-farm conservation to on-farm management of agro-biodiversity. Such participatory breeding work will be linked to training in seed technology, including aspects of post-harvest technology. The project, initiated in June 1998, took into account the tribal and farm family status and environment and chose the crops for PPB in three target sites: Jeypore Tract of Orissa (rice), Wayanad district of Kerala (medicinal rice, pepper) and Kolli Hills of Tamil Nadu (minor millets). PPB provides benefits to a specific user and builds farmer skill to enhance farmer selection and seed production efforts. Participatory Crop Improvement (PCI) involves farmers in different stages of selection and evaluation of future varieties, and proposes several levels of involving farmers in decision making (Sperling *et al.*, 2001). In PPB, farmers are actively involved in the breeding process, from setting goals to selecting variable, early-generation material. In PVS, farmers are given a wide range of new cultivars to test for themselves in their own fields. In our PPB programmes we have exploited the results of PVS by using identified cultivars as parents of crosses.

*Need of participatory plant breeding (PPB)*

1) To build a better linkage between users' demand and breeders' offer (spontaneous in private breeding)
2) To help the farmers to get a better output from poorly controlled cropping environments (thanks to more diverse and locally adapted genetic material)
3) To facilitate knowledge and know-how sharing between users and scientists
4) To contribute to *in situ* managing the genetic resources valuable for the local communities

It is based on the principle of sufficient knowledge of farmers' specific production needs and of the advantages and disadvantages of the local varieties they use. When farmers are involved from the first segregating generation ($F_2$ or $F_3$) and when selection is to be carried out in their own fields, PPB should necessarily be conducted with relatively few plants. That is, the number of either crosses or plants from the $F_2$ generation should be smaller than that used in a conventional programme on an experiment station. Considering this limitation, some authors consider that PPB should involve a strategy that makes only a few crosses, rigorously selects parental varieties and uses a large number of plants from the $F_2$ generation (Witcombe and Virk, 2001). Another PPB strategy comprises population improvement methods associated with recurrent selection, using a narrow genetic base and well-selected specific germplasm. It can be applied to autogamous crops when several traits are being selected and the proportion of desired recombination is expected to increase, thus limiting the risks of a strategy of few crosses (Witcombe and Virk, 2001). Progress from plant breeding has been slow in some marginal environments. Conventional or formal plant breeding (FPB) programs conducted by international agricultural research centers or national programs in developing countries have been criticized for ignoring indigenous germplasm, failing to breed for conditions facing poor farmers, and emphasizing selection for broad versus local adaptation.

A suite of techniques, referred to as participatory plant breeding (PPB) and including farmer-participatory or farmer-led selection, on-farm evaluation, and use of local landraces, has been advocated in response to this critique. PPB programs are diverse in scope and approach, but often rely heavily on farmer visual evaluation or phenotypic mass selection to select for simply-inherited traits, with limited replicated yield testing in multiple-environment trials (MET), one of the main tools of Conventional Plant Breeding. Prediction equations derived from selection theory can be used to examine the conditions under which idealized versions of CPB and PPB may be expected to achieve genetic progress for traits such as yield. The effectiveness of any selection environment is determined by both the genetic correlation between genotype performance in it and the target environment ($r_G$) and the heritability of genotypic differences in the selection environment ($H_s$). The $r_G$ is a measure of the accuracy with which performance of genotypes in the selection environment predict performance in the target environment; $H_s$ is a measure of precision with which performance differences among genotypes can be measured in the selection environment. In this paper we use this framework to compare FPB and PPB with respect to these determinants of selection response. Particular areas examined include: (i) selection for broad and specific adaptation; (ii) on-station versus on-farm selection; and (iii) selection under high-yield versus low-yield conditions.

In general, PPB systems attempt to maximize gains through the use of on-farm evaluation and the skills of farmer-selectors to maximize $r_G$. FPB exploits METs to maximize Hs. PPB is most likely to develop cultivars that out-perform the products of FPB when it is applied in low-yield cropping systems, because it is in such situations that $r_G$ between high-yield breeding nurseries and low-yield target environments is likely to be low or negative. To make continued gains, and to compete with internationally-supported CPB programs, PPB systems will need to counter the obscuring effects of uncontrollable within-field, site-to-site, and year-to-year heterogeneity. Simple and robust designs for on-farm METs are needed for this purpose.

Modern plant breeding stands among the greatest scientific and human success stories of all time. Yet the fruits of major advances in agricultural science, such as those from the Green Revolution, have by passed hundreds of millions of farmers in developing countries, most of whom operate small farms under unstable and difficult growing conditions. The adoption of new plant varieties by this group has been abysmally low. For years, this gap has haunted scientists, development workers, governments, donors, and all others with a stake in agricultural progress and the fight against poverty. But, beginning in the 1980's, it also stimulated the creation of a novel and promising set of research methods collectively known as participatory plant breeding (PPB).

*Goal of PPB*

Over the last decade, PPB has been applied as a crop improvement strategy primarily in non-commercial crops and in very unpredictable, stressed production environments. A range of other goals have also been defined within PPB programs i.e., enhancing biodiversity and germplasm conservation; developing adapted germplasm for especially disadvantaged user groups (example: women, poor farmers); making breeding programs more cost efficient, particularly through decentralization of programs which target more niches. As partners usually have to accept trade-offs in reaching certain goals, it is important at the very beginning of a PPB collaboration for those concerned scientists, farmers, development /NGO personnel- to discuss explicitly primary and secondary goals, and the minimal agreed-upon outcomes for which collaborators are aiming.

*Key assumptions for PPB*

1) Farmers are interested in participating in plant breeding
2) Farmers and scientists can successfully collaborate
3) It will not fail because:
a) The parents of crosses include locally adapted material;
b) Selection is in the local environment; and
c) Varieties are selected by farmers for the traits farmers consider important.

*Types of PPB*

PPB can be consultative and collaborative. The approach used will depend on the crop and the availability of resources.

*Consultative*

Farmers are consulted at every stage - for example, in setting the breeding objectives, choosing the appropriate parent, and by making joint selections with breeders from material grown by breeders. Hence, until there is a finished product from the breeding programme for farmers to test in PVS trials, farmers are not involved in growing material in their fields.

*Collaborative*

Farmers grow the variable PPB material in their own fields and select the best plants from it. Scientists can then obtain seed from farmers to test their selections in research station and participatory trials.

*Steps for participatory plant breeding*

1) Set the breeding objectives: a) Crop-focused PRAs, b) Analyze results of participatory varietal selection.
2) Identify the parent material: a) From local landraces, b) From varieties tested by PVS, c) From high-yielding varieties with complementary characteristics.
3) Decide on the model (consultative/collaborative): a) On the basis of available resources, b) On the basis of the crop (collaborative participation is simpler in an inbreeding crop).
4) Enter the best participatory plant breeding lines in PVS trials and facilitate their entry in normal on-station trials.
5) Prepare release proposal, if success is achieved

*Possible Outcomes/Benefits of PPB*

1) Production gains: yield increases; increases in stability of yield; faster uptake; wider diffusion; and higher market value of products.
2) Biodiversity enhancement: communities have wider access to germplasm; wider access to related knowledge; and increased inter- and intra-varietal diversity.
3) Cost-efficiencies and effectiveness: fewer researches dead-end; more opportunities for cost-sharing in research; and less expensive means of diffusing varieties.
4) Effective meeting of user needs: higher degree of farmer satisfaction; broader range of users reached,

including marginal farmers; and promotion of group learning through farm walks.

*Advantages of participatory over conventional breeding methods*

1) At least one parent in any cross is well adapted to the local environment.
2) Genotype × environment interactions is used positively because breeding is done in the target environment.
3) The impact of genotype × year interaction is probably reduced because local parental materials have adapted to local year-to-year variations.
4) Only a few crosses are made, so large $F_2$ and $F_3$ populations can be grown to increase the likelihood of selecting desirable segregants.

*Effects of PPB on adoption*

Although PPB and PVS programs are relatively recent, there are already some examples of impact. For examples there are cases where varieties preferred by farmers were identified in environments where no improved varieties have ever been available to farmers, such as the rice variety combining the frost tolerance of a landrace with the higher yield of a modern variety which has been adopted in the mountains of Nepal. A bean variety combining disease resistance with a desirable coat color has been adopted in northeastern Brazil before the variety could be formally recommended. Other examples are provided by rainfed rice varieties in India, potatoes in Ecuador, maize in Ethiopia, and, surprisingly, irrigated wheat and rice in Gujarat, in India, demonstrating that even in the areas where formal plant breeding has been particularly successful, farmer participation can identify desirable varieties at an earlier stage than in conventional breeding. One of the best examples of fast adoption through farmers' participation in variety testing (PVS), is the spreading of the rice variety Kalinga III in India. Seed of this variety, which has neither been recommended nor released, was initially made available to farmers in three villages in Rajasthan in 1993 through a farmer-managed participatory research trial. By 1997 in one of the villages 65 per cent of the area was planted with Kalinga III, while the lowest adoption was about 20 per cent of the area. The variety also spread to other villages with very high rates of spread, and with the number of villages growing Kalinga III increased by a factor of 2.3 to 7.0.

1. Participatory research (PR) increases the benefits and is more effective at reaching women and the poor.
2. PR improves research efficiency. One of the efficiency gains from including participatory approaches in plant breeding is based on the extent to which breeding priorities or research practices are reoriented in ways that save time and/or money. Here too it is useful to first look at expert opinion from the PRGA survey: 82 per cent of the respondents concluded that PR led to the formation of feedback links and 54 per cent considered that this led to changes in priorities. Another 75 per cent considered that PR led to a change in breeding methods.
3. PR accelerates adoption. The incorporation of participatory approaches consistently enables breeding programs to "break through" adoption bottlenecks caused by low levels of acceptability of new varieties to poor farmers.
4. PR changes cost structures of breeding. One of the main concerns of conventional breeding programs about the inclusion of participatory approaches into their portfolio of breeding methodologies is that PR looks very time intensive and therefore costly. Many aspects of PPB seem likely to increase costs: on farm testing begins earlier, more seed is needed of experimental varieties, the trials are dispersed outside the experiment stations, different kinds of personnel may be needed to interact effectively with farmers.

*Participatory plant breeding in maize*

A case study in Gujrat describes how plant breeders and farmers worked together to produce improved varieties of maize for the low-resource farmers of the Panchmahals district of Gujarat, India. Initially, farmers tested a range of maize varieties in a participatory varietal selection (PVS) programme. However, none of these proved to be very popular with farmers, although farmers who had more fertile fields adopted the variety Shweta from Uttar Pradesh. Hence, in 1994, a participatory plant breeding (PPB) programme was begun to generate new, more appropriate varieties. Yellow- and white-endospermic maize varieties were crossed that had been either adopted to some extent following PVS or had attributes, such as very early maturity, that farmers had said were desirable. In subsequent generations, the population was improved by mass selection for traits identified by farmers. In some generations, farmers did this in populations which were grown by breeders on land rented from a farmer. Soil fertility management was lower than that normally used on the research-station. The breeding programme produced several varieties that have performed well in research-station and on-farm trials. One of them, GDRM-187, has been officially released as GM-6 for cultivation in hill areas of Gujarat state, India. It yielded 18 per cent more than the local control in research-station trials, while being seven days earlier to silk. In farmers' fields, where average yields were lower, the yield advantage was 28 per cent and farmers perceived GDRM 187 to have better grain quality than local landraces. PPB produced a variety that was earlier to mature than any of those produced by conventional maize breeding, and took fewer years to do so. A case in point was the maize breeding program at Pantnagar, one of the leading agricultural universities in India. On-farm research was a prime component of that program, and a routine was developed to enhance the odds that information generated in the on-farm stage would be utilized in decision-making on breeding priorities (Agarwal, 1979; Biggs, 1983).

In China, a project team has been established in which local farmers cooperate with the Centre for Chinese Agricultural Policy, which is part of the Chinese Academy of Sciences, and the Guangxi Maize Research Institute. The multidisciplinary research team carries out trials in six villages and on-station using both PPB and PVS experiments. The trials allow for comparison in terms of locality, approach, objectives and the types of varieties tested. Varieties include landraces, open-pollinated varieties, so-called waxy maize varieties and varieties introduced by CIMMYT. Some of the CIMMYT varieties have been locally improved through crossings and selections. Agronomic traits, yields, taste and palatability of these improved varieties are satisfactory. They are showing better adaptation to the local environments. Varietal diversity is increasing. The project team supports farmers' groups by bringing them into contact with formal system actors through training, network building and raising awareness about markets. Since the early 1990s, scientists in CAZS-NR (Centre for Arid Zone Studies) at the University of Wales, Bangor, in the United Kingdom have worked in participatory variety selection and participatory plant breeding. For PPB, they have focused on cereals, mostly rice and maize, in marginal regions of South Asia, mainly India, Nepal, and, most recently, Bangladesh.

A major PPB project is being carried out in Guangxi province in south-west China and follows up on an impact study carried out from 1994 to1998 by the International Maize and Wheat Improvement Centre (CIMMYT) to assess the impact of CIMMYT's maize germplasm on poor farmers in south-west China (Yiching Song, 1998). One of the key findings of the impact study was that the systematic separation between the formal and the farmers' seeds system resulted in inadequate variety development, poor adoption of formally bred modern varieties, an increasingly narrow genetic base for breeding and a decrease in genetic biodiversity in farmers' fields.

*Participatory plant breeding programme (Kharif 2018)*

A client oriented comprehensive initiative was undertaken to address location-specific issues pertaining to maize cultivation in various maize growing pockets of intermediate altitude zones of valley since 2015. In this regard 22 trials were constituted comprising of advanced pipeline genotypes of maize (Table 1).

The validation trials were laid in 14 villages across various districts of the valley and one set of trial each type was also laid out at the station for metric trait estimation and validity of findings. The basic aim of the trials was to access the performance of cultivars in actual farm conditions and to accumulate feedback from the farming masses so that a redefined and comprehensive strategy can be devised for the betterment of crop productivity on long term basis.

The information generated from the trials was collected in the form of questionnaire pertaining to diverse facets of maize and rajmash cultivation, utilization and farming pattern of the individual farmers. The cumulative average results of the trials are appended based on individual experience of the farmers. Further the farmer's preference ranking of most promising genotypes (cumulative average) is also appended in the document (Table 2-5).

**Table 1**. Description of the trials

| S.No. | Crop | Trial Type | Entries involved | Checks | Locations |
|---|---|---|---|---|---|
| 1 | Maize | Baby trial | H 17, KDM 72, KDM 322 and KDM 111 | SMC 4, Local Check | 13 |
| 2 | Maize | Mother trial | SKUA-MH-46, SKUA-MH-47, SKUA-MH-49, SKUA-MH-50 | Local Check | 9 |

**Table 2.** Kernel profiling of the test entries

| Entry | Test weight (g) | Specific gravity | Protein (%) | Tryptophan in protein (%) |
|---|---|---|---|---|
| KDM 322 | 253.7 | 1.15 | 7.59 | 0.35 |
| KDM 72 | 250.0 | 1.16 | 7.97 | 0.42 |
| H 17 | 399.0 | 1.17 | 12.95 | 0.52 |
| KDM 111 | 289.2 | – | – | – |
| SMC-4 | 356.0 | 1.19 | 5.91 | 0.65 |

**Table 3.** Average coordinated performance of test entries

| Test Entry | Trial | Year | Average yield (kg/ha) |
|---|---|---|---|
| KDM 322 | 103 | 2010 | 5503 |
| KDM 72 | 103 | 2012 | 7199 |
| H 17 | 103 | 2008 | 5402 |

*Future prospects of PPB*

1) PPB should take a broader approach to solve the critical problems arising out of modern agriculture such as economic viability of crop production, food and nutritional insecurity, degradation of land, destruction of biodiversity and unsustainable agricultural practices.
2) PPB should recognize and value the knowledge and important role of women in conservation of biodiversity and agriculture and should address gender imbalances in participation, decision-making and benefit sharing.
3) PPB should be based on principles that emphasis deposition of problems to be addressed, symmetrical relationships among partners and constant evaluation and monitoring of the success of participatory improvement programmes.

**Table 4.** Average performance of test entries in station trials (*Kharif* 2013)

| Entry name | Days to 50% pollen shed | Days to 50% silking | Days to 75% dry husk | Plant height (cm) | Ear height (cm) | Shelling (%) | Grain yield (t/ha) |
|---|---|---|---|---|---|---|---|
| KDM 72 | 77 | 80 | 139 | 215 | 115 | 79 | 6.087 |
| KDM 322 | 75 | 78 | 138 | 190 | 85 | 78 | 6.321 |
| KDM 111 | 80 | 83 | 142 | 165 | 75 | 79 | 5.834 |
| H 17 | 77 | 80 | 141 | 195 | 75 | 81 | 6.917 |
| SMC 4 | 78 | 82 | 138 | 185 | 85 | 80 | 5.526 |
| SKUA-MH-46 | 77 | 80 | 137 | 200 | 100 | 78 | 7.259 |
| SKUA-MH-47 | 76 | 79 | 137 | 195 | 85 | 78 | 7.424 |
| SKUA-MH-49 | 84 | 87 | 145 | 190 | 85 | 79 | 5.908 |
| SKUA-MH-50 | 78 | 81 | 139 | 185 | 80 | 78 | 7.039 |

**Table 5.** Farmer's preference ranking for different traits and future intensions with respect to best possible test entries involved in the PPB trials over all locations (117 farmers) during *Kharif* 2013

| Entry | Maturity Fitness | Productivity performance | Crop feel | Will grow again | Taste | Realized farm yield |
|---|---|---|---|---|---|---|
| KDM 72 | 4 | 5 | 5 | Yes | 4 | 49.71 |
| KDM 322 | 5 | 4 | 3 | Yes | 3 | 51.23 |
| KDM 111 | 4 | 3 | 3 | Yes | 2 | 42.73 |
| H 17 | 5 | 5 | 5 | Yes | 2 | 57.71 |
| SMC 4 | 4 | 3 | 3 | – | 3 | 41.31 |
| SKUA-MH-46 | 4 | 5 | 4 | Yes | 4 | 59.19 |
| SKUA-MH-47 | 3 | 4 | 4 | Yes | 3 | 56.47 |
| SKUA-MH-49 | 1 | 2 | 3 | No | 2 | 35.13 |
| SKUA-MH-50 | 2 | 4 | 4 | Yes | 2 | 55.78 |

Rating Scale: 5= Excellent, 4= Good, 3= Average, 2= at par with local check, 1= below local check

4) PPB should be into account farmer's gendered priorities and practices, and strive for social and gender equity in conservation.
5) Participatory breeding need to be cost-effective, work on downstream technology and breed site-specific varieties capable of sustaining production.
6) Farmers' gendered ITK and wisdom need to be recognized rewarded and incorporated in participatory initiatives.
7) Scientific analysis of PPB data should be a course of strength for farmers for making optimal decisions.
8) PPB options should not deal only with improving yield of farmers' varieties, but also provide a strategic frame for saving grains and seeds and gainful marketing, as provided in the protection of plant varieties and farmers' Right Act, 2001. Government should make it a policy to procure farmers' landraces at a remunerative price and distribute them through the Public Distribution System (PDS).
9) Participatory research has been shown to be efficient and accelerate adoption of acceptable varieties.
10) Novel genetic combinations incorporating drought and salinity resistance need to be properly programmed into participatory breeding taking care to assign proper weight age to phenotypic expression.
11) Indigenous varieties like Indian rice with special traits like *Njavara, Kalajeera* and *Kalanamak* should have their place in participatory improvement. It is essential to characterize them for bimolecular that are relevant to the medicinal properties.

## Conclusion

Most of the data on different types of farmer participation in selection of most suitable plant ideotype suggest that there is little to lose and much to gain by involving farmers, and more generally the users, in the process of plant breeding. Decentralized-participatory plant breeding should not be seen as "an alternative" type of plant breeding somewhat opposed to the formal plant breeding, but rather as an approach to specifically address situations such as marginal environments where GE interactions are repeatable and large, precluding the adaptation of one or few varieties, or where there is a variety of different requirements (quality, crop duration, management, etc.). One specific advantage of decentralized participatory plant breeding is to rapidly adapt the crops to a changing agronomic management. Eventually, PPB could be the only possible type of breeding for crops grown in remote regions, for crops which required a high level of diversity within the same farm, or for those crops locally important but globally considered as minor crops and therefore neglected by formal breeding. In the past, researchers have relied heavily on the extension department of the Ministry of Agriculture to transfer technologies to the farmers. Without intensive interaction with farmers in the development of maize varieties, there is a possibility of ignoring certain characteristics that would be of importance to farmers in their decision making process of whether to adopt a variety or not. In the very poor, rain-fed rice-growing areas of South Asia that the Green Revolution passed by, participatory plant breeding is now paying off with strong early adoption of farmer selected varieties that provide 40% higher yields in farmers' fields. The approach needs to be more widely tested in the heterogeneous rain-fed environments of Africa, where involving farmers, especially women farmers, in selecting varieties has shown early successes for beans, maize and rice. The cost effectiveness of the approach for wider use also needs to be evaluated.

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