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# Rhamnolipid biosurfactant could be a green approach for the management of maydis leaf blight of maize

Robin Gogoi<sup>1</sup> · Sunaina Bisht<sup>1</sup> · Kirti Rawat<sup>1</sup> · Deeksha Joshi<sup>1</sup> · Nilam Sarma<sup>2</sup> · Suresh Deka<sup>2</sup>

**Abstract:** Southern corn leaf blight or maydis leaf blight (MLB) caused by *Cochliobolus heterostrophus* (Drechs.) [(anamorph *Bipolaris maydis* (Nisikado and Miyake) Shoemaker)] is an important disease of maize. Current pest management strategy relies mostly on chemical pesticides becomes a public concern due to environmental pollution. Hence use of green compounds (GRAS) has been emphasized for pest management so as to achieve sustainable agricultural productivity. Recently, biosurfactants are receiving much attention and brought in to use for plant pathogen elimination and also for increasing the bioavailability of nutrients for beneficial plant associated microbes. Biosurfactants are a diverse group of surface active molecules produced mostly on microbial cell surfaces and they can be excreted extracellularly. The present study was carried out to evaluate the efficacy of rhamnolipid (RL) biosurfactant produced by newly isolated *Pseudomonas aeruginosa* strain SS14 as an antifungal agent against *B. maydis* in *Zea mays* L. *In vitro* experiment was conducted by adopting poison food technique at different concentrations (25, 50 and 100 ppm). The studies indicated that the biosurfactant produced by this bacterial strain inhibits the growth of *B. maydis* by 39 per cent at a concentration of 100 ppm. *In planta*, treatment of maize seeds with a RL concentration of 100 mg/lit with foliar spray of 100

mg/lit resulted in improved biomass compared to those of healthy control plants and complete suppression of characteristic disease symptoms of MLB disease. The results demonstrated the possibility to develop a sustainable and eco-friendly management measure against the necrotrophic fungal pathogen *B. maydis*.

**Keywords:** Maize · *Bipolaris maydis* · Maydis leaf blight (MLB) · Rhamnolipid (RL) · Biosurfactants · Management

## Introduction

Maydis leaf blight (MLB) of maize, also known as southern corn leaf blight (SCLB) caused by ascomycetous fungus *Cochliobolus heterostrophus* (Drechs.) [(anamorph *Bipolaris maydis* (Nisikado and Miyake) Shoemaker)] synonym *Helminthosporium maydis* (Nisikado) is common in tropical and subtropical regions (Singh and Singh, 1963; Singh and Srivastava, 2012). Symptoms are exhibited in the form of small diamond shaped spots (2-6 × 3-22 mm). The center of each lesion was straw colored to light brown, surrounded by a dark brown margin. These lesions elongate as they mature, although growth of the lesions is restricted by leaf veins. Symptoms may be confined to leaves or may develop on sheaths, stalks, husks, ears and cobs (Manamgoda *et al.*, 2014). The incidence of this disease was first time reported by Drechsler (1925) from United States and by Munjal and Kapoor (1960) from Maldah (West Bengal, India). The disease is presently prevalent in almost all the agro-climatic zones in India (Agarwal *et al.*, 2022). Payak and Renfro (1968) reported that disease epidemics at an early stage causing premature death of blighted leaves which lose their value as fodder.

✉ Robin Gogoi: r.gogoiari@gmail.com;

✉ Sunaina Bisht: sunaina39486@gmail.com

<sup>1</sup>Division of Plant Pathology, ICAR-Indian Agricultural Research Institute, New Delhi-110012, India

<sup>2</sup>Institute of Advanced Study in Science and Technology (DST, GoI), Guwahati-781035, Assam, India

The MLB pathogen found on seed and sporulates on seedlings from infected seeds (Singh *et al.*, 1974). White and Ellett (1971) concluded that severely infected seeds from MLB failed to germinate and loss in plant population is directly correlated with the percentage of infected seed. It is considered to be one of the most devastating diseases as it appears in sizeable form resulting in reduction of grain yield of maize by 28 to 91 per cent (Payak and Sharma, 1980). Severe losses in grain yield due to epiphytotic have been reported in several parts of India and these losses vary from 25 to 90 per cent depending upon the severity of the disease (Bisht, 2015). Cultivation practices favoring high humidity and moderate temperature conditions may influence the development of maydis blight. MLB disease management primarily relies on the screening, identification and use of resistant varieties (Vasmatkar *et al.*, 2022; Hooda *et al.*, 2012) and initially the use of fungicide was rare, but during the last few decades an increase in use of foliar fungicides has been noticed. A variety of fungicides was reported to use for management of this disease, but only few have crop specific label claim for maize in India (Bisht, 2015; ICAR-IIMR, 2016). Further, due to their (fungicides) environmental hazards, high cost and sometime unavailability in the local market, limits the widespread use of effective chemicals by the maize growers.

Several workers reported that apart from fungicides, some botanicals (Debnath *et al.*, 2020) and biocontrol measures may also play an important role in preventing the growth of the necrotrophic pathogen *B. maydis* (Deng *et al.*, 2014; Bisht, 2015). Fluorescent pseudomonads are widely used for the purpose of biocontrol, the antifungal properties of pseudomonads against many pathogens have been reported earlier by other researchers but still are not widely exploited to control MLB of maize. Pseudomonads produce many secondary metabolites which include antibiotics and biosurfactants that are inhibitory to plant pathogens (Haas and Defago, 2005). Various types of biosurfactants have been reported to produce by Pseudomonads based on their physico-chemical properties as glycolipids, lipopeptides, neutral lipids, phospholipids, fatty acids, and polymeric compounds (Cameotra *et al.*, 2010; Goswami *et al.*, 2015; Borah *et al.*, 2016). The biosurfactants exhibit antibacterial, antifungal, and antiviral activities, which make them relevant molecules for applications in combating many diseases of crop plants (Gudina *et al.*, 2010; Kiran

*et al.*, 2015). Rhamnolipids (RLs) are one of the most widely studied biosurfactants over the years (Abdel-Mawgoud *et al.*, 2010; Randhawa and Rahman, 2014). RLs are having some antibacterial and antifungal properties which were demonstrated by many workers against a wide range of pathogens (Kiran *et al.*, 2015; Lahkar *et al.*, 2015; Borah *et al.*, 2016). In the present study, we evaluated the antifungal effect of RLs produced by *Pseudomonas aeruginosa* SS14 as a prospective antifungal molecule against MLB under laboratory and field conditions.

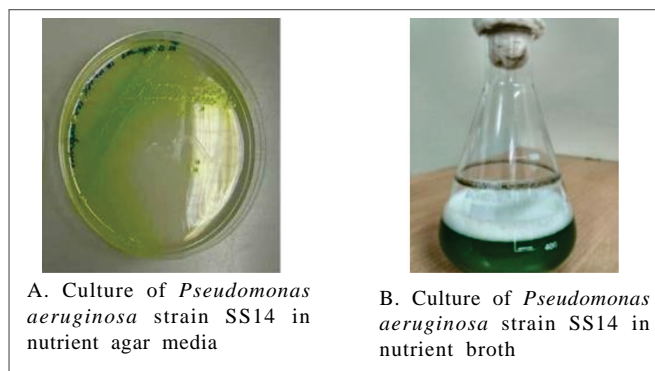
## Materials and methods

### Preparation of culture media

Mineral salt media (MSM) was prepared with the compositions (g/L)  $\text{Na}_4\text{NO}_3$ -4,  $\text{KCl}$ -0.1,  $\text{KH}_2\text{PO}_4$ -0.5,  $\text{K}_2\text{HPO}_4$ -1.0,  $\text{CaCl}_2$ -0.01,  $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ -0.5,  $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ -0.01, yeast extract -0.01 with trace element solution (1%) containing (g/L)  $\text{H}_3\text{BO}_3$ -0.26,  $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ -0.5,  $\text{MnSO}_4 \cdot \text{H}_2\text{O}$ -0.05,  $(\text{NH}_4)_6\text{Mo}_7\text{O}_{24} \cdot 4\text{H}_2\text{O}$ -0.06 and  $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ -0.7 where glucose was used as the sole carbon source (Lahkar *et al.*, 2015). The chemicals used in preparation of MSM were purchased from HiMedia. The pH of the media was adjusted to  $7.0 \pm 0.2$  (Borah *et al.*, 2015).

### Preparation of cell free supernatant (CFS)

The bacterial strain *P. aeruginosa* strain SS14 (GenBank accession no. KF031434, NCBI) was isolated from crude oil contaminated soil of upper Assam, India. A 5% inoculum from active culture of SS14 was transferred aseptically in 100 ml NB in a 250 ml Erlenmeyer flask (Figure 1) in shaking incubator (Scigenics Biotech, ORBITEK LJEIL, India) for 24 hrs at 150 rpm where temperature was maintained at  $35 \pm 1^\circ\text{C}$ . Fresh culture of SS14 from NB was inoculated (5%) in to mineral salt medium (MSM) under sterile condition and incubated in a shaker incubator at 150 rpm for 72 hrs at a temperature of  $35 \pm 1^\circ\text{C}$  (Borah *et al.*, 2016). After every 24 hrs till 72 hrs, surface tension (ST) of the bacterial cultures in MSM was measured in K1 tensiometer (Kruss, Germany) using plate method and optical density (OD) was measured using an UV visible spectrophotometer. After 72 hrs, the bacterial culture was centrifuged for a period of 15



**Figure 1.** Culture of *Pseudomonas aeruginosa* strain SS14 in different media

minutes at 10000 rpm at 4°C in a cooling centrifuge (Remi) and the cell free supernatant (CFS) was obtained. The ST of the CFS of all the bacterial cultures was measured.

#### *Production of crude biosurfactant*

For extraction of crude biosurfactant, the CFS of cultures of SS14 obtained by centrifugation was put to deproteinize at 10°C for 15 min followed by acidification with 6N HCl till pH of the CFS set at 2.0 (Lakhar *et al.*, 2015). The acidified CFS was kept at 4°C for overnight for precipitation of biosurfactant. After all the treatment, the precipitated biosurfactant was mixed with the solvent ethyl acetate (ACS) in a ratio 1:1 and shook vigorously. The solvent was then evaporated using rotary evaporator (Equitron 63R-D) at a temperature of 60°C and reduced pressure (Goswami *et al.*, 2016). The crude biosurfactant obtained so was collected in a glass beaker and dried to find out the yield in g/L unit. The production of crude biosurfactant is represented in Figure 2.

#### *Purification of column purified biosurfactant*

Silica gel (HiMedia) of mesh size 60-20 was dissolved in chloroform (ACS Mark) and a glass column of length

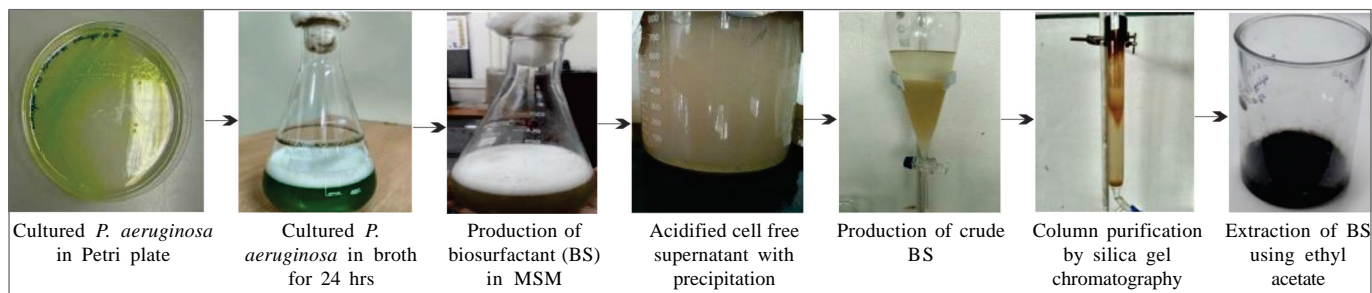
500 mm, diameter 25 mm was loaded with silica. For purification, 1g of crude biosurfactant was dissolved in 5 ml of methanol (ACS grade, Sigma Aldrich) and was allowed to elute through the glass column. A solvent system comprised of methanol and chloroform of the gradient 3:50, 5:50 and 50:50 was used for total elution of the biosurfactant. The eluted mixture was evaporated in rotary evaporator at 40°C under reduced pressure (Goswami *et al.*, 2015) and the extracted column purified biosurfactant was dried.

#### *Isolation of Bipolaris maydis*

The fungus *B. maydis* was isolated from leaf lesions obtained from maize field of the Division of Plant Pathology, ICAR-IARI, New Delhi using modified method of Sharma *et al.* (2005). Symptomatic tissues from leaves were cut into 5 mm pieces and surface-sterilized with 1% sodium hypochlorite (NaOCl) for 30–60 s. The tissues were then rinsed with three changes of sterile distilled water, dried on sterilized blotting paper and aseptically transferred to potato dextrose agar (PDA) medium in Petri plates with three pieces of tissues per plate. Plates were incubated at 25±2°C for 5 days. The mycelium growing out of the plant tissue was sub-cultured to PDA and incubated at 25±2°C again for 5 days. Purification was done by single spore isolation and identified the pathogen on the basis of cultural characters and morphological features like shapes and size of conidia and conidiophores.

#### *In vitro evaluation of biosurfactant against Bipolaris maydis*

Efficacy of the RL biosurfactant at 25, 50 and 100 ppm concentrations was studied against *B. maydis* by using poisoned food technique (Sharvelle, 1961). Crude extract of biosurfactant dissolved in acetone was determined as



**Figure 2.** Production of crude rhamnolipid biosurfactant from *Pseudomonas aeruginosa* strain SS14

stock solution of 10000 ppm concentration. Required amount of the solution was added into 50 ml flask containing 50 ml of the sterilized molten PDA to get final concentrations of 25, 50 and 100 ppm. The culture medium was mixed thoroughly before plating and subsequently poured in three Petri plates. Media mixed with acetone was poured into separate Petri plates were kept as a negative control and plates with unamended media as positive control. After solidification of media a 2 mm mycelia disc of 10 days old culture of the test pathogen was cut out with sterile cork borer and placed at the centre of each Petri plate. The Petri plates were incubated at  $25\pm 2^{\circ}\text{C}$ . Antagonistic activity was expressed in terms of percentage inhibition of mycelial growth (Vincent, 1947) measured after 10 days of incubation when the positive control plates were completely covered by the mycelia of *B. maydis* and the radial growth was compared with the test plates under observation.

#### *Field evaluation of biosurfactant for efficacy against maydis leaf blight*

##### *Field preparation*

Three field experiments were conducted to study the effect of RLs against MLB of maize for three years during *kharif* (summer), 2017-19 in the research field of ICAR-IARI, New Delhi. The experiments were laid out in randomised block design (RBD) maintaining plot size 3 m x 3 m with spacing of 20 cm and replicated thrice. One MLB susceptible cultivar of sweet corn was used for the study. The package of practices standardized by ICAR-Indian Institute of Maize Research (Parihar *et al.*, 2011) was followed during the cropping period.

##### *Inoculum preparation, inoculation and field experimentation*

Inoculum of *B. maydis* was prepared by mass multiplication of its pure culture on sorghum seeds by following the method of Ahuja and Payak (1978). Little quantity of the inoculum powder was taken in between fingers and placed into the central whorl of 30 days old (4-6 leaf stages) maize plants in the evening to allow successful infection when moisture and ambient temperature are optimum (Payak and Sharma, 1983). After 8-10 days, the plants were inoculated for the second time to avoid failure of disease establishment.

Three treatments were applied for the management of MLB of maize, *viz.*, seed treatment with biosurfactant (100 mg/kg) (T1), biosurfactant seed treatment along with foliar spray (100 mg/l) (T2), biosurfactant foliar spray (100 mg/l) (T3), Mancozeb foliar spray (2.5 g/l) (T4) and untreated control (T5). The seed treatment was given for overnight and foliar spray was performed one week after the appearance of first lesions. The per cent disease progress was recorded using blight assessment keys of the standard 1-5 scale and percent disease index (PDI) was calculated (Payak and Sharma, 1983). The observation of the effect of different treatments of RL on germination and grain yield (q/ha) were also recorded.

##### *Statistical analysis*

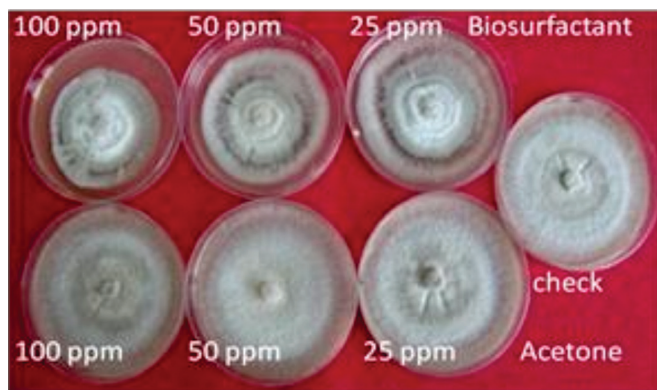
The data represented arithmetical averages of three replicates and the error bars indicate the standard deviations. Statistical analyses were performed with PASW-18 software (SPSS Inc.). Pearson's correlation was used to analyze the correlation between biosurfactant production and percentage inhibition ( $p < 0.05$ ). Results for the effect of RL on germination per cent, percent disease index and yield were analyzed using one-way ANOVA with *PostHoc* pair wise Least Significant Difference (LSD) comparison and Duccan Multiple Range Test (DMRT) at a significance level of 0.05.

## **Results**

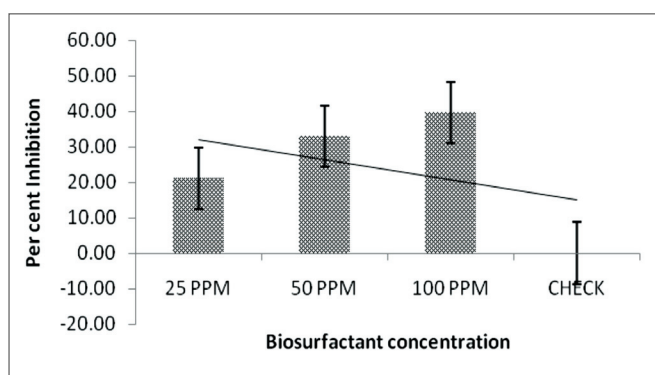
#### *In vitro efficacy of rhamnolipids against Bipolaris maydis*

The antagonistic effect of different concentrations of RL biosurfactant on mycelial growth of *B. maydis* was evaluated by calculating percentage inhibition in comparison to the control on tenth day of inoculation of the fungus (Figure 3). The assessment of antifungal activity of RL biosurfactant produced by newly isolated *P. aeruginosa* strain SS14 against *B. maydis* revealed significant inhibition in mycelial growth ( $p < 0.05$ ,  $df_{2,6}$ ; F 24.9) at all the concentrations. The RL biosurfactant exhibited 39 per cent inhibition of *B. maydis* at the concentration of 100 ppm which was followed by 33% at 50 ppm and 21% at 25 ppm (Table 1). The percentage of inhibition of the mycelium growth was gradually increased with the increase in concentration of RL biosurfactant (Figure 4).





**Figure 3.** Inhibition of mycelial growth of *Bipolaris maydis* at different concentrations of rhamnolipid biosurfactant



**Figure 4.** Percent inhibition of mycelial growth of *Bipolaris maydis* at different concentrations of biosurfactant

**Table 1.** Effect of biosurfactant on mycelial growth of *Bipolaris maydis* at different concentrations

Concentrations of biosurfactant	Mycelial growth (mm)*	Inhibition (%)*
25 ppm	7.1±0.17 <sup>c</sup>	21±1.7 <sup>a</sup>
50 ppm	6.0±0.45 <sup>b</sup>	33±5.0 <sup>b</sup>
100 ppm	5.4±0.21 <sup>a</sup>	39±2.1 <sup>c</sup>
Control	9±0.0 <sup>d</sup>	-

\* After 10 days of incubation; “mm”; values of mycelial growth (MG) and % inhibition is mean of three replications; ± values represent standard deviations (SD); Different letters within each column indicates significantly different values according to LSD at alpha = 0.05.

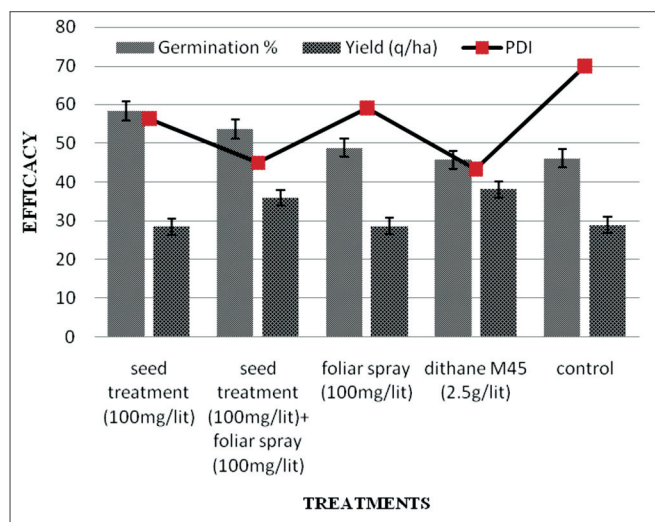
*Field evaluation of biosurfactant for efficacy against maydis leaf blight*

The RL biosurfactant was found effective in improving germination percent of sweet corn variety of maize (Table 2; Figure 5). As per observation, the highest 58.3 percent germination was recorded in the maize seeds treated with a concentration of 100 mg/l of biosurfactant and 53.6 per cent germination was in seed treatment (100 mg/l) coupled later with foliar spray (100 mg/l) treatment. Seed treatment with RL biosurfactant exhibited similar results

**Table 2.** Effect of rhamnolipid biosurfactant on seed germination, percent disease index of maydis leaf blight and grain yield of maize during *kharif* (summer) 2017-2019

Rhamnolipid biosurfactant	Germination (%)				PDI (%)				Yield (Q/ha)			
	2017	2018	2019	Pooled*	2017	2018	2019	Pooled*	2017	2018	2019	Pooled*
Seed treatment (100 mg/l) (T1)	65.0	50.0	60.0	58.3±7.6 <sup>a</sup>	67.5	67.5	65.5	56.4±17.4 <sup>a</sup>	15.9	35.0	34.5	28.5±10.8 <sup>a</sup>
Seed treatment (100 mg/l) + Foliar spray (100 mg/l) (T2)	62.0	55.0	44.4	53.6±9.0 <sup>a</sup>	49.8	49.8	50.1	44.9±8.6 <sup>a</sup>	18.4	46.9	42.5	35.9±15.3 <sup>a</sup>
Foliar spray (100 mg/l) (T3)	60.3	46.7	40.3	48.8±10.3 <sup>a</sup>	67.5	67.5	64.5	59.1±12.0 <sup>a</sup>	14.4	34.8	37.1	28.8±12.3 <sup>a</sup>
Mancozeb (2.5 g/l) (T4)	66.7	40.0	30.6	45.7±18.7 <sup>a</sup>	47.3	47.3	50.1	43.2±9.5 <sup>a</sup>	20.8	48.9	44.4	38.0±15.0 <sup>a</sup>
Control (water) (T5)	60.0	45.0	33.3	46.1± 13.3 <sup>a</sup>	86.7	86.7	75.6	70.0 ±20.0 <sup>a</sup>	11.1	33.1	40.0	28.0 ±15.9 <sup>a</sup>

\*Data are means of three replicates ± SD. Different letters within each column indicate significantly different values according to LSD at alpha = 0.05. ANOVA was performed for germination percent (F<sub>4,9</sub> = 0.47), PDI (F<sub>4,10</sub> = 1.7), Yield (F<sub>4,10</sub> = 0.32)



**Figure 5.** Effect of rhamnolipid biosurfactant on seed germination, percent disease index of maydis leaf blight and grain yield of maize during *Kharif* (summer) 2017-2019

( $F_{4,9} = 0.47$ ,  $p < 0.05$ ) with that of mancozeb. It is interesting to note that, both seed treatments (T1 and T2) with RL biosurfactant induced better germination of the maize seeds than the rest treatments (T3, T4 & T5).

Reduction in the percent disease index (PDI) of MLB was achieved due to application of RL biosurfactant (Figure 5). Seed treatment (100 mg/l) combined with foliar spray (100 mg/l) of biosurfactant (T2) exhibited lowest PDI (approx. 45%,  $F_{4,10} = 1.7$ ,  $p < 0.05$ ) which was at par with mancozeb spray (43.2%). The three years data showed significant difference among all the treatments in comparison to plain water spray ( $p < 0.05$ ).

Among all the treatments, maize grain yield (q/ha) was maximum (35.9 q/ha) in seed treatment (100 mg/kg) combined with foliar spray (100 mg/l) of biosurfactant (T2) which exhibited comparable result ( $F_{4,10} = 0.32$ ,  $p < 0.05$ ) with that of the fungicide spray (38.0 q/ha). It was followed by the treatment of foliar spray (100 mg/l) which contributed grain yield nearing about 29 q/ha. In case of seed germination percent and yield of consecutive three years, no significant difference was found among the treatments in comparison to the recommended fungicide mancozeb application, but significant difference was observed among all the treatments in comparison to the control (water) (LSD  $p < 0.05$ ).

## Discussion

Biosurfactants are reported to produce by bacteria, yeasts, and fungi which can serve as green surfactants. The

rhamnolipid biosurfactants are considered as less toxic and eco-friendly, and thus several types of biosurfactants encompass the potential to produce commercially for extensive applications in pharmaceuticals, cosmetics, and food industries (Cardoso *et al.*, 2010; Saikia *et al.*, 2012). Such biosurfactants synthesized by microorganisms also show evidence of promising role in the agricultural industry (Canova *et al.*, 2010). Several reports had highlighted the role of biosurfactants for plant pathogen elimination and for increasing the bioavailability of nutrients to the beneficial plant associated microbes (Sachdev and Cameotra, 2013; Goswami *et al.*, 2015).

In the present study, management of maydis leaf blight disease in a sweet corn variety of maize was studied by using biosurfactant. The biosurfactant was produced by newly isolated *P. aeruginosa* strain SS14 using mineral salt media. The crude biosurfactant was identified as rhamnolipids (Borah *et al.*, 2016; Goswami *et al.*, 2014). The rhamnolipid (RL) biosurfactant exhibited inhibition of 39 per cent of *B. maydis* at concentration of 100 ppm which was followed by 33% inhibition at 50 ppm and 21% at 25 ppm. The percentage inhibition in mycelium growth was gradually increased with the increase in concentration of RL biosurfactant. The results of the present study were truly anticipated as the reports of previous workers opined that rhamnolipids can be a potential antifungal agent to control various diseases of several crop plants (Borah *et al.*, 2016; Goswami *et al.*, 2015; Yang *et al.*, 2014). The unique analogues present in the RLs produced by the strain SS14 seemed to enhance the antifungal activity as reported earlier (Benincasa and Accorsini 2008; Borah *et al.*, 2016). Biological control with bacteria and the biosurfactant produced by them offers a simple and cost-effective strategy for managing various phytopathogens (Pacwa *et al.*, 2011; Sachdev and Cameotra, 2013). The biosurfactant was very effective in inhibiting the mycelial growth of the fungus and suppressing the disease symptoms as well as found highly effective in increasing crop growth (Ngullie, 2010; Gamalero and Glick, 2011).

Commonly used surfactants are divided on the basis of polar and non-polar groups into synthetic surfactants and biosurfactants (Banat *et al.*, 2010; Zhong *et al.*, 2016). The mechanism of RL action against microorganisms is not yet elucidated, but, it is supposed that the cell membrane is the target. Some studies showed that biosurfactants can increase the permeability of

microbial cells (Sotirova *et al.*, 2008), cell surface charge and CSH (Kaczorek *et al.*, 2010; Kim *et al.*, 2015). Liu *et al.* (2012) observed that rhamnolipids can modify the CSH and the surface charge of *Penicillium simplicissimum* due to the change of the cell surface functional groups (increased the hydrophobic functional group) and element concentrations. This will alter the membrane structure and character, which will amend the major membrane functions (such as matter transport, energy generation and membrane permeability, and cell surface properties (Sotirova *et al.*, 2008; Banat *et al.*, 2010). Sotirova *et al.* (2009) observed that RLs increased the membrane permeability of *Bacillus subtilis* and *Pseudomonas aeruginosa*, resulting in metabolite leakage leading to the change of cell surface properties. It was also reported that surfactants can induce increase of membrane lipids and led to the decrease of membrane fluidity of *Arthrobacter* sp. strain Sphe3 (Kallimanis *et al.*, 2007). The effects of RLs on cell surface properties depend on the concentration and types of rhamnolipids, the species of microorganism, and environmental conditions etc. (Yuan *et al.*, 2007). However, no correlation with the antimicrobial activity was demonstrated.

The RL biosurfactant was found successful in improving germination and grain yield of maize. The maize seeds treated with RL biosurfactant showed better rate of germination which exhibited comparable results with the recommended fungicide. Foliar spray of this RL biosurfactant affected expression of the disease symptom on the leaves. The external symptoms were appeared in the form of small diamond shaped spots (2-6 × 3-12 mm). The center of each lesion was straw colored to light brown, surrounded by a dark brown margin and restricted only on the affected leaves. Otherwise, diseased lesions appear on the entire leaf lamina under severe condition and also may develop on the sheaths, stalks, husks, ears and cobs (Manamgoda *et al.*, 2014). Keeping consistency with this, the percent disease index in the present study was evaluated in terms of symptomatic leaf infections using blight assessment keys (Payak and Sharma, 1983). Further, yield (q/h) was evaluated to monitor the effect of disease on plant growth and health and ultimately on the cobs. The use of RL biosurfactants for plant growth promotion was also well documented owing to their detrimental effect on pathogens (Gamalero and Glick, 2011). On account of the same reason, RL

biosurfactant could effectively reduce intensity of MLB disease which was comparable with the performance of mancozeb, a fungicide recommended in the package of practice of maize cultivation. Among the entire treatments considerably higher yield (q/ha) was obtained by practicing seed treatment and subsequent foliar spray with RL biosurfactant @ 100mg/l. The enhanced grain yield resulted from dual application of RL biosurfactant has indicated about its role in encouragement of better plant health and suppression of the MLB diseases more effectively.

## Conclusion

In conclusion, the results of the present investigation reveal that the RL biosurfactants produced by the bacterial strain *P. aeruginosa* strain SS14 have strong antifungal property against the necrotrophic fungal pathogen *B. maydis*. The performance of biosurfactant was similar with the recommended fungicide mancozeb with respect to MLB disease control. However, its mode of action against the target pathogen needs to be determined. Biosurfactants as a whole are highly biodegradable under natural environmental conditions. They are non-toxic and safe for the environment, which may offer a possibility of their application as an alternative to the synthetic fungicides to control the fungal diseases of crops like maydis leaf blight of maize.

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## ***In vitro* studies on bio efficacy of native isolates of *Trichoderma viride* against maize stalk rot disease incited by *Fusarium verticillioides***

Mamatha Ch. · Mallaiah B. · Vidyasagar B. · Bhadru D. · M. V. Nagesh Kumar · K. Vanisree

**Abstract:** *Fusarium verticillioides* is a soil born plant pathogen causing stalk rot diseases in several plants including maize (*Zea mays* L.) and management of stalk rot diseases are of very difficult with existing chemical methods. So, the main objective of this experiment is to identify effective *Trichoderma* isolates against *Fusarium verticillioides*. In this connection *in vitro* experiments are conducted to evaluate the anti-fungal activity of ten native isolates of *Trichoderma viride*, collected from different locations in Telangana state against maize stalk rot pathogen incited by *Fusarium verticillioides*. Among the isolates evaluated, Tv-8 isolate collected from Gundlapally village of Karimnagar, Dist. recorded the maximum (77.8 %) inhibition of mycelial growth (1.96 cm) of the pathogen at 10 days after inoculation and it was followed by Tv-6 isolate collected from Sitaramapuram village of Warangal, Dist. and Tv-3 isolate collected from Lachhigudem village of Khammam, Dist. with 73.9 and 71.6 per cent inhibition of mycelial growth of the pathogen over control respectively. Tv-1 isolate collected from Tanikella village of Khammam, Dist. was found to record the minimum inhibition of mycelial growth of the pathogen (5.83 cm) at ten days after incubation which counted for 34.1 per cent growth reduction over control. These results signifies the importance of exploration of native bio agents for effective management of soil born diseases like maize stalk rots.

**Keywords:** Biological control, *Fusarium verticillioides*, Maize, *Trichoderma viride*

✉ Mallaiah B.: mallyagrigo@gmail.com

Department of Plant Pathology, Maize Research Centre, PJTSAU, Rajendranagar, Hyderabad 500030, Telangana, India

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### **Introduction**

Maize (*Zea mays* L.) is one of the most versatile emerging crops having wider adaptability under varied agro-climatic conditions. It is cultivated in tropics, sub tropics and temperate regions under irrigated and rain fed conditions. Globally, maize is known as queen of cereals, because it has the highest genetic yield potential among the cereals.

Maize is affected by various biotic and abiotic stresses. Among the biotic stresses, fungal diseases are one of the major constraints in realizing the potential yields of this crop. Of the fungal diseases, post flowering stalk rots poses a major threat to the productivity of maize crop. Post flowering stalk rot is a complex disease which occurs at post flowering stage of the crop in both *kharif* and *rabi* season. In India, eight fungi and three bacteria were reported to cause stalk rots (Raju and Lal, 1976). Among all, *Fusarium* stalk rot (*Fusarium verticillioides*), Charcoal rot (*Macrophomina phaseolina*) and late wilt (*Cephalosporium maydis*) are more prevalent and destructive in India (Khokhar *et al.*, 2014). Among the stalk rots, *Fusarium* stalk rot caused by *F. verticillioides* was first reported from USA by Pammel (1914) as a serious root and stalk disease. Later, Valleau (1920) reported that *F. moniliforme* was a primary cause of root and stalk rot of maize. In India, the disease was first reported from Mount Abu, Rajasthan (Arya and Jain, 1964) and prevalent in most of the maize growing areas of country where water stress occurs at the flowering stage of the crop. The disease becomes apparent when crop enters senescence phase and severity increases during grain filling stage. The rotting extends from the infected roots to the stalk and causes premature drying, stalk breakage and ear dropping and thus resulting in

reduction of maize yields (Colbert *et al.*, 1987). The disease causes internal decay and discoloration of stalk tissues, directly reducing yield by blocking translocation of water and nutrients, thus resulting in death and lodging of the plant (Dodd, 1980). The fungus survives on crop residues in the soil or on the soil surface.

Use of chemicals is expensive and the heavy usage of chemicals is hazardous to the environment. Biological control of plant pathogens using microorganisms has been considered as more natural and an environmentally acceptable alternative to the chemical controlling methods. *Trichoderma* spp. have been found as an effective BCA against many soil borne pathogens (Eziashi *et al.*, 2006, Jat *et al.*, 2018). *Trichoderma* controls pathogens in an indirect way by producing several groups of antibiotics that inhibit the growth of the pathogen. Apart from that, they act directly by showing antagonism against the pathogen which is called mycoparasitism. *Trichoderma* species can also inhibit or reduce the growth of plant pathogens especially fungi, through competition for space, enzyme substrates, nutrients, and or oxygen (Sanchez *et al.*, 2006). Therefore, *Trichoderma* species have been used as BCAs for phyto-pathogenic fungi to control plant diseases and they have very good potential in controlling *F. verticillioides* successfully.

Keeping in view the importance of stalk rot disease in maize and native isolates of *Trichoderma* species the present experiment was formulated to evaluate the efficiency of different isolates collected from different places in Telangana state.

## Materials and methods

### *Isolation of native Trichoderma spp.*

Antagonistic fungi were isolated from the rhizosphere soil collected from different crops grow in various places of Telangana. The plants were pulled out gently with intact roots and the excess soil adhering on roots was removed gently. Ten grams of rhizosphere soil collected from different crops was transferred to 250 ml Erlenmeyer flask containing 100 ml of sterile distilled water separately. After thorough shaking, the antagonist present in the suspension was isolated by serial dilution plate method. From the final dilutions of  $10^{-3}$ ,  $10^{-4}$ ,  $10^{-5}$  and  $10^{-6}$ . One ml of each aliquot was pipetted out poured into sterilized

petri dishes containing *Trichoderma* selective medium (TSM), they were gently rotated clockwise and anticlockwise for uniform distribution and incubated at room temperature ( $28 \pm 2^\circ\text{C}$ ) for 24 hours. Colonies with characteristics of *Trichoderma* spp. was isolated from TSM medium and the culture was purified in PDA medium. The pure cultures of the antagonists were maintained on respective agar slants of  $4^\circ\text{C}$  respectively.

### *Screening of fungal antagonists*

Isolates of *Trichoderma viride* were screened against *Fusarium verticillioides* by dual culture method (Dennis and Webster, 1971). A nine mm mycelial disc of *Fusarium verticillioides* and each isolate of *T. viride* were placed opposite to each other near the periphery of the petridish separately and incubated at room temperature ( $28 \pm 2^\circ\text{C}$ ). The petridishes with pathogen alone were kept as control. After 10 days of incubation, mycelial growth of the pathogen was measured in each petridish separately and expressed in cm. Per cent inhibition of mycelial growth of the pathogen by different isolates of antagonists was calculated using the formula suggested by Vincent (1947). Complete randomized design was used for statistical data analysis.

## Results and discussion

### *Isolation and identification of Trichoderma*

Ten isolates of *Trichoderma* were isolated from the collected samples of different locations of Telangana state (Table 1, Plate 1). The identification was carried out according to the morphological and microscopic characteristics. The conidia were globose to sub globose with light green colour with flask shaped phialides in a divergent group of 2-4.

Identification of *Fusarium verticillioides*: Microconidia were hyaline, oval to club shaped with a flattened base and measured  $5.12-7.11 \times 2.04-3.18$  (L×W). They were formed from monophialides and were found in long chains. Macroconidia observed were sickle shaped, hyaline with apical cell curved and tapered, and basal cell notched. They were typically 4-6 celled with 3-5 septa and measured  $20.01-31.12 \times 2.01-3.21$  (L×W) chlamydo spores were globose, inter calary, solitary or in chains.

**Table 1.** Efficacy of isolates of *Trichoderma viride* against mycelial growth of *Fusarium verticillioides* (F-ISO-7) *in vitro*

S.No.	Place of collection	District	Isolate	Mycelial colonization (cm)* at 10 DAI	(%) Growth inhibition over control
1	Tanikella	Khammam	Tv-1	5.83	34.1
2	Narasimhapuram	Khammam	Tv-2	3.99	54.9
3	Lacchigudem	Khammam	Tv-3	2.51	71.6
4	Wyra	Khammam	Tv-4	4.46	49.6
5	Pallipadu	Khammam	Tv-5	3.41	61.5
6	Sitaramapuram	Warangal	Tv-6	2.31	73.9
7	Arepally	Warangal	Tv-7	4.22	52.3
8	Gundlapalli	Karimnagar	Tv-8	1.96	77.8
9	Thimmapur	Karimnagar	Tv-9	4.90	44.6
10	Rajendranagar	Rangareddy	Tv-10	5.20	41.3
11	-	-	Control	8.86	-
			CD (P=0.05)	0.034	-
			SE(m) ±	0.012	-
			C. V.	0.607	-

\*Mean of five replication DAI– Days after incubation

**Plate 1.** *Trichoderma* spp. isolated from the different rhizosphere soils

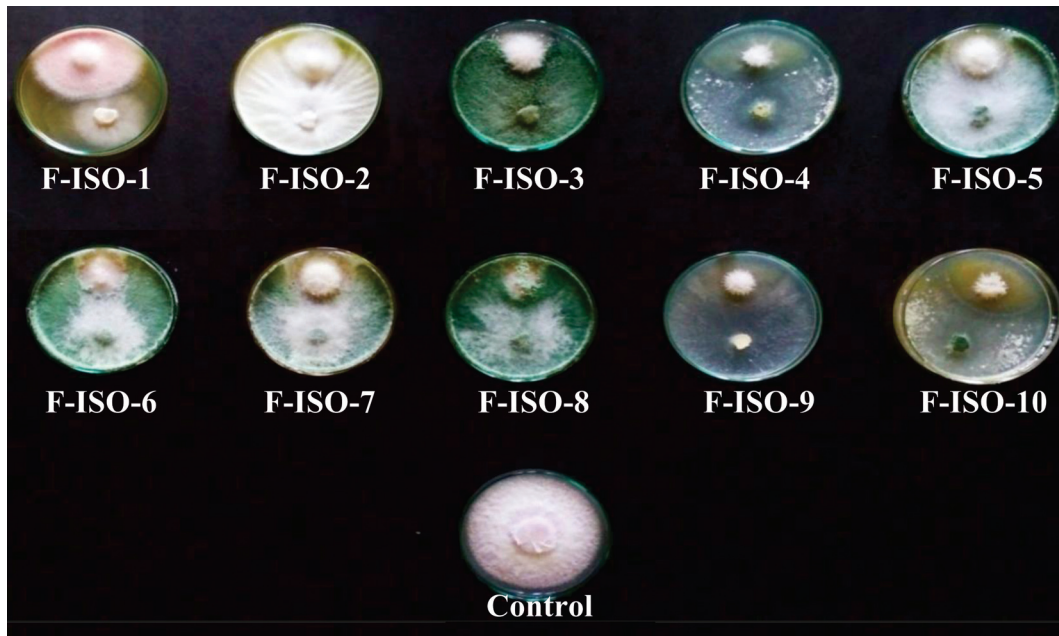
### Efficacy of isolates of *Trichoderma viride* against *Fusarium verticillioides*

Ten isolates of *Trichoderma viride* were tested for their antagonistic activity against *Fusarium verticillioides* by dual culture technique (Plate 2). Among the isolates tested, Tv-8 isolate recorded the maximum (77.8%) inhibition of mycelial growth (1.96 cm) of the pathogen at 10 days after inoculation and it was followed by Tv-6 and Tv-3 with 73.9 and 71.6 per cent inhibition of mycelial growth of the pathogen over control respectively. Tv-1 was found to record the minimum inhibition of the pathogen (5.83

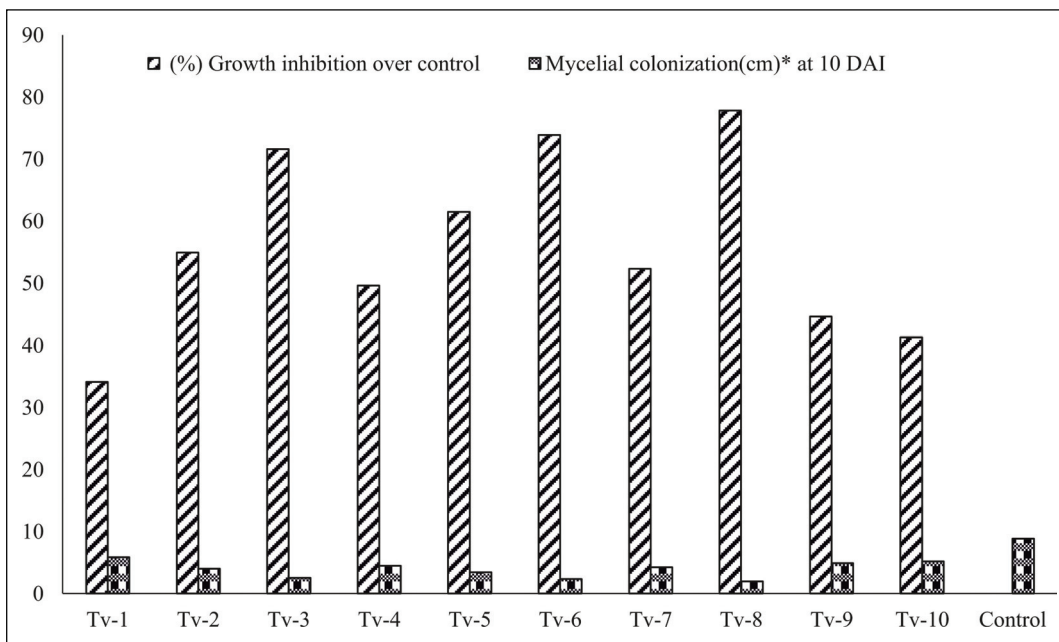
cm) at ten days after incubation which counted for 34.1% growth reduction over control (Table 1) (Figure 1).

Bunbury-Blanchette and Walker (2018) isolated seven native *Trichoderma* species and reported antagonism against *Fusarium oxysporum* f.sp. *cepae* (FOC) in dual culture. Among them *Trichoderma hamatum* and *T. harzianum* most frequently elicited multiple signs of antagonism. Dubey *et al.* (2006) evaluated ten isolates belonging to three species of *Trichoderma* against *Fusarium oxysporum* f.sp. *ciceris* in dual culture and reported antagonistic activity due to production of volatile and non-volatile inhibitors.





**Plate 2.** Efficacy of isolates of *Trichoderma viride* against *Fusarium verticillioides* (F-ISO-7) *in vitro*



**Figure 1.** Efficacy of different isolates of *T. viride* against mycelial growth of *F. verticillioides* *in vitro*

The *T. viride* isolated from Ranchi followed by *T. harzianum* (Ranchi) and *T. viride* isolated from Delhi recorded maximum mycelial growth of the pathogen. Meki *et al.* (2011) also evaluated 32 *Trichoderma* isolates against *Fusarium oxysporum* f.sp. *ciceris*, among them isolates T3, T10 and T23 were fastest in their colony growth, it suggested a high potential for competition for nutrients and space against the pathogen. This appeared to be true in a dual culture test, where most *Trichoderma* isolates showed a high level of inhibition of the pathogen.

Sravankumar *et al.* (2016) screened 100 strains of *Trichoderma* as potential biocontrol agents against *Fusarium* wilt in cucumber both under *in vitro* and *in vivo* methods, among them 10 isolates inhibited the growth of the pathogen with more than 85 per cent inhibition and in greenhouse trials 1 strain *Trichoderma asperellum* was able to decrease the severity of *Fusarium* wilt by 71.67 per cent. Anitha *et al.* (2010) tested efficacy of six isolates of *T. harzianum* against *M. phaseolina* and found that Hyderabad isolates of *T. harzianum* caused

maximum inhibition (62.3%) of radial growth of *M. phaseolina* and regarded as potential bio control agent for minimizing PFSR incidence on maize. The results of present study provide a strong basis for further development of these isolates as bio inoculants to attain the desired disease management on maize crop.

## Conclusion

This investigation sheds new light on the ways for the use of *Trichoderma* spp. in controlling the plant pathogen *Fusarium verticillioides* which is one of the serious fungal pathogens which cause wilting in maize plants. The precise benefits and consequences of the present findings open several avenues for future research in the field of bio control for sustainable crop yields.

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## Effect of zinc-based nano fertilizer foliar application with varying fertility levels on yield attributes and yield of single cross maize hybrid (*Zea mays* L.)

Piyush Choudhary<sup>1</sup> · D. Singh<sup>2</sup> · D. P. Singh<sup>3</sup> · Ramniwas<sup>4</sup> · Shankar Lal Kumawat<sup>5</sup> · Gograj Ola<sup>6</sup>

**Abstract** An experimental field study was carried out during the consecutive *kharif* seasons of 2020 and 2021 at the Instructional Farm of Rajasthan College of Agriculture, Udaipur using single cross maize hybrid. The study encompassed two main factors: the foliar application of nanofertilizer and different fertility levels with three replications. Factor A (Foliar Application of Nanofertilizer) serve the parameters like control receiving water spray, application at the knee-high stage (0.1%), application at the 50 per cent tasseling stage (0.1%), and combined application at both the knee-high and 50 per cent tasseling stages (0.1%) however Factor B represents focusing on fertility levels, included the control, 100 per cent recommended dose of fertilizer (RDF), 90 per cent RDF, and 80 per cent RDF. Significantly highest grain, stover and biological yield (51.90, 82.32 and 134.21 q ha<sup>-1</sup>) were recorded with the dual foliar application of nanofertilizer at knee high stage and at 50 per cent tasseling stage over single stage foliar application. Among the different fertility levels, the application of 90 per cent RDF significantly increased grain, stover and biological yield. Yield attributing characters viz., grains cob<sup>-1</sup>, grain

weight cob<sup>-1</sup> (g) and 1000 grain weight (g) were significantly higher with the dual foliar application of nanofertilizer at knee high stage and at 50 per cent tasseling stage and application of 90 per cent RDF in maize. Similarly, the significantly highest protein content of maize (11.13% and 10.97%) was found in dual foliar application of nanofertilizer and 90 per cent RDF, respectively. These findings emphasize the importance of strategic nanofertilizer application and optimal RDF levels in enhancing maize yield and protein content.

**Keyword:** Maize · Zinc · Nano fertilizer · yield

### Introduction

Maize is a versatile crop that integrates effectively into existing cropping systems. In India, it holds significance not only due to its extensive acreage but also because of its adaptability across a wide range of agro-climatic conditions. Currently, maize is cultivated over 9.72 m ha area with 28.64 mt production and an average yield of 29.45 q ha<sup>-1</sup> In Rajasthan, the crop occupies 0.97 m ha area with an annual production of 2.70 mt and average yield of 27.69 q ha<sup>-1</sup> (Govt. of India, 2021). Amongst major nutrients, (N) is the most vital nutrient element and performs as a component of many organic compounds viz. proteins, amino acids, nucleic acids and nucleotides, enzymes, vitamins, hormones, alkaloids etc. (P) is the second most essential primary macronutrient, crucial for crop growth and yield. Among micronutrients, (Zn) is the most important micronutrient for crops as per its imperative role in the plant's enzyme system as a co-factor. Presently deficiency of Zn has become so widespread that it ranks next to N and P in Rajasthan as

✉ Piyush Choudhary: piyushdudi@gmail.com

<sup>1,2</sup>Department of Agronomy, Rajasthan College of Agriculture, MPUAT, Udaipur, Rajasthan, India

<sup>3</sup>Department of Soil Science and Agricultural Chemistry, Rajasthan College of Agriculture, MPUAT, Udaipur, Rajasthan, India

<sup>4</sup>ICAR-Indian Institute of Maize Research, New Delhi-110012, India

<sup>5</sup>Junagadh Agriculture University, Junagadh Gujarat, India

<sup>6</sup>Swami Keshwanand Rajasthan Agriculture University, Bikaner, Rajasthan, India

well as many other states under intensive cropping systems (Zhu *et al.*, 2012). Thus, a higher yield of a single cross-maize hybrid can be obtained through the judicious use of N, P and Zn. Nanotechnology holds significant potential in modern agriculture, addressing both plant diseases and the limited availability of important plant nutrients (Parisi *et al.*, 2015). A variety of nanomaterials, mostly metal and carbon-based nanomaterials has been studied in maize and wheat for plant growth (Yang *et al.*, 2015; Choudhary *et al.*, 2022a). Recent discoveries reported that various nanomaterials applied to different crops can be serve as nano nutrients. Assessing the potential of nanomaterials to efficiently fulfill plant nutrient requirements, as compared to conventional fertilizers, is a prudent approach (Raliya *et al.*, 2018; Choudhary *et al.*, 2022b). Chitosan-based nanomaterials like Cu, Zn and salicylic acid (SA) exhibit great potential to be used in crop plants as fertilizers (Choudhary *et al.*, 2019).

## Materials and methods

The experiment was conducted during *kharif* 2020 and 2021 at the Instructional Farm, Rajasthan College of Agriculture, MPUAT. The climate in the region exhibits sub-tropical characteristics, marked by mild winters and moderate summers. Notably, the monsoon period, spanning from July to September, brings about higher relative humidity. The experiment was laid out in Factorial Randomized Block Design (RBD) with three replications. It comprised sixteen treatment combinations, encompassing two factors. The first factor involved four levels of foliar nanofertilizer application: a *control* treated with water spray, application at the knee-high stage (0.1%), application at the 50 per cent tasseling stage (0.1%), and combined application at both the knee-high and 50 per cent tasseling stages (0.1%). The second factor pertained to fertility levels, encompassing the control, 100 per cent RDF, 90 per cent RDF, and 80 per cent RDF. The recommended fertilizer dosage comprised 120, 60 kg, and 25 kg ha<sup>-1</sup> N, P<sub>2</sub>O<sub>5</sub> and Zn respectively. Treatment application was structured as follows: the entire phosphorus quantity was applied at sowing, while nitrogen was split into three applications: one-third at sowing, one-third at the knee-high stage, and the remaining third at the 50 per cent tasseling stage. The Zn-chitosan NPs were formulated at the Department of Molecular Biology

and Biotechnology, Rajasthan College of Agriculture, Udaipur, utilizing ionic gelation between chitosan and sodium tripolyphosphate, as described by Saharan *et al.* (2015) and Kumar *et al.* (2020). The application of Zn-chitosan NPs in a 0.1 per cent solution sprayed at the knee-high stage and the 50 per cent tasseling stage, aligning with the designated treatments. The Zn-chitosan NPs 0.1 per cent solution was sprayed at knee high stage and at 50 per cent tasseling stage as per treatments. For the cultivation of maize, the Pratap Hybrid Maize-3 variety was sown with a seed rate of 20 kg ha<sup>-1</sup>, coinciding with the commencement of rainfall on July 4<sup>th</sup> in 2020 and July 5<sup>th</sup> in 2021. The crop was spaced at dimensions of 60 x 25 cm.

## Results and discussion

### *Yield attributes*

Number of grains cob<sup>-1</sup> (322.8), grain weight cob<sup>-1</sup> (134.39), 1000 seed weight (236.24), cob length (19.28 cm), girth of cob (16.14 cm) and cob height (96.15 cm) of maize were significantly influenced by the foliar application Zn based nanofertilizer at dual stage *viz.*, at knee high stage and at 50 per cent tasseling stage as compared to single stage application (Table 1). This is strongly support that sink strength are advocated by higher sink activity and sink size (Choudhary *et al.*, 2019). Based on these outcomes, the study hypothesizes that slow release of Zn outdoes the plant's antagonistic response by awfully synchronizing the need for zinc at the most ravenous-dynamic development (reproductive and grain filling) stages of maize plant. Furthermore, availability of N and P at the aforementioned crucial stages is also essential for growth and development of maize plants (Razaq *et al.*, 2017). Zn based chitosan nanofertilizer used in present study contain 7.40 per cent N as -NH<sub>2</sub> group in chitosan backbone and 4.89 per cent P as -PO<sub>4</sub> group in TPP cross-linker (Sharma *et al.*, 2020). Therefore, study can't rule out the potential contribution of N and P from zinc chitosan based nanofertilizer to complement source-activity and sink-strength of plants. Outcomes of the study exclusively establish the fact that an conducive cellular environment is imperative for sustained source-activity and sink-strength and the slow release of Zn plays an elite role in enhancing the cellular homeostasis of maize plant for

**Table 1.** Effect of foliar application of zinc based nanofertilizer and varying fertility levels on yield attributes of maize

Treatment	Cob length (cm)	Girth of cob (cm)	Grains cob <sup>-1</sup>	Cob height (cm)	Grain weight cob <sup>-1</sup> (g)	1000 grain weight (g)
<i>Foliar application</i>						
Control	15.6	13.8	263.3	88.8	105.6	198.0
At knee high stage	17.5	15.6	301.3	92.2	121.4	225.2
At 50% tasseling stage	17.2	15.4	297.8	91.2	115.5	219.9
Both stages	19.2	16.1	322.8	96.1	134.3	236.2
SEm±	0.18	0.14	3.44	0.78	1.21	1.11
C.D. (P = 0.05%)	0.52	0.40	9.72	2.20	3.43	3.15
<i>Fertility levels (N, P and Zn kg ha<sup>-1</sup>)</i>						
Control	14.9	13.4	242.2	88.4	96.6	193.9
80% RDF	16.7	15.1	281.9	89.8	114.4	214.0
90% RDF	18.7	16.0	326.5	94.1	131.4	234.2
100% RDF	19.2	16.4	334.7	96.0	134.5	237.2
SEm±	0.18	0.14	3.44	0.78	1.21	1.11
C.D. (P = 0.05%)	0.52	0.40	9.72	2.20	3.43	3.15

higher growth, yield and nutrient content in grain and stover (Choudhary *et al.*, 2019). Application of 90 per cent RDF significantly influenced the all-yield attributing characters as compared to 80 per cent RDF and control. The level of Zn in the leaves increased via foliar application of Zn based chitosan nanofertilizer. These improvements suggest greater availability of metabolites and nutrients synchronized to demand for growth and development of each reproductive structure. The better availability of inputs as evident from N, P, and Zn content present in leaves showed reduce competition of these between developing structure. This, in turn, enhances the functional activities of each vegetative structure and results in robust individual plant growth, as evidenced by increased yield attributes.

The positive response of maize under N, P and Zinc fertilization is in close conformity with the findings of different researchers *i.e.*, Sharma *et al.* (2020); Van *et al.* (2020); Kumar *et al.* (2020) and Choudhary *et al.* (2022c).

### Yield

The grain, stover and biological yield (51.9, 82.3 and 134.2 kg ha<sup>-1</sup>) of maize significantly increased to the tune of 15.1, 19.6 and 17.8 per cent with the foliar application of 0.1% zinc based nanofertilizer at knee high stage and 50 per cent tasseling stage over control, respectively (Table 2). Application of 90 per cent RDF

had significant influenced on grain, stover and biological yield of maize over 80 per cent RDF and control, although highest yield was recorded with 100 per cent RDF. The grain yield depends on the synthesis and accumulation of photosynthates and their distribution among various plant parts. The synthesis, assembly, and translocation of photosynthates depend upon the efficient photosynthetic structure and the extent of translocation into the sink (grains) and plant growth and development during the

**Table 2.** Effect of foliar application of zinc based nanofertilizer and varying fertility levels on yield of maize

Treatment	Yields (q/ha)		
	Grain	Stover	Biological
<i>Foliar application</i>			
Control	45.1	68.8	113.9
At knee high stage	47.8	75.4	123.2
At 50% tasseling stage	46.6	71.9	118.6
Both stages	51.9	82.3	134.2
SEm±	0.62	0.93	1.04
C.D. (P = 0.05%)	1.74	2.63	2.94
<i>Fertility levels (N, P and Zn kg/ha)</i>			
Control	34.1	53.2	87.3
80% RDF	48.8	75.9	124.8
90% RDF	53.7	84.1	137.8
100% RDF	54.8	85.2	140.0
SEm±	0.62	0.93	1.04
C.D. (P = 0.05%)	1.74	2.63	2.94

early crop growth stages (Bihmidine *et al.*, 2013). Zn, among micronutrients, is indispensable for plants as it acts as a structural, catalytic and co-catalytic component in many enzymes (Singh *et al.*, 2015; Choudhary *et al.*, 2022a). Overall, it is assumed that application of Zn based chitosan nanofertilizer enhanced cellular homeostasis and positively contribute source-activity and contributes to sink-strength *viz.*, yield attributes and yield (Lemoine *et al.*, 2013; Choudhary *et al.*, 2022b). The study thus affirms that application of nanofertilizer both at knee high stage followed by 50 per cent tasseling stage significantly increases the activity of major enzymes of starch biosynthetic pathways and is accountable for higher starch accumulation in grains.

## Conclusion

The results of the current study suggest that the most effective approach for enhancing yield attributes and overall maize yield is the foliar application of a 0.1% nanofertilizer containing zinc at both the knee-high stage and the 50 per cent tasseling stage. In the coming years, nanofertilizers hold great promise for revolutionizing modern agriculture through enhanced nutrient delivery and eco-friendly practices.

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## Declaration

The authors do not have any conflict of interest.

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# Performance evaluation of quality protein hybrid maize under different planting and weed control methods

Bhumitra Awasthi · V. S. Hooda · Meenakshi Sangwan · S. Singh · A. Yadav

**Abstract:** A field experiment was conducted at Agronomy research farm, CCS Haryana Agricultural University, Hisar, Haryana during *kharif* 2012. Three main plot treatments and six sub plot treatments were tested using a split plot design with four replications. In main plots, three planting methods namely Conventional Tillage (CT), Zero Tillage (ZT) and Furrow Irrigated Raised Bed System (FIRBS) were evaluated. In the sub-plots, the evaluation encompassed three pre-emergence herbicides atrazine @ 750 g/ha, atrazine @ 500 g/ha + hand weeding at 35 DAS and pendimethalin @ 1000 g/ha. Additionally post-emergence herbicides, tembotrione (Laudis 42% SC) @ 120 g /ha + S 1000 ml/ha (10-15 DAS/2-4 leaf stage) were compared with weed free and weedy check conditions. Important weed species observed in the experimental plots were *Cyperus rotundus*, *Echinochloa colona*, *Trianthema portulacastrum* and *Digera arvensis*. Application of atrazine (750 g/ha) recorded significantly reduced weed population and weed dry weight. This treatment also exhibited higher weed control efficiency and resulted in improved crop growth and higher crop yields. Whereas, in planting methods, zero tillage recorded lowest weed population, dry weight of weeds and higher weed control efficiency with higher crop growth and crop yield. Pre-emergence application of atrazine (750 g/ha) under zero tillage was found effective for controlling weeds with higher crop yield in *kharif* maize.

**Keywords:** Atrazine · Hybrid maize · Planting methods · Tembotrione · Weed control efficiency

✉ Bhumitra Awasthi: bhumiputra89@gmail.com

Department of Agronomy, CCSHAU, Hisar, Haryana, India

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## Introduction

Cereals play a vital role as a primary source of nutrition for a significant portion of the global population, particularly in regions such as Southeast Asia and Sub-Saharan Africa. Among these cereals, wheat, rice, and maize stand out as the predominant staples, serving as essential dietary components for millions of people in these areas. Maize is known as the Queen of cereals due to its high yield potential and wider utilization for diverse purposes (Sethi *et al.*, 2021). The global consumption pattern of maize is: feed-61 per cent, food-17 per cent, and industry-22 per cent. Currently, about 1151.7 million MT of maize is being produced together by over 170 countries from an area of 200.96 m ha with average productivity of 5.73 t/ha (Anonymous, 2023). India produced 36 million MT of maize in an area of 10.1 m ha during 2022-23 with average productivity of 3.56 t/ha (Anonymous, 2023). The primary maize-producing states in India, including Karnataka, Madhya Pradesh, Maharashtra, Tamil Nadu, West Bengal, Rajasthan, Bihar, Andhra Pradesh, Uttar Pradesh, Telangana, Gujarat, Punjab, Haryana, and Odisha, collectively contribute to more than 95 per cent of the country's total maize production. Haryana state has an ample scope to further increase acreage and productivity of maize as it can be a strong candidate in the drive of crop diversification to displace puddled transplanted rice.

The maize cultivation occurred throughout the year in all states of the country for various purposes including grain, fodder, green cobs, sweet corn, baby corn and pop corn in peri-urban areas. Maize is renowned for its broad adaptability and its versatility in being used for food, animal feed, and various industrial applications (Murdia *et al.*, 2016). Maize is an important source of



vitamins and minerals like Ca, P, S and small amounts of Na. Its flour is considered to be a good diet for heart patients due to its low gluten (protein) content (Rasool and Khan, 2016). The productivity of maize in India is relatively very low compared to developed countries of world and the yield can be increased with many agronomic practices. Among the various agronomic practices that can impact maize productivity, weed management is widely recognized as a crucial factor in achieving higher maize yields (Gharde *et al.*, 2018).

Weeds are serious problem in *kharif* maize since its sowing time coincides with the commencement of monsoon rains and congenial growing conditions i.e., high temperature and humidity. Addition of farm yard manure (FYM) during *kharif* season also encourages weed growth in this crop. Initial slow growth and wider row spacing coupled with congenial environmental conditions allows luxuriant weed growth, with yield reduction from 28-100 per cent (Dass *et al.*, 2012). Weed infestation is supreme importance among the biotic factors that are responsible for low maize grain yield. Weed control practices in maize resulted in 77 to 96.7 per cent higher grain yield than the weedy check (Rani *et al.*, 2020). Weeds can exert a detrimental influence on maize growth and yield by competing for essential resources like nutrients, light, and water. In this context, it becomes imperative to implement effective weed management strategies to mitigate these adverse effects and optimize maize production. Achieving this goal typically involves a comprehensive approach that combines mechanical, chemical, and cultural methods of weed control (Alptekin *et al.*, 2023). Manual weeding is most popular among the farmers but it is expensive, laborious and time-consuming. Herbicides usage for weed control is an important alternative to manual weeding because they are cheaper, faster and gives better weed control (Rani *et al.*, 2020).

Maize crop management involves decision making on several cultural practices aimed to maximize grain yields like planting methods, improved varieties and proper nutrient management. Interventions in the form of new resource conservation technologies (RCTs) like zero-tillage and furrow irrigated raised bed system (FIRBS) coupled with crop diversification by including maize in place of rice may be a viable solution (Hossain *et al.*, 2020). The mean grain yield of hybrid genotypes grown on FIRBS yielded 44 per cent more than conventional practice and

12 per cent with no-till practices (Jat *et al.*, 2007). Higher maize yield on bed planting as compared to conventional tillage was also reported by Basavanneppa *et al.* (2017). The total water use in maize under FIRBS and no-tillage were remarkably reduced by 84.3 and 62.8 per cent, respectively compared to conventional tillage practice. Weeds are accountable for causing a substantial global loss of approximately 37 per cent in total maize production. In the contemporary agricultural landscape, chemical methods have gained prominence and are now considered the predominant approach for weed management in maize cultivation (Sharma and Rayamajhi, 2022). Efficacy of different herbicides may also vary with different planting methods in any crop including maize. Most of the pre-emergence (PRE) herbicides (most commonly and widely used herbicides atrazine or pendimethalin) provide only narrow spectrum weed control in maize (Tetarwal *et al.*, 2022), and therefore, an attempt was done to find out an effective and economical method for weed control in maize using post emergence (POST) herbicides.

## Materials and methods

A field experiment was conducted at Agronomy research farm, CCS Haryana Agricultural University, Hisar, Haryana during *kharif* 2012. The experimental site is located at 29°16'N latitude and 75°7'E longitude at the mean sea elevation of 215.2 m in north-west part of India. The climate of the area is semiarid type, with very hot summers (40-46 °C) and relatively cool winters (1.5-4 °C) and main characteristics of climate in Hisar are dryness, extremes of temperature, and scanty rainfall (around 450 mm).

Figure 1 show that temperature ranged between 16.2 to 37.8 °C, average relative humidity fluctuated between 53.5 to 88.5 per cent, bright sunshine hours ranged from 1.4 to 10.1 hrs/day and crop received 393.6 mm of rainfall during the entire crop growing season.

The texture of the surface soil of the experimental field was sandy loam, that was low in organic carbon and nitrogen (230 kg/ha), medium in available phosphorus (19 kg/ha), high in potassium (310 kg/ha) and slightly alkaline (pH 7.9) in reaction. Hybrid maize HQPM-1 was planted on July 19, 2012, using the dibbling method. The spacing between rows was set at 70 cm, and the distance between individual plants within the row was 22.5 cm. The seeds were sown at a depth of 5-6 cm. Harvesting of the maize crop was completed on October 23, 2012.

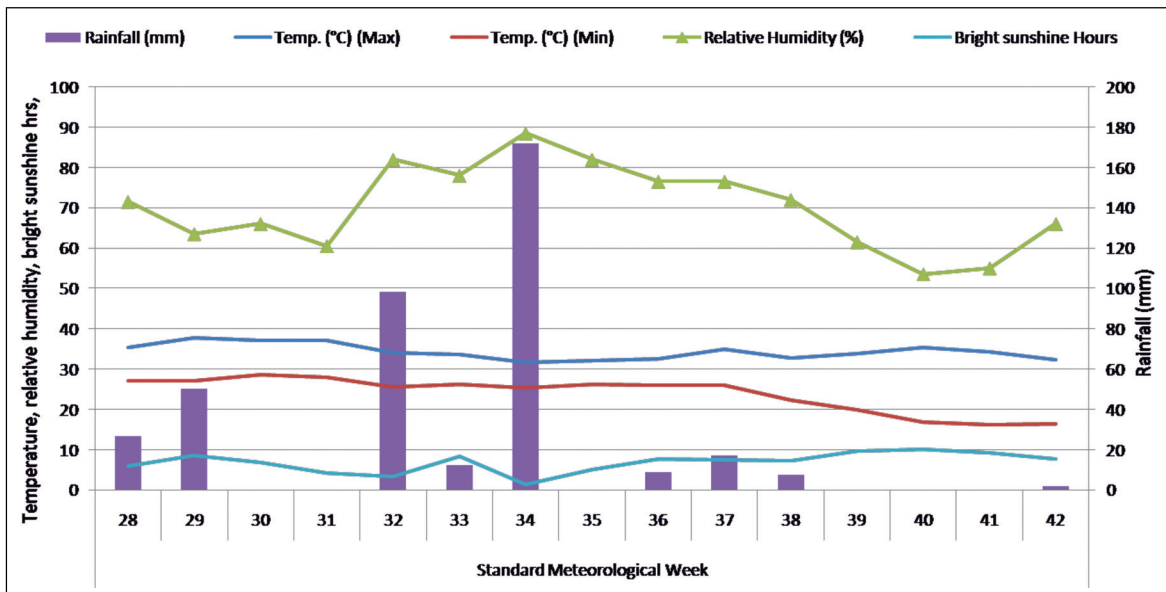


Figure 1. Weekly meteorological observatory data during experimental year

Three main plot treatments [planting methods including CT, ZT and FIRBS] and six sub plot treatments [three pre emergence herbicides *viz.*, atrazine 750 g/ha, atrazine 500 g/ha + hand weeding at 35 DAS and pendimethalin 1000 g/ha and one post-emergence herbicides *viz.*, tembotrione (Laudis 42% SC) 120 g/ha + S 1000 ml/ha (10-15 DAS/2-4 leaf stage) along with weed free and weedy check] were tested using a split plot design with four replications. Herbicide atrazine and pendimethalin were applied as pre-emergence (PRE) spray to the soil surface as per treatment on the day of sowing and tembotrione was applied as post emergence (POST) spray to the foliage as per treatment at 15 DAS. Herbicides were applied through knapsack sprayer fitted with flat fan nozzles delivering 700 L water/ha (for PRE and 325 L water/ha for POST application) and sufficient moisture was maintained in the soil at the time of application.

Observations for total weed density were recorded by randomly placing quadrat (0.25 m<sup>2</sup>) in each plot at maturity. Important weed species observed in the experimental plots were *Cyperus rotundus*, *Echinochloa colona*, *Trianthema portulacastrum* and *Digera arvensis*. For calculating dry matter accumulation (g/m<sup>2</sup>) of weeds, the weeds taken with a quadrat were dried in oven at 65 ± 5 °C. The dried samples were weighed and expressed as g/m<sup>2</sup>. Weed control efficiency was calculated with the help of formula:

$$\text{WCE (\%)} = \frac{W_2 - W_1}{W_2} \times 100$$

Where,  $W_2$  = Dry weight of weeds in weedy check plot  
 $W_1$  = Dry weight of weeds in treatment plot

The plant height was measured from the base of the plant to collar of the flag leaf and expressed in cm. Three plants were randomly selected from each plot and carefully uprooted to take dry matter accumulation. These samples were dried at 70 °C in oven to achieve a constant weight. The number of cobs per meter row length was recorded in each plot at physiological maturity. Grain number per cob was calculated from the grain weight per cob and the corresponding 100 grain weight as follows:

$$\text{Grain number per cob} = \frac{\text{Grain weight per cob (g)}}{\text{Weight of 100 grains (g)}} \times 100$$

After harvesting the net area of individual plot, the bundles were sun dried for 4-5 days and weight was taken from each plot before threshing. After sun drying, the biomass, representing the biological yield, was threshed and subsequently weighed to determine the grain yield (kg/ha). The grain yield so obtained was deducted from the biomass of the harvested crop to compute the stover yield. Harvest index, which is the ratio of economic yield to biological yield (expressed in per cent), was worked out by the following formula:

$$\text{Harvest index (\%)} = \frac{\text{Grain yield (kg/ha)}}{\text{Biological yield (kg/ha)}} \times 100$$

The experimental data were statistically analyzed by the methods of analysis of variance (ANOVA) as described by Panse and Sukhatme (1978).

## Results and discussion

### Effect on weed density

There was significant variation in weeds density in maize due to different planting and weed control methods. ZT recorded the lowest number of total weeds density compared to FIRBS and CT (Table 1). This might be due to lower weed population due to crop residue of previous crop which acted as mulch resulting into lower crop-weed competition in ZT than other treatments. These results are in line with Khedwal *et al.* (2017) where they observed lower grassy weeds in zero tillage as compared to raised bed planting method. Dahal and Karki (2014) have also reported that zero tillage had significantly lowered density and dry weight of grassy weeds and broad leaf weeds as compared to conventional tillage in maize crop. Ghosh *et al.* (2023) also reported that zero tillage reduced weed densities compared with conventional tillage in maize-wheat-mungbean system. In the present study, significantly lower density of weeds was also observed in raised seed bed as compared to conventional tillage.

Gaurav *et al.* (2018) also reported lesser weeds in raised beds as compared to the conventional system.

Maximum weed density was observed in weedy check while in weed free plots, which received hand weeding at regular intervals, indicated that complete weed control was possible only by local control methods. These results align with the research conducted by Alptekin *et al.* (2023). The lowest weed density was observed in plots treated with atrazine at a rate of 750 g/ha compared to other herbicide treatments. Conversely, among the various herbicidal treatments, the highest weed density was recorded in plots treated with atrazine at a rate of 500 g/ha along with hand weeding at 35 days after sowing (1HW 35 DAS). This weed density was comparable to the weed densities observed in plots treated with tembotrione at 120 g/ha along with S at 1000 ml/ha and pendimethalin at 1000 g/ha. Importantly, these herbicide treatments exhibited a significant difference in weed density compared to the weedy check plots. These results are supported by Kaur *et al.* (2020) who observed that significantly less density of weeds, including *C. rotundus*, was recorded under atrazine at dose of 0.8 and 1.0 kg/ha with or without residue in Punjab. Barla *et al.* (2016) also reported lower weed density (including sedges) with application of atrazine 1.0 kg/ha as pre-emergence in maize crop. Similarly, Kakade *et al.* (2016) and Iqbal *et*

**Table 1.** Effect of different planting methods and weed control treatments on density of weeds in maize

Treatments	Density of weeds (No./m <sup>2</sup> ) at maturity			
	<i>C. rotundus</i>	<i>E. colona</i>	<i>T. portulacastrum</i>	<i>D. arvensis</i>
<i>Planting methods</i>				
Conventional tillage (CT)	4.08 (15.62)	3.41 (10.66)	3.15 (8.91)	2.37 (4.62)
Zero tillage (ZT)	3.79 (13.37)	3.15 (8.95)	2.87 (7.25)	1.85 (2.41)
Furrow irrigated raised bed system (FIRBS)	4.06 (15.50)	3.29 (9.83)	3.07 (8.45)	2.25 (4.08)
SEM±	0.10	0.08	0.07	0.07
CD (p=0.05)	0.26	0.25	0.18	0.19
<i>Weed control treatment</i>				
Atrazine 750 g/ha (PRE)	3.87 (14.00)	3.11 (8.66)	2.71 (6.33)	1.89 (2.58)
Atrazine 500 g/ha (PRE)+ 1 HW 35 DAS	4.10 (15.83)	3.38 (10.42)	3.11 (8.66)	2.10 (3.41)
Pendimethalin 1000 g/ha (PRE)	4.07 (15.58)	3.41 (10.66)	3.17 (9.08)	2.14 (3.58)
Tembotrione 120 g/ha + S 1000 ml/ha (POST)	4.07 (15.58)	3.44 (10.83)	3.17 (9.08)	2.61 (5.83)
Weed free	1.00 (0.00)	1.00 (0.00)	1.00 (0.00)	1.00 (0.00)
Weedy check	5.39 (28.00)	4.40 (18.33)	4.13 (16.08)	2.80 (6.83)
SEM±	0.07	0.08	0.10	0.07
CD (p=0.05)	0.19	0.24	0.29	0.20

Original data given in parenthesis were subjected to square root ( $\sqrt{x+1}$ ) transformation before analysis

al. (2020) reported decreased weed concentration and dry weight of the weed with pre-emergence application of atrazine @ 1.0 to 1.2 kg/ha.

#### Effect on weed dry weight and WCE

Weed dry weight also differed significantly due to various planting and weed control methods and significantly lower dry weight of weeds along with higher WCE was recorded under the ZT compared to CT and FIRBS (Table 2). This might be due to the fact that ZT recorded the lowest number of total weeds density compared to FIRBS and CT. This aligns with the findings of Gaurav *et al.* (2018), where researchers observed a tillage operation resulted in significant reduction in dry weight of weeds at all the growth stages in maize. Dahal and Karki (2014) have also reported that zero tillage had significantly lowered dry weight of grassy weeds and broad leaf weeds as compared to conventional tillage in maize crop.

Among weed control treatments, significantly higher weed dry weight was recorded under weedy check compared to other methods. In the present study, this was mainly due to higher and uninterrupted growth of weeds which made best use of the growth resources. Among weed control methods, atrazine @ 750 g/ha attained lowest weed dry weight as compared to other treatments. Weed free treatment attained highest WCE,

while, lowest was in weedy check. Maximum WCE was recorded under atrazine @ 750 g/ha compared to rest of the treatments. The higher WCE with these treatments could be attributed to the lower weed population and total weed dry weight. These results are also in consensus with Barla *et al.* (2016) who reported lower weed dry weight with application of atrazine 1.0 kg/ha as pre-emergence in maize crop. Another report by Kaur *et al.* (2020) also observed significantly less dry weight of weeds with application of atrazine 0.8 to 1.0 kg/ha as pre-emergence in maize crop. Kakade *et al.* (2016) also reported that, among all the herbicidal treatments, application of 50 per cent WP atrazine at 1.0 kg/ha as pre-emergence demonstrated the highest weed control efficiency with a range of values extending from 75.52 per cent to 83.10 per cent except for *Cyperus* spp., which showed only 2.84 per cent efficiency. Iqbal *et al.* (2020) reported that the maximum weed control efficiency, along with remarkably decreased weed concentration and dry weight of the weed, was registered in the pre-emergence application of atrazine at 1.2 kg/ha.

#### Effect on crop growth and yield attributes

Among planting methods, maximum plant height was attained in ZT which was at par with FIRBS but significantly higher than CT at maturity (Table 3). The

**Table 2.** Effect of different planting methods and weed control treatments on dry weight of weeds and WCE in maize

Treatments	Dry weight of weeds (g/m <sup>2</sup> ) at maturity				WCE (%) at maturity
	<i>C. rotundus</i>	<i>E. colona</i>	<i>T. portulacastrum</i>	<i>D. arvensis</i>	
<i>Planting methods</i>					
Conventional tillage (CT)	4.42 (18.55)	3.25 (9.56)	2.79 (6.79)	2.07 (3.29)	42.66
Zero tillage (ZT)	4.20 (16.66)	3.01 (8.06)	2.50 (5.26)	1.69 (1.87)	52.18
Furrow irrigated raised bed system (FIRBS)	4.39 (18.31)	3.13 (8.82)	2.69 (6.22)	1.99 (2.98)	45.45
SEM <sub>±</sub>	0.06	0.05	0.05	0.04	–
CD (p=0.05)	0.16	0.16	0.14	0.12	–
<i>Weed control treatment</i>					
Atrazine 750 g/ha (PRE)	4.26 (17.17)	2.97 (7.81)	2.41 (4.80)	1.73 (2.01)	52.27
Atrazine 500 g/ha (PRE)+ 1 HW 35 DAS	4.47 (18.94)	3.22 (9.35)	2.74 (6.49)	1.87 (2.50)	44.02
Pendimethalin 1000 g/ha (PRE)	4.48 (19.05)	3.24 (9.52)	2.76 (6.64)	1.89 (2.56)	43.29
Tembotrione 120 g/ha + S 1000 ml/ha (POST)	4.44 (18.70)	3.28 (9.74)	2.75 (6.58)	2.30 (4.30)	40.96
Weed free	1.00 (0.00)	1.00 (0.00)	1.00 (0.00)	1.00 (0.00)	100.00
Weedy check	5.85 (33.19)	4.18 (16.47)	3.61 (12.04)	2.43 (4.90)	0.00
SEM <sub>±</sub>	0.07	0.08	0.08	0.07	–
CD (p=0.05)	0.17	0.22	0.24	0.19	–

Original data given in parenthesis were subjected to square root ( $\sqrt{x+1}$ ) transformation before analysis

plant dry matter accumulation in ZT was significantly higher than CT and FIRBS which were at par with each other. Whereas, minimum values of these parameters were in CT which was followed by FIRBS. Number of cobs per meter row length and number of grains per cob were not affected by different planting. Results are also supported by the findings of Khedwal *et al.* (2017).

Among various weed control treatments, weed free treatment had maximum plant height, plant dry matter accumulation and number of grains per cob which remained significantly higher than all herbicidal treatments (which remained at par with each other) while significantly lower values of these parameters were recorded in weedy check as compared to other treatments. Different weed control treatments failed to affect number of cobs per meter row length. These results are also supported by the finding of Barla *et al.* (2016). Kaur *et al.* (2020) and Iqbal *et al.* (2020) also recorded the higher values for all the growth and yield parameters with the weed free treatments, followed by application of atrazine as pre-emergence as compared to the untreated control.

#### *Effect on crop yield and harvest index*

Crop yield (grain yield, stover yield and biological yield) and harvest index of maize were significantly affected by

different planting and weed control methods (Table 4). Zero tillage provided maximum harvest index and crop yield which was higher than FIRBS and CT but it was statistically at par with both. Lowest grain yield was obtained by CT compared to ZT and FIRBS. This might be due to better plant stand and more number of grain/cob, and lesser weed infestation under ZT. Similarly, Khedwal *et al.* (2017) also noticed that tillage operation resulted in significant effect on yield of maize.

Among various weed control treatments, significantly higher crop yield was observed in weed free (presented in Table 4). It was mainly due to minimum crop-weed competition through out the crop growth period, thus enabling the crop for maximum utilization of nutrients, moisture, light and space which had influence on growth components and yield attributes. Among the herbicidal treatments, maximum crop yield was recorded in atrazine @ 750 g/ha. Kakade *et al.* (2016) also recorded the higher maize yield with application of 50 per cent WP atrazine at 1.0 kg/ha as pre-emergence as compared to the untreated control. In current study, lowest yield was recorded in weedy check as a consequence of greater removal of nutrients and moisture by weed and severe crop weed competition resulting in poor sources and sink development with poor yield attributes. These results are conformity with finding of Barla *et al.* (2016) and Kaur *et al.* (2020). Iqbal *et al.* (2020) also reported that

**Table 3.** Effect of different planting methods and weed control treatments on growth and yield attributes of maize

Treatments	Plant height (cm) at maturity	Plant dry matter accumulation (g/m <sup>2</sup> ) at maturity	No. of cobs per meter row length	No. of grains per cob
<i>Planting methods</i>				
Conventional tillage (CT)	182.42	305.59	5.75	471.94
Zero tillage (ZT)	187.17	316.82	5.95	477.72
Furrow irrigated raised bed system (FIRBS)	185.25	306.54	5.87	472.76
SEM <sub>±</sub>	1.00	1.26	0.12	3.47
CD (p=0.05)	3.55	4.46	NS	NS
<i>Weed control treatment</i>				
Atrazine 750 g/ha (PRE)	186.22	313.56	6.00	478.66
Atrazine 500 g/ha (PRE)+ 1 HW 35 DAS	186.85	312.45	5.83	477.57
Pendimethalin 1000 g/ha (PRE)	186.78	312.20	5.75	477.59
Tembotrione 120 g/ha + S 1000 ml/ha (POST)	186.63	312.06	5.83	477.77
Weed free	190.81	323.99	6.16	518.78
Weedy check	172.40	283.64	5.58	414.47
SEM <sub>±</sub>	0.56	1.95	0.20	7.26
CD (p=0.05)	1.61	5.57	NS	20.77

**Table 4.** Effect of different planting and weed control treatments on yields and harvest index of maize

Treatments	Grain yield (q/ha)	Stover yield (q/ha)	Biological yield (q/ha)	Harvest index (%)
<i>Planting methods</i>				
Conventional tillage (CT)	48.50	64.53	113.03	42.91
Zero tillage (ZT)	52.43	68.76	121.19	43.26
Furrow irrigated raised bed system (FIRBS)	49.60	65.70	115.3	43.02
SEm±	0.87	0.71	1.42	-
CD (p=0.05)	2.42	2.02	4.38	-
<i>Weed control treatment</i>				
Atrazine 750 g/ha (PRE)	51.86	66.73	118.59	43.73
Atrazine 500 g/ha (PRE)+ 1 HW 35 DAS	49.76	66.00	115.76	42.99
Pendimethalin 1000 g/ha (PRE)	51.46	65.63	117.09	43.95
Tembotrione 120 g/ha + S 1000 ml/ha (POST)	49.40	65.96	115.36	42.82
Weed free	54.30	70.46	124.76	43.52
Weedy check	44.30	63.90	108.2	40.94
SEm±	0.46	0.20	0.33	-
CD (p=0.05)	1.46	1.89	2.84	-

the maximum maize yield was registered in the pre-emergence application of atrazine at 1.2 kg/ha.

## Conclusion

Planting of maize in zero tillage condition resulted in significantly higher crop yield with lower weed density and their dry weight as compared to conventional tillage and FIRBS methods of planting. Pre-emergence application of atrazine (750 g/ha) under zero tillage was found effective for controlling weeds with higher weed control efficiency and getting higher crop yield in *khariif* maize. This study concluded that zero tillage for maize cultivation, holds great promise for enhancing agricultural productivity and sustainability.

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# Effect of sources and levels of zinc on growth rate, grain yield and economics in shalimar QPMH-1 hybrid

Hadiya Kounsar<sup>1</sup> · Raihana Habib Kanth<sup>2</sup> · Tauseef A. Bhat<sup>1</sup> · Bilkees Jamsheed<sup>1</sup> · Suhail Ahmad Bhat<sup>3</sup> · Zahoor Ahmad Dar<sup>4</sup> · Amal Saxena<sup>1</sup> · Rasool Fasil<sup>4</sup> · Mohd Rafiq<sup>1</sup>

**Abstract:** Zinc deficiency can result in significant losses in the yield of maize. Therefore, an experimental field trial was conducted at the Agronomy Research Farm, located within the Faculty of Agriculture, Wadura, Sopore SKUAST-Kashmir during *kharif* season 2021 to study the impact of various sources and levels of zinc on growth and yield performance of Quality Protein maize. The experiment consisted of two sources of Zinc *viz.* Zinc Oxide, Zinc Sulphate and five levels of Zinc *viz.* control, 0.25%, 0.50%, 0.75% and 1.0%, the setup followed a factorial complete block design, consisting of 3 replications. Zinc was applied as foliar spray @ 50 DAS. The findings of the experiment revealed significant variations in terms of growth, yield and economics for various treatments under investigation. Among the various treatments, application of zinc oxide at a concentration of 1%, exhibited superior performance in terms of plant growth attributes, yield components, and overall yield. It recorded the highest grain yield amounting to 53.92 q/ha. Additionally, the application of zinc oxide @ 1%

resulted in the highest net returns and a favourable benefit-to-cost (B:C) ratio of 1.75 followed by application of zinc oxide @ 0.75%) and zinc sulphate @ 1%. Among all the treatments, lowest plant growth, yield and economic suitability was recorded with control (ZnSO<sub>4</sub>). Hence, application of zinc oxide @ 1% was found economically suitable and sustainable followed by application of zinc oxide @ 0.75% and zinc sulphate @ 1%.

**Keywords:** Foliar application · Growth · QPM · ZnO · ZnSO<sub>4</sub> · Yield

## Introduction

In India and the rest of the globe, maize (*Zea mays* L.) holds position as the third most important cereal crop, following wheat and rice. Maize is called a “miracle crop” since it has greater productivity potential than any other cereal crop and can be grown in both tropical and temperate climates. It is an essential staple diet for those residing in Asia, Africa and Latin America as well as the most significant fodder cereal for cattle.

Lysine and tryptophan, the two crucial amino acids are deficient in maize grain thereby poses the risk of nutritional deficiencies that can cause neurological disorders, kwashiorkor disease, appetite loss, defective skeletal development, growth retardation and abnormal behaviour. Researchers detected several mutants in maize, specifically Opaque-2, which are responsible for increased lysine and tryptophan concentration. The cultivars that showed improved agronomic performance and protein quality were termed as Quality Protein Maize (QPM) (Vivek *et al.*, 2008).

✉ Raihana Habib Kanth: raihana\_k@rediffmail.com;

✉ Tauseef A. Bhat: tauseekk@gmail.com

<sup>1</sup>Faculty of Agriculture, <sup>2</sup>Dean, Sher-e-Kashmir University of Agricultural Sciences and Technology of Kashmir-193201, J&K, India

<sup>3</sup>School of Agriculture, Lovely Professional University, Punjab-144411, India

<sup>4</sup>Dryland Agriculture Research Station, Sher-e-Kashmir University of Agricultural Sciences and technology of Kashmir-190025, J&K, India



All across East Asia, pervasive zinc (Zn) deficiency has been identified, with India and Pakistan accounting for between 50 and 70% of cases. Cereal grains are found to be severely deficient in Zinc, particularly in areas where soils contain insufficient plant-available Zn (Shivay and Prasad, 2012). The majority of micronutrient deficiency issues are made worse by the use of high yielding crop varieties, which quickly exhaust the scarce soil nutrients (Martens and Lindsay, 1990). As a result, correcting Zn shortage in farming systems based on cereal is made more difficult.

As Zn is involved in auxin metabolism, enhanced assimilate partitioning towards reproductive portions, grain production depends on it in a critical way. Zn fertilization therefore enhances plant development and growth, yield characteristics, and yield. Zn contributes to protein biosynthesis, which is a key component in the growth of the maize crop since Zn is required for the structural and functional integrity of around 2,800 proteins (Broadley *et al.*, 2007). Better uptake and utilization of Zn can be achieved through proper method of nutrient application which includes the foliar spray of micronutrients that can efficiently enhance the crop productivity. To optimize the potential crop yield, one effective method is the application of micronutrients through foliar spraying. This approach offers a straightforward and rapid means to enhance the nutritional status of maize plants. Zn is delivered directly to the foliage in foliar sprays, which accounts for its efficacy. Numerous deciding factors, including as nutrient penetration, absorption, and translocation, influence plant response to the foliar application.

The application of Zn as foliar spray on quality protein maize may help to cater with nutritional deficiencies present in the crop. With all of these aspects in mind, it was thought worthwhile to conduct research on our hybrid to assess the performance under different zinc sources and levels.

## Material and methods

The field investigation entitled “Performance of quality protein maize under different sources and levels of zinc” was conducted during the *khariif* season of 2021 at the Experimental Farm of the Division of Agronomy, Sopore, Sher-e-Kashmir University of Agricultural Sciences & Technology of Kashmir.

The experimental site, geographically, is situated at coordinates 34°21' N and 74°23' E, with an elevation of 1590 meters above mean sea level. The soil of the research trial exhibited a silty clay loam texture analysed by hydrometer method (Byoucouc *et al.*, 1962). It had moderate levels of organic carbon (0.76%), estimated with Walkley and Black's rapid titration method (Walkley and Black, 1934), sufficient amounts of available nitrogen, phosphorus, and potassium (308, 16.75, 177.65 kg/ha, respectively) estimated with Alkaline permanganate method (Subbiah and Asija, 1956), Olsen's method  $\text{NaHCO}_3$  (Olsen *et al.*, 1954) and Ammonium acetate extract method (Jackson, 1973), respectively. However, it displayed deficient levels of available Zn (0.68 mg/kg) which was evaluated with DTPA (diethylenetriaminepentaacetic acid) extractable method (Lindsay and Norvell, 1978). The pH of the soil was neutral (6.74) with normal EC (0.37dS/m) determined with the help of Digital glass electrode pH meter (Jackson, 1973) and Solou-bridge conductivity meter (Jackson, 1973).

The data indicated that the weekly minimum temperature ranged from 7.3°C to 19.4°C, while the maximum temperature fluctuated between 24.4°C and 32.6°C. Throughout the entire cropping period in 2021, the total precipitation accumulated to 190.7 mm (Figure 1).

The experiment was set up with two factors with ten treatments ( $T_1$ : ZnO @ 0.25%,  $T_2$ : ZnO @ 0.50%,  $T_3$ : ZnO @ 0.75%,  $T_4$ : ZnO @ 1.0%,  $T_5$ : ZnO Control,  $T_6$ :  $\text{ZnSO}_4$  @ 0.25%,  $T_7$ :  $\text{ZnSO}_4$  @ 0.50%,  $T_8$ :  $\text{ZnSO}_4$  @ 0.75%,  $T_9$ :  $\text{ZnSO}_4$  @ 1.0%,  $T_{10}$ :  $\text{ZnSO}_4$  Control). The experiment was performed using a randomized complete block design and replicated three times. A regional hybrid called Shalimar QPMH-1 developed by SKUAST-Kashmir was used in the study. Different observation recorded with respect to growth and yield are as under:

*Plant height:* Plant height was assessed as mean of five randomly chosen and labelled plants, beginning from the base and extending up to the end tip of the flag leaf in net plot area of each treatment at 20, 40, 60, 80, 100 DAS and at harvest.

*Relative growth rate:* The relative growth rate was determined by using the given formula and expressed in mg/g/day (Watson *et al.*, 1952).

$$\text{RGR} = \frac{\ln w_2 - \ln w_1}{T_2 - T_1}$$

Where,  $W_1$  and  $W_2$  = plant dry weight (g) (at time  $T_1$  and  $T_2$ , respectively)

$T_2 - T_1$  = time interval of sampling (in days)

Ln = natural log

*Crop growth rate:* The crop growth rate was determined by using the provided formula (Redford, 1967) and was expressed in  $g/m^2/day$ .

$$CGR = \frac{w_2 - w_1}{T_2 - T_1}$$

Where,  $W_1$  and  $W_2$  are the plant dry weights (g) at times  $T_1$  and  $T_2$ , respectively.

$T_2 - T_1$  = time interval of sampling in days

*Days taken to phenological stages:* The number of days required from the sowing date to attain 50% of knee height, 50% of tasseling, 50% of emergence of silk were examined, counted and recorded from each treatment.

*Cob diameter:* Cob girth of five observational cobs in each plot was measured and the value was divided by 3.14. The results were averaged average and expressed as cob diameter in centimetres.

*Number of kernel rows per cob:* After summing the rows on each cob under observation, the mean value of kernel rows per cob was determined.

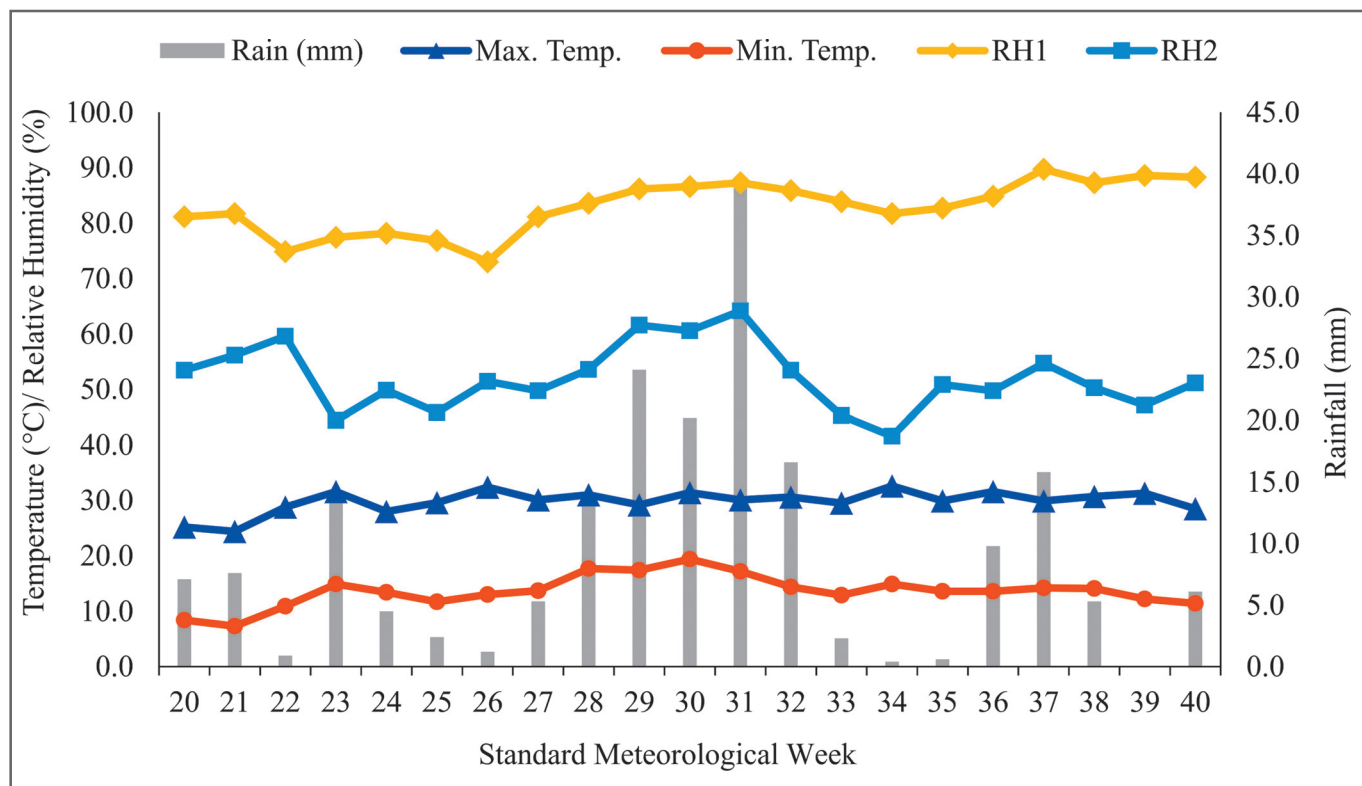
*Grain yield:* The cobs were manually shelled and properly sun dried after being collected from net plots. The cumulative weight was measured in kilograms and then demonstrated as grain yield in q/ha.

*Harvest index:* The ratio of economic yield (grain yield) to the biological yield (grain + stover yield) was used to determine the harvest index, represented as a percentage (Donald and Hamblin, 1976).

$$\text{Harvest index (HI)} = \frac{\text{Economic yield}}{\text{Biological yield}} \times 100$$

*Relative economics:* For each treatment, the cultivation costs and gross profits were estimate during the prices of the current market. The following formula was used to calculate the benefit cost ratio:

$$\text{B:C ratio} = \frac{\text{Net returns}}{\text{Total cost of cultivation}}$$



\*RH1 and Rh2 are morning and evening relative humidity

**Figure 1.** Mean weather condition during crop growing period (kharif 2021)

To assess the impact of zinc sources and levels upon the growth and productivity of QPM (Quality Protein Maize), the experimental data was evaluated through statistical analysis by applying analysis of variance (ANOVA) within a randomized complete block design (RCBD) in a factorial arrangement. The significance of treatments was evaluated adopting the F-test at 0.05 level of significance. The critical difference was estimated to compare the effects of various treatments in situations where statistically significant data were obtained  $p < 0.05$ .

## Results and discussion

### Growth parameters

Both sources and levels of zinc significantly influenced almost all the growth characters except in different phenological phases in case of sources of zinc and control treatment. Zinc oxide measured significantly greater plant height at 60 DAS (158.56 cm), 80 DAS (188.50 cm), 100 DAS (194.05 cm), and at harvest (195.36 cm) respectively, compared to zinc sulphate application, which recorded significantly lower plant height of 153.10 cm, 182.26 cm, 188.22 cm, 190.07 cm at 60 DAS, 80 DAS, 100 DAS and at harvest, respectively. Zinc application observed significantly higher plant height at 60 DAS, 80 DAS, 100 DAS and at harvest (166.21 cm, 197.79 cm, 203.85 cm and 204.44 cm) with the application of 1% solution than all other concentrations except with 0.75%. Lower plant height of 146.41 cm, 176.10 cm, 180.27

cm and 180.87 cm among the zinc levels was reported in control treatment at 60 DAS, 80 DAS, 100 DAS and at harvest, respectively.

Application of zinc oxide recorded significantly higher CGR at 40-60 DAS (26.76 g/m<sup>2</sup>/day), 60-80 DAS (26.91 g/m<sup>2</sup>/day) whereas, zinc sulphate application registered significantly lower CGR at 40-60 DAS (24.21 g/m<sup>2</sup>/day), 60-80 DAS (26.39 g/m<sup>2</sup>/day). Among the levels, significantly maximum value for CGR of 31.61 g/m<sup>2</sup>/day at 40-60 DAS, 5.87 g/m<sup>2</sup>/day at 80-100 DAS was recorded with application of 1% zinc. At 60-80 DAS significantly higher CGR of 28.55 g/m<sup>2</sup>/day was obtained with the application of 0.50% of zinc solution in terms of levels of zinc.

Application of zinc oxide registered significantly higher RGR of 39.26 mg/g/day at 40-60 DAS, 1.30 mg/g/day at 100-120 DAS. At 60-80 DAS and at 80-100 DAS application of zinc sulphate recorded higher value for RGR of 22.72 mg/g/day and 3.58 mg/g/day, respectively. Significantly lower value for RGR was registered with application of zinc sulphate at 40-60 DAS (36.99 mg/g/day) and at 100-120 DAS (mg/g/day). With the application of zinc oxide, significantly lower RGR was observed at 60-80 DAS and 80-100 DAS, with values of 22.02 mg/g/day and 3.19 mg/g/day, respectively. Among the levels of zinc, significantly higher value for RGR of 44.18 mg/g/day at 40-60 DAS was registered with 1% zinc concentration followed by application of 0.25% at 60-80 DAS (23.80 mg/g/day), control at 80-100 DAS (3.65 mg/g/day) and 0.25% at 100-120 DAS (1.34 mg/g/day).

**Table 1.** Effect of different sources and levels of zinc on plant height (cm) at different growth stages of QPM

Treatments	Plant height (cm)					
	20 DAS	40 DAS	60 DAS	80 DAS	100 DAS	120 DAS
<i>Sources of zinc</i>						
ZnO	22.17	63.84	158.56	188.50	194.05	195.36
ZnSO <sub>4</sub>	22.19	61.44	153.10	182.26	188.22	190.07
SEm±	2.74	2.54	1.60	1.11	1.47	1.58
C.D(P≤0.05)	NS	NS	4.81	3.33	4.40	4.74
<i>Levels of zinc</i>						
Control	21.89	59.77	146.41	176.10	180.27	180.87
0.25%	22.12	61.55	151.60	178.76	184.36	187.71
0.50%	22.11	62.33	154.94	184.27	189.83	192.52
0.75%	22.39	63.38	159.99	189.99	197.37	198.05
1.0%	22.39	66.16	166.21	197.79	203.85	204.44
SEm±	4.33	4.02	2.54	1.75	2.32	2.50
C.D(P≤0.05)	NS	NS	7.61	5.26	6.96	7.50

**Table 2.** Effect of different sources and levels of zinc on CGR (g/m<sup>2</sup>/day)

Treatments	CGR (g/m <sup>2</sup> /day)				
	20-40 DAS	40-60 DAS	60-80 DAS	80-100 DAS	100-120 DAS
<i>Sources of zinc</i>					
ZnO	15.28	26.76	26.91	4.91	2.14
ZnSO <sub>4</sub>	15.08	24.21	26.39	5.36	1.99
SEm±	0.11	0.77	0.18	0.20	0.18
C.D(P≤0.05)	NS	2.31	0.49	NS	NS
<i>Levels of zinc</i>					
Control	15.08	18.07	22.49	4.69	1.82
0.25%	15.08	24.01	27.83	5.04	2.12
0.50%	15.08	25.47	28.55	5.04	2.12
0.75%	15.33	28.25	27.64	5.05	2.12
1.0%	15.33	31.61	26.77	5.87	2.12
SEm±	0.14	1.22	0.33	0.25	0.22
C.D(P≤0.05)	NS	3.65	1.04	0.78	NS

**Table 3.** Effect different sources and levels of zinc on RGR (mg/g/day)

Treatments	RGR (mg/g/day)				
	20-40 DAS	40-60 DAS	60-80 DAS	80-100 DAS	100-120 DAS
<i>Sources of zinc</i>					
ZnO	58.59	39.26	22.02	3.19	1.30
ZnSO <sub>4</sub>	58.54	36.99	22.72	3.58	1.26
SEm±	0.66	0.62	0.22	0.07	0.01
C.D(P≤0.05)	NS	1.87	0.61	0.18	0.03
<i>Levels of zinc</i>					
Control	57.22	29.79	22.21	3.65	1.34
0.25%	58.99	37.07	23.80	3.33	1.34
0.50%	58.66	38.59	23.60	3.23	1.29
0.75%	58.96	41.01	21.94	3.16	1.25
1.0%	58.97	44.18	20.31	3.57	1.20
SEm±	1.04	1.02	0.33	0.11	0.02
C.D(P≤0.05)	NS	3.01	0.90	0.29	0.07

The enhancement in the growth characteristics viz., plant height, CGR, RGR recorded in present study could be attributed to activation of auxin metabolism due to zinc leads to vigorous growth of meristematic tissue through cell elongation and cell enlargement at the shoot apex. Similar results have been obtained by Krishnaraj *et al.* (2020) in maize. Enhanced plant height due to zinc oxide spray could be attributed to the reason that zinc oxide provides more zinc to the plant than zinc sulphate does. Torabian *et al.* (2016) reported that foliar zinc oxide spray enhanced the plant height in sunflower. Foliar zinc oxide spray improved the shoot length of plants as reported by

Sadak and Bakry (2020) in flax. Similar outcomes were found by Tariq *et al.* (2021) in onion.

Since zinc is an important co-factor of various enzymes, higher doses of zinc may favorably effect energy and carbohydrate metabolism, stress responses and internodal distance which may contribute to taller plants with application of higher doses of zinc and improved values of tryptophan, a precursor of auxin and indole acetic acid, may lead to increase in growth. The increased plant height could be attributed to involvement of zinc in growth and development due to its catalytic impact in the metabolism of nearly all crops as reported by

Ehsanullah *et al.* (2015). Yasin *et al.* (2017) reported that 1% foliar application of zinc considerably increased plant height in maize.

#### *Duration of phenological stages*

Days taken to attain knee height, 50% tasseling, and 50% silking did not significantly vary with the application of various sources of zinc. However, a significant impact was noted in days taken to attain seed maturity. Zinc sulphate application observed significantly less days to reach seed maturity (112.26) than zinc oxide application, which noted significantly more days (113.86).

Application of 1% solution of zinc took longest number of days to achieve 50% tasseling (68.66), 50% silking (72.81) and seed maturity (115.83) compared to all other levels of zinc application except with 0.75% which was statistically at par with 1%. Significantly lesser number to days were taken to reach 50% tasseling (64.61) and 50% silking (70.18) and seed maturity (108.83) under control.

The administration of various sources of zinc had no significant influence on the number of days needed to reach distinct phenological phases, such as 50% tasseling and 50% silking, with the exception of seed maturity, where significant influence of zinc sources was noticed. These results are in coherence with Noreen and Kamran (2019). This could be due to the fact that foliar zinc treatment improved zinc absorption which could lead to higher uptake of nitrogen causing enhanced vegetative

growth of plant prior to the reproductive stage. These findings obtained are in coherence with the Gul *et al.* (2011). The various phenophases took less number of days to complete under control, which could be attributed to the crop sensitivity to the zinc deficiency, thus reducing the calendar days of different phenological stages.

#### *Yield attributes*

Zinc oxide application recorded significantly greater cob diameter of 6.05 cm than zinc sulphate application which realised significantly lower cob diameter of 5.69 cm. However, number of kernel rows per cob was found non-significant with numerically higher number of rows per cob (12.50) was found with application of zinc oxide than with the application of zinc sulphate (12.13). Among the levels of zinc, 1% concentration recorded significantly more number of kernel rows per cob (12.77) and higher cob diameter (6.55 cm) compared to all other levels of zinc. Significantly lower number of kernel row per cob (11.99) and cob diameter (4.39 cm) was observed in absolute control. The foliar treatment of various sources of zinc using zinc oxide as opposed to zinc sulphate had a significant impact on cob diameter. However, the number of kernel rows per cob were not significantly influenced by the various zinc sources. It is possible that the higher values observed in yield-related parameters could be attributed to the stimulation of photosynthesis-related enzymes, assimilation, and by the transfer of photosynthates from source to sink. Enhanced crop

**Table 4.** Effect of different sources and levels of zinc on number of days taken to different phenological stages of QPM

Treatments	Phenological Stages			
	Knee high	Tasseling	Silking	Seed maturity
<i>Sources of zinc</i>				
ZnO	37.04	67.10	72.08	113.86
ZnSO <sub>4</sub>	35.81	66.79	71.81	112.26
SEm±	0.66	0.13	0.11	0.42
C.D(P≤0.05)	NS	NS	NS	1.27
<i>Levels of zinc</i>				
Control	35.60	64.61	70.18	108.83
0.25%	36.77	66.22	71.90	112.33
0.50%	36.21	67.09	72.09	113.50
0.75%	35.72	68.13	72.74	114.83
1.0%	37.83	68.66	72.81	115.83
SEm±	1.04	0.20	0.18	0.67
C.D(P≤0.05)	NS	0.61	0.54	2.02

metabolic activity, growth, and development as well as improved source activity brought on by zinc treatment to the foliage could also be attributed to the improved yield characteristics. Priya *et al.* (2020) has reported similar findings in maize crop. Among the different zinc levels, treatment with 1% solution observed significantly highest value for cob diameter and kernel rows per cob. Significantly lower values for these parameters were recorded under control treatment. Higher percentage of zinc might have enhanced partitioning of assimilates towards the reproductive part, improving the yield attributes. Anjum *et al.* (2017) and Peddapuli *et al.* (2021) both found comparable findings in maize.

#### Grain yield and harvest index

Zinc oxide application resulted in significantly higher grain (53.92 q/ha) yield than zinc sulphate which reported significantly lower grain (50.75 q/ha) yield. Application of 1% zinc solution produced significantly increased grain (59.10 q/ha) yield in comparison to all other concentrations of zinc. Lower grain (41.9 q/ha) yield was registered under control treatment.

Sources of zinc significantly influenced the harvest index of maize. Application of zinc oxide recorded higher harvest index value (41.87) than with the application of zinc sulphate (41.32). Among the zinc levels, application of 0.75% zinc solution resulted in significantly higher value of harvest index (42.23) which was noted to be

statistically at par with all the other treatments except control treatment. The harvest index of control treatment recorded significantly lower value (39.54).

The foliar application of various sources and concentrations of zinc had a significant influence on the grain yield of QPM. Notably, the use of zinc oxide as compared to zinc sulphate resulted in significantly increased grain yield produce along with harvest index. The larger proportion of zinc in zinc oxide compared to zinc sulphate might be the cause of the improved yield brought on by the application of zinc oxide. Sangma *et al.* (2017) reported that zinc oxide along with urea improved the yield in maize. The enhanced availability of zinc could be responsible for initiating pyrophosphates activities and enzymatic reactions (Anjum *et al.*, 2017) in maize. Foliar treatment of zinc increases the nitrogen absorption and protein quality which may improve the growth and yield parameters of the maize crop (Potarzycki and Grzebisz, 2009; Peddapuli *et al.*, 2021).

In terms of levels of zinc, application of 1% zinc recorded statistically higher grain yield than other concentrations except harvest index which was recorded highest with 0.75%. Control treatment resulted in significantly lower yield. Ruffo *et al.* (2015) reported that zinc application at higher levels recorded higher yield compared to control treatment in maize crop. The observed improvement in yield might be due to the optimization of zinc dosage in conjunction with a balanced application of NPK (nitrogen, phosphorus, and potassium),

**Table 5.** Effect of different sources and levels of zinc on yield attributes and yield of QPM

Treatments	Yield attributes		Yield	
	Cob diameter (cm)	No. of kernel rows/cob	Grain yield (q/ha)	Harvest index (%)
<i>Sources of zinc</i>				
ZnO	6.05	22.60	53.92	41.87
ZnSO <sub>4</sub>	5.69	21.61	50.75	41.32
SEm±	0.05	0.27	0.32	0.14
C.D(P≤0.05)	0.16	0.81	0.97	0.44
<i>Levels of zinc</i>				
Control	4.39	21.66	41.90	29.54
0.25%	6.02	22.07	50.25	41.77
0.50%	6.13	22.31	53.35	41.95
0.75%	6.28	22.53	57.10	42.23
1.0%	6.55	23.37	59.10	42.10
SEm±	0.07	0.58	0.51	0.34
C.D(P≤0.05)	0.22	NS	1.54	1.02
Interaction	NS	NS	NS	NS

**Table 6.** Effect of different sources and levels of zinc on relative economics of QPM

Treatments	Total cost (000 Rs./ha)	Gross returns (000 Rs./ha)	Net returns (000 Rs./ha)	B:C ratio
<i>ZnO</i>				
Control	60.199	120.149	59.950	1.00
0.25%	60.574	146.941	86.367	1.43
0.50%	60.949	157.549	96.600	1.58
0.75%	61.324	166.161	104.837	1.71
1.0%	61.699	170.023	108.324	1.75
<i>ZnSO<sub>4</sub></i>				
Control	60.199	117.703	57.504	0.96
0.25%	60.304	135.841	75.537	1.25
0.50%	60.409	142.061	81.652	1.35
0.75%	60.514	154.190	93.676	1.55
1.0%	60.619	161.725	101.106	1.67

Input cost: Seed= Rs. 120 kg/ha, Urea= Rs. 7 kg/ha, MOP= Rs. 19 kg/ha, DAP= Rs. 24 kg/ha, ZnO= Rs. 250 kg/ha, ZnSO<sub>4</sub>= Rs. 90 kg/ha, labour= Rs. 400/day Output Cost: Grain= Rs. 25/kg, Stover= Rs. 2 kg/ha

which might improve developmental factors and yield characteristics. Yasin *et al.* (2017) specified that foliar application of zinc @ 1% considerably increased the yield of maize. These results are lined up with those of Amanullah *et al.* (2016); Waseem *et al.* (2011) in maize crop and by Dawar *et al.* (2022) in wheat.

#### Relative economics

The different sources and levels of zinc resulted in a significant impact on various economic parameters. Specifically, when 1% zinc oxide was applied, it led to higher cost of cultivation, gross and net returns along with highest benefit-to-cost (B:C) ratio of 1.75. However, it is important to note that these values were statistically similar to those obtained with 0.75% zinc oxide and 1% zinc sulphate. Compared to other treatments, lowest B:C ratio was observed with zinc sulphate control. Higher biomass accumulation, better utilization of zinc along with RDF and effective translocation to the reproductive portions due to a sufficient supply of zinc fertilizers might be the cause of the increased crop yield, which results in the highest possible gross and net returns. Sangma *et al.* (2017) reported that zinc oxide along with urea had a synergistic effect on the yield of maize which might lead to increased return of the crop. Kumar and Bohra (2014) also reported that application of zinc as zinc oxide along with RDF registered maximum gross returns, net returns and benefit cost ratio. Similar outcomes were also reported by Rodinpuia *et al.* (2019); Venkateswarlu *et al.* (2022) in maize.

#### Conclusion

Zinc is an important mineral involved in catalytic activity, energy and carbohydrate metabolism. The present study validated the premise that zinc fertilization could be favourably used to enhance yield and its components in maize. It is recommended that zinc oxide @ 1% should be incorporated in the package and practices for maize zinc fertilization for maize to get higher yield and yield components with better profitability in case of QPM hybrid.

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# Influence of planting geometry and nutrient levels on growth dynamics, quality and cob yield of sweet corn in north western India

Amit Bhatnagar<sup>1</sup> · Sailesh Deb Karjee<sup>2</sup> · Gurvinder Singh<sup>1</sup> · Dinesh Kumar Singh<sup>1</sup> · Veer Singh<sup>3</sup>

**Abstract:** A field experiment was conducted at G.B. Pant University of Agriculture and Technology, Pantnagar, Uttarakhand during *kharif* season, 2018 to find out the response of sweet corn to varied planting geometry and nutrient doses. The experiment was laid out in split plot design keeping four planting geometries (60 cm × 25 cm, 60 cm × 30 cm, 75 cm × 25 cm and 75 cm × 30 cm) in main plots and three doses of NPK (120: 60: 40, 150: 75: 50 and 180: 90: 60 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha) in sub plots. Dry matter accumulation, LAI and net assimilation rate were significantly higher in the narrowest planting geometry (60 cm × 25 cm). This geometry also recorded significantly higher green cob yield (13325 kg/ha) than the other planting geometries. Nutrient level 180: 90: 60 being at par with 150: 75: 50 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha produced significantly taller plants, leaf area per plant, dry matter accumulation per plant, LAI, CGR, RGR and NAR than 120: 60: 40 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha. Green cob yield was maximum (12687 kg/ha) in 180: 90: 60 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha but did not vary due to nutrient levels.

**Keywords:** Growth · NPK dose · Planting geometry · Sweet corn

## Introduction

The demand of sweet corn (*Zea mays* L. *saccharata*) in India has been increased during past years due to its taste and multi use. The major agronomic constraints of sweet corn production in *kharif* season in north western India are erratic plant stand, excess soil moisture, high leaching loss of nitrogen, heavy infestation of weeds, more incidences of disease and pest etc. Among these constraints, optimum population and balanced nutrition are of prime importance. There is need to minimize the competition among the plants to get a healthy cob. Spatial arrangement of plants governs the shape and size of the leaf area per plant, affects interception of radiant energy by the canopy and influences intra and inters plant competition for growth resources. Planting geometry also affects proliferation and growth of roots and their activity. Cob yield of maize is decided by number of cobs per unit area and cob size. Higher plant population may increase the number of cobs per unit area but individual cob size may be reduced because of intense competition among the plants. On the other hand, cob size increases with lower plant population but the number of cob per unit area become low (Bhatt *et al.*, 2012). Thus, planting pattern of sweet corn should be adjusted in such a way that the highest cob yield can be obtained by a compensatory balance between number of cobs per unit area and cob size. Even at same plant population, intra and inter plant competition under different planting geometry affect crop performance. Thus, not only plant population but planting geometry also influence cob yield. An adequate and balanced supply of nutrients is essential for proper growth and development of crop. Rational use of nutrients needs priority in sweet corn production.

✉ Amit Bhatnagar: bhatnagaramit75@gmail.com

<sup>1</sup>Professor Agronomy, <sup>2</sup>Ex-PG Scholar, <sup>3</sup>Professor Soil Science, Department of Agronomy, College of Agriculture, G.B. Pant University of Agriculture & Technology, Pantnagar, Uttarakhand, India

Nitrogen is an integral part of chlorophyll and proteins, which are essential for survival of plants. Yield and quality of green cobs are influenced by nitrogen fertilization to a great extent. Numerous field experiments have shown that nitrogen is one of the important growth-limiting factors (Khan *et al.*, 2018). Therefore, its proper dose is must to realize higher yield potential. Phosphorus is known to influence growth and vigour of plant as well as root growth and improves the growth of crop. Phosphorus nutrition plays a key role in plant metabolism as it provides energy through ATP. It is an essential component of nucleic acids, phosphorylated sugars and lipids, which control all life processes (Grazia *et al.*, 2003). Potassium is important in the synthesis and metabolism of carbohydrates and also contributes in photosynthetic activity. Potassium is essential for many plant processes such as enzyme activation, protein synthesis, photosynthesis, osmoregulation, stomatal movements, solute phloem transport, electrical neutralization, regulation of membrane potential, co-transport of sugars and the maintenance of cation-anion balance in the cytosol as well as in the cell sap (Srikanth *et al.*, 2009). Nutrient demand of a crop may vary with plant population. To exploit the yield potential of a crop applied nutrient amount should fulfill nutritional requirement of crop. Therefore, study was conducted to analyse the growth of sweet corn at different planting geometry under varying nutrient levels.

## Materials and methods

A field experiment was conducted during the *kharij* season, 2018 at the Norman E. Borlaug crop research centre of G.B. Pant University of Agriculture and Technology, Pantnagar, district Udham Singh Nagar, Uttarakhand. Pantnagar lies in the *Tarai* belt and is situated at 29° N latitude, 79.5° E longitude and altitude of 243.83 m above mean sea level in the foot hills range of the Himalaya. The soil of experiment plot was silty clay loam in texture, neutral in reaction (pH 7.2), medium in organic carbon (0.69%), low in available nitrogen (216.3 kg/ha), medium in available phosphorus (21.5 kg P/ha) and available potassium (222.3 kg K/ha). The experiment consisted of four planting geometries (60 cm × 25 cm, 60 cm × 30 cm, 75 cm × 25 cm and 75 cm × 30 cm) in main plots and three doses of NPK (120: 60: 40, 150: 75: 50 and 180: 90: 60 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha) in sub plots

was set in split plot design with three replications. The field was pulverized by three cross harrowings with the tractor mounted disc harrow. Thereafter, the field was leveled with leveler. Gross plot size was 6.0 m × 4.5 m. N, P and K were applied through NPK mixture (12: 32: 16), urea and muriate of potash. The 20 per cent of nitrogen and full dose of phosphorous and potassium were applied as basal. Remaining 80 per cent nitrogen was applied in 3 splits as 20, 30 and 30 per cent at 4 leaf-stage, knee height stage and tasseling, respectively. Nutrient dose was applied as per treatment in respective plots. Sweet corn variety 'Sugar-75' was sown on 29 June 2018 and harvested on 16 September 2018. The furrows were opened at 4-5 cm depth by hand operated furrow opener as per treatments in respective plots. Seeds were sown in flat beds manually and were placed at the distance as per treatment. Atrazine @ 1.0 kg a.i./ha in 500 liters of water was sprayed on next day after sowing in all treatments by using knapsack sprayer. One manual weeding was done at 25 days after sowing to manage the weeds. During the crop period no irrigation was given as the crop water demand was fulfilled by adequate rainfall of 1378.5 mm at regular interval.

Randomly five selected plants were tagged in each net plot and their height was measured at vegetative stage (30 and 45 DAS) from ground surface to the tip of newly emerged leaf. At harvest plant height was measured up to basal node of tassel with the help of meter scale. The values were averaged and expressed in cm. At the time of harvesting, stem girth was measured just below the point of emergence of cob with the help of thread and thread length was reported in cm. The plant assimilatory material accumulation pattern was computed by using growth analysis formula as reported by Radford (1967). All the leaves from five selected plants were removed and categorized in to three groups *viz.*, small, medium and large. The representative three leaves from each category were taken out to measure length and width. Average values of leaf length and width was multiplied to get leaf area of each respective category. Leaf area recorded from each category was multiplied by the total number of leaves of respective category and summed up to get the leaf area of sample. Average leaf area per plant was computed by dividing the number of sampled leaves. The whole value was multiplied by correction factor of 0.75 (Montgomery, 1911). Five plants from sample row were selected and cut just above the

ground level with the help of sickle. These cut plants were allowed to sun drying for 48 hours. After sun drying, these plants were dried in the oven at  $65\pm 5^\circ\text{C}$  temperature for 72 hours or till the samples attained a constant weight and then average weight was expressed in gram per plant. At harvest stage, the weight of three plants along with cobs from net plot was recorded and added in biological yield. Dry matter per plant (g) was multiplied by plant population in each net plot and was divided by net plot area ( $\text{m}^2$ ) to get dry matter accumulation per square metre. The days taken for 50 per cent tasseling and 50 per cent silking were calculated by taking the difference of date of sowing and dates of 50 per cent plants have tassel and silk. At milk stage cobs from the net plot area were harvested and weighed, without removing husk. The value was expressed on hectare basis in kg. Grains from middle portion of randomly selected cob were used. Hand refractometer was used to measure total soluble solids. Protein content in grain was worked out by multiplying nitrogen content of grains with a factor 6.25 (A.O.A.C., 1965).

## Results and discussion

The increase in plant height was slower in first 30 days after sowing (DAS) but a rapid increase in plant height was observed between 30 and 45 DAS (Table 1). Rate of increase was slower between 45 DAS to harvest stage. The varying planting geometries did not influence plant height significantly at different growth stages. However,

a trend of increase in plant height was found with increase in row to row and plant to plant spacing. Where the widest planting geometry *i.e.* 75 cm  $\times$  30 cm registered numerically the highest plant height at all growth stages (73.8, 134.0 and 155.4 cm, respectively). Plant height increased with increasing nutrient dose at all crop growth stages but differences were non-significant at 30 DAS. At 45 DAS and at harvest, application of 180: 90: 60 kg N:  $\text{P}_2\text{O}_5$ :  $\text{K}_2\text{O}$ /ha being at par with 150: 75: 50 kg N:  $\text{P}_2\text{O}_5$ :  $\text{K}_2\text{O}$ /ha resulted into significantly higher plant height (131.5 and 153.9 cm, respectively) than 120: 60: 40 kg N:  $\text{P}_2\text{O}_5$ :  $\text{K}_2\text{O}$ /ha. An increase in nutrient dose from 120: 60: 40 to 180: 90: 60 kg N:  $\text{P}_2\text{O}_5$ :  $\text{K}_2\text{O}$ /ha led to 5.8 and 8.6 per cent increment in plant height at 45 DAS and harvest stage, respectively.

Numerically more plant height under wider plant geometries may be attributed to less competition for water, nutrient, space, solar radiation etc. compared to narrow spacing. These factors helped in improving the growth in terms of plant height. The results are in conformity with the findings of Kurne *et al.* (2017) who reported taller plants under wider planting geometry. Nutrients play important role in plant growth and development. An adequate supply of N is associated with high photosynthetic activity and more cell division, cell elongation and nucleus formation. Similarly, P has significant role in energy storage and transfer and required for various physiological process. It also helps in N metabolism, biosynthesis of proteins and nucleic acids. K is required for enzyme activation for biochemical

**Table 1.** Effect of planting geometry and nutrient levels on plant height, dry matter and leaf area index (LAI) of sweet corn

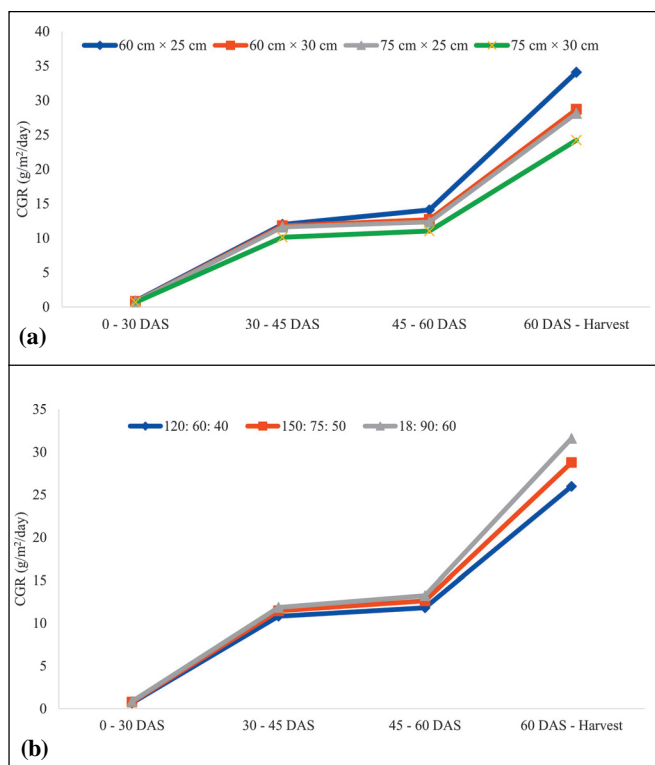
Treatment	Plant height (cm)			Dry matter ( $\text{g}/\text{m}^2$ )				LAI			
	30 DAS	45 DAS	Harvest	30 DAS	45 DAS	60 DAS	Harvest	30 DAS	45 DAS	60 DAS	Harvest
<i>Planting geometry</i>											
60 cm $\times$ 25 cm	67.9	123.3	141.7	23.5	183.9	376.9	935.7	1.59	3.39	2.78	2.20
60 cm $\times$ 30 cm	70.4	127.8	148.1	21.3	183.2	354.3	820.5	1.47	3.15	2.71	2.14
75 cm $\times$ 25 cm	71.1	128.7	149.1	20.7	180.3	350.3	815.3	1.44	3.08	2.69	1.94
75 cm $\times$ 30 cm	73.8	134.0	155.4	18.9	158.7	311.5	716.4	1.31	2.76	2.46	1.69
SEm $\pm$	2.1	2.9	2.1	0.7	6.3	10.6	19.5	0.03	0.12	0.06	0.08
CD at 5 %	NS	NS	NS	2.6	22.3	37.5	68.8	0.12	0.41	0.22	0.29
<i>Nutrient levels (kg N: <math>\text{P}_2\text{O}_5</math>: <math>\text{K}_2\text{O}</math>/ha)</i>											
120: 60: 40	68.5	124.3	141.7	19.0	164.7	324.1	786.1	1.39	2.98	2.51	1.87
150: 75: 50	71.2	129.7	150.2	20.9	177.6	350.2	822.1	1.46	3.07	2.65	2.02
180: 90: 60	72.6	131.5	153.9	23.4	190.2	370.6	856.3	1.51	3.23	2.80	2.09
SEm $\pm$	2.2	1.0	1.3	0.5	4.6	7.5	17.4	0.03	0.06	0.05	0.06
CD at 5 %	NS	2.9	4.7	1.4	13.9	22.7	52.8	0.09	0.17	0.16	0.18

reaction in plants. More availability of these nutrients in high nutrient dose treatments resulted into higher plant height. These results are in close agreement with the findings of Kurne *et al.* (2017) who noted that high dose of nutrients increased all the growth parameters.

Dry matter accumulation per square metre progressively increased with the advancement of crop age and reached to be the maximum at harvest stage (Table 1). The different planting geometry of sweet corn significantly influenced dry matter production at different growth stages. Dry matter accumulation per square metre decreased with increase in geometry. Planting geometry of 60 cm × 25 cm resulted into significantly higher dry matter accumulation (23.5 g/m<sup>2</sup>) at 30 DAS than 75 cm × 25 cm and 75 cm × 30 cm, respectively but was statistically at par with 60 cm × 30 cm. At 45 and 60 DAS, crop sown at 60 cm × 25 cm had significantly more dry matter accumulation (183.9 and 376.9 g/m<sup>2</sup>, respectively) than that of 75 cm × 30 cm but was at par with 60 cm × 30 cm and 75 cm × 25 cm. Significantly highest dry matter accumulation (935.7 g/m<sup>2</sup>) was recorded at the narrowest geometry of 60 cm × 25 cm. The lowest value was recorded from the widest planting geometry *i.e.* 75 cm × 30 cm at all growth stages. The per cent decrease in dry matter accumulation per square metre under 75 cm × 30 cm was 19.6, 13.7, 17.4 and 23.4 per cent over 60 cm × 25 cm at 30, 45 and 60 DAS and at harvest, respectively. Nutrient levels exerted significant effect on dry matter accumulation per square metre at all growth stages. An increasing trend in dry matter was observed with increase in nutrient dose. At 30 DAS, application of 180: 90: 60 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha produced significantly highest dry matter accumulation (23.4 g/m<sup>2</sup>). Crop fertilized with 180: 90: 60 being at par 150: 75: 50 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha had significantly higher dry matter accumulation than 120: 60: 40 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha at 45 and 60 DAS and at harvest (190.2, 370.6 and 856.3 g/m<sup>2</sup>, respectively). This treatment attained 23.2, 15.5, 14.3 and 8.9 per cent more value than 120: 60: 40 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha at 30, 45, and 60 DAS and at harvest stage, respectively. Dry matter accumulation in unit area depends on dry matter accumulation per plant and number of plants in unit area. Though narrow geometry produced less dry matter accumulation per plant than wider geometry but it occupied more numbers of plants per square metre. Therefore, narrow planting geometry produced more dry matter accumulation per

square meter compared to wider planting geometry. Similar results were also reported by Arvadiya *et al.* (2012). More dry matter accumulation per square metre under high nutrient dose was the result of more dry matter accumulation per plant under these treatments. This was the reason of more dry matter accumulation per square metre. Similar findings were reported by Jat *et al.* (2009).

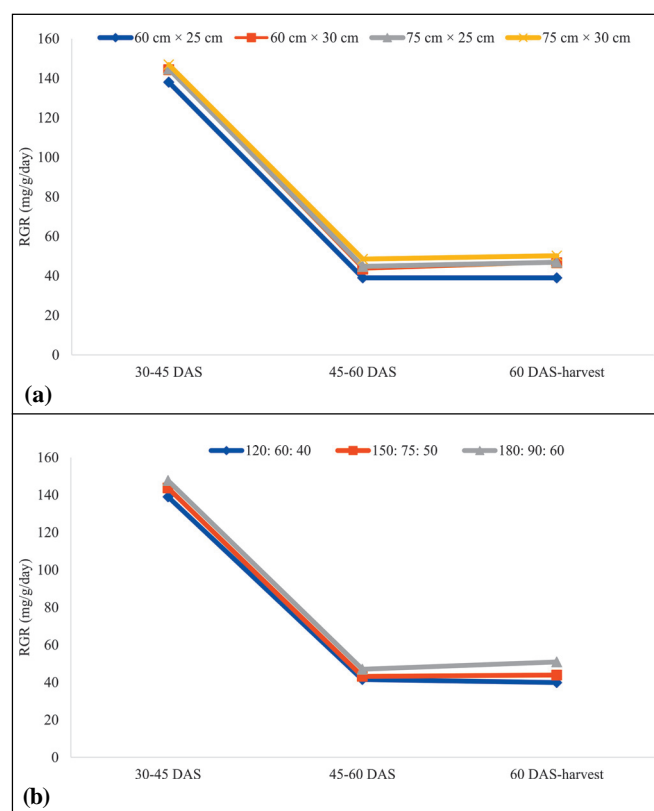
LAI increased with advancement of the crop age up to 45 DAS and declined thereafter (Table 1). Widening of planting geometry from 60 cm × 25 cm to 75 cm × 30 cm significantly reduced LAI at all growth stages. At 30 DAS, 60 cm × 25 cm planting geometry being at par with 60 cm × 30 cm recorded significantly more LAI (1.59) than other geometries. Crop grown under 60 cm × 25 cm planting geometry also exhibited significantly more LAI at 45 and 60 DAS and at harvest stage (3.39, 2.78 and 2.20, respectively) than 75 cm × 30 cm but did not differ significantly with other planting geometries. Significantly lower values were found under the widest planting geometry of 75 cm × 30 cm at all crop growth stages. Increase in nutrient supply brought significant enhancement in LAI of sweet corn at all growth stages. Crop fertilized with 180: 90: 60 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha being at par with 150: 75: 50 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha recorded significantly higher LAI than 120: 60: 40 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha at 30, 45 and 60 DAS and harvest stage (1.51, 3.23, 2.80 and 2.09, respectively). The lowest value of LAI (1.39, 2.98, 2.51 and 1.87, respectively) was recorded with minimum NPK dose at all growth stages. Leaf area index is ratio of leaf area per plant to ground area available per plant. Though, wider geometry produced more leaf area per plant than narrow plant geometry but it occupied more ground area per plant. Therefore, wider planting geometry showed low leaf area index compared to narrow geometry. The results are in close agreement with Verma & Tomar (2014). More LAI under higher dose of nutrients might be attributed to the adequate supply of NPK which helped in more leaf production and maintained the leaves active for longer period. Besides, more availability of nutrients in high nutrient level treatments increased leaves size by improving metabolic activities as evident by leaf area per plants. Because of these reasons, more LAI was recorded in high nutrient level treatments. Similar findings were also reported by Bhatt (2012). Data indicated that  $\overline{CGR}$  increased with the advancement of crop age (Figure 1). The maximum CGR was observed



**Figure 1.** Influence of different planting geometry (a) and nutrient levels (b) on crop growth rate (CGR) of sweet corn at different growth stage intervals

in the narrowest planting geometry of 60 cm × 25 cm at all growth stage intervals. At 0–30 and 30–45 DAS, this geometry was significantly superior (0.84 and 11.98 g/m<sup>2</sup>/day, respectively) to 75 cm × 30 cm but remained at par with 60 cm × 30 cm and 75 cm × 25 cm. At 45 - 60 DAS, 60 cm × 25 cm was also significantly higher (14.09 g/m<sup>2</sup>/day) than other geometries except 60 cm × 30 cm. At 60 DAS – harvest stage, significantly highest CGR was noticed (34.09 g/m<sup>2</sup>/day) at 60 cm × 25 cm planting geometry. Significantly lower  $\overline{CGR}$  (0.68, 10.12, 11.02 and 24.22 g/m<sup>2</sup>/day, respectively) was recorded in widest planting geometry 75 cm × 30 cm at all growth stages intervals. Increase in nutrient dose from 120: 60: 40 to 180: 90: 60 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha significantly enhanced  $\overline{CGR}$ . Crop fertilized with 180: 90: 60 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha produced significantly more  $\overline{CGR}$  (0.84, 11.85, 13.2 and 31.59 g/m<sup>2</sup>/day at 0–30 DAS, 30–45 DAS, 45–60 DAS and 60 DAS–harvest stage, respectively) than 120: 60: 40 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha but remained at par with 150: 75: 50 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha. Crop growth rate denotes dry matter accumulation in unit land area per day. Dry matter accumulation per unit area is product of dry matter per plant and plant population per unit area. Crop grown under narrow geometries though had less

dry matter accumulation per plant compared to that of grown under wider geometries but owing to more plant population led to higher CGR under these treatments. These results are in close agreement with the findings of Bhatt (2012). Crop fertilized with high levels of nutrients accumulated more dry matter per plant. Because of this reason, more CGR was obtained under high nutrient level treatments. These results are in accordance with Singh (2014). RGR exhibited significant variations due to different planting geometries and nutrient levels (Figure 2). An increase in RGR was observed with increase in planting geometries. Difference in RGR in relation to different planting geometries was significant during 45–60 DAS and 60 DAS – harvest stage. During 30–45 DAS, different planting geometry did not affect RGR significantly. However, crop sown under 75 cm × 30 cm geometry recorded numerically maximum RGR (146.9 mg/g/day). Planting geometry of 75 cm × 30 cm exhibited significantly higher RGR (48.5 and 50.2 mg/g/day at 45–60 DAS and 60 DAS – harvest, respectively) than 60 cm × 25 cm spacing but remained at par with 60 cm × 30 cm and 75 cm × 25 cm. RGR increased with increase



**Figure 2.** Relative growth rate (RGR) of sweet corn as influenced by different planting geometry (a) and nutrient levels (b) at different growth stage intervals

in nutrient dose and differed significantly at 45–60 DAS and 60 DAS – harvest stage. But at 30–45 DAS, nutrient levels failed to show significant effect in RGR while numerically the highest value (147.7 mg/g/day) was noted in 180: 90: 60 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha. Application of 180: 90: 60 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha recorded significantly maximum RGR (47.0 and 50.9 mg/g/day at 45–60 DAS and 60 DAS – harvest stage, respectively) whereas 120: 60: 40 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha recorded significantly lower values (41.5 and 39.9 mg/g/day, respectively). NAR reached maximum at 60 DAS – harvest stage (Figure 1). NAR decreased significantly with increase in plant geometries at all growth stage interval of crop. During the period of 30–45 DAS, 45–60 DAS and 60 DAS – harvest significantly higher NAR (5.89, 4.57 and 13.82 g/m<sup>2</sup>/day, respectively) was observed in 60 cm × 25 cm plant geometry than 75 cm × 25 cm and 75 cm × 30 cm but it was remained at par with 60 cm × 30 cm. Nutrient levels also significantly influenced NAR at all growth stages. Crop nourished with 180: 90: 60 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha being at par with 150: 75: 50 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha attained significantly higher NAR at 30–45 DAS, 45–60 DAS and 60 DAS – harvest stage (5.85, 4.67 and 13.77 g/m<sup>2</sup>/day, respectively) than 120: 60: 40 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha. NAR is governed by leaf area and dry matter production per unit leaf area in unit time. Not only leaf area but also dry matter production per plant is important in determining NAR. Both leaf area per plant and dry matter accumulation per plant were obtained significantly higher under the widest plant geometry of

75 cm × 30 cm but due to less number of leaves owing to less number of plants per unit area caused low NAR. On the other hand, the narrowest planting geometry of 60 cm × 25 cm produced less leaf area and dry matter accumulation per plant but due to more number of plants per unit area resulted into more NAR.

Planting geometry did not differ significantly for days taken to 50 per cent tasseling (Table 2). However, the narrowest (60 cm × 25 cm) and the widest (75 cm × 30 cm) geometry have taken minimum and maximum number of days to reach 50 per cent tasseling, respectively. Crop fertilized with different nutrient doses did not exhibit significant variations on days taken to reach 50 per cent tasseling. However, numerically the highest (51.6) and the lowest (51.2) days were noted in 180: 90: 60 and 120: 60: 40 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha, respectively. Different planting geometries and nutrient levels failed to bring significant difference in days taken to 50 per cent silking (Table 2). However, numerically the earliest silking (55.3 days) was recorded under the narrowest planting geometry with 60 cm × 20 cm.

Among the nutrient doses, days required to reach 50 per cent silking was minimum (55.3) under 120: 60: 40 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha. Phenology of maize plant is largely determined by genetic characters and not expected to be changed with external factors such as planting geometry and optimum nutrient dose. These results are in close conformation of Singh (2014) who also reported non-significant difference among various nutrient levels in days to reach 50 per cent tasseling and 50 per cent silking.

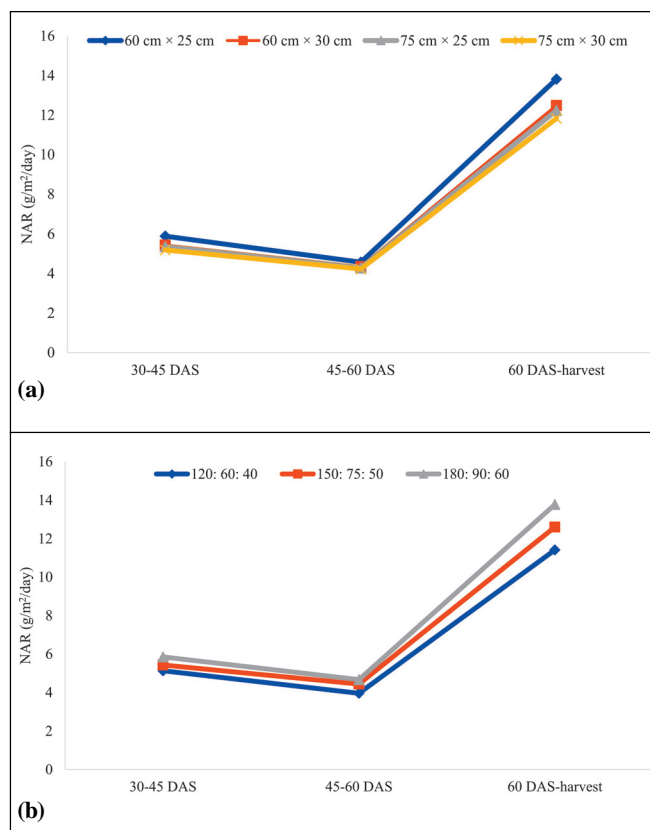
**Table 2.** Effect of planting geometry and nutrient levels on stem girth, flowering, quality and green cob yield of sweet corn

Treatment	Stem girth (cm)	Days to 50%		TSS (degree brix)	Protein content (%)	Green cob yield (kg/ha)
		Tasseling	Silking			
<i>Planting geometry</i>						
60 cm × 25 cm	4.8	51.2	55.3	15.2	7.96	13325
60 cm × 30 cm	5.2	51.3	55.5	15.4	8.14	12285
75 cm × 25 cm	5.3	51.3	55.5	15.5	8.38	12096
75 cm × 30 cm	5.6	51.5	55.7	15.6	8.52	11114
SEm ±	0.2	0.7	0.5	0.3	0.16	410
CD at 5 %	0.5	NS	NS	NS	NS	1447
<i>Nutrient levels (kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha)</i>						
120: 60: 40	5.0	51.2	55.3	15.4	7.81	11711
150: 75: 50	5.3	51.3	55.5	15.5	8.31	12217
180: 90: 60	5.4	51.6	55.7	15.5	8.63	12687
SEm ±	0.1	0.5	0.5	0.4	0.14	258
CD at 5 %	0.1	NS	NS	NS	0.43	NS

Stem girth significantly increased with increase in planting geometry from 60 cm × 30 cm to 75 cm × 30 cm (Table 2). Plants grown at wider geometry at 75 cm × 30 cm produced significantly more stem girth (5.6 cm) than that of 60 cm × 25 cm (4.8 cm) but were at par with other geometries. The increase in stem girth under planting geometry at 75 cm × 25 cm was to the tune of 16.7 per cent over 60 cm × 25 cm planting geometry. An increase in nutrient dose resulted into significant increase in stem girth. Application of 180: 90: 60 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha being at par with 150: 75: 50 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha exhibited significantly more stem girth (5.4 cm) than 120: 60: 40 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha (5.0 cm). Under wider planting geometry, plants have less inter plant competition which helped in proper growth and thus more stem girth was obtained in these treatments.

Different planting geometries were failed to bring significant variations in total soluble solids at milk stage (Table 2). However, numerically the maximum (15.6 degree brix) and minimum (15.2 degree brix) TSS were observed in 75 cm × 30 cm and 60 cm × 25 cm, respectively. Different nutrient levels also did not exhibit significant variations in TSS. However, numerically the maximum TSS (15.5 degree brix) was noted in 180: 90: 60 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha followed by 150: 75: 50 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha. An increase in protein content in grain from 7.96 to 8.52 per cent was observed with increase in spacing but variations were non-significant (Table 2). Protein content in grain differed significantly due to increase in nutrient levels. Crop fertilized with 180: 90: 60 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha being at par with 150: 75: 50 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha showed significantly more protein content (8.63 %) in grain than that of 120: 60: 40 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha. Protein content depends on nitrogen availability to plants and its assimilation into amino acids. More availability of nitrogen in high nutrient level treatments resulted into its more uptake by crop. High nutrient level also provided more phosphorus and potassium which might help in nitrogen assimilation into protein via energy supply and enzyme activation, respectively. Similar findings were also reported by Khan *et al.* (2018).

Significant difference in green cob yield was noted due to different planting geometries (Table 2). A progressive reduction in green cob yield was recorded with increase in planting geometry. Crop grown at 60 cm × 25 cm spacing being at par with 60 cm × 30 cm



**Figure 3.** Influence of different planting geometry (a) and nutrient levels (b) on net assimilation rate (NAR) of sweet corn at different growth stage intervals

and 75 cm × 25 cm produced significantly higher green cob yield (13325 kg/ha) than that of 75 cm × 30 cm. Planting geometry of 75 cm × 30 cm had 16.6 per cent lower cob yield than 60 cm × 25 cm. Increase in nutrient dose from 120: 60: 40 to 180: 90: 60 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha increased green cob yield but difference was not significant. Crop fertilized with 180: 90: 60 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha resulted into numerically the maximum green cob yield (12687 kg/ha). Wider planting geometry attained less number of cobs per hectare under this treatment due to low plant population which led to low cob yield. These results corroborate the findings of Kar *et al.* (2006) and Kurne *et al.* (2017).

## Conclusion

On the basis of present study it can be inferred that sweet corn in north western plain of India during *kharif* season should be sown at planting geometry 75 cm × 25 cm for its better growth and higher green cob yield. It should be fertilized with 120: 60: 40 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha.

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# Influence of planting density and nitrogen management on growth and productivity of maize in eastern India

Indrani Saha<sup>1,2,3</sup> · S. L. Jat<sup>2</sup> · Preeti Singh<sup>1</sup> · Smruti Ranjan Padhan<sup>3</sup> · Radheshyam<sup>3</sup> · Abhijit Mandal<sup>3</sup> · Ramniwas<sup>2</sup> · Manish Karkraliya<sup>2</sup>

**Abstract:** A field experiment was conducted during *kharif* 2022 for exploring the effect of planting density and N management on hybrid maize (*Zea mays* L.) in eastern India at the experimental farm of ICAR-Indian Agricultural Research Institute, Gauria Karma, Jharkhand. The treatments comprised of three planting densities *viz.*, 67.5 cm × 20 cm (D<sub>1</sub>), 67.5 cm × 22 cm (D<sub>2</sub>) and 67.5 cm × 25 cm (D<sub>3</sub>) in main-plots and five nitrogen management practices *viz.*, control, farmers practices (FP), RDN-conventional, 75 per cent RDN-SSB (sub-surface band placement) and RDN-SSB in sub-plots and replicated thrice. The results of the study indicated that the growth parameters, *viz.* plant height, leaf area index (LAI), dry matter accumulation and net assimilation rate was significantly higher at D<sub>1</sub>. However, crop growth rate, relative growth rate and net assimilation rate was obtained highest in D<sub>2</sub> and D<sub>3</sub>, respectively. Similarly, these growth parameters were enhanced by RDN-SSB. However, significantly higher grain yield was obtained with D<sub>2</sub> and RDN-SSB. Further, statistically at par growth parameters as well as grain yield was obtained under RDN-conventional and 75 per cent RDN-SSB which shows that saving of 25 per cent N could be achieved through sub-surface band placement. It was concluded that of growing of maize with 67.5 cm × 22 cm spacing and

fertilization with recommended dose of N as sub-surface band placement is recommended for yield maximization and saving of 25 per cent N can be achieved with sub surface band placement without any yield penalty and benefit-cost ratio is also higher at 67.5 cm × 22 cm in Eastern region of India.

**Keywords:** Dry matter accumulation · Nitrogen saving · Planting density · Sub surface band placement · Maize

## Introduction

Maize (*Zea mays* L.) being one of the most adaptable crops, has a wide range of adaptation under the various agro-climatic situations worldwide. Maize is referred to as the “queen of cereals” internationally due to the highest yield potential among all the cereals. It was grown in 202 m ha worldwide with production of 1162 mt with a productivity of 5.8 t/ha (FAOSTAT, 2022). In India during 2020-21, 9.9 m ha of maize were cultivated and producing 30 mt of maize with a productivity of over 3 t/ha despite the challenging *kharif* season environment over 82 per cent of the land. Due to its competitive advantage over C<sub>3</sub> plants as a C<sub>4</sub> plant, maize has an advantage over other crops in terms of the scenario of climate change and the sustainability of natural resources (Dass *et al.*, 2012; Padhan *et al.*, 2023). Poultry feed, which accounts for 47 per cent of all maize consumption and has increased over the past five years with a CAGR of 11 per cent (FICCI, 2022) is the most significant usage and demand driver of maize. The eastern India, specifically known for higher winter maize productivity whereas the productivity during the *kharif* season is below the national average despite grown under good agro-climatic conditions with adequate rainfall. The inappropriate

✉ S. L. Jat: sliari2016@gmail.com

<sup>1</sup>ICAR-Indian Agricultural Research Institute, Gauria Karma, Jharkhand, India

<sup>2</sup>ICAR-Indian Institute of Maize Research, Delhi Unit, Pusa Campus, New Delhi, India

<sup>3</sup>Post Graduate School, ICAR-Indian Agricultural Research Institute, New Delhi, India

adoption of crop management practices specifically planting density, nutrient and weed management leads to lower maize productivity in this region during *kharif* season.

Agro techniques *viz.*, Planting density and nutrient management are particularly important for boosting of the maize yield. The selection of inappropriate cultivars and inadequate plant population in the field are two of the major factors that contribute to low crop production (Yao *et al.*, 2016; Battaglia *et al.*, 2019). The contribution of nitrogen, phosphate, and potassium fertilisers are between 40 and 45 per cent (Khan *et al.*, 2014), and their use must be optimised to increase the production of the maize crop by improved placement methods. The split application of nitrogen fertilizer was a best practise because it's minimised the losses and led to greater dry matter formation and plant development compare than solitary application (Harikrishna *et al.*, 2005). When nitrogen was applied at low rates, maize grain output decreased by 43–74 per cent and the number of grains per plant increased by 33–65 per cent (Andrea *et al.*, 2006). The point placement of the N in maize has increased the yield and nutrient use efficacy significantly (Nayak *et al.*, 2022).

The planting density × nitrogen management optimization is required in maize in order to reduce bareness under high density environment and to increase per plant yield under lower planting densities. Hence, a study was planned with a hypothesis that planting density or nitrogen placement alone cannot optimize maize productivity but their synergistic or antagonistic effect need to accounted for yield maximization with lower environmental footprints.

## Materials and methods

An experiment was conducted during *kharif* 2022 at the Gauria Karma experimental farm of the ICAR-Indian Agricultural Research Institute, Jharkhand (24.2852°N, 85.360E and 228.6 m above the mean sea level) under irrigated conditions. The rainfall was unevenly distributed and most of it is received between July and September. The experiment was laid out in split plot with treatments consisted of three planting densities; 67.5 cm × 20 cm (D<sub>1</sub>), 67.5 cm × 22 cm (D<sub>2</sub>) and 67.5 cm × 25 cm (D<sub>3</sub>) were in the main-plots and five nitrogen management practices *viz.*, control, farmers practices (FP), RDN-conventional, 75 per cent RDN-SSB and RDN-SSB in sub-plot and replicated thrice. In farmers practice,

124.3:24.8:0 while in RDN, 150:26.2:33.2 kg NPK/ha was applied in our study. The sources used for applying N, P and K were urea, single super phosphate, diammonium phosphate (adjusted for its N content) and muriate of potash, respectively. Fertilizer application was made as per the treatment. Full dose of phosphorus and potassium and 1/3<sup>rd</sup> or 30 per cent N dose were applied at the time of sowing by drilling fertilizer in crop rows at ~4-5 cm below the seeding depth. The remaining N was given in two equal splits in farmers practice and RDN-conventional at knee high and tassel initiation stages as top dressing. In the SSB treatment, the N split at knee high stage was band placed along the crop rows by opening furrows with hand plough and the third split was applied at tassel initiation stages as top dressing. The maize hybrid CP 858 was used in our study. At harvest, the plants were counted from net plot area and expressed in thousands/ha as final plant stand. The plant height obtained from five tagged plants were averaged from each experimental unit. Three plants were randomly sampled at different growth stages (30, 60 and 90 DAS) from each experimental unit from designated rows outside net plot area (not from border) and samples were sun-dried and then oven-dried at 65°C for 72 hr and dry weight was recorded using electronic balance. The above-ground dry matter was averaged to get dry matter accumulation as g/plant and then converted to per square meter. Leaf area index was computed with formula given by Watson (1947) as follows:

$$\text{Leaf area index} = \frac{\text{Leaf area per plant (sq.cm)}}{\text{Ground area per plant (sq.cm)}}$$

The crop growth rate (CGR) was worked out at 30 days interval on the basis of dry matter accumulation at 30, 60 and 90 DAS and at harvest by using following equation:

$$\text{CGR (g/plant/day)} = \frac{W_2 - W_1}{T_2 - T_1}$$

Where, W<sub>1</sub>: dry weight at first stage (g), W<sub>2</sub>: dry weight at second stage (g), T<sub>1</sub>: Days at first stage, T<sub>2</sub>: Days at second stage

The relative growth rate (RGR) was calculated from the measurements taken at time T<sub>1</sub> and T<sub>2</sub> at 30 days interval. The RGR value was calculated by using following equation:

$$RGR \text{ (mg/g/day)} = \frac{\text{Loge}W_2 - \text{Loge}W_1}{T_2 - T_1}$$

Similarly, the net assimilation rate (g/cm<sup>2</sup> leaf area/day) was calculated by using the following formula:

$$NAR = \frac{W_2 - W_1}{T_2 - T_1} \times \frac{\text{Loge}L_2 - \text{Loge}L_1}{L_2 - L_1}$$

Where, L<sub>1</sub> and L<sub>2</sub> are leaf area at stage 1 and 2, respectively.

Data were statistically analysed using the analysis of variance technique applicable to the split-plot design. The significance of the treatment effect was determined using F-test; the means of the treatments compared using the least significant difference (LSD) at 5% probability level.

## Results and discussion

### Growth parameters

There was a consistent increase in plant height up to 60 DAS, after that the rate of increase in plant height was marginal. Plant height was significantly higher in D<sub>1</sub> by 4.5 per cent over D<sub>2</sub> at 60DAS and D<sub>1</sub> is on par with D<sub>2</sub>

at 30 and 90 DAS and higher by 7.4, 7.8 and 7.7 per cent over D<sub>3</sub> at 30, 60 and 90 DAS, respectively (Table 1). The lowest plant height was recorded with D<sub>3</sub> at all crop growth stages. When plants are more densely spaced, more auxin is secreted in the shaded areas due to shading (Alene *et al.*, 2000; Kumar *et al.*, 2012). On the other hand, it prevents the degradation of auxin and grows higher due to the increased concentration. However, the N-management practices have significant effect on plant height and the treatment RDN-SSB had statistically higher plant stand in our field experiment. The significantly lowest plant stand was recorded at harvest with control treatment. The plant height at 90 DAS was increased by 3.5, 6.6, 4.6 and 8.0 per cent with Farmers practice, RDN-conventional, 75 per cent RDN-SSB and RDN-SSB, respectively over control. The better availability of N under SSB increased chlorophyll content, which increased the rate of photosynthesis and extension of stem resulting in increased plant height.

Similarly, the planting densities and nitrogen management practices have significant effect on the leaf area index (LAI) of maize at various growth stages. The leaf area index was higher (4.60) with D<sub>1</sub> at 30, 60 & 90 DAS. The increase in LAI with increase in plant density may be due to a greater number of plants per

**Table 1.** Effect of different planting density and nitrogen management practices on the plant height and leaf area index at various growth stages of *kharif* maize

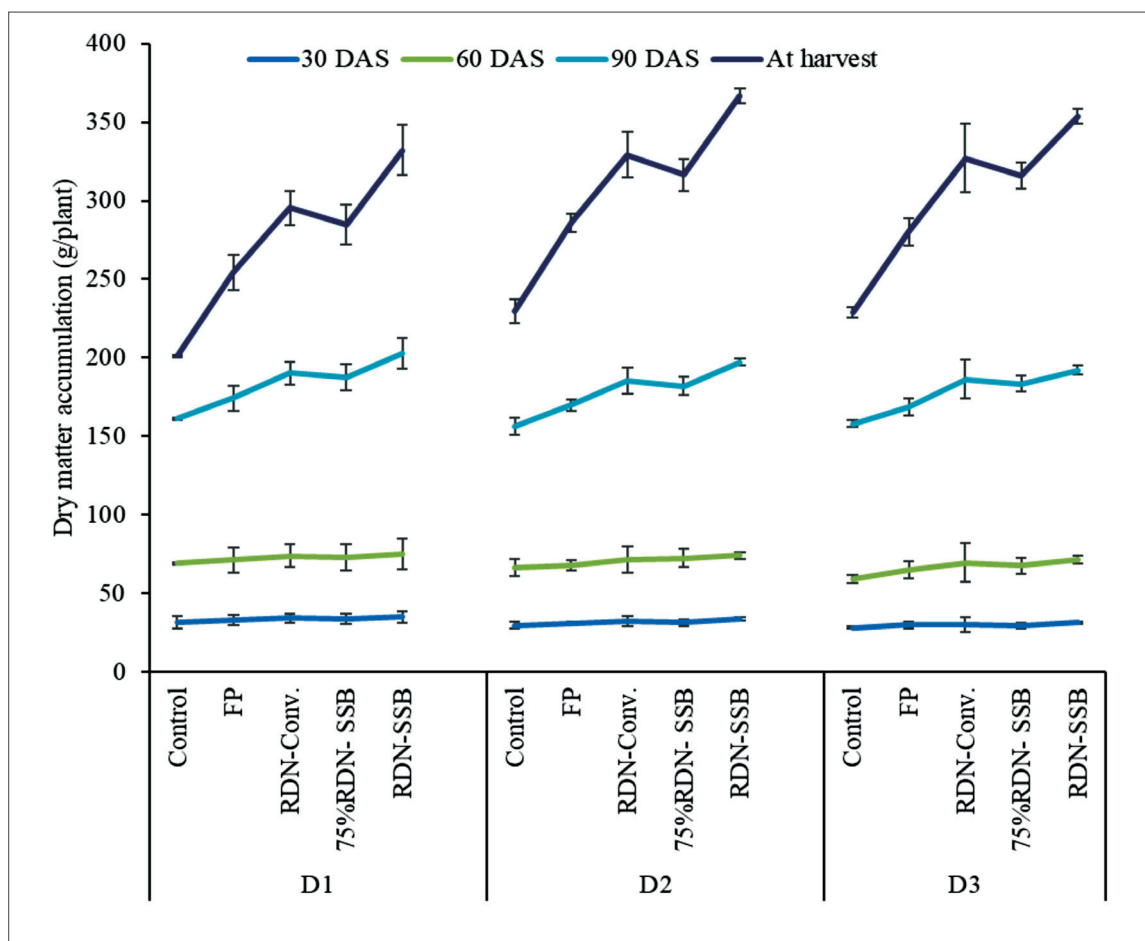
Treatments	Plant height (cm)			Leaf area index		
	30 DAS	60 DAS	90 DAS	30 DAS	60 DAS	90 DAS
<i>Planting density (plants'000/ha)</i>						
D <sub>1</sub> : 74 (67.5 cm × 20.0 cm)	90.1	212.2	215.1	2.21	4.80	4.60
D <sub>2</sub> : 67 (67.5 cm × 22.0 cm)	89.4	203.1	205.5	1.75	4.36	4.09
D <sub>3</sub> : 59 (67.5 cm × 25.0 cm)	83.9	196.8	199.7	1.42	3.67	3.26
SEm±	0.90	1.93	2.48	0.014	0.032	0.041
LSD (p=0.05)	3.52	7.57	9.73	0.054	0.127	0.162
<i>Nitrogen management (Tor N)</i>						
N <sub>1</sub> : Control	76.7	192.5	197.8	1.35	2.81	2.50
N <sub>2</sub> : Farmers practice	83.3	202.3	204.7	1.66	4.14	3.83
N <sub>3</sub> : RDN-Conventional	92.7	208.7	210.9	1.98	4.73	4.42
N <sub>4</sub> : 75% RDN-SSB	90.6	204.4	206.8	1.90	4.66	4.37
N <sub>5</sub> : RDN-SSB	96.6	212.1	213.6	2.08	5.06	4.80
SEm±	1.96	4.38	3.56	0.039	0.096	0.093
LSD (p=0.05)	5.71	12.79	10.40	0.115	0.280	0.272
<i>Interaction</i>						
SEm±	3.386	7.59	6.17	0.068	0.166	0.162
LSD (p=0.05)	NS	NS	NS	NS	NS	NS

\* Control: only P and K, Farmers practice: 124.3:24.8:0; RDN: 150:26.2:33.2 kg NPK/ha; DAS: days after sowing, SSB; Sub-surface band placement.

unit area. Similar observation was also made by Muniswamy *et al.* (2007); Suryavanshi *et al.* (2008) and Kumar *et al.* (2012). Amongst the N management practices, significantly higher LAI was recorded in RDN-SSB which was found to be at par with RDN-conventional whereas it was significantly higher over the Farmers practice (Table 1). In case of RDN-SSB treatment, it is 92 per cent higher over the control treatment at 90 DAS. This might be due to higher nitrogen content stimulates protein synthesis, which in turn improves vegetative growth and increases photosynthetic surface area, resulting in longer and wider leaves. The plant height as well as in RDN-conventional and 75 per cent RDN-SSB was found to be statistically similar which reflects that similar key growth parameters can be achieved in maize with a saving of 25 per cent N by band placement of N to achieve that of conventional one. This could be due to the higher losses of N through N-volatilization in surface application in case of RDN-conventional whereas better

N placement in 75 per cent RDN-SSB improved soil N availability and provided adequate available N throughout the growing season, resulting in favourable increases in plant height, girth, leaf area and finally dry matter accumulation (Biradar *et al.*, 2013; Sinha, 2016; Nayak *et al.*, 2022).

Due to changes in plant density, dry matter accumulation (DMA) of maize exhibited considerable variation at the 30, 60, and 90 DAS developmental stages. The DMA/m<sup>2</sup> was significantly higher in D<sub>1</sub> by 21.3, 17.8, 24.7 and 11.6 per cent over D<sub>3</sub> at 30, 60, 90 and at harvest whereas it was at par with D<sub>1</sub> (Table 2). The increased plant population might have led in enhanced DMA per unit area in our study (Figure 1). The more space plant with increased LAI/plant could be primarily responsible for the higher dry matter production due to higher availability of the resources (sunlight, water, nutrient, space, etc.) (Valadabadi *et al.*, 2010; Siamak *et al.*, 2014).



**Figure 1.** Interaction effect of plant densities and nitrogen management practices on the dry matter accumulation of *kharif* maize at various growth stages. The vertical bars represent the standard error (N=3). [D<sub>1</sub>: 7.4 (67.5 cm × 20.0 cm); D<sub>2</sub>: 6.7 (67.5 cm × 22.0 cm); D<sub>3</sub>: 5.9 (67.5 cm × 25.0 cm); FP: Farmers practice: 124.3:24.8:0; RDN conv. (conventional): 150:26.2:33.2 kg NPK/ha; DAS: days after sowing; SSB: Sub-surface band placement)]

The N management practices significantly affected the DMA at all growth stages except at 30 DAS and significantly higher DMA either per plant or per m<sup>2</sup> was recorded in RDN-SSB at 60, 90 DAS, and at harvest as compared to other practices. At harvest, the RDN-SSB increased the DMA in maize by 30.9, 56, 49 and 77.3 per cent over the control, farmers practice, RDN-conventional and 75 per cent RDN-SSB, respectively. The DMA by RDN-conventional and 75 per cent RDN-SSB in all the stages were statistically at par which indicates possibilities of the 25 per cent N saving through sub-surface placement. However, interaction effects between plant density and N management practices were found non-significant for all the growth parameters studied at various growth stages in our study.

### Grain yield

The grain yield of the maize was significantly affected by density and N management methods and their interaction in our study. Compared to the wide spacing (67.5 cm × 25 cm) and closer spacing (67.5 cm × 20 cm), the yield performance at 67.5 cm × 22 cm spacing was primarily higher due to better optimization of space above and below ground leading to improved availability

of resources such as sunshine, air movement, and nutrient availability. The grain yield was higher at planting density of D<sub>2</sub> (6478 kg/ha) over D<sub>1</sub> and D<sub>3</sub> whereas the lowest grain yield (5872 kg/ha) was recorded with D<sub>3</sub> treatment. The grain yield at D<sub>2</sub> and D<sub>1</sub> treatments was increased by 10.3 and 3.5 per cent over D<sub>3</sub>, respectively. Similarly, the grain yield was increased by 91.7, 122.6, 120.6 and 170.8 per cent by farmers practice, RDN-conventional, 75 per cent RDN-SSB and RDN-SSB, respectively over control. However, The RDN-SSB treatment increased the grain yield of maize by 258.1, 54.9, 24.9 and 24.9 per cent over control, farmers' practice, RDN-conventional and 75 per cent RDN-SSB, respectively. (Table 2). The grain yield obtained by RDN-conventional and 75 per cent RDN-SSB was found to be statistically at par which indicates the possibility of 25 per cent N saving through sub-surface placement of the first conventional top dressing without any yield penalty. The increased growth parameters of maize under RDN-SSB as well as better growth parameters with increased plant stand resulted in higher yield of maize under these treatment in our study. The increased yield due to better N placement in maize was also recorded by Nayat *et al.* (2022).

**Table 2.** Effect of planting densities and nitrogen management practices on dry matter accumulation of *khariif* maize at various growth stages

Treatments	Dry matter accumulation (g/m <sup>2</sup> )				Grain yield (kg/ha)
	30 DAS	60 DAS	90 DAS	At harvest	
<i>Planting density (plants'000/ha)</i>					
D <sub>1</sub> : 74 (67.5 cm × 20.0 cm)	217.2	497.2	1327.0	1798.1	6080
D <sub>2</sub> : 67 (67.5 cm × 22.0 cm)	215.1	492.0	1244.2	1802.2	6478
D <sub>3</sub> : 59 (67.5 cm × 25.0 cm)	179.1	422.0	1064.4	1611.6	5872
SEm±	3.37	5.70	14.11	18.21	49.6
LSD (p=0.05)	13.24	22.38	55.40	71.50	195
<i>Nitrogen management</i>					
N <sub>1</sub> : Control	191.3	436.5	1071.6	1217.9	2433
N <sub>2</sub> : Farmers practice	201.3	459.0	1152.3	1594.3	5624
N <sub>3</sub> : RDN-Conventional	208.1	482.9	1262.9	1900.0	6975
N <sub>4</sub> : 75% RDN-SSB	203.2	477.7	1242.7	1814.5	6973
N <sub>5</sub> : RDN-SSB	215.2	495.8	1329.8	2159.9	8712
SEm±	5.42	10.70	27.64	38.48	118.5
LSD (p=0.05)	NS	31.23	80.66	112.32	345.8
<i>Interaction</i>					
SEm±	9.39	18.53	47.87	66.65	205.3
LSD (p=0.05)	NS	NS	NS	NS	NS

**Table 3.** Effect of different planting densities and nitrogen placement practices on the physiological indices at various growth stages of the *kharif* maize

Treatments	CGR (g/m <sup>2</sup> /day)				RGR (mg/g/day)			NAR (g/cm <sup>2</sup> leaf area/day)	
	0-30 DAS	30-60 DAS	60-90 DAS	90 DAS harvest	30-60 DAS	60-90 DAS	90 DAS harvest	30-60 DAS	60-90 DAS
<i>Planting density</i>									
D <sub>1</sub> : 7.4 (67.5 cm × 20.0 cm)	8.29	8.38	24.29	22.13	23.25	29.86	13.96	0.12	0.29
D <sub>2</sub> : 6.7 (67.5 cm × 22.0 cm)	7.14	7.41	23.29	25.39	23.71	31.67	16.76	0.10	0.26
D <sub>3</sub> : 5.9 (67.5 cm × 25.0 cm)	5.90	6.58	20.91	23.15	24.88	32.79	17.20	0.09	0.25
SEm±	0.16	0.14	0.86	0.37	0.65	0.67	0.48	0.00	0.01
LSD (p=0.05)	0.61	0.55	NS	1.47	NS	NS	1.88	NS	NS
<i>Nitrogen management</i>									
Control	6.73	6.02	18.30	11.68	21.31	29.86	10.80	0.10	0.25
Farmers practice	7.02	7.03	21.08	20.80	23.18	30.67	15.62	0.09	0.25
RDN-Conventional	7.18	7.99	24.37	27.13	25.08	32.01	17.51	0.11	0.27
75% RDN-SSB	7.05	7.84	23.62	25.15	25.13	31.79	16.80	0.11	0.25
RDN-SSB	7.58	8.39	26.79	33.03	25.04	32.87	19.13	0.12	0.31
SEm±	0.20	0.37	0.91	1.46	1.18	0.98	0.93	0.01	0.01
LSD (p=0.05)	NS	1.09	2.66	4.27	NS	NS	2.73	NS	0.04
<i>Interaction</i>									
SEm±	0.35	0.65	1.58	2.53	2.04	1.70	1.62	0.01	0.03
LSD (p=0.05)	NS	NS	NS	NS	NS	NS	NS	NS	NS

### Physiological indices

Planting density of D<sub>1</sub> recorded significantly highest crop growth rate (CGR) followed by D<sub>2</sub> and D<sub>3</sub> at all crop growth stages except 60-90 DAS (Table 3). The rate of CGR increased till 60-90 DAS and afterwards it decreased and in RGR D<sub>1</sub> recorded higher followed by D<sub>2</sub> and D<sub>3</sub> in our study. Planting density D<sub>2</sub> had the highest RGR and NAR at 60-90 DAS. Generally, increased planting density had negative effects on CGR, NAR and RGR in our experimentation. The decrease in CGR, RGR and NAR with increasing planting density may be due the decrease in light interception and LAI at low than at high plant density.

However, CGR was significantly influenced by N management practices at all the stages except during early stage of sowing-30 DAS. The RDN-SSB recorded significantly highest CGR which was at par with RDN-conventional and 75 per cent RDN-SSB at 90 DAS-harvest. The RGR was significantly influenced by N management practices at 60-90 DAS. Similarly, NAR was significantly affected by N management only at 60-90 DAS. Among the N management practices, NAR in RDN-SSB was on par with RDN-conventional and 75 per cent RDN-SSB at 60 DAS-90 DAS.

### Conclusion

It was concluded that planting of maize at 67.5 cm × 20 cm increased dry matter/m<sup>2</sup> and plant height whereas higher grain yield was obtained at 67.5 cm × 22 cm. Among the nitrogen management practices, growth parameters were recorded significantly higher with application of RDN-SSB. Also, RDN-conventional and 75 per cent RDN-SSB were statistically at par for all parameters including yield which indicates saving of 25 per cent N through sub-surface placement and reducing the cost of production and environmental footprints. Therefore, with an optimized spacing of 67.5 cm × 22 cm coupled with sub-surface N-placement can be adopted in eastern region of India and similar agro-ecologies for higher growth and productivity of *kharif* maize.

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# Effect of front line demonstration of improve maize varieties and bio-fertilizer in central Gujarat

D. M. Rathod<sup>1</sup> · B. N. Thakker<sup>2</sup> · M. B. Patel<sup>3</sup> · K. H. Patel<sup>4</sup> · H. S. Varma<sup>5</sup>

**Abstract:** Maize (*Zea mays* L.) is one of the most important cereal grains grown worldwide in a wider range of environments because of its greater adaptability. It is mainly used as a food source and now has become the most important raw material for animal feed. Attempts are made to improve productivity and to increase the area under maize by adopting improved maize varieties. To compare local check total of 165 FLDs were carried out in a systematic manner on the farmer's field to show the worth of new maize varieties. It was observed the improved maize varieties recorded a higher average yield of 2114.35 kg/ha (GAYMH-1), 2134.32 (GAWMH-2) and 3339 kg/ha (GAWMH-2) in *kharif* and 3906.47 kg/ha (GAYMH-1) and 3318.56 kg/ha (GAWMH-2) in *rabi*, respectively. The per cent increase was found to increase 17.58 per cent, 13.00 per cent and 12.42 per cent in *kharif* and 26.01 per cent and 15.10 per cent in *Rabi*, respectively. Frontline demonstrations can promote new crops and technologies and improve their popularity. Such improvements in crop technology will help farmers in reducing yield gaps and will improve their knowledge, attitude and skill.

**Keywords:** FLDs · Improved maize varieties

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✉ D. M. Rathod: dm Rathod@aau.in

<sup>1,4,5</sup>Assistant Research Scientist, MMRS, AAU, Godhra-389001, Gujarat, India

<sup>2</sup>Assistant Research Scientist, ARS, AAU, Sansoli-387130, Gujarat, India

<sup>3</sup>Unit Head & Associate Research Scientist, MMRS, AAU, Godhra-389001, Gujarat, India

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## Introduction

One of the most adaptable new commercial crops is maize (*Zea mays* L.), also known as 'corn' and having a wide range of climatic tolerance. It is referred to be the queen of cereals everywhere. After rice and wheat, maize is the third-most significant food crop in India. Maize is one of the principal sources of cereals for food, fodder, and processed industrial products. Around 790 million tonnes of maize are produced worldwide each year, and in some nations, it serves as a staple meal, accounting for more than one-third of the calories and proteins consumed. The developing world's need for maize is expected to treble by 2050, and it will likely surpass other crops in terms of worldwide production (Chaudhari *et al.*, 2017). It has assumed greater significance due to its demand for food, feed and industrial utilization. The global production of maize is next to wheat and rice. India produces between 10 to 14 million tonnes of maize annually, 80 to 90 per cent of which is harvested during the *kharif* season (Rai *et al.*, 2018). Due to high nutritional content of maize and the entire plant, it is widely used as fodder in several Indian states. By using improved high-yielding cultivars and suggested scientific practices, maize productivity per unit could be raised. In light of the aforementioned consideration, frontline demonstrations were regularly conducted in farmer's fields to highlight the value of new varieties and persuade them to use maize to enhance productivity (Rai *et al.*, 2016).

The productivity of maize may be improved by applying scientific and sustainable management production practices. This is usually achieved by adopting high-yielding varieties. Frontline demonstrations of crop cultivation mainly aim at advertising the newly released



crop production technologies and its management practices directly in the farmer's fields. These are conducted in fields ranging from varying agro-climatic zones. Furthermore, frontline demonstrations help to examine the factors assisting in higher crop production, field constraints during production, as well as generating production data and farmer's feedback information. The study aimed to find out yield, and percent increase effecting the adoption of new improved variety. Therefore, a study was carried out on "Effect of front line demonstration of improve maize varieties and bio-fertilizer in central Gujarat".

## Materials and methods

To show the effect of FLDs of improved maize varieties, a study was carried out in the central Gujarat districts of Panchmahal, Dahod and Mahisagar. These districts were selected purposively because FLDs were given to these districts and maize was the main crop grown in these districts.

The main purpose of FLDs was given to above districts is that to increase the maize productivity and to show the effect of FLDs of improved maize varieties and make farmers aware about the new hybrids and improved technologies. Purposive sampling technique was used for the selection of farmers. Front line demonstrations were planned and conducted at the farmer's fields under ICAR-AICRP project. Each demonstration was conducted in an area of 0.4 ha and adjacent to the farmer's fields in which the crop was cultivated with farmer's practice/local variety. The package of practices included were improved hybrids, maintenance of optimum plant stand, recommended fertilizers dose, plant protection measures. The spacing followed was at 60 x 20 cm sown with the seed rate of 8 kg/acre. All the participating farmers were trained on all aspects of maize production management. A total of 32 farmers were selected as respondent through

proportionate sampling. There is 12.8 ha area was covered by the frontline demonstration.

In addition, 133 farmers received FLDs through the STATE project. In this total, 53.2 ha area was covered in frontline demonstration. In this demonstration only Bio-fertilizer was given as fertilizer with package of practices included were improved maize varieties, maintenance of optimum plant stand, spacing followed was at 60 x 20 cm sown with the seed rate of 8 kg/acre. Production information was gathered and analyzed, including the average yield of maize hybrids and average yield of local varieties, and the percentage increase over FLDs and farmer's practices.

### Sampling technique

Purposive sampling technique was used for selection of farmers.

## Results and discussion

A comparison of frontline demonstration based on farmers' practice was analyzed and presented under the ICAR-AICRP project in Table 1. In this total 12.8 ha area was covered in front line demonstration in *kharif* and *rabi* season. During the period of study, it was observed that in a frontline demonstration, the improved maize varieties recorded a higher average yield of 3339 kg/ha (GAWMH-2) in *kharif* and 3906.47kg/ha (GAYMH-1) in *rabi* as compared to local check 2840 kg/ha in *kharif* and 3102.65 kg/ha in *rabi*, respectively. Similarly, the per cent increase in improved maize varieties was found to increase by 17.58 % in *kharif* and 26.01 per cent in *rabi* was reported by Chaudhari *et al.* (2017) and Kumar *et al.* (2010).

Besides, A Comparison of frontline demonstrations based on farmers' practices was analyzed and presented under the State improvement project in different districts of central Gujarat in Table 2. In this total, 53.2 ha area

**Table 1.** Yield and % increase in FLD under AICRP over local check

S.No.	Season	District	Variety	No. of FLDs	Total area (ha)	Average yield of FLDs (kg/ha)	Average yield local check (kg/ha)	% increase over local check
1	<i>Rabi</i> 2021-2022	Panchmahal, Mahisagar and Dahod	GAYMH-1	17	6.8	3906.47	3102.65	26.01
2	<i>Kharif</i> 2022	Panchmahal	GAWMH-2	15	6	3339	2840	17.58

Note: Hybrid cultivar: GAYMH-1 (Gujarat Anand Yellow Maize Hybrid-1, GAWMH-2 (Gujarat Anand White Maize Hybrid-2)

**Table 2.** Yield and % increase in FLDs along with bio-fertilizer under State project over local check

S.No.	Season	District	Variety	No. of FLDs	Total area (ha)	Average yield of FLDs (kg/ha) + bio-fertilizer	Average yield local check (kg/ha)	% increase over local check
1	Rabi 2021-2022	Panchmahal, Mahisagar and Dahod	GAWMH-2	73	29.2	3318.56	2883.49	15.10
2	Kharif 2022	Mahisagar	GAYMH-1	23	9.2	2114.35	1880.65	12.42
		Panchmahal, Mahisagar	GAWMH-2	37	14.8	2134.32	1888.78	13.00

Note: Local check = farmer's own stored seed

was covered in frontline demonstration in *Kharif* and *rabi* season to show the impact of Bio-fertilizer as only fertilizer. During the period of study, it was observed that in a front-line demonstration, the improved maize varieties with Bio-fertilizer recorded a higher average yield of 2114.35 kg/ha (GAYMH-1) and 2134.32 (GAWMH-2) in *kharif* and 3318.56 kg/ha (GAWMH-2) in *rabi* as compared to local check 1880.65 kg/ha and 1888.78 kg/ha in *kharif* and 2883.49 kg/ha in *rabi*, respectively. Similarly, the per cent increase in improved maize varieties was found to increase by 12.42 & 13.00 per cent in *kharif* and 15.10 per cent in *rabi* was reported by Rai *et al.* (2018 & 2016); Kumar *et al.* (2010) and Chaudhari *et al.* (2017).

From these results, it is revealed that the performance of improved maize varieties was found better than the local check and local practices.

## Conclusion

The Study revealed that the productivity, production and area under maize cultivation can be improved by adopting improved maize varieties which can be popularized through front-line demonstration in different districts of central Gujarat. It was observed that improved maize varieties recorded a higher average yield of 2114.35 kg/ha (GAYMH-1), 2134.32 (GAWMH-2) and 3339 kg/ha

(GAWMH-2) in *kharif* and 3906.47 kg/ha (GAYMH-1) and 3318.56 kg/ha (GAWMH-2) in *rabi*, respectively. The per cent increase was found to increase 17.58 per cent, 13.00 per cent and 12.42 per cent in *kharif* and 26.01 per cent and 15.10 per cent in *rabi*, respectively. New crop varieties and technologies can be promoted with frontline demonstrations. These provide good platforms for advertising the benefits with proven results. Further, these platforms help to improve the knowledge, attitude and skills of farmers, which will help reduce the prevailing yield gaps.

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## Maize composite Vijay: A landmark variety

Baldev S. Dhillon<sup>1</sup> · Surinder K. Sandhu<sup>2</sup>

Maize is a unique crop plant which has contributed more than others towards the science of Genetics and methods of Plant Breeding. It is the crop in which the concept of breeding hybrid cultivars, the cultivars with better performance than alternatives types, was developed, which is now-a-days is being adopted in almost all crop plants. Above all, maize cytogeneticist Barbara McClintock earned Nobel Prize in 1983 for her discovery of mobile genetic elements.

All-India Coordinated Research Projects (AICRPs) working under the auspices of Indian Council of Agricultural Research (ICAR) serve as a backbone of the National Agricultural Research System. The first AICRP was established on maize in 1957 (initially named as All India Coordinated Maize Breeding Scheme), (Dhillon *et al.*, 2006). Its grand success led to extension of the concept by ICAR to other field crops, horticultural crops, animal sciences, natural resource management, agricultural engineering and home science. At present, ICAR has more than 80 AICRPs including Network Projects, a shorter form of AICRP. The AICRP on Maize released nine hybrids during 1961 to 1964 (Dhillon *et al.*, 2006). These included seven double-cross hybrids: ‘Ganga 1’, ‘Ganga 101’, ‘Ranjit’ and ‘Deccan’ released in 1961, ‘VL 54’ in 1962 and ‘Ganga 3’ and ‘Him 123’ in 1964; and two double top-cross hybrids namely, ‘Ganga Safed 2’ and ‘Hi-Starch’ in 1963. ‘Ganga Safed 2’ became a popular hybrid in *Kharif* season, and ‘Hi-Starch’ played a historical role by laying the foundation of winter maize cultivation in Bihar and South India.

In spite of remarkable success of ‘Ganga Safed 2’ and ‘Hi-Starch’, hybrid maize cultivation did not make an impact on the expected scale. During 1963-64 to 1967-68, the coverage under hybrid maize has been rather slow and about 5% of the total area of 15 lakh acres under maize was sown with the hybrid varieties (Punjab Agricultural University (PAU) (Package of Practices of *Kharif* Crops, 1969). The most serious limitations identified were cumbersome hybrid seed production requiring long term planning and that the farmers have to purchase fresh hybrid seed for every planting. To overcome this, AICRP on Maize gave the concept of breeding composite cultivars, a type of open pollinated cultivar (OPC), and reoriented the breeding programme accordingly. The protocol for producing breeder seed of a composite and retaining its open-pollinated produce to use that as seed by the farmers for two to three generations was also developed (PAU Package of Practices of *Kharif* Crops, 1967).

A composite cultivar is developed by recurrent selection in an open-pollinating base population developed by bulking seed of a number of phenotypically selected genotypes. It differs from a synthetic variety, another OPC, which is produced by crossing *interse* a number of genotypes, selected for good combining ability, in all possible hybrid combinations. AICRP on Maize released six composites, namely ‘Amber’, ‘Jawahar’, ‘Kisan’, ‘Vijay’, ‘Vikram’ and ‘Sona’ for commercial cultivation in different parts of the country in 1967 (Dhillon *et al.*, 2006). Of these six composites, ‘Vijay’ was developed by PAU, Ludhiana. The genetic base of ‘Vijay’ was selection of open-pollinated ears from 14 yield trials, having Caribbean, Mexican, US and Indian materials, grown in *Kharif* season 1961 at Nucleus Seed Farm, Narainagarh, now known as University Seed Farm of PAU. The seeds of the selected ears were bulked and the

✉ Surinder K. Sandhu: suindersandhu@pau.edu

<sup>1</sup>Former Vice Chancellor, <sup>2</sup>Principal, Maize Breeder, Punjab Agricultural University, Ludhiana-141004, Punjab, India

resulting population allowed to random pollinate in subsequent generations accompanied by selection. Subsequently, it has been maintained in isolation in 1964 and 1965. It was initially known as 'Naraingarh Complex' and was evaluated over years and locations under the name 'J 1'

'Vijay' was recommended for cultivation for grain production across the country and was also grown in certain regions like Punjab, Jammu and Kashmir, Himachal Pradesh and North-eastern region even for green fodder. With time, 'Vijay' was replaced by new high yielding hybrids and composites but it continued to be cultivated in some niches in northern hill region (Jammu and Kashmir, Himachal Pradesh, Uttarakhand and North-east) and Punjab. It is evident from the allocation of breeder seed as per BSP I up to 2019 (Table 1). 'Vijay' was also released in Pakistan under its experimental name 'J1' and in Nepal as 'Rampur Yellow' which further testifies its

outstanding performance and wide adaptation. It was also a parent of two varietal crosses the advanced generations of which served as the base populations to develop composites 'Jawahar' and 'Sona'. 'Vijay' is only Indian maize cultivar which continued to be cultivated in the country for nearly half a century as well as was commercially cultivated outside India, that too in two countries, namely Pakistan and Nepal. The PAU continued to play a stellar role in Indian maize research programme on cultivar development and production technology. 'Ageti 76' is the first early maturing, high yielding variety in the country. It was released for cultivation in *Kharif* season in Punjab in 1976 and at the national level in 1982 (Khehra *et al.*, 1977, 1986). In 1995, PAU took the lead to develop India's first high yielding single cross hybrid 'Paras' (Dhillon *et al.*, 1995), which gave a new orientation to maize breeding programmes in the country.

The pioneering studies started in 1978 by PAU made it possible to commercially cultivate maize during non-traditional winter/spring season. Two high yielding, cold tolerant composites, 'Partap' and 'Partap-1' were released for cultivation in 1983 along with their production technology package for winter season comprising seed rate, sowing date, application of nutrients, irrigation schedule, etc. (Dhillon *et al.*, 1984). The technology package developed by PAU was adopted as such by the Haryana Agricultural University (now Chaudhary Charan Singh Haryana Agricultural University), Hisar and Rajasthan Agricultural University (Now Swami Keshwanand Rajasthan Agricultural University), Bikaner. Further, 'Ageti 76' laid the foundation for maize cultivation (after potato) during non-traditional spring season in Punjab in early 1980s, and maize has now emerged as an important crop in spring season.

**Table 1.** Year wise details of breeder seed allocation of maize composite Vijay

Year	Actual Allocation as per BSP I target (q)	Actual Production as per BSP IV (q)
1995	3.20	4.00
1996	2.20	-
1997	-	-
1998	0.50	1.70
1999	1.70	3.50
2000	1.80	-
2001	1.00	1.00
2002	0.80	1.50
2003	1.50	4.00
2004	1.37	2.00
2005	1.37	2.00
2006	0.62	2.20
2007	-	-
2008	0.57	1.50
2009	0.59	2.00
2010	0.32	4.00
2011	0.70	2.90
2012	1.00	2.40
2013	0.10	0.20
2018	0.50	0.50
2019	0.40	0.40

Source: Annual Reports, All India Coordinated Research Project on Maize

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# Utilization of corn for ethanol production: An analysis of production, economy, and grain type requirements

**Bijender Pal**

## Introduction

Corn ethanol production has gained significant traction as a renewable and environmentally friendly alternative to fossil fuels. Ethanol, a biofuel derived from corn, holds the promise of reducing greenhouse gas emissions, promoting energy security, and bolstering rural economies (Padhan *et al.*, 2023). This note aims to delve into the process of corn ethanol production, the economic implications, and the specific grain type requirements involved. Government of India has launched programs on ethanol blending patrols (EBP) in the year of 2003. 10% (E10) of the blended targets so far has been met from the sugarcane and broken rice. In order to achieve E20 target, we need nearly 14 million liters bio-ethanol by 2025-26. A call has been taken up to use water efficient crop “Corn” for bioethanol. Scientific community hears need to be geared up to develop the high yielding corn hybrids with preferred grain types for various segments. It is critical to start developing or introducing the germplasm which can enable to develop the hybrid with the grain texture referred by the ethanol industry. The product concept notes and trait prioritization has to be relooked and given equal emphasis of the grain types suitable for the ethanol production. In this article, efforts have been made to put some of the important facts which can be considered by the Breeders and policymakers for better use of corn as feedstock in ethanol production.

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✉ Bijender Pal: bijendrapal127@gmail.com

Bioseed Research India, Hyderabad-500033, Telangana, India

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## *Corn ethanol production process*

The production of corn ethanol involves several stages, beginning with the cultivation of corn, harvesting, and then it undergoes milling, where it is ground into a fine powder (Kurambhatti *et al.*, 2019). The next step is the conversion of starch into sugar, achieved through the process of enzymatic hydrolysis. Enzymes break down the corn starch into simple sugars, mainly glucose. Fermentation is the subsequent crucial step. Yeast is added to the sugar-rich liquid, and under controlled conditions, it consumes the sugars and produces ethanol and carbon dioxide. Distillation follows fermentation, concentrating the ethanol and removing impurities, resulting in a high-purity ethanol product. Lastly, dehydrating the ethanol to remove any remaining water content is necessary to achieve the required fuel grade.

## *Economic significance*

The production of corn ethanol has significant economic implications, both on a regional and national scale. The industry provides job opportunities across the supply chain, from farmers growing corn to workers in ethanol production plants. This contributes to the overall economic growth of rural areas and helps support local communities. Moreover, the reduction in dependence on fossil fuels through the use of ethanol can positively impact energy security. As a renewable resource, corn can be continuously grown, unlike finite fossil fuels. Furthermore, domestic production of ethanol may reduce reliance on foreign oil.

## *Grain type requirements*

While corn is the primary feedstock for ethanol production, not all corn varieties are equally suitable. The

most commonly used type is dent corn, known for its high starch content. Starch is the primary component that gets converted into sugar during the production process. Other types of corn, such as sweet corn or popcorn, are less suitable for ethanol production due to their lower starch content. In recent years, there has been growing interest in developing advanced technologies to produce ethanol from other parts of the corn plant, such as corn stover (leaves, stalks, and cobs) and even corn kernel fiber. These advancements could potentially increase overall ethanol production efficiency and reduce the pressure on using high-quality grain for fuel purposes. As technology and research continue to advance, there are ongoing developments in the field of corn ethanol production, aiming to enhance efficiency, sustainability, and cost-effectiveness.

#### *Breeding and product developments*

Breeding corn for better ethanol productivity is a very important activities of this mission. Till now, starch content was not a targeted trait in the most of the breeding programs in India, that is why the starch content in most of the breeding materials in India as below 70%. Low starch content and hard grain texture may be the undesirable and uneconomical for ethanol production. Breeding groups need to work and accumulate the Germplasm which has high starch content and developing the product accordingly. This is a long process (explained below) and also so critical to develop the hybrids with grain type and textures which is required for ethanol production (Gong *et al.*, 2022).

- 1) Germplasm collection – introduction of temperate material
- 2) Characterization of germplasm
- 3) Understanding heritability of traits
- 4) Trait introgression in the local Germplasm
- 5) Developing the trait base gene pools as pre breeding activity
- 6) Line development
- 7) Hybrids development / identification of the high yielding hybrids with high starch contents.

#### *Other considerations*

- 1) Second-Generation Ethanol Production: Second-generation ethanol production involves using the non-

food parts of the corn plant, such as corn stover (leaves, stalks, and cobs). These non-grain components contain cellulose and hemicellulose, which can be broken down into sugars and further converted into ethanol (Li *et al.*, 2016). This approach allows for better resource utilization and reduces the competition between food and fuel production.

- 2) Improved Fermentation and Yeast Strains: Research is ongoing to develop more efficient and robust yeast strains for ethanol fermentation. These new strains are engineered to withstand higher ethanol concentrations, tolerate contaminants, and have higher conversion rates, resulting in increased ethanol yields and faster production processes.
- 3) Energy-Efficient Processing: Scientists and engineers are continually seeking ways to improve the energy efficiency of corn ethanol production. This includes optimizing the use of water, electricity, and other resources during various stages of the production process. Energy-saving measures not only reduce the environmental footprint but also improve the economic viability of ethanol production.
- 4) Co-Products Utilization: Corn ethanol production generates several valuable co-products, such as distillers' dried grains with soluble (DDGS). DDGS is a protein-rich byproduct that is used as livestock feed. Researchers are exploring ways to further enhance the value of these co-products, such as refining them for human consumption or developing novel applications in other industries.
- 5) Sustainable Farming Practices: Sustainable farming practices are being promoted to reduce the environmental impact of corn cultivation. These practices include precision agriculture techniques, improved irrigation methods, and reduced chemical usage (Nisar *et al.*, 2021). Sustainable farming not only benefits the environment but also supports the long-term viability of corn as a biofuel feedstock.
- 6) Integrated Bio-refineries: Integrated bio-refineries represent an innovative approach where multiple bio-based products are produced from a single feedstock, such as corn. Alongside ethanol, these facilities may produce bio-chemicals, bio-plastics, and other renewable materials. This integrated approach maximizes resource utilization and diversifies revenue streams, making ethanol production more economically viable.

## Conclusion

The use of corn for ethanol production has emerged as a vital component of the renewable energy landscape. The process of producing ethanol from corn involves various stages, from cultivation to fermentation and distillation, with the specific grain type requirements emphasizing the need for high-starch varieties like dent corn. The economic benefits of corn ethanol production are multi-faceted, ranging from job creation to energy security and reduced greenhouse gas emissions. However, continuous research and technological advancements will play a crucial role in further improving efficiency, sustainability, and the overall viability of corn-based ethanol as a significant energy source. The development and implementation of new technologies and practices are transforming corn ethanol production, making it more sustainable, economically viable, and efficient. From the use of second-generation feedstock like corn stover to advanced fermentation techniques and integrated bio-refineries, these developments hold the promise of further reducing the environmental impact of bio-fuel production

and contributing to a greener, more sustainable energy future. Continuous research and innovation will play a vital role in unlocking the full potential of corn-based ethanol and other renewable fuels.

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