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Growth and Yield of Hybrid Maize as Influence by Levels of Nitrogen and Biofertilizer

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A B S T R A C T

A field experiment entitled "Influence of bio-fertilizer and nitrogen on growth and yield of hybrid maize (*Zea mays* L.)" was conducted in 2012 at the Agronomy Experimental Farm of the College of Agriculture, Central Agricultural University, Imphal. The experiment consisting of four nitrogen levels (0, 60, 120 and 180 kg N/ha) and three source of bio-fertilizers (control, *Azotobacter* and *Azospirillum*) was laid out in factorial randomized block design with three replication. The required quantity of nitrogen (60,120 and 180 kg/ha), phosphorus (60 kg/ha) and potash (40 kg/ha) were applied for each plot using Urea, SSP and MOP. Application of different levels of nitrogen and sources of bio-fertilizers exhibited significant differences in growth characters, yield contributing characters and yield. Application of nitrogen up to 120 kg/ha showed significant increase in yield contributing characters as well as the grain yield with an exception in test weight. Among the bio-fertilizers, *Azotobacter* resulted in significantly higher growth characters, yield contributing characters and yield than *Azospirillum* and control. The interaction effect between nitrogen and bio-fertilizer was found to be significant in all the characters considered. The treatment combination of 180 kg N/ha + *Azotobacter* or *Azospirillum* inoculation recorded higher grain and stover yield which remained at par to 120 kg N/ha + *Azotobacter*. Based on the results obtained from the experiment, it can be concluded that application of 120 kg N/ha and *Azotobacter* can be recommended for hybrid maize to get high yield.

Introduction

Maize (*Zea mays* L.) is one of the most important cereal crops in the world's agricultural economy both as food for man and feed for animals. It has very high yield potential, there is no cereal on the earth which has so immense potentiality and that is why it is called 'queen of cereals'. Maize crop is utilised in many ways and over 85 per cent of maize produced in the country is consumed as human food. It is also a good feed for poultry, piggery and other animals. It ranks below wheat and sorghum but considerably above rice in nutrition.

Introduction of high yielding varieties/hybrids of maize and adoption of improved production technologies enhanced the productivity of maize that resulted in more turn-over of the nutrients from the soil. With almost twice the quantity of plant nutrients being removed from the soil than what is added through fertilizers, the growing plant nutrient imbalance poses a major threat to sustain soil health and crop productivity in India. The need for continued increase in food and fodder production to meet the ever expanding human and livestock population and inadequate domestic fertilizer supplies coupled with the inability of chemical fertilizers to maintain long-term soil health and crop productivity in intensive cropping systems have underlined the need for integrated nutrient supply system. This involves the combined use of different nutrient sources such as chemical fertilizers, organic manures, bio-fertilizers, etc.

Maize is an exhaustive crop and therefore, needs adequate supply of nutrients particularly nitrogen for better growth and yield. Nitrogen is an integral part of chlorophyll, which is the primary absorber of light energy needed for photosynthesis and also imparts vigorous vegetative growth, dark green colour to plants. Inside the plant, Nitrogen converts to amino acids, the building blocks for proteins. These amino acids are then used in forming protoplasm, which is used in cell division. These amino acids are also utilized in producing necessary enzymes and structural parts of the plant and can become part of the stored proteins in the grain. In the first 25 days of a corn plants growth, it will absorb 8 per cent of its total nitrogen. This is critical because the corn plant determines the number of kernel rows in the first 25 days of growth. The number of kernel rows can dramatically influence yield, so placement in the root zone is extremely important. Nitrogen deficiency shows up in the yellowing or chlorosis of the plant leaves. Plants will typically be shorter or stunted and grow slower than plants with sufficient nitrogen. Nitrogen stress also reduces the amount of protein in the seed and plant.

Nitrogenous fertilizer, synthesized from fossil fuel, is a major limiting factor in agricultural productivity due to its depleting reserves. Therefore, an alternate source of nitrogen will have to be found out for crop plants. Bio-fertilizers have emerged as an important component of the integrated nutrient supply systems and hold a great promise to improve crop yields through environmentally better nutrient supplies. *Azotobacter* a free-living soil microbes will play an important role in the nitrogen cycle in nature, binding atmospheric nitrogen, which is inaccessible to plants, and releasing it in the form of ammonium ions into the soil. The aerobic bacteria *Azotobacter chroococcum* known to fix considerable quantity of nitrogen in the range of 20-40 kg of nitrogen per hectare in the rhizosphere in non-leguminous crops. The bacterium produces growth-promoting substances like Indole acetic acid, gibberellins, pantothenic acid, thiamine and niacin which promotes root proliferation and improve the plant growth and yield. It increases the rootlet density and root branching resulting in the increased uptake of mineral and water.

Effectiveness of inoculation on plant growth enhancement and crop yields depends upon its ability to survive and multiply in soils and is influenced by many abiotic and biotic factors including texture, pH, temperature, moisture content, soil type, soil amendment, nutritional status of the plant, plant species, plant age, microbial competition and predation.

The high cost of nitrogen fertilizers is often prohibitive to the small and marginal farmers. It is therefore important to explore alternative sources of nitrogen to partly meet the nitrogen requirement. Thus, the use of bio-fertilizers would be more remunerative and cost effective for getting higher returns with considerable fertilizer economy and better soil health.

Materials and Methods

The experiment was conducted during the pre-*kharif* season of 2012 at the Agronomy Research Farm of the College of Agriculture, Central Agricultural University, Imphal. The experimental site is situated at 24º 45' N latitude and 93 º 56' E longitude and at an altitude of 790 m above sea level. The mean minimum and maximum temperature recorded during the cropping season were 14.6 and 29.5 °C, respectively. The total rainfall recorded was 18 mm. The average relative humidity in the morning hours was 79.9 % and in the evening 56%.

The average sunshine hour was 6.3 and wind speed recorded 5 km/hr. The soil of the experimental site was clay in texture with a pH of 5.9 and available nitrogen 301.0 kg N/ha (medium), phosphorus 13.8 kg P_2O_5/ha (medium) and potash 355.4 kg K_2O/ha (high). Hybrid maize variety Varsha was chosen for this study. The prime objective of the experiment was to study the influence of bio-fertilizer and nitrogen on growth parameters and yield of hybrid maize.

The field experiment was laid out in Factorial randomized block design with three replications. The treatment consists of four nitrogen levels (0, 60, 120 and 180 kg N/ha) and three sources of bio-fertilizers (control, *Azotobacter* and *Azospirillum*). A uniform dose of 60 kg phosphorus (through SSP, 16% P_2O_5) and 40 kg potash (through MOP, 60% K_2O) per hectare and required amount of nitrogen as per treatment was applied in furrows, made 60 cm apart one day before sowing at 4-5 cm depth. Half of the total nitrogen, full dose of P_2O_5 and K_2O were applied as basal dose just a day before sowing of seeds and well mixed with the soil. The remaining half dose of nitrogen was applied in two equal splits at knee high stage and tasseling stage. Seeds were inoculated with *Azotobacter* and *Azospirillum* (20 g/kg seed) just before sowing as per the treatment. A slurry of inoculant and jaggery solution was prepared and crop seed mixed with it to have uniform coating over the seeds, which were dried for 30 minutes under shade, then seeds were sown immediately. All other agronomic practices were followed as per the standard. The hybrid variety of maize 'Varsha' was sown on 23rd February 2012 and harvested on 30th June 2012.

The observations on growth parameters viz., plant height, fresh and dry weight of plant and leaf area index were measured at 30, 60, 90 and 120 days after sowing. The yield parameters number of cobs per plant, cob length, cob girth, number of grain per cob and test weight was recorded.

Results and Discussion

Plant height (cm)

Plant height is reliable index of growth and developments representing the infrastructure build up by the plant over a period of time. The data recorded at various growth stages revealed that there was significant increase in plant height due to application of nitrogen (Table 1). In all the stages of recording with an exception at 60 DAS the plant height increased consistently and significantly up to 120 kg N/ha. However, at 60 DAS the plant height increased significantly up to 180 kg N/ha. Adequate nitrogen application increases the cell division, cell elongation, nucleus formation as well as green foliage. It also encourages the shoot growth. Therefore, higher doses of nitrogen increased the chlorophyll content which increased the rate of photosynthesis and extension of stem resulting increased plant height (Diallo *et al.*, 1996 and Thakur *et al*., 1998). Shivay and Singh (2000) also observed that application of nitrogen increased plant height by increasing length and number of internodes.

Inoculations of *Azotobacter* showed significantly taller plant over that of *Azospirillum* and control at all the growth stages. This might be due to the growth promoting effect showed by the beneficial microbe. This finding is also supported by Wu *et al.,* (2005).

Fresh weight and dry weight (g/plant)

The fresh and dry weights of plant were significantly influenced at various stages by the application of nitrogen. Though maximum fresh and dry weight was observed with 180 kg N/ha, however, the increase was significant up to 120 kg N/ha (Table 2 and 3). Beneficial effect of nitrogen application on fresh and dry weight of plant was also reported by Kumar (2008) and Siam *et al*., 2008. Raju *et al.,* (1997) also reported that higher doses of nitrogen applied to maize increased its availability and uptake, resulting in production of more photosynthates in terms of dry matter.

Remarkably fresh weight and dry weight of plant was recorded with the inoculation of *Azotobacter*. *Azotobacter* can affect plant growth not only by fixing nitrogen but also by altering microbial balance, suppression of pathogenic microorganisms, metabolism of soil phosphate and by providing metabolites that stimulate plant development after germination (Meshram and Shende, 1982). The effect on dry weight of plant with *Azotobacter* inoculation are in line with the earlier report of Jarak *et al.,* (2012) in maize.

Leaf area index (LAI)

LAI is an important plant growth index which determines the capacity of plants to trap solar energy for photosynthesis. The LAI increased progressively with the increase in age and reached maximum at 90 DAS and declined thereafter till harvest (Table 4). The maximum value of LAI was recorded with application of 180 kg N/ha at 30 and 60 DAS. However, at later stages it remained at par to 120 kg N/ha. This might be due to increase in leaf number and size, photosynthetic pigment in leaves with proper supply of nitrogen at higher dose resulting in more and larger photosynthetic apparatus of the crop which consequently influence the assimilated product. The beneficial effect of nitrogen on LAI was in conformity with the earlier findings of Sharar *et al.,* (2003), Bindhani *et al.,* (2007) and Meena *et al.,* (2007). Similarly, Oscar and Tollenaar (2006) concluded that LAI of maize increased with the application of higher rate of nitrogen and decline in lower doses.

Irrespective of the growth stages, inoculation of *Azotobacter* remarkably enhanced the LAI as compared to *Azospirillum* and control. Maximum LAI was observed at 90 DAS and decline thereafter, this might be due to increase in aging of leaves.

Yield contributing characters and yield

Number of cobs per plant

The number of cobs per plant increased significantly with every increase in the level of nitrogen up to 120 kg N/ha (Table 5). Higher LAI recorded in this level of nitrogen might have helped the crop to capture more solar radiation thereby leading to increased number of cobs per plant. Bangarwa *et al.,* (1988) and Kumar, 2008 also reported that adequate supply of nitrogen resulted in easy and greater availability of nitrogen to the crop which consequently increase the number of cobs. A significant increase in number of cobs per plant as a consequence of seed inoculation with *Azotobacter* in the present investigation is attributed to an improvement in nutrition status of the soil and creation of congenial environment for better root growth through secretion of growth promoting substances such as Gibberellin, cytokinin and auxin and availability of nitrogen fixed by the micro-organisms (Singh and Totawat, 2002).

Cob length (cm)

The maximum length of cob was recorded with application of 120 kg N/ha. This might be due to better nitrogen uptake and increased translocation of photosynthates from source to sink with adequate supply of nitrogen. The findings are in agreement with those of Sahoo and Mahapatra (2004) and Ananthi *et al.,* (2011).

The highest cob length was observed with *Azotobacter* inoculation which was superior to *Azospirillum* and control. The possible reason could be better absorption of essential nutrients, synthesize phytohormones that promote the ear length. Similar positive effect of *Azotobacter* was also reported by Biari *et al.,* (2008) on cob length of maize.

Cob girth (cm)

The cob girth increases with each increment in nitrogen up to 120 kg N/ha. Increase in cob girth of maize and baby corn with increasing levels of nitrogen was observed by Selvaraju and Iruthayaraj (1994) and Bindhani *et al.,* (2007). Inoculation of seed with *Azotobacter* was significantly superior to *Azospirillum* and control in respect of cob girth. The increase in cob girth with the inoculation of *Azotobacter* in maize was reported by Abbass and Okon (1993).

Number of grains per cob

Significant increase in number of grains per cob was recorded with increasing dose of nitrogen up to 120 kg N/ha. The lesser number of grains in lower dose of nitrogen might be due to nitrogen deficiency which reduced biomass production traits of the plant and ultimately reflecting to number of grains per cob. The results are in agreement with the earlier findings of Hammad *et al.,* (2011) and Dawadi and Sah (2012)

Azospirillum and control resulted in lesser number of grains/cob as compared to *Azotobacter*. The increase in grains per cob with *Azotobacter* inoculation has been reported by Sharifi *et al.,* (2011) in maize.

Test weight (g)

The test weight increases with each increment in nitrogen levels up to 120 kg N/ha. Such positive role of nitrogen in enhancing the test weight of maize is also recorded by Arif *et al.,* (2010) and Salam *et al.,* (2010).

Inoculation of *Azotobacter* showed higher value of test weight over control but remained at par to *Azospirillum*. The findings are in agreement with those of Biari *et al.,* (2008) in maize.

Grain yield (q/ha)

The grain yield was significantly affected by nitrogen application. Application of 120 kg N/ha showed a significant superiority over that of control and 60 kg N/ha and was at par with 180 kg N/ha in respect of grain yield (Table 5).

Such a positive yield response of nitrogen application is obvious when it is deficient in the growing medium. The soil sample analysed before the start of the experiment also showed that the available nitrogen status of soil in the experimental site was in medium range (301.0 kg N/ha). Application of nitrogen, therefore, provided better nutrition to maize which resulted in higher grain yield.

Table.1 Effect of nitrogen and biofertilizer on plant height (cm)

Treatment	30 DAS				60 DAS				90 DAS				120 DAS			
	B_0	B_1	B_2	Mean	B_0	B_1	B_2	Mean	B_0	B_1	B_2	Mean	B_0	B_1	B_2	Mean
N_0	21.73	25.37	23.57	**23.56**	81.33	103.20	91.70	**92.08**	124.50	158.30	143.80	**142.20**	161.40	195.07	175.80	**177.42**
N_1	24.03	27.53	25.80	**25.79**	99.07	114.13	107.40	**106.87**	156.10	185.17	166.77	**169.34**	179.27	210.90	194.07	**194.74**
N_2	25.77	30.53	28.30	**28.20**	109.57	128.73	117.00	**118.43**	184.93	218.33	192.47	**198.58**	199.63	225.07	211.93	**212.21**
N_3	27.80	32.47	30.17	**30.14**	119.70	141.13	131.53	**130.79**	194.73	229.83	213.90	**212.82**	210.20	236.83	229.53	**225.52**
Mean	**24.83**	**28.98**	**26.96**		**102.42**	**121.80**	**111.91**		**165.06**	**197.91**	**179.23**		**187.63**	**216.97**	**202.83**	
	N	B	N x B		N	B	N x B		N	B	N x B		N	B	N x B	
SEd+-	**1.02**	**0.89**	**1.77**		**4.60**	**3.98**	**7.96**		**7.59**	**6.58**	**13.15**		**7.38**	**6.39**	**12.78**	
CD (P=0.05)	**2.12**	**1.84**	**3.67**		**9.54**	**8.25**	**16.51**		**15.74**	**13.65**	**27.27**		**15.31**	**13.25**	**26.51**	

Table.2 Effect of nitrogen and biofertilizer on fresh weight of plant (g/plant)

Treatment	30 DAS				60 DAS				90 DAS				120 DAS			
	B_0	B_1	B_2	Mean	B_0	B_1	B_2	Mean	B_0	B_1	B_2	Mean	B_0	B_1	B_2	Mean
N_0	2.33	2.65	2.55	**2.51**	46.00	61.03	55.67	**54.23**	130.00	153.27	151.67	**144.98**	156.13	205.00	178.07	**179.73**
N_1	2.59	2.94	2.73	**2.75**	78.33	89.33	81.77	**83.14**	154.47	188.10	160.00	**167.52**	186.23	221.67	208.33	**205.41**
N_2	2.90	3.48	3.17	**3.18**	92.20	117.00	103.80	**104.33**	163.37	210.00	184.63	**186.00**	210.00	256.47	224.93	**230.47**
N_3	3.23	3.67	3.50	**3.47**	108.33	133.03	120.33	**120.57**	186.60	218.33	205.00	**203.31**	228.87	273.33	256.67	**252.96**
Mean	**2.76**	**3.19**	**2.99**		**81.22**	**100.10**	**90.39**		**158.61**	**192.43**	**175.33**		**195.31**	**239.12**	**217.00**	
	N	B	N x B		N	B	N x B		N	B	N x B		N	B	N x B	
SEd+-	**0.10**	**0.09**	**0.18**		**4.58**	**3.97**	**7.93**		**8.61**	**7.45**	**14.91**		**11.60**	**10.05**	**20.10**	
CD (P=0.05)	**0.21**	**0.19**	**0.38**		**9.50**	**8.23**	**16.45**		**17.85**	**15.46**	**30.92**		**24.06**	**20.84**	**41.69**	

Table.3 Effect of nitrogen and biofertilizer on dry weight of plant (g/plant)

Treatment	30 DAS				60 DAS				90 DAS				120 DAS			
	B_0	B_1	B_2	Mean	B_0	B_1	B_2	Mean	B_0	B_1	B_2	Mean	B_0	B_1	B_2	Mean
N_0	0.450	0.497	0.453	**0.467**	8.69	13.02	11.63	**11.11**	45.43	51.63	46.70	**47.92**	53.30	66.67	58.33	**59.43**
N_1	0.473	0.653	0.610	**0.579**	11.63	17.34	15.57	**14.89**	56.50	65.17	58.33	**60.00**	64.87	80.07	72.70	**72.54**
N_2	0.593	0.777	0.643	**0.671**	14.54	19.67	16.97	**17.06**	60.10	76.67	69.77	**68.84**	73.33	95.03	89.93	**86.10**
N_3	0.700	0.829	0.753	**0.761**	17.20	21.33	19.12	**19.22**	63.37	83.30	77.27	**74.64**	80.70	111.67	95.00	**95.79**
Mean	**0.554**	**0.689**	**0.615**		**13.05**	**17.84**	**15.82**		**56.35**	**69.19**	**63.02**		**68.05**	**88.36**	**78.99**	
	N	B	N x B		N	B	N x B		N	B	N x B		N	B	N x B	
SEd+-	**0.034**	**0.029**	**0.058**		**0.90**	**0.78**	**1.55**		**3.36**	**2.91**	**5.82**		**4.69**	**4.06**	**8.13**	
CD (P=0.05)	**0.071**	**0.060**	**0.120**		**1.87**	**1.61**	**3.21**		**6.97**	**6.04**	**12.07**		**9.73**	**8.42**	**16.86**	

Table.4 Effect of nitrogen and biofertilizer on LAI

Treatment	30 DAS				60 DAS				90 DAS				120 DAS			
	B_0	B_1	B_2	Mean	B_0	B_1	B_2	Mean	B_0	B_1	B_2	Mean	B_0	B_1	B_2	Mean
N_0	0.207	0.357	0.280	**0.281**	1.100	1.333	1.267	**1.233**	1.57	1.90	1.70	**1.72**	0.500	0.700	0.567	**0.589**
N_1	0.327	0.453	0.360	**0.380**	1.367	1.600	1.500	**1.489**	1.80	2.23	2.03	**2.02**	0.633	0.833	0.733	**0.733**
N_2	0.483	0.570	0.533	**0.529**	1.700	2.267	1.900	**1.956**	2.07	2.70	2.33	**2.37**	0.800	1.133	0.867	**0.933**
N_3	0.510	0.650	0.619	**0.593**	1.933	2.433	2.233	**2.200**	2.37	2.83	2.60	**2.60**	0.900	1.167	1.067	**1.044**
Mean	**0.382**	**0.508**	**0.448**		**1.525**	**1.908**	**1.725**		**1.95**	**2.42**	**2.17**		**0.708**	**0.958**	**0.808**	
	N	B	N x B		N	B	N x B		N	B	N x B		N	B	N x B	
SEd+-	**0.031**	**0.027**	**0.053**		**0.084**	**0.073**	**0.146**		**0.11**	**0.10**	**0.20**		**0.053**	**0.046**	**0.093**	
CD (P=0.05)	**0.064**	**0.056**	**0.110**		**0.174**	**0.151**	**0.303**		**0.23**	**0.21**	**0.41**		**0.110**	**0.095**	**0.193**	

Table.5 Effect of nitrogen and biofertilizer on yield attributing characters, grain and stover yield

Treatment	No. of cobs/plant	Cob length(cm)	Cob girth(cm)	No. of grains/cob	Test weight(g)	Grain yield(q/ha)	Stover yield(q/ha)
Levels of Nitrogen							
N_0	1.489	14.20	14.30	434.11	330.30	40.50	62.37
N_1	1.733	16.18	16.14	483.44	337.76	47.45	75.11
N_2	2.122	18.17	16.84	517.44	351.31	57.97	102.13
N_3	2.244	19.33	17.29	530.56	356.70	60.19	111.53
SEd+-	0.069	0.56	0.26	8.43	4.76	1.58	5.78
CD (p=0.05)	0.143	1.16	0.54	17.48	9.87	3.28	11.99
Source of biofertilizer							
B_0	1.750	15.51	15.27	473.83	334.70	47.13	78.76
B_1	2.042	18.41	16.83	508.75	352.80	55.51	96.79
B_2	1.900	16.99	16.33	491.58	344.55	51.96	87.79
SEd+-	0.060	0.49	0.22	7.30	4.12	1.37	5.00
CD (p=0.05)	0.124	1.02	0.46	15.14	8.54	2.84	10.37
SEd+-(NxB)	0.120	0.98	0.45	14.60	8.25	2.74	10.00
CD for NxB (p=0.005)	0.249	2.03	0.93	30.28	17.11	5.68	20.74

Increase in grain yield with the application of nitrogen may be attributed to better growth of plant as expressed in terms of plant height, fresh weight and dry weight of plant and LAI which were favourably affected by nitrogen fertilization. The improvement in growth resulted in significant increase in yield attributes like number of cobs per plant, grains per cob, cob length and cob girth and test weight which ultimately contributed to higher grain yield with the application of nitrogen. The beneficial effect of nitrogen on grain yield of maize has also been reported by Sahoo and Mahapatra (2004), Kumar (2008) and Srikanth *et al.,* (2009). The grain yield of maize was significantly higher with *Azotobacter* seed inoculation compared to *Azospirillum* and control. Again *Azospirillum* recorded higher grain yield over that of control. The reported increase in grain yield due to bio-fertilizer may be on account of its direct role in nitrogen fixation and also due to production of phytohormones like Indole acetic acid, Gibberellins and Cytokinin. Thus, adequate and continuous supply of nitrogen to the crop resulted in better growth and consequently higher yield. The higher grain yield associated with *Azotobacter* may also be attributed to more number of cobs per plant, cob length and girth and grains per cob. This finding is also supported by Mala *et al.,* (2010) and Farboodi *et al.,* (2011).

Stover yield (q/ha)

Stover yield increased significantly with the application of nitrogen up to 120 kg N/ha. The higher stover yield associated with the application of nitrogen can be attributed to better growth of the plant as expressed in terms of plant height, fresh and dry weight of the plant and LAI. This finding confirms the earlier report of Meena *et al.,* (2007) and Siam *et al.,* (2008).. The results of the present investigation have clearly indicated that application of bio-fertilizer had remarkably increased the stover yield. Between the bio-fertilizer significantly higher stover yield was recorded with the inoculation of *Azotobacter* as compared to *Azospirillum*. Higher stover yield with the inoculation of *Azotobacter* have also been reported by Patil *et al.,* (1992).

Interaction

The Interaction effect of 180 kg N/ha + *Azotobacter* showed similar trend with 180 kg N/ha + *Azospirillum* and 120 kg N/ha + *Azotobacter* in all the characters studied with few exception. Maize growth and yield were markedly increased when bio-fertilizers were combined with nitrogenous fertilizer. This finding was supported by Patil *et al.,* (1992) and Rout *et al.,* (2001).

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