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Effect of Consortium of Endophytic Nitrogen Fixing Bacteria on Plant Nutrient Concentration of Seasonal (Suru) Sugarcane under Drip Irrigation.

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Abstract: *A field experiment was conducted on "Effect of consortium of endophytic nitrogen fixing bacteria on yield and quality of seasonal (Suru) sugarcane (Saccharum officinarum) under drip irrigation" was carried out at AICRP on Water Management, M.P.K.V., Rahuri during 2014-15. The experiment was laid out in Randomized Block Design with six treatments and four replications. There were four levels of nitrogen (100%, 50%, 25% and 0%) with P_2O_5, K_2O, PSB, FYM and foliar application of consortium of endophytic nitrogen fixing bacteria with and without combination and set treatment of Acetobacter diazotrophicus. The results of this experiment revealed that foliar application of consortium of endophytic bacteria @ 3 L ha^{-1} at 60 days after planting given stage-wise results in concentration in leaves and cane.*

I. Introduction

Sugarcane is a sub-tropical crop which is botanically known as *Saccharum officinarum* (2n = 80). Sugarcane is the pride crop of Maharashtra State and plays a vital role in a Agriculture economy. India ranks first in production of sugarcane. It has occupied 40.75 lakh ha area in India, while 7.36 lakh ha in Maharashtra state. Sugarcane is a C4 intermediate short day crop. It is long duration i.e. perennial gramminous plant and it is favourably adaptable to various soil conditions. Among the sugarcane growing states, Maharashtra has a very congenial climate for the growth of sugarcane. The optimal temperature for sugarcane cultivation is between 20 and 35°C and the minimum rainfall requirement is 1,200mm per year (Ando. 2010., [1]). Area under this crop has been nearly doubled during the last two decades due to establishment of large number of sugar factories. Sugar industry is the largest agro based processing industry after textiles in India. Most of the sugar obtained in world i.e. 60% is from sugarcane. Sucrose content of sugarcane is 20%. . Sugarcane being a long duration crop produces huge amount of biomass, and requires large quantity of water (1100-2200 mm) and is mostly grown as an irrigated crop using surface irrigation. The drip irrigation adoption in sugarcane increases water use efficiency (60-200%), saves water (20-60%), reduces fertilization requirement (20-33%) through fertigation, produces better quality crop and increases yield (7-25%) as compared with conventional irrigation. However, if not installed properly, it may result in wastage of water, time, money and yield. Adoption of drip irrigation (surface or subsurface) system in sugarcane is technically feasible and economically viable and needs to be vigorously followed (Kaushal *et al.* 2012., [2]*)*. Nitrogen is primary nutrient, required by all crops in large amount. However, nitrogen fertilizer added in soil get leached out or washed out. It not only causes economic loss but also gives invitation to soil pollution, water pollution and environmental pollution. It causes harm to soil health as well as human health. The use of biofertilizers some extent useful and ecofriendly, option to overcome these problems. Nitrogen is an essential element in plant development and a limiting factor in plant growth. It represents about 2 per cent of the total plant dry matter that enters the food chain. Nevertheless, plants cannot directly access dinitrogen gas, which makes up about 80 per cent of the atmosphere. Plants absorb the available nitrogen in the soil through their roots in the form of ammonium and nitrates. The limited bio-availability of nitrogen and the dependence of crop growth on this element have spawned a massive N-based fertilizer industry worldwide (Dobermann, 2007., [3]; Westhoff, 2009., [4]). Endophytic nitrogen fixing bacteria are associative type nitrogen fixers. They fix nitrogen by staying in tissues. Mostly they are present in sugar containing plants, but are also present in non sugar plants. *Pennisetum purpureum*, *Ipomea batatas* (Doberiner *et al.* 1993., [5]), *Coffea Arabica* (Jimenez – Salgado *et al.* 1997., [6]), *Eleusine coracana* (Loganathen *et al.* 1999., [7]) and *Ananascomosus* (Tapia – Hernandez *et al.* 2000., [8]) these are some examples of non sugar plants in which endophytic bacteria stay. Consortium of endophytic bacteria include various bacteria like, *Acetobacter, Agrobacterium, Burkhloderia, Azospirillum, Herbaspirillum, Azoarcus* etc. Such endophytes, which are defined as microorganisms living inside of plant organs and tissues without causing disease symptoms, have become highly interesting models to study plant microbe interactions. *Acetobacter diazotrophicus* now a day's known as *Glucanoacetobacter diazotrophicus* is an acid loving bacterium requiring pH of 4.0 to 4.5 for growth and N fixation. It showed positive growth at 25^0, 30^0 and 40^0C temperature. Among the biofertilizers, the endophytic bacteria *Gluconacetobacter* and *Herbaspirillum* are gaining more importance, since they fix atmospheric nitrogen endosymbiotically.

II. Material And Methods

The investigation was carried out at AICRP on Water Management, M.P.K.V., Rahuri during 2014-15. Soil was medium black it belongs to Inceptisols. The climate of this area is semi-arid. The seedling material of sugarcane Co.M -0265 (Phule - 0265) was procured from Chief Scientist, AICRP on Water Management, Department of Agronomy, M.P.K.V., Rahuri. The experiment was laid out in RBD design with four replications and six treatments including one control and one recommended dose of fertilizers. Basal dose of nitrogen, phosphorus, and potassium i.e. recommended dose (250:115:115 kg of N, P_2O_5 and K_2O ha^{-1}) along with organic manures i.e. full dose of FYM, *Acetobacter diazotrophicus* and PSB as per the different treatment details per plot and replication wise were given before transplanting of seedlings. Remaining doses of fertilizers were applied at 6-8, 12-14, 18-20 weeks after transplanting i.e. top dressing. Consortium of Endophytic Bacteria foliar spray were taken at 60 days after transplanting for treatments 4 and 5 in all the four replications. Two hand weedings were carried out to keep plots free from weeds. Irrigation schedule for drip at alternate day as per 100% ETc was fixed.

2.1, Details of treatments

T_1 - Absolute control (No fertilizers),
T_2 - RDF (100% N, P_2O_5 and K_2O)
T_3 - 50% N + *Acetobacter diazotrophicus* @ 10 kg.ha^{-1} (set treatment)
T_4 - 25% N + Consortium of Endophytic Bacteria @ 3 L ha^{-1} (500L water ha^{-1}) [foliar spray at 60 DAT]
T_5 - 0% N + Consortium of Endophytic Bacteria @ 3 L ha^{-1} (500 L ha^{-1}) [foliar spray at 60 DAT]
T_6 - 0% N + without Consortium of Endophytic Bacteria.
NOTE :- 75% P_2O_5, 100% K_2O, 20 t.ha^{-1} FYM, 1.25 kg.$ha^{-1,}$
PSB common to all treatments except T_1.
The Consortia of Endophytic Nitrogen fixing bacteria was applied as foliar application @ 3 lit ha^{-1}. (500 lit water) at 60 days after transplanting.

2.2, Sampling technique and analysis

The cane and leaf samples were collected at harvest and were dried in the oven at 60^oC till constant weight is achieved. The samples were ground to fine powder with the help of whilly mill and were further digested in Kjeldahl digestion unit (48 tubes set) at required temperature as suggested by (Parkinson and Allen 1975., [9]). Then acid extract was used for determining the concentration of N, P, K by using standard methods of analysis (Table 1) The data was analyzed statistically by using randomized block design as per procedure described by (Panse and Sukhatme 1985., [11]).

III. Results And Discussion:

3.1, Plant nutrient Concentration

3.1.1, Nutrient concentration at 90 days of seasonal sugarcane.

The data regarding the effect of consortium of endophytic nitrogen fixing bacteria on nutrient concentration of leaf at 90 days of seasonal sugarcane is given in the Table 2.

3.1.1.1, Nitrogen concentration

The nitrogen concentration in the treatment T_3 was found to be the highest (2.49%) and it was at par with all other treatments except T_1. The lowest nitrogen concentration (1.20%) was recorded in treatment T_1.

3.1.1.2, Phosphorus concentration

The highest concentration of phosphorus (0.74%) was found in the treatment T_3 and it was at par with all other treatments except T_1. The lowest value of phosphorus concentration (0.54%) was recorded in treatment T_1.

3.1.1.3, Potassium concentration

The treatment T_5 recorded the highest potassium concentration (1.64%) and it was at par with the treatment T_2 (1.30%) and T_3 (1.46%), the lowest potassium concentration at 90 days (0.72%) was recorded in treatment T_1.

3.1.2, Nutrient concentration at 180 days of seasonal sugarcane.

The data regarding the effect of consortium of endophytic nitrogen fixing bacteria on nutrient concentration of leaf at 180 days of seasonal sugarcane is given in the Table 3.

3.1.2.1, Nitrogen concentration
The highest concentration of nitrogen at 180 days (2.10%) was found in the treatment T_4 and it was at par with treatments T_2 (1.93%), T_5 (2.07%) and T_6 (1.72%), respectively. The lowest (1.23%) was recorded in treatment $T_{1.}$
3.1.2.2, Phosphorus concentration
The highest concentration of phosphorus (0.68%) was found in the treatment T_4 and it was at par with treatments T_2 (0.65%), T_3 (0.63%) and T_6 (0.64%), respectively. The lowest value (0.56%) was recorded in treatment T_1.
3.1.2.3, Potassium concentration
The highest concentration of potassium (1.13%) was found in the treatment T_2 and it was at par with all other treatments except T_1. The lowest Potassium concentration (0.61%) was recorded in treatment T_1.
3.1.3, Nutrient concentration at 270 days of seasonal sugarcane.
The data regarding the effect of consortium of endophytic nitrogen fixing bacteria on nutrient concentration of leaf at 270 days of seasonal sugarcane is given in the Table 4.
3.1.3.1, Nitrogen concentration
The treatment T_4 recorded the highest (2.45%) concentration of nitrogen in leaf at 270 days and it was at par with treatment T_2 (2.00%). The lowest value of nitrogen (1.23%) was recorded in treatment T_1.
3.1.3.2, Phosphorus concentration
The highest value of phosphorus concentration was recorded in treatment T_2 (0.66%) and it was at par with all other treatments except T_1. The lowest value (0.52%) was recorded in treatment T_1.
3.1.3.3, Potassium concentration
At 270 days the treatment T_5 recorded the highest (1.56%) potassium concentration and it was at par with treatments T_2 (1.46%) and T_4 (1.43%). The lowest potassium concentration (1.00%) was recorded in T_1.
3.1.4, Nutrient concentration at 360 days of seasonal sugarcane.
The data regarding the effect of consortium of endophytic nitrogen fixing bacteria on nutrient concentration of leaf at 360 days of seasonal sugarcane is given in the Table 5.
3.1.4.1, Nitrogen concentration
The treatment T_4 recorded the highest (2.98%) concentration of nitrogen in leaf at 360 days and it was at par with treatment T_2 (2.91%). The lowest nitrogen concentration (2.21%) was recorded in T_1.
3.1.4.2, Phosphorus concentration
At 360 days the treatment T_5 recorded the highest (0.67%) phosphorus concentration in leaf and it was at par with treatments T_2, T_3 and T_4. The lowest phosphorus concentration (0.54%) was recorded in treatment T_1.
3.1.4.3, Potassium concentration
The highest concentration of potassium (2.60%) was found in the treatment T_5 and it was at par with all other treatments except T_1. The lowest value (1.50%) was recorded in the treatment T_1. The nutrient concentration in the leaves at different stages of growth was more in the treatments with combined application of N through urea and consortia of endophytic N fixing bacteria. This tends to the effect that, the growth of the plant and roots was luxurious, which could absorb more of nutrients which lead to increase in concentration in leaves.

3.2, Nutrient concentration of cane at harvest
The data regarding the effect of consortium of endophytic nitrogen fixing bacteria on nutrient concentration of cane at harvest of seasonal sugarcane is given in Table 6.
3.2.1, Nitrogen concentration
The treatments T_2 and T_4 jointly recorded the highest concentration (2.98%) of nitrogen at harvest in cane which was at par with the treatment T_3 (2.70%). The lowest N concentration (2.21%) was recorded in the treatment T_1.
3.2.2, Phosphorus concentration
At harvest the treatment T_2 recorded the highest (0.76%) concentration of phosphorus in cane and it was at par with all other treatments except T_1. The lowest phosphorus concentration (0.59%) was recorded in the treatment T_1.
3.2.3, Potassium concentration
At harvest the highest concentration of potassium (2.48%) was recorded in both the treatments T_4 and T_5 which was at par with all other treatments except T_1. The lowest potassium concentration (1.55%) was recorded in the treatment T_1. The increase in stage wise total nitrogen, phosphorus and potassium content in sugarcane might be due to application of organic and inorganic recommended dose of fertilizer was also reported by (Soomro *et al.* 2013., [12]). The increase in concentration in sugarcane might be due to better nutrient supply to plants due to application of nutrients either through soil application of fertilizers and consortium foliar spray. The results are in conformity with (Muthukumarasamy *et al*. 2006. [13]).

IV. Tables

Table 1. Methods adopted for plant analysis

1.	Total N (%)	Microkjeldahl method	(Piper 1966., [10])
2.	Total P (%)	Vanadomolybdate phosphoric acid yellow colour	(Piper 1966., [10])
3.	Total K (%)	Flame photometry	(Piper 1966., [10])

Table 2. Effect of consortium of endophytic nitrogen fixing bacteria on leaf nutrient concentration at 90 days of seasonal sugarcane.

Sr. No	**Treatments**	**Leaf nutrient concentration at 90 days (%)**		
		N	P	K
T_1	Absolute control	1.20	0.54	0.72
T_2	RDF (100% N,P_2O_5,K_2O)	2.45	0.71	1.30
T_3	50% N + *Acetobacter diazotrophicus*	2.49	0.74	1.46
T_4	25% N + consortium of endophytic bacteria foliar spray	2.21	0.72	1.10
T_5	0% N + consortium of endophytic bacteria foliar spray	2.42	0.69	1.64
T_6	0% N + without consortium of endophytic bacteria foliar spray	2.31	0.73	1.18
	S.Em. ±	**0.22**	**0.03**	**0.15**
	CD at 5 %	**0.69**	**0.10**	**0.45**
	Mean	**2.18**	**0.69**	**1.23**

Table 3. Effect of consortium of endophytic nitrogen fixing bacteria on leaf nutrient concentration at 180 days of seasonal sugarcane.

Sr. No.	**Treatments**	**Leaf nutrient concentration at 180 days (%)**		
		N	P	K
T_1	Absolute control	1.09	0.56	0.61
T_2	RDF (100% N,P_2O_5,K_2O)	1.93	0.65	1.13
T_3	50% N + *Acetobacter diazotrophicus*	1.47	0.63	0.89
T_4	25% N + consortium of endophytic bacteria foliar spray	2.10	0.68	1.11
T_5	0% N + consortium of endophytic bacteria foliar spray	2.07	0.62	0.91
T_6	0% N + without consortium of endophytic bacteria foliar spray	1.72	0.64	0.93
	S.Em. ±	**0.18**	**0.02**	**0.09**
	CD at 5 %	**0.55**	**0.05**	**0.26**
	Mean	**1.73**	**0.63**	**0.93**

Table 4. Effect of consortium of endophytic nitrogen fixing bacteria on leaf nutrient concentration at 270 days of seasonal sugarcane.

Sr. No	**Treatments**	**Leaf nutrient concentration at 270 days (%)**		
		N	P	K
T_1	Absolute control	1.23	0.52	1.00
T_2	RDF (100% N,P_2O_5,K_2O)	2.00	0.66	1.46
T_3	50% N + *Acetobacter diazotrophicus*	1.82	0.65	1.25
T_4	25% N + consortium of endophytic bacteria foliar spray	2.45	0.65	1.43
T_5	0% N + consortium of endophytic bacteria foliar spray	1.82	0.64	1.56
T_6	0% N + without consortium of endophytic bacteria foliar spray	1.75	0.62	1.24
	S.Em. ±	**0.18**	**0.02**	**0.09**
	CD at 5 %	**0.55**	**0.06**	**0.28**
	Mean	**1.84**	**0.62**	**1.32**

Table 5. Effect of consortium of endophytic nitrogen fixing bacteria on leaf nutrient concentration at 360 days of seasonal sugarcane.

Sr. No	**Treatments**	**Leaf nutrient concentration at 360 days (%)**		
		N	P	K
T_1	Absolute control	2.21	0.54	1.50
T_2	RDF (100% N,P_2O_5,K_2O)	2.91	0.61	2.55
T_3	50% N + *Acetobacter diazotrophicus*	2.52	0.63	2.48
T_4	25% N + consortium of endophytic bacteria foliar spray	2.98	0.65	2.55
T_5	0% N + consortium of endophytic bacteria foliar spray	2.56	0.67	2.60
T_6	0% N + without consortium of endophytic bacteria foliar spray	2.31	0.59	2.25
	S.Em. ±	**0.08**	**0.02**	**0.19**
	CD at 5 %	**0.24**	**0.06**	**0.58**
	Mean	**2.58**	**0.62**	**2.32**

Table 6. Effect of consortium of endophytic nitrogen fixing bacteria on nutrient concentration of cane at harvest of seasonal sugarcane.

Sr. No	Treatments	Cane nutrient concentration at harvest (%)		
		N	P	K
T_1	Absolute control	2.21	0.59	1.55
T_2	RDF (100% N,P_2O_5,K_2O)	2.98	0.76	2.38
T_3	50% N + *Acetobacter diazotrophicus*	2.70	0.70	2.28
T_4	25% N + consortium of endophytic bacteria foliar spray	2.98	0.74	2.48
T_5	0% N + consortium of endophytic bacteria foliar spray	2.63	0.71	2.48
T_6	0% N + without consortium of endophytic bacteria foliar spray	2.38	0.73	2.00
	S.Em. ±	**0.09**	**0.03**	**0.17**
	CD at 5 %	**0.28**	**0.08**	**0.51**
	Mean	**2.64**	**0.70**	**2.19**

V. Conclusion

Effect Of Consortium Of Endophytic Nitrogen Fixing Bacteria On Plant Nutrient Concentration Of Seasonal (Suru) Sugarcane Under Drip Irrigation.

Plant nutrient concentration *viz.,* N, P, K. at 90 DAT the treatment 50% N + *Acetobacter diazotrophicus* recorded significantly higher leaf nutrient concentration, at 180, 270, 360 DAT. The treatment 25% N + consortium of endophytic bacteria foliar spray recorded significantly higher leaf nutrient concentration. The treatment RDF (100% N, P_2O_5 and K_2O) recorded significantly higher N, P concentration at harvest and regarding K the treatment 25% N + consortium of endophytic bacteria foliar spray recorded significantly higher concentration. Application of 25% N + consortium of bacteria foliar spray recorded the maximum leaf nutrient concentration of N at harvest of the crop. Application of 0% N + consortium of bacteria foliar spray recorded the maximum leaf nutrient concentration of P and K at harvest of the crop. Nutrients like N and K were recorded highest in 25% N + consortium of bacteria foliar spray in cane at harvest of the crop. The application of 25% N + foliar application of consortium of endophytic bacteria @ 3 L ha^{-1} 60 days after transplanting of sugarcane was found beneficial for saving of 75 per cent nitrogen. In view of the above, it is concluded that, the application of 25% N + foliar application of consortium of endophytic bacteria @ 3 L ha^{-1} 60 days after planting of sugarcane was found beneficial in increasing stage wise total N concentration in leaves and cane. The results obtained in the present investigation are based on one year experimentation and needs to be validated for final recommendations.

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