International Journal of Chemical Studies

P-ISSN: 2349–8528
E-ISSN: 2321–4902
IJCS 2018; 6(2): 1030-1033

Received: 16-01-2018
Accepted: 18-02-2018

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Effect of consortium of Endophytic nitrogen fixing bacteria on soil properties of seasonal (Suru) sugarcane under drip irrigation

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Abstract
The present investigation entitled "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 the experiment conducted revealed that The higher value of organic carbon, N and K was recorded in treatment 25% N + consortium of endophytic bacteria foliar spray, soil available P was maximum in treatment 50% N + *Acetobacter diazotrophicus*.

Keywords: phosphorus solubulizing bacteria (PSB), farm yard manure (FYM)

Introduction
Sugar cane (Saccharum spp.) is a perennial grass (family Poaceae, subfamily Panicoide), and is cultivated in tropical and warm-temperate regions between 35°N and 35°S and from sea level to altitudes of 1,000 m in a wide variety of soil types (Reis *et al.*, 2007) [11]. 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]. Sugarcane production is highest in Brazil (734 million tons), followed by India (342 million tons), and China (115 million tons). Sugarcane was cropped over an area of 25 million hectares all over the world; the average yield was 70.5 tons per hectare. It has occupied 40.75 lakh ha. area in India, while 7.36 lakh ha. in Maharashtra state. 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. 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) [7]. Nitrogen is one of the most important key element in the agricultural production of various crop plants, sugarcane crop responds well to the nitrogenous fertilizers in terms of their yield and economic produce. Sugarcane being a very exhaustive and extracting crop, removes about 205 kg nitrogen for yielding a crop of 100 $t.ha^{-1}$ and for sustaining productivity, nitrogen is replenished through chemical fertilizer @ 340 $kg.ha^{-1}$ in sugarcane. However, nitrogenous fertilizers added in soil get leached out or washed out. It not only causes economic loss but also leads to soil pollution, water pollution and environmental pollution. It causes harm to soil health as well as human health. The use of biofertilizers to some extent are useful and ecofriendly option to overcome these problems. 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 *viz. Pennisetum purpureum*, *Ipomea batatas* (Dobereiner *et al.*, 1993) [4], *Coffea Arabica* (Jimenez – Salgado *et. al.*, 1997) [6], *Eleusine coracana* (Loganathan *et al.* 1999) [8] and *Ananas comosus* (Tapia – Hernandez *et al.*, 2000) [13] these are some examples of non-sugar plants in which endophytic bacteria stay. Biological nitrogen fixation is the potential biological process that maintains the soil nitrogen status under normal conditions.

In recent ears nitrogen fixation by *G. diazotrophicus* in sugar rich crops has been well established. Biological nitrogen fixation effectively supplemented the need of nitrogen and minimizes the cost of production by reducing doses of nitrogenous fertilizers. Endophytic nitrogen fixation concept has been recently gaining momentum. Cultivation in the tropical regions entails least input package of practices and soils are generally low in the nutrient status (Dakora and Phillips, 2002) [2] thus biologically fixed nitrogen can supplement the nitrogen requirement of the crops. Major studies of endophyic diazotrophs are focused on sugarcane and kallar grass (Dobbelaeve *et al.*, 2003) [3] thus indentification of diazotrophs from other economically important plants is of considerable value for sustainable agriculture. It has been suggested to be an endophytic contributor of nitrogen to sugar rich crops, as it fixes nitrogen in culture medium under acidity levels, sugar concentration and micro aerobic conditions that resemble those inside the plants and it fixes or accumulates nitrogen to sugar rich plants between 10 to180 kg per hectare per season. Among the biofertilizers, the endophytic bacteria *Gluconacetobacter* and *Herbaspirillum* are gaining more importance, since they fix atmospheric nitrogen endosymbiotic ally. The costs of nitrogen fertilizers are increasing day by day. Therefore, biological nitrogen fixation and phosphorus solubilization through microorganisms has been found very economical. So, *Acetobacter* and other endophytic bacteria are cheaper biofertilizers. Production technique is simple as well as method of application is easy. These endophytic bacteria will definitely help to reduce chemical nitrogen fertilizer doses. But, the work on endophytic bacteria and their consortium is very meager. The results received by the scientist on different crops has shown that endophytic bacteria can help in getting yields to a certain level, but the work on sugarcane has not been initiated to a greater extent.

Material and Methods

The present investigation entitled "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 soil belonged to Inceptisols. Climatically, this area falls in semi-arid, subtropical zone with annual rainfall varying from 307 mm to 619 mm. The average annual rainfall is 520 mm. Most of the rainfall is being received through South-West monsoon from June to September months. 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 seedling materials were prepared by taking the prominent buds of sugarcane, the buds were separated from cane with the help of bud chipper and grown in plastic trays by filling half the portion of the plastic tray cavities with sawdust, coco-pith, vermi compost and then they were fully filled with mixture after placing one prominent bud in the tray cavities. Before filling the trays the sugarcane buds/sets were treated with *Acetobacter diazotrophicus* for the treatment three only. After filling, the trays were kept one over the other inside the shade net, and a plastic sheet was placed over them for 2 days by tight covering for increasing temperature and avoiding entry of water, air or sunlight into trays and then spread evenly in the shade net. Watering to the trays (seedlings) was done in the evenings using rose cans. After appearance of two leaves, the application of water was increased gradually, depending on the moisture level in the trays. About 35-40 days after sowing nursery, the seedlings were ready for transplanting.

The experiment was laid out in RBD design with four replications and six treatments including one control and one recommended dose of fertilizers. The details of present investigation conducted during the year 2014-15 are as below.

Crop	:	Sugarcane
Variety	:	Co.M -0265 (Phule -0265)
Season	:	Seasonal (suru)
Design	:	RBD
Treatments	:	6
Replications	:	4
Plot size	:	Gross: 7.5 × 8.0 m Net: 4.5 × 7.0 m
Spacing	:	150 × 60 cm (Ridges and Furrows)
Recommended dose of Fertilizers	:	Suru crop – 250:115:115 kg N,P_2O_5 and K_2O ha^{-1}
Date of transplanting	:	11.2.2014
Date of harvesting	:	07.2.2015

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.

The following endophytic bacteria are the components of consortia:

1. *Acetobacter*
2. *Agrobacterium*
3. *Burkhloderia*
4. *Azospirrillum*
5. *Herbaspillum*
6. *Azoarcus*

Fertilizers Application

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,

Initial soil analysis

A composite soil sample (0-30 cm) was collected randomly from the experimental field for initial soil analysis. The chemical parameters like pH, EC, organic carbon, available N, P, K were determined using standard methods

After harvest of the crop

Composite soil samples (0-30 cm) were collected randomly from each plot after harvest of the crop. The soil samples were shade dried, pounded in wooden mortar and pestle and finally sieved for chemical analysis of parameters pH, EC, organic carbon, available N, P and K

Table 1: Methods adopted for chemical

Sr. No.	Parameter	Method used	Reference
	Soil analysis		
1.	pH (1:2.5)	Potentiometric	Jackson (1973) [5]
2.	EC (1:2.5)	Conduct metric	Jackson (1973) [5]
3.	Organic carbon (%)	Wet Oxidation	Nelson and Sommers (1982) [9]
4.	Available N (kg ha^{-1})	Modified alkaline permanganate	Subbiah and Asija (1956) [12]
5.	Available P (kg ha^{-1})	0.5 M $NaHCO_3$ (pH 8.5)	Olsen *et al.*, (1954) [10]
6.	Available K (kg ha^{-1})	N N NH_4OAC	Jackson (1973) [5]

Results and Discussion

Soil properties

The data regarding the effect of consortium of endophytic nitrogen fixing bacteria on soil properties of seasonal sugarcane is given in the Table 2.

There was no significant effect of consortium of endophytic nitrogen fixing bacteria on the value of pH and EC. The highest value of pH (8.50) and EC (0.17 dS m^{-1}) was recorded in treatment T_5 – 0% N + consortium of endophytic bacteria foliar spray. The lowest value of pH (8.33) was recorded in T_6 – 0% N + without consortium of endophytic bacteria foliar spray and lowest EC value was recorded in both the treatments T_1 and T_3 (0.14 dS m^{-1}).

The highest value of organic carbon (0.69%) was recorded in treatment T_4 - 25% N + consortium of endophytic bacteria foliar spray which was at par with T_2 – RDF (0.63 %) and the lowest value (0.48%) was recorded in treatment T_6.

The Treatment T_4 - 25% N + consortium of endophytic bacteria foliar spray recorded the highest available nutrients in soil for N and K (169.34 and 644.10 kg ha^{-1}), and lowest available N content was recorded in treatment T_6 (116.03 kg ha^{-1}), lowest available K content in soil was in treatment T_1 (530 kg ha^{-1}) respectively.

The treatment T_3 - 50% N + *Acetobacter diazotrophicus* recorded the highest available P content (18.48 kg ha^{-1}) in soil being at par with all other treatments except T_1. The lowest value of P (14.82 kg ha^{-1}) was observed in treatment T_1 (absolute control).

Table 2: Effect of consortium of endophytic nitrogen fixing bacteria on soil properties of seasonal sugarcane at harvest.

S. No	Treatments	PH (1:2.5)	EC (dS m^{-1})	Organic carbon (%)	Available nutrients in soil (kg ha^{-1})		
					N	P	K
T_1	Absolute control	8.49	0.14	0.49	119.17	14.82	530
T_2	RDF (100% N,P_2O_5,K_2O)	8.36	0.16	0.63	153.66	18.11	638
T_3	50% N + *Acetobacter diazotrophicus*	8.36	0.14	0.53	125.44	18.48	636
T_4	25% N + consortium of endophytic bacteria foliar spray	8.41	0.15	0.69	169.34	17.80	644
T_5	0% N + consortium of endophytic bacteria foliar spray	8.50	0.17	0.59	137.98	18.16	621
T_6	0% N + without consortium of endophytic bacteria foliar spray	8.33	0.15	0.48	116.03	18.26	621
	Mean	8.41	0.15	0.57	136.93	17.60	615
	S.Em. ±	0.05	0.01	0.02	4.18	0.62	19.98
	CD at 5 %	N.S	N.S	0.07	12.60	1.87	60.22
	Initial observations	8.70	0.14	0.42	134.8	18.59	694

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