J. Indian bot. Soc. e-ISSN:2455-7218, ISSN:0019 - 4468 Vol. 96 (3) 2017 :216-222

GROWTH RESPONSE OF *GLYCINE MAX* (L.) MERRILL VARIETIES TO MICROBIAL INOCULANTS

ARVIND KUMAR

Department of Botany, T.N.B. College , T.M. Bhagalpur University , Bhagalpur 812007 , Bihar , India

Email: akarvindkumar863@gmail.com

Date of online publication: 30th September 2017

In recent years use of microbial inoculants as a source of bio-fertilizers has become a hope for most of countries, as far as economical and environmental view point are concerned. The present paper deals with the comparative adaptability of two varieties of *Glycine max* (L.) Merrill (BSS-2 and RAUS-5) after seed dressing with *Rhizobium japonicum* (SB-16 strain) and *Azotobacter chroococcum* (S-41 strain) with respect to various morphological, derived and reproductive growth parameters. The growth parameters were calculated such as relative growth rate (RGR) , net assimilation rate (NAR) ,leaf area ratio (LAR) , specific leaf area (SLA) ,leaf weight ratio (LWR) ,shoot/root ratio. *Rhizobium* inoculated plants gave the highest values for vegetative, growth indices, fruit and yield traits as compared with the *Azotobacter* inoculated plants and non-inoculated (control) plants .Early flowering and weight of 100 seeds were noticed in *Rhizobium* treated plants. RAUS-5 variety performed better than BSS-2 in different microbial inoculants treatments.

Key Words : Microbial inoculants , Growth parameters , Early flowering , Yield

In developing country like India, use of chemical fertilizers is very common to increase the food production but these are economically not feasible for poor farmers. The environmental and health hazards associated with chemical fertilizer further make their possible use difficult. In recent years use of microbial inoculants as a source of biofertilizers has become a hope for most of countries, as far as economical and environmental view point are concerned. In recent years, free living bacteria (*Azotobacter*), associated (*Azosprillium*) and symbiotic (*Rhizobium*) bacteria are gaining much popularity. Such practices are being encouraged to save the chemical fertilizers, national economy and the environmental. Effects of rhizobial culture on yield of different pulses are made under All India Co-ordinated Pulse Improvement Research programme of ICAR, New Delhi. The present study attempts to study the effects of microbial inoculants (*Rhizobium japonicum* and *Azotobacter chroococcum*) on the growth behaviour of two varieties of *Glycine max* (L.) Merrill (BSS-2 and RAUS-5) with respect to various morphological, derived and reproductive growth parameters.

MATERIALS AND METHODS

Seeds of two varieties of *Glycine max* (L.) Merrill (BSS-2 and RAUS-5) were procured from the Birsa Agricultural University, Ranchi (Jharkhand). Microbial inoculants were obtained from Department of Microbiology, Bihar Agriculture University, Sabour, Bhagalpur. Seeds of both varieties were treated with of *Rhizobium japonicum* (SB-16 strain) and *Azotobacter chroococcum* (S-41 strain) to assess their germination and growth performance . For each variety nearly 200gm. of jaggary was dissolved in 500ml. of warm water. The syrup was cooled down to room temperature and *Rhizobium* and *Azotobacter* strains were added to it to make a thick paste. The seeds were well smeared with these inoculums separately and were spread on blotting paper in shade to get rid of extra moisture before sowing in pots.

Three sets of experiments were designed as follows:

UT :Non-inoculated (control) seeds.

RZ :Seeds inoculated with *Rhizobium.*

AZ : Seeds inoculated with *Azotobacter.*

Altogether 6 weekly harvests were taken. At each harvests 3 plants were randomly selected

Received on March 25, 2017 www.indianbotsoc.org Accepted on April 10, 2017

for each treatment and variety and labelled by tagging. The monoliths were washed with fine jet of water to remove soil particles. Root, Shoot and leaves were separated with scissors. Cotyledonary leaves were collected with normal ones. Outline of leaves were drawn on a graph paper and the leaf area were found out. Plants parts (root, stem and leaves) were dried separately in butter-paper bags at $80^0 \pm 2^0$C in an oven for 48 hrs. and stored in a desiccator before weighing. The 7th harvest was taken under each treatment at their full maturity for the estimation of fruit and seed yield per plant.

With the help of dry weights of roots, stems and leaves ,the growth performance including dry matter accumulation, leaf area, RGR, NAR, SLA, LWR and S/R ratio were worked out as per Evans (1972) and data were analysed statistically for significance test by analysis of variance (ANOVA-single factor) as between harvests and within treatments at 95% confidence level.

RESULTS AND DISCUSSION

From the result it is evident that both varieties attained maximum height, Number of branches/plant and number of leaves/plant in *Rhizobium* inoculated plants followed by *Azotobacter* inoculated plants and non-inoculated (control) plants respectively (Table 1). From the result it can be emphasised that BSS-2 performed better than RAUS-5 in different microbial inoculated conditions. These results confirm the finding of Venkatech *et al.* (1988) ,Vessey (2003) , Amit *et al*. (2015) and Dixit (2013) .Plants treated with microbial inoculants increased the rhizospheric population of bio-fertilizers in those plants and they supply hormones to the roots. The supply of hormones eventually leads to an increases in root and shoot length.

The dry matter production and leaf area (Table 2) was maximum in *Rhizobium* treated plants in both the varieties and minimum in control. The RAUS-5 had higher dry matter accumulation than BSS-2. BSS-2 showed higher leaf area than RAUS-5 in *Rhizobium* treated plants and it was higher in *Azotobacter* treated plants in case of RAUS-5. Leaf area increased with the advancement of age of the plants. Inoculation with *Rhizobium* might more increase the nodulation and there by fixed more atmospheric N_2 which was used by the plant. Higher N_2 avaibility in plants through *Rhizobium* and *Azotobacter* inoculation might have increased the uptake of all other nutrients, which in turn helped the crop for better performance (Kader 2002, Bhunia *et al*. 2006, Minaxi *et al*. 2013). The plant growth promoting rhizobacteria enhance plant growth by direct or indirect mechanisms .These enhance plant growth by producing different metabolites which are responsible for the growth promotion of plant by direct mechanism, such as production of plant hormones like IAA, Gibberellic acid, Cytokikines and production of siderophores (Husen 2003). Ardhapurkar and Manwar

Table 1: Morphological growth attributes of soybean varieties (BSS-2 and RAUS-5) under different microbial inoculants

Harvest	varieties	Height (cm)			Branch No./ Plant			Leaf No. / Plant		
		Cont.	Az	Rz	Cont.	Az	Rz	Cont.	Az	Rz
1	BSS-2	35.30	37.70	40.10	1	1	1	8.00	8.50	9.00
	RAUS-5	32.05	36.45	39.05	1	1	1	6.50	7.50	8.50
2	BSS-2	44.80	47.25	50.85	1	1	1	9.50	12.00	14.00
	RAUS-5	40.25	42.45	46.10	1	1	1	10.00	12.00	12.50
3	BSS-2	103.25	106.00	110.50	1	1	1	19.00	20.50	21.00
	RAUS-5	69.75	110.50	76.25	1	1	1	16.00	17.50	18.00
4	BSS-2	108.95	112.00	114.50	1	1	1	26.00	27.50	28.00
	RAUS-5	101.50	105.00	108.00	1	1	1	25.50	27.50	29.50
5	BSS-2	118.00	121.50	124.00	4.50	6.00	7.50	41.00	43.50	46.00
	RAUS-5	116.50	119.00	122.00	4.00	6.50	7.50	36.00	39.00	42.00
6	BSS-2	138.00	142.00	145.50	9.00	10.00	11.00	66.00	70.00	72.50
	RAUS-5	126.75	129.50	132.00	11.00	11.00	11.50	59.5	62.00	66.00

Table 2: Morphological growth attributes of soybean varieties (BSS-2 and RAUS-5) under different microbial inoculants

Harvest	Varieties	Dry wt. of plant (mg)			Leaf area (cm^2)		
		Cont.	Az	Rz	Cont.	Az	Rz
1	BSS-2	390± 0.00	410± 0.01	440± 0.01	41.30± 12.73	44.95± 15.40	51.32± 24.95
	RAUS-5	490± 0.00	610± 0.10	660± 0.01	65.57± 9.20	70.41± 24.24	76.93± 15.26
2	BSS-2	1100± 0.36	1200± 0.07	1260± 0.17	81.58± 32.58	105.37± 9.20	125.29± 25.30
	RAUS-5	1370± 0.33	1630± 0.89	1740± 0.20	79.03± 12.35	102.84± 20.69	115.94± 18.18
3	BSS-2	3490± 7.26	3980± 4.66	4610± 7.16	372.61± 10.31	417.79± 9.30	468.97± 20.37
	RAUS-5	3530± 8.15	4570± 21.51	4750± 8.15	307.88± 16.83	355.65± 8.77	396.75± 8.06
4	BSS-2	6080± 11.73	6680± 7.16	7430± 13.11	620.08± 60.08	652.27± 12.73	702.26± 23.68
	RAUS-5	7080± 27.69	8.03± 12.47	8.02± 28.45	627.90± 8.87	698.93± 20.21	738.71± 8.56
5	BSS-2	9830± 18.33	11060± 27.88	11470± 31.98	1028.51± 9.09	1095.13± 26.76	1164.75± 20.21
	RAUS-5	9290± 26.76	10350± 24.95	11460± 43.09	1091.56± 24.95	1135.76±21.1 8	1187.27± 10.20
6	BSS-2	16360± 10.31	18390± 12.60	20240± 26.76	1385.95± 16.83	1495.55± 21.51	1560.86± 16.83
	RAUS-5	19080± 28.07	20490± 24.95	20750± 17.42	1452.15± 27.50	1520.15± 11.49	1599.84± 6.24

(2011) also reported growth promotion of soybean by plant growth promoting rhizobacteria (PGPR).

The relative growth rate (Table 3) was also higher in Rhizobium inoculated plants and minimum in non-inoculated plants. BSS-2 showed higher RGR than RAUS-5 in all the treatments. Rana *et al*. (2013) reported increased RGR of soybean with Rhizobium inoculation plants. Many workers like Singh and Pareek (2003) also found similar results.

The NAR values (Table 3) in both the varieties increased at the initial stage but after 3-4 harvest the average values decreased. In later stage higher NAR was reported in Rhizobium treated plants of BSS-2 and Azatobacter treated plants of RAUS-5 than control. However, Luthra *et al.* (1983) stated that total nodule nitrogen activity in pigeon pea increased with plant growth to reach maximum at flowering and decreased thereafter until maturity. Assimilates produced after flowering were exported to the reproductive structure at the expense of the nodules. It is suggested that decreased availability of photosynthate to nodules decrease nitrogen fixation.

Both varieties showed higher LAR in *Rhizobium* and *Azotobacter* treated plants than non-treated plants (Table 4). Starting from the higher LAR in both the varieties declined steadily with the increase of plant age. The data were found insignificant in all the harvest at 95% confidence level .Wallace and Munger (1965) also reported higher LAR during early vegetative stage but later it decreased rapidly with the advancement of age.

The highest value for SLA was found at the first stage of growth in both the varieties and in all the treatment. SLA was found to decrease uniformly (Table 4). BSS-2 showed higher SLA in *Rhizobium* inoculated plant. The overall SLA was higher in RAUS-5 than BSS-2. Investigation on Cowpea, Mungbean and Bengal gram showed that nodulation keeps

Table 3: Derived growth parameters of soybean varieties (BSS-2 and RAUS-5) between harvests under different microbial inoculants

Harvest	Varieties	RGR (mg/mg/week)			NAR (mg/cm²/week)		
		Cont.	Az	Rz	Cont.	Az	Rz
1 - 2	BSS-2	1.04	1.10	1.05	11.99	11.11	9.87
	RAUS-5	1.03	0.98	0.97	12.42	11.95	11.35
2 - 3	BSS-2	1.15	1.20	1.30	12.48	12.28	12.87
	RAUS-5	0.95	1.03	1.00	12.84	14.42	13.18
3 - 4	BSS-2	0.56	0.52	0.48	5.34	5.18	4.84
	RAUS-5	0.70	0.56	0.52	7.88	6.85	5.93
4 - 5	BSS-2	0.48	0.50	0.43	4.68	4.85	4.46
	RAUS-5	0.27	0.25	0.36	2.62	2.60	3.60
5 - 6	BSS-2	0.50	0.51	0.57	5.45	5.67	6.48
	RAUS-5	0.72	0.68	0.59	6.87	7.85	7.26

Table 4: Derived growth parameters of soybean varieties (BSS-2 and RAUS-5) under different microbial inoculants

Harvest	Varieties	LAR (cm²/mg)			SLA (cm²/mg)		
		Cont.	Az	Rz	Cont.	Az	Rz
1	BSS-2	0.11	0.11	0.12	0.30	0.32	0.34
	RAUS-5	0.13	0.12	0.12	0.50	0.50	0.48
2	BSS-2	0.07	0.09	0.09	0.20	0.26	0.30
	RAUS-5	.06	0.06	0.07	0.15	0.18	0.19
3	BSS-2	0.11	0.10	0.10	0.26	0.26	0.28
	RAUS-5	0.09	0.08	0.08	0.27	0.23	0.21
4	BSS-2	0.10	0.09	0.09	0.27	0.25	0.25
	RAUS-5	0.08	0.09	0.09	0.22	0.23	0.23
5	BSS-2	0.09	0.10	0.11	0.26	0.27	0.27
	RAUS-5	0.12	0.11	0.10	0.32	0.30	0.30
6	BSS-2	0.08	0.09	0.09	0.23	0.21	0.21
	RAUS-5	0.08	0.10	0.09	0.20	0.19	0.19

Table 5: Derived growth parameters of soybean varieties (BSS-2 and RAUS-5) under different microbial inoculants

Harvest	Varieties	LWR (mg/mg)			S/R ratio		
		Cont.	Az	Rz	Cont.	Az	Rz
1	BSS-2	0.35	0.34	0.34	1.18	1.08	1.07
	RAUS-5	0.27	0.23	0.24	2.00	2.46	2.25
2	BSS-2	0.35	0.36	0.37	2.18	1.82	1.83
	RAUS-5	0.39	0.34	0.34	1.47	1.51	1.46
3	BSS-2	0.38	0.39	0.40	1.27	1.17	1.02
	RAUS-5	0.33	0.35	0.40	1.43	1.43	1.35
4	BSS-2	0.39	0.40	0.42	1.04	1.10	1.11
	RAUS-5	0.40	0.40	0.43	1.19	1.32	1.38
5	BSS-2	0.40	0.43	0.40	1.64	1.34	1.34
	RAUS-5	0.37	0.38	0.40	1.90	1.89	1.71
6	BSS-2	0.40	0.42	0.42	2.04	1.78	1.57
	RAUS-5	0.38	0.39	0.40	2.02	1.93	1.80

Table 6: Effect of different microbial inoculants on reproductive growth attributes of soybean varieties (BSS-2 and RAUS-5)

Varieties	Days of Flowering primordia after sowing			No. of flowers/plant			No. of pods/ plant			No. of seeds / plants			Wt. of 100 Seeds (gm)		
	Con	Az	Rz	Con	Az	Rz	Con	Az	Rz	Con	Az	Rz	Con	Az	Rz
BSS-2	57	56	55	4	5	7	140	155	158	364	370	410	12.76	13.13	13.99
RAUS-5	55	55	54	5	6	8	154	156	160	401	407	418	13.54	13.97	14.23

pace with plant growth until flowering starts (Singh *et al*. 1978). The photosynthetic rates were higher in these crops at the seedling and pre-flowering stages. SLA values decreased with increasing plant dry weight was reported by Chanda *et al.* (1987).

The leaf weight ratio (Table 5) was maximum in *Rhizobium* treated plants in both the varieties and minimum in control . Almost similar results due to different factors were reported by Sivakumar *et al*. (1979) and Rabindranath *et al.* (1983) in different crops. Pandey *et al.* (1981) stated that senescence and abscission of the older leaves might cause the depletion of LAI at the later stages of growth.

From the data on S/R ratio (Table 5), it was evident that *Rhizobium* inoculated plants have higher S/R ratio than *Azotobacter* inoculated and non-inoculated plants. RAUS-5 shows better result than BSS-2 in all the treatments. The present finding is in line with those of Husen (2003) and Bhunia *et al.* (2006). The increase in root fresh weight in consequence of applying biofertilizers may be due to nitrogen fixation, increase the uptake of nutrients and released some growth regulators which stimulating establishment and vegetative growth, hence increasing root and foliage fresh weight. There are many investigators confirming this conclusion like Anjum *et al.* (2006) and Madhavan *et al. (*2012).

Floral initiation was noticed on day 56th, 55th and 57th in BSS-2 and on 55th, 54th and 55th days in RAUS-5 in *Azotobacter* inoculated, *Rhizobium* inoculated and non-inoculated plants respectively (Table 6). The early floral initiation was noticed in *Rhizobium* treated plants. The number of flowers per plant, number of pod per plant, number of seeds per plant and weight of 100 seeds were also higher in *Rhizobium* treated plants. The seed weight was higher in RAUS-5 than BSS-2 in *Rhizobium* treated plants. Biofertilizer inoculations improve days of flowering, pod/plant, seed yield over control which might be accorded to the better translocation of photosynthesis Kumravat *et al*. (1997). *Rhizobium* showed its superiority over *Azotobacter* inoculated and control plant. Annapurna *et al.* (1998) also stated that *Rhizobium japonicum* inoculation with the seeds is beneficial to increase the soybean yield. Many investigators like Govindan and Thirumurugan (2005), Gupta *et al* .(2006), Singh *et al*. (2007), Dhami *et al*.(2009) , Javaid and Mahmood (2010), Babaogli *et al*. (2012) and Rana *et al.* (2013) also reported similar findings.

On the basis of findings of the present work it was concluded that the RAUS-5 variety of soybean performed better than BSS-2 in morphological and reproductive growth attributes under various edapho-climatic stress conditions. In this respect *Rhizobium* inoculated plants showed its superiority over *Azotobacter* inoculated and control plant.

Author is thankful to UGC (ERO) , Kolkata , India for providing financial help for the research project on soybean and also grateful to Head , Department of Botany, T.N.B. College ,Bhagalpur and P.G. Department of Botany ,T.M .Bhagalpur University, Bhagalpur for their constant encouragement and providing laboratory facilities.

REFERENCES

Amit A , Nalawde and Satish A Bhalerao 2015 Comparative account of Effect of Biofertilizers on the growth and biochemical parameters of *Vigna mungo* (L.Hepper*). Int J Adv Res Biol Sci* **2(5):** 62–66

Anjum M S , Ahmed Z I , Rauf C A 2006 Effect of Rhizobium Inoculation and Nitrogen Fertilizer on Yield and Yield Components of Mungbean. *International Journal of Agriculture and Biology* **8:** 1560-8530

Annapurna K , Kama AK, Shende, Rasika and Balasundaram VR 1998 Plant growth promoting rhizobacteria: effects on growth and nitrogen fixation of soybean(*Glycine max*). 39th Annual Conference-Association of microbiologists of India, Manglore. Dec.5 to7, AM-13:250

Ardhapurkar N A and Manwar A V 2011 Study on direct mechanism of growth promotion of soybean. *Asian J Bio Sci* **6(2)** 191-193.

Babaogli F, Rahimzadeh Khoeai F and Yarnia M 2012 Effect of biological fertilizer on yield and yield components of corn (*Zea mays* L.) CV. S. C. 504 in drought condition. *J Appl Environ Biol Sci* **2 (3)** 117-122

Bhunia S R, Chauhan Yadav, B S and Bhati A S 2006 Effect of phosphorus, irrigation and rhizobium on productivity, water nutrient uptake in fenugreek (*Trigonella foenum-graecum*). *Indian Journal of Agronomy,* **51(3)** 239-241.

Chanda S V, Joshi A K , Vishnav P P and Singh Y D 1987 Growth analysis using classical and curve fitting methods in relation to productivity in pearl millet (*Penniselum americanum* L.). *J Agron Crop Sci* **159** 312-319.

Dixit S 2013 Impact of Bio-fertilization on morphological parameters of *Vigna mungo* (L.) Hepper: *International Journal of Research in Plant Science* **3(1)** 10-13

Dhami N and Prasad BN 2009 Increase in root nodulation and crop yield of soybean by native *Bradyrhizobium japonicum* strains. *J Plant Sci* **6** 1-3

Govindan K and Thirumurugan V 2005 Synergistic association of Rhizobium with phosphate-solubilising bacteria under different sources of nutrient supply on productivity and soil fertility in soybean (*Glycine max*). *Indian J of Agron* **50(3)** 214-217.

Gupta A, Sharma V K , Sharma G D and Chopra P 2006 Effect of biofertilizers and phosphorus levels on yield attributes, yield and quality of urdbean (*Vigna mungo*). *Indian J of Agron* **51(2)** 142-144

Husen Edi 2003 Screening of soil bacteria for plant growth promotion activities in vitro. *Indinesian J Agric Sci* **4(1)** 27-31.

Kader M A 2002 Effects of Azotobacter inoculants on the yield and nitrogen uptake by wheat. *Journal of Biological Science* **2** 259-261.

Kumrawat B , Dighe J M , Sharma R A and Katti G V 1997 Response of soybean to biofertilizers in black clay soils. *Crop Research* **14(2)** 209-214.

Luthra Y P , Sheoram I S and Singh R 1983 Ontogenic interaction between photosynthesis and symbiotic nitrogen fixation in *Pigeon pea Ann Appl Biol* **103** 549-556.

Madhavan S , Karnan M , Senthilkumar G , Kulothungan S , Panneerselvam A 2012 Effect of biofertilizers on morphological and physiological parameters of cowpea (*Vigna unguiculata). Advances in Applied Science Research* **3** 3269-3272.

Minaxi, Saxena J, Chandra S and Nain L 2013 Synergistic effect of phosphate solubilizing rhizobacteria and arbuscular mycorrhiza on growth and yield of wheat plants. *J Soil Sci Plant Nutr* **13** 511-525.

Rana M , Pathania P and Khaswan S L 2013 Effect of biofertilizers and phosphorus on productivity and nutrient uptake of soybean (*Glycine max L.*). *Ann. Agric. Res. New Series,* **34(3)** 245-247.

Pandey R P, Nair P K R, Tiwari J P 1981 Correlation of morphophysiological and sink parameters in cow pea. *Indian J Agric Sci* **51** 221-224

Rabindranath P, Shiv Raj A 1983 Effect of moisture stress on growth, yield and yield components of field-grown sorghum varieties having glossy and non-glossy leaves. *Indian J Agric Sci* **53** 428-430.

Sivakumar T , Ravikumar M , Prakash M and Thamizhmani R 2013 Comparative effect on bacterial biofertilizers on growth and yield of green gram (*Phaseolus radiata L.*) and cow pea (*Vigna siensis Edhl.*) *Int J Curr Res Aca Rev* **1(2*)*** 20-28

Singh S D 1978 Effect of Rhizobium inoculation on nodulation and yield of moong (*Vigna radiata* L. Wilczek). *Ann Arid Zone* **16** 79-84

Singh S R , Najar G R and Singh U 2007. Productivity and nutrient uptake of soybean (*Glycine max*) as influenced by bioinoculants and farmyard manure under rainfed conditions. *Indian Journal of Agronomy* **52(4)** 325-329.

Singh RP , Gupta SC and Yadav AS 2008 Effect of levels and sources of phosphorus and PSB on growth and yield of blackgram (*Vigna mungo* L. Hepper). *Legume Research* **31(2)** 139-141.

Singh B and Pareek R G 2003 Studies on phosphorus and bio-inoculants on biological nitrogen fixation, concentration, uptake, quality and productivity of mungbean. *Annals of Agricultural Research* **24** 537-541.

Tilak KVBR , Ranganayaki N, Pal KK, De R, Saxena AK, Nautiyal CS, Mittal S, Tripathi AK, Johri BN 2005. Diversity of plant growth and soil health supporting bacteria. *Current Science* **89 (Suppl 1)** 136-150.

Venkatech A , Vanagamudi M , Vanangamudi K, Ravichandran V and Rai RSV 1998 Impact of biofertilizers on morphophysiological attributes in pungam *(Pongamia pinnata*) seedling. *Trop, Agric.Res. Extension*. **1** 7-11.

Vessey K 2003 Plant growth promoting rhizobacteria as biofertilizers. *Plant Soil* **255** 571-586.

Wallace D M and Munger H M 1965 Studies on the physiological basis of yield differences. I. Growth analysis of six dry bean varieties. *Crop Sci* **5** 343-348.