

International Journal of Current Microbiology and Applied Sciences ISSN: 2319-7706 Volume 12 Number 11 (2023) Journal homepage: <u>http://www.ijcmas.com</u>



## **Original Research Article**

https://doi.org/10.20546/ijcmas.2023.1211.006

# Field Evaluation of Bacterial Consortia on Growth, Yield and Quality parameters of Soybean (*Glycine max*)

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# ABSTRACT

#### Keywords

Soybean, Bio fertilizer, phosphate solubilizing, Bacillus, nodulation

Article Info

Received: 15 September 2023 Accepted: 22 October 2023 Available Online: 10 November 2023

# Introduction

Soybean is known as the golden bean now a day and emerged as an important commercial oilseed crop. Nutrient management is important in soybean productivity. Application of bio-fertilizers along with manures and balanced inorganic fertilizers into the soil leads to improvement in productivity of the crop and sustain the soil health management (Manna *et al.*, 2007). Rhizobia is symbiotic nitrogen fixing

The use of bio fertilizers is important for sustainable agriculture and the use of nodule bacteria, p-solubilizing bacteria, K-mobilizing bacteria and PGPR is an appropriate way to enhance germination percentage, plant growth and yield. This study tested the effects of a bacterial consortia (Bio-Release KONCERT) containing Rhizobium sp, Bradyrhizobium sp, Bacillus sp. and Pseudomonas sp. for the combined effect on Soybean crop. Experiment designed for seed treatment of soybean during kharif 2022-2023, to check the efficacy of consortia product at different doses (5, 10, 15, and 20 ml/kg of seeds) with RDF, local strain as check and control for comparing the data. Seed treatment observed uniform in all the treated seeds with different range of doses. Treating seeds with Bio-release KONCERT @ 20 ml kg<sup>-1</sup> of seeds was found superior in enhancing the soybean growth parameters, soil biological activity in terms of soil enzymes, physiological parameters and yield attributes. This is found to be at par with Biorelease KONCERT @ 15 ml kg<sup>-1</sup> of seeds and Bio-release KONCERT @ 10 ml kg<sup>-1</sup> seeds. Compared with the non-inoculated seeds, application of Bio-release KONCERT observed superior results in all concentrations. This bio fertilizer which significantly promoted plant growth, nodulation, nitrogen fixation, nutrient uptakes, and seed yield in soybean. Use of bio fertilizers can be reducing the application of chemical fertilizer in soybean production.

> bacteria with legumes (Subba Rao, 1982). As most of our soils are deficient in organic matter, give poor response to fertilizers. Under this conditions inoculation with microorganism like PSB, KMB and PGPR can be advantageous. The application of beneficial microbes need to decide established to a high level of effectiveness.

> Nitrogen is important for improving crop growth and yield. Excessive use of chemical fertilizer

affects the environment (Dacko *et al.*, 2016), including soil fertilizer degradation, soil organic matter deterioration, decrease the nutrient holding capacities and nutrients use efficiency. (Baligar *et al.*, 2001; He *et al.*, 2005). Efficient rhizobia N<sub>2</sub>fixing bacteria alone or together inoculants are improving leguminous crop production and reduction in environmental problems by decreasing the use of chemical fertilizers (Alves *et al.*, 2004).

Plant growth-promoting bacteria (PGPB) enhance plant growth through the availability of nutrients originating from processes such as biological nitrogen fixation, phosphate solubilisation and potassium mobilization. Microbes are producing phytohormones, enzymes, organic acids and siderophores (Souza *et al.*, 2015; Sinha *et al.*, 2010).

A bio fertilizer containing living microorganisms when applied to seeds or soil, colonize the rhizosphere or interior of the plant and promote growth by increasing the supply or availability of nutrients to the host plant (Vessey, 2003).

Bio fertilizers are important for integrated nutrient management play key roles in the productivity, sustainability of soil are cost-effective and ecofriendly.

Considering the importance of soybean in production, nutritional importance and status of biological fertilizers in sustainable agriculture, studies were planned to see the impact of bio fertilizer on growth, yield and yield components of soybean. Combination of  $N_2$ -fixing bacteria, PSB, KMB and PGPR bacteria which help to improve the productivity and soil health management were used for pre sowing seed treatment.

# **Materials and Methods**

A field experiment was conducted during *kharif* 2022 in deep black soil at Block E, Plot No. 118 of the MARS, UAS, DHARWAD. The place is situated at 15°24' 41.8 N latitude and 74°59' 13.1E longitude with an altitude of 697 meters above the

mean sea level. The experimental plot soil was clay, deep black soil (pH 7.33), low in available N (224 kg ha<sup>-1</sup>), medium in available P (35 kg ha<sup>-1</sup>) and medium in available K (274 kg ha<sup>-1</sup>). The organic C content in soil was 0.68 %.The experiment consisted of six treatments namely T<sub>1</sub>-Bio release KONCERT 5 ml/kg<sup>-1</sup> of seeds, T<sub>2</sub>– Bio release KONCERT 10 ml/kg<sup>-1</sup> of seeds, T<sub>3</sub>- Bio release KONCERT 15 ml/kg<sup>-1</sup> of seeds, T<sub>4</sub>- Bio release KONCERT 20 ml/kg<sup>-1</sup> of seeds, T<sub>5</sub>-Local *Bradyrhizobium* sp, (as per POP)and T<sub>6</sub>- Untreated control and nutrients applied as per recommended dose of NPK for soybean was used as per package of practices.

# Soil Biological activities of experimental site

Microbial biomass carbon (186  $\mu$ g g<sup>-1</sup>of soil), Dehydrogenase activity (12.71  $\mu$ g TPF formed g<sup>-1</sup> soil d<sup>-1</sup>), Phosphatase activity (19.34  $\mu$ g pnp released g<sup>-1</sup> soil h<sup>-1</sup>), Urease activity (7.47  $\mu$ g NH<sub>4</sub><sup>+</sup>-N g<sup>-1</sup> soil day<sup>1</sup>), Free living N<sub>2</sub> fixing bacteria 2.7 X 10<sup>4</sup> CFU g<sup>-1</sup> of soil, P- solubilizing bacteria 1.6 X 10<sup>3</sup>CFU g<sup>-1</sup> of soil and Fluorescent *Pseudomonads* 0.6 X 10<sup>3</sup> CFU g<sup>-1</sup> of soil.

# **Results and Discussion**

# Growth parameters

Plant height (cm) was measured from the base to the tip of the plant on 30 and 60 days after sowing in each treatment in five randomly selected plants. The observation recorded on plant height indicated that the treatment with Bio-release KONCERT 20 ml kg<sup>-1</sup> of seeds improved the plant height (20.49 cm), followed by Bio-release KONCERT 15 ml kg<sup>-1</sup> of seeds (19.74 cm), Bio-release KONCERT 10 ml kg<sup>-1</sup> of seeds (19.26 cm).

However, the results were statistically at par with each with the treatment received Bio-release KONCERT @ 10, 15 and 20 ml kg<sup>-1</sup> of seeds. Compared with local *Brady rhizobium* strain (18.00 cm) and Un-inoculated seeds treatments (13.76 cm). Similar trend of results was also recorded at 60 DAS (Table 1).

#### Shoot and Root length (cm)

At 30 DAS, the maximum root length was recorded with Bio-release KONCERT 20 ml kg<sup>-1</sup> of seeds, followed by Bio-release KONCERT 15 ml kg<sup>-1</sup> of seeds (17.04 and 16.87 cm, respectively). At 30 DAS, the highest shoot length was recorded with Bio-release KONCERT 20ml kg<sup>-1</sup> of seeds, followed by Bio-release KONCERT 15 ml kg<sup>-1</sup> of seeds (14.23 and 13.12 cm, respectively).

## Number of Branches

The results at 60 DAS have revealed that the highest branches were observed in the plants received Biorelease KONCERT 20 ml kg<sup>-1</sup> of seeds (24.00) while the second highest was with Biorelease KONCERT 15 ml kg<sup>-1</sup> of seeds (23.70), however the results were at on par with each other.

Stefan *et al.*, (2010) stated that inoculation of soybean by *Bacillus pumilus* significantly increased plant height, leaf number, leaf area, grain protein and nodulation. The seed treatment with KONCERT green indicated similar impact on growth of plants.

#### Soil enzyme activity

The results pertaining to the soil enzyme activity viz., dehydrogenase, phosphatase and urease activity are presented in Table 2. The observation recorded on soil dehydrogenase activity at flowering stage in the soybean rhizosphere revealed that the highest dehydrogenase activity was recorded in the soybean inoculated with Bio-release KONCERT 20 ml kg<sup>-1</sup> of seeds (24.60  $\mu$ g TPF formed g<sup>-1</sup> soil d<sup>-1</sup>), followed by Bio-release KONCERT 15 ml kg<sup>-1</sup> of seeds (24.52  $\mu$ g TPF formed g<sup>-1</sup> soil d<sup>-1</sup>) and Bio-release KONCERT 10 ml kg<sup>-1</sup> of seeds (23.81  $\mu$ g TPF formed g<sup>-1</sup> soil d<sup>-1</sup>), however, the results obtained were statistically on par with each other. The lowest dehydrogenase activity was recorded with uninoculated control (13.91  $\mu$ g TPF formed g<sup>-1</sup> soil d<sup>-1</sup>). The phosphatase activity at flowering stage

was recorded with Bio-release KONCERT 20 ml kg<sup>-1</sup> of seeds  $\mu$ g (31.88  $\mu$ g pnp released g<sup>-1</sup> soil h<sup>-1</sup>), followed by Bio-release KONCERT 15 ml kg<sup>-1</sup> of seeds (31.17  $\mu$ g pnp released g<sup>-1</sup> soil h<sup>-1</sup>) and Bio-release KONCERT 10 ml kg<sup>-1</sup> of seeds (30.95  $\mu$ g pnp released g<sup>-1</sup> soil h<sup>-1</sup>), the lowest dehydrogenase activity was recorded with un inoculated control (22.02  $\mu$ g pnp released g<sup>-1</sup> soil h<sup>-1</sup>).

The maximum urease activity was observed for the seed treatment with Bio-release KONCERT 20 ml kg<sup>-1</sup>which recorded the highest urease activity (12.89  $\mu$ g NH<sub>4</sub> + N g<sup>-1</sup>soil day<sup>-1</sup>), followed by Bio-release KONCERT 15 ml kg<sup>-1</sup> of seeds (12.36  $\mu$ g NH<sub>4</sub> + N g<sup>-1</sup> soil day<sup>-1</sup>).

The enzyme activity was observed with the un inoculated control soybean rhizosphere (6.29  $\mu$ g NH<sub>4</sub> + N g<sup>-1</sup>soil day<sup>-1</sup>). The result thus indicate that soil enzyme activity is increased by use of Biorelease KONCERT.

#### **Nodule parameters**

The nodule parameters were recorded at flowering stage, nodule number, nodule dry weight and leg haemoglobin content was highest in the treatment Bio-release KONCERT 20 ml kg<sup>-1</sup> (23.07, 1.29 g/plant and 0.66mgg<sup>-1</sup> FW nodules, respectively) followed by Bio-release KONCERT 15 ml kg<sup>-1</sup> of seeds (23.06, 1.02 g/plant and 0.61mgg<sup>-1</sup> FW nodules, respectively) and Bio-release KONCERT 10 ml kg<sup>-1</sup> of seeds (23.00,1.04 g/plant and 0.61 mgg<sup>-1</sup> FW nodules, respectively), compared to un inoculated the control (14.04, 0.60 g/plant and 0.39 mgg<sup>-1</sup> FW nodules, respectively), it was also better over local *Bradyrhizobium* sp.,(Table:3).

Results were concominent with Zhang *et al.*, (2002)who reported that *B. japonicum* bacteria increased number of pods per plant, number of seeds per plant, hundred seed weight, grain protein, total protein and development of plant leaves in two soybean cultivars.

Tr No	Treatment	Plant height (cm)		Number of branchesRoot length (cm)		Shoot length (cm)		
		30 DAS	60 DAS	60 DAS	30 DAS	60 DAS	30 DAS	60 DAS
<b>T</b> <sub>1</sub>	Bio-release KONCERT (5 ml)	17.00	40.30	20.13	14.36	20.13	9.57	26.06
$T_2$	Bio-release KONCERT (10 ml)	19.26	49.67	22.60	16.64	23.50	12.80	32.22
<b>T</b> <sub>3</sub>	Bio-release KONCERT (15 ml)	19.74	49.88	23.70	16.87	23.80	13.12	32.30
$T_4$	Bio-release KONCERT (20 ml)	20.49	49.27	24.00	17.04	24.00	13.23	32.65
<b>T</b> <sub>5</sub>	Local <i>Bradyrhizobium</i> sp (as per POP)	18.00	42.82	21.19	15.37	21.19	9.82	27.54
T <sub>6</sub>	Control (Untreated Seeds)	13.76	37.79	18.63	12.35	18.63	7.56	23.84
S.EM±		0.45	0.23	0.62	0.22	0.27	0.38	0.21
CD (5%)		1.41	0.68	1.96	0.66	0.81	1.15	0.63

Table.1 Growth parameters of soybean as influenced by Bio-release KONCERT at varied level

Table.2 Soil enzyme activity as influenced by Bio-release KONCERT in the soybean rhizosphere.

Tr	Treatment	Soil enzymes (At flowering stage)			
No		Dehydrogenase (µg TPF formed g <sup>-1</sup> soil d <sup>-1</sup> )	Phosphatase (µg pnp released g <sup>-1</sup> soil h <sup>-1</sup> )	Urease (µg NH4 + N g <sup>-1</sup> soil day <sup>-1</sup> )	
<b>T</b> <sub>1</sub>	Bio-release KONCERT (5 ml)	19.22	26.85	9.84	
$T_2$	Bio-release KONCERT (10 ml)	23.81	30.95	12.12	
<b>T</b> <sub>3</sub>	Bio-release KONCERT (15 ml)	24.52	31.17	12.36	
<b>T</b> <sub>4</sub>	Bio-release KONCERT (20 ml)	24.60	31.18	12.44	
<b>T</b> <sub>5</sub>	Local <i>Bradyrhizobium</i> sp (as per POP)	23.00	29.20	11.31	
<b>T</b> <sub>6</sub>	Control (Untreated Seeds)	13.91	22.02	6.29	
S.EM±		0.32	0.16	0.14	
<b>C.D</b> (5%)		0.96	0.49	0.41	

# Table.3 Nodule parameters as influenced by Bio-release KONCERT in the soybean

Tr No	Treatment	Nodule number	Nodule dry weight (g plant <sup>-1</sup> )	Leg haemoglobin content (mgg <sup>-1</sup> FW nodules)
<b>T</b> <sub>1</sub>	Bio-release KONCERT (5 ml)	18.05	0.68	0.52
$T_2$	Bio-release KONCERT (10 ml)	23.00	1.04	0.61
<b>T</b> <sub>3</sub>	Bio-release KONCERT (15ml)	23.06	1.02	0.61
T <sub>4</sub>	Bio-release KONCERT (20 ml)	23.07	1.09	0.63
<b>T</b> 5	Local Bradyrhizobium sp (as per POP)	22.05	0.62	0.56
<b>T</b> <sub>6</sub>	Control (Untreated Seeds)	14.04	0.60	0.39
	S.EM ±	0.08	0.02	0.01
	<b>C.D</b> (5 %)	0.23	0.08	0.04

Tr No	Details	Relative chlorophyll content (SPAD Value)		
			60 DAS	
<b>T</b> <sub>1</sub>	Bio-release KONCERT (5 ml)	39.54	37.03	
<b>T</b> <sub>2</sub>	Bio-release KONCERT (10 ml)	41.28	42.23	
<b>T</b> <sub>3</sub>	Bio-release KONCERT (15 ml)	41.29	42.54	
<b>T</b> <sub>4</sub>	Bio-release KONCERT (20 ml)	41.35	42.58	
<b>T</b> 5	Local Bradyrhizobium sp (as per POP)	40.77	37.59	
T <sub>6</sub>	Control (Untreated Seeds)	27.58	29.95	
S.EM±		0.15	0.21	
	C.D (5 %)	0.46	0.63	

# Table.4 Relative chlorophyll content as influenced by Bio-release KONCERT in the soybean

# Table.5 Plant dry biomass as influenced by Bio-release KONCERT in the soybean at harvest stage

Tr No	Treatments	Plant dry biomass at Harvest stage (g plant <sup>-1</sup> )			
		Shoot dry biomass Root dry biomass		Total dry biomass	
<b>T</b> <sub>1</sub>	Bio-release KONCERT (5 ml)	10.54	2.65	13.19	
<b>T</b> <sub>2</sub>	Bio-release KONCERT (10 ml)	12.77	3.55	16.32	
<b>T</b> <sub>3</sub>	Bio-release KONCERT (15 ml)	12.67	3.60	16.27	
<b>T</b> 4	Bio-release KONCERT (20 ml)	12.80	3.83	16.63	
<b>T</b> 5	Local <i>Bradyrhizobium</i> sp (as per POP)	10.61	2.51	13.12	
T <sub>6</sub>	Control (Untreated Seeds)	9.60	2.03	11.63	
S.EM±		0.08	0.11	0.24	
<b>C.D</b> (5 %)		0.26	0.34	0.71	

Table.6 Yield and yield attributes as influenced by Bio-release KONCERT in the soybean at harvest stage

Tr No	Treatments	1000 seed weight (g)	No of pods (per plants)		Yield (Kg/ha)
			<b>60 DAS</b>	At Harvest	At Harvest
<b>T</b> <sub>1</sub>	Bio-release KONCERT (5 ml)	117.14	13.67	15.00	1038.00
$T_2$	Bio-release KONCERT (10 ml)	123.99	19.18	20.32	1224.00
<b>T</b> <sub>3</sub>	Bio-release KONCERT (15 ml)	125.04	19.33	20.63	1238.00
<b>T</b> 4	Bio-release KONCERT (20 ml)	127.61	20.67	21.66	1254.68
<b>T</b> 5	Local Bradyrhizobium sp (as per POP)	122.13	14.36	18.68	1078.00
T <sub>6</sub>	Control (Untreated Seeds)	102.21	11.34	13.62	995.13
S.EM±		1.46	0.63	0.69	5.22
CD (5%)		4.60	1.98	2.18	17.97

#### **Relative Chlorophyll content (SPAD values)**

The relative chlorophyll content was recorded on 30 and 60 DAS are presented in (Table 4). Highest relative chlorophyll content was recorded with Biorelease KONCERT 20 ml kg<sup>-1</sup> at 30 and 60 DAS (41.35 and 42.58 respectively) followed by Biorelease KONCERT 15 ml kg<sup>-1</sup> seeds (41.29 and 42.54 respectively) and Bio-release KONCERT 10 ml kg<sup>-1</sup> of seeds (41.28 and 42.23 respectively), while lowest relative chlorophyll content was recorded in the un inoculated control (27.58 and 29.95 respectively). The Bio-release KONCERT observed improvement in relative chlorophyll over local treatment of *Bradyrhizobium* sp.

## Plant dry biomass (g plant<sup>-1</sup>)

The highest shoot dry biomass, root biomass and total dry biomass was recorded with the treatment Bio-release KONCERT 20 ml kg<sup>-1</sup> (12.80, 3.83 and 16.63 gm/plant respectively), followed by Bio-release KONCERT 15 ml kg<sup>-1</sup> of seeds (12.67, 3.60 and 16.27 gm/plant respectively) and Bio-release KONCERT 10 ml kg<sup>-1</sup> of seeds (12.77, 3.55 and 16.32 gm/plant respectively), compared with the Local *Bradyrhizobium* sp., (10.61,2.51 and 13.12 gm/plant respectively) and un inoculated control (9.60, 2.03 and 11.63 gm/plant respectively) (Table 5). Olivera *et al.*, (2004) had also reported the positive effect of combined inoculation of bean by phosphate solubilizing bacteria and *Bradyrhizobium japonicum* bacteria on dry weight.

#### Yield and yield attributes as influenced by Biorelease KONCERT in Soybean

The influence of Bio-release KONCERT on soybean yield and yield parameters are presented in Table 6.The highest 1000 seed weight, number pods and seed yield kg ha<sup>-1</sup> was found in the treatment Bio-release KONCERT @ 20 ml kg<sup>-1</sup> of seeds compared to rest of the treatments and followed by Bio-release KONCERT @ 15 ml kg<sup>-1</sup> of seeds which is at par with Bio-release KONCERT @ 10 ml kg<sup>-1</sup> of seeds and better compared with local strain and un

inoculated control treatments. Kazemi *et al.*, (2005) reported that soybean seed inoculation by rhizobia bacteria significantly increased the number of pods per plant, number of seeds per plant, thousand grain weights and finally the yield of soybean. Rosas *et al.*, (2002) reported that combined inoculation of soybean by symbiotic bacteria of soybean and phosphate solubilizing bacteria improved dry weight of soybean. These similar trend is recorded with seed treatment with Bio-release KONCERT.

The study was conducted to see the impact of seed treatment of bio-fertilizer Bio-release KONCERT at different doses used to seed treatment on soybean growth and yield as well as soil health. Bio fertilizer enhances the nodulation and nitrogen uptake of plants, P-solubilizing bacteria improved the uptake of phosphorus and K-mobilizing bacteria helped to improve uptake of potassium in soil which increased the growth and yield of soybean. The results indicated that seed treatment is key component in soybean productivity to improve the germination and nutrient management. In this study the efficacy of Bio-release KONCERT was studied and observed significantly higher plant growth nodulation, enzyme activity yields and yield attributes over untreated seeds.

#### Acknowledgement

Financial assistance for testing the product provided by AGRI SEARCH (INDIA) PVT. LTD. Nasik, Maharashtra, INDIA. No. DR/T-3/ Agri Search/24/2022-23/Dtd:12-04-2022.

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# How to cite this article:

Basavesha, K. N., Satish Bhonde and Jones P. Nirmalnath. 2023. Field Evaluation of Bacterial Consortia on Growth, Yield and Quality parameters of Soybean (*Glycine max*). *Int.J.Curr.Microbiol.App.Sci.* 12(11): 55-61. doi: https://doi.org/10.20546/ijcmas.2023.1211.006