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RESPONSE OF SOYBEAN (*Glycinemax*L. Merrill) PRODUCTIVITY TO BIOFERTILIZER INOCULANTS COMBINED WITH ORGANIC FERTILIZERUNDER SANDY SOIL CONDITIONS

Abo El-Soud, A.A.;Rabab A.A.Amer; Hebe B.A.Kandil; Amani M. Ahmed;Gihan M. Salem and Sonya H. Mohamed*

Department of Agricultural Microbiology; Soils, Water and Environmental Research Institute; Agricultural Research Center; Giza; Egypt. P.O. Box, 12619
*Corresponding author: Mohamed Sonya H. (E-mail: drsonyasweri@gmail.com)

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ABSTRACT

One of the most important food sources for humans is the soybean crop because it is an important source of protein and oil, which makes it important to improve its productivity under stress conditions, especially in sandy lands. Therefore, the study aimed to improve soybean productivity by using biological fertilizers in the presence of organic fertilizer and low concentration of mineral fertilizer under new sandy soil conditions.The experiment was carried out in two summer seasons of 2020 and 2021 at the Ismailia Research Station, Ismailia Governorate. The used inoculates in the experiment were,an identified *Streptomyces* strain,*Bacillus megatherium*, (SEWRI) and *Bradyrhizobiumjaponicum* (Okaden) as,biofertilizer agents.Activated dose of N-fertilizer was used in seven different treatments. Results showed that Bradyrhizobia+*Bacillus*+actinomycetes+20 kg nitrogen fertilizer/fed was effective in the presence of organic fertilizer compared to organic fertilizer-free soil or un-inoculated+70 kg N/fed. Bradyrhizobia+ *Bacillus*+Actino+20 kg N/fed (T14) was the most effective treatment in the presence of organic fertilizer compared to organic fertilizer-free soil or uninoc.+70 kg N/fed. Promising expected yields of each of seeds and straw (Ton/fed) of soybean plants cultivated under different fertilizers and biofertilizers conditions expected was clear from increasing the yield seed up to 1.8400Ton/fed in the presence of organic fertilizer (T14) compared to 1.4565 when soybean was cultivated in the absence of organic fertilizer (T01). This could be supported by the amounts of NPK, N-percentage, and protein content, numbers of branches and pods and dehydrogenase activity in soil. Significant differences between the straw yield in the presence and absences of organic fertilizer as well as biofertilizers were recorded. Organic fertilizer application recorded the highest significant increases in nodulation status, growth and straw nitrogen content as well as harvest parameter of soybean compared with those un-received organic fertilizer.

Key words: Soybean, SEWRI and Okaden, *Bradyrhizobiumjaponicum*, Actinomycetes,*Bacillus megatherium*, Organic fertilizer, dehydrogenase activity, Crude protein percentage.

INTRODUCTION

The cultivation of soybeans in Egypt began in 1970 with an area not exceeding 3000 Faddenand an average production of 3000 kg per Fadden. An area of 15, 233 ha producing 43 342 tons with an average seed yield of 2.84 tons ha^{-1} was cultivated in Egypt (**FAO, 2020**).Soybean (*Glycine max* L. Merrill), originated from eastern Asia was domesticated~ 4000 years ago, and has been cultivated ever since.Soybean cv. Giza 111 is a hybrid cultivar, resistant to the cotton leaf worm, ripens after about 115-120 days of planting, and its cultivation is successful in all Governorates of the Republic up to the New Valley, and new soils in the south of the valley, and it is not recommended to delay its cultivation as for the end of May. Its productivity ranges between 1.5-1.7 tons per Feddan in the old soils and 1.2-1.4 tons / Feddan in the new soils (**AbouElkhair et al., 2014)**.

Biofertilizers are microorganisms that enrich the nutrient quality of soil. The main sources of biofertilizers are bacteria, fungi, and cyanobacteria (blue-green algae) (**Chittora et al., 2020**). The most striking relationship that these have with plants is symbiosis, in which the partners derive benefits from each other (**Al Abboud et al., 2014**). Biofertilizers are products applied on the surface of a plant, seedsor in soil and contain live microorganisms that promote plant growth and development. These products may include bacterial species such as *Rhizobium*, *Azotobacter*, and *Azospirilium* as well as blue green algae (BGA) (**Kumar et al., 2017** and**Noufal et al., 2018).** Biofertilizers such as *Rhizobium*, *Azotobacter*, *Azospirilium* and blue green algae (BGA) have been in use a long time. Rhizobium inoculant is used for leguminous crops. *Azotobacter* can be used with crops like wheat, maize, mustard, cotton, potato and other vegetable crops (**Kumari et al., 2017**and**Kumari et al., 2019)**.

The need for N fertilizers could be reduced by biological nitrogen fixation (**Nicolás et al., 2006**) in other mean bio-fertizers (**Ewees and Abdel Hafeez, 2010**and **Al Abboud et al., 2014**). An essential role in crop establishment and yield, was reported as a result of using biological nitrogen fixation, where N fertilizer was not apply, and it save the most needed nitrogen of plants (**Chen, 2006**).**)** An increase in germination of seeds appears as a direct result to improving soil productivity by adding plant growth-promoting rhizobacteria (PGPR) which considered as a group of free-living bacteria that colonize the rhizosphere and benefit the root growth (**Oad et al., 2004)**. Nitrogen fixation and plant growth promotion by rhizobacteria are important criteria for an effective biofertilizer**(Kumari et al., 2019**).

Ștefuanescu and Palanciuc (2000) found greater seed yield of soybean crop due to *Rhizobium japonicum* inoculation. The amount of required nitrogen of this plant is fulfilled by a establishing a N_2-fixing nodule symbiosis with rhizobia, which its inoculation plays an important role in yielding enhancement of soybean plants.

Tran et al., (2001) found that the nutrient contents of soybean plants, mainly N, P and K as well as soil available P and K were significantly improved by the application of bio-fertilizer (*Rhizobium fredii*and*Bradyrhrizobium* sp.). Rhizobium seed inoculation alone significantly increased soil nitrogen content and soil available phosphorus compared to the control in both seasons (**Hatim, 2013**).

AbouElkhair et al., (2014) conducted two field experiments during summer seasons of 2012 and 2013, at Seds Agric. Res. Station in BeniSewif Governorate to study the effect of some bio-fertilizers as PGPRs combined with mineral N, P and K fertilizer on the soybean cultivars (Giza cvs. 21, 35 and 111) productivity. A number of five microbial inoculants, combined with 1/3 mineral N and 1/2 P fertilizers were experimented randomized complete block design with three replications was applied. Results recommend using bio-fertilizers and reduce the mineral fertilizers and cultivation soybean Giza 111 and 35 under the same conditions.

Liu et al., (2020) investigated soil water use efficiency and crop yield of millet and soybean under nine fertilization regimes in the Loess Plateau, China. Combined N and P fertilization resulted in the greatest increase in crop yield and WUE, followed by the single P fertilizer application, and single N fertilizer application. The control treatment, which consisted of neither P nor N fertilizer application, had the least effect on crop yield. The combined N and P fertilization increased soil organic fertilizer(SOM) and soil total N, while soil water consumption increased in all treatments. SOM and total N content increased significantly when compared to the control conditions, by 27.1–81.3%, and 301.3–669.2%, respectively, only under combined N and P application. The experiment aimed at imp-roving soybean productivity by using biofertilizers in the presence as well as absences of organic fertilizerunder the conditions of new sandy soils.

MATERIALS AND METHODS

In this studytwo consequence experiments were carried out in two summer seasons of 2020 and 2021 at the Ismailia Research Station (Section No. 9), Ismailia Governorate,Egypt.

Source of soybean seeds: Soybean seeds (cv. Giza 111) were kindly obtained from Field Crops Institute, (ARC), Giza, Egypt. Seeds were sown at 25 cm distance between plants and 50 cm between rows as recommended by **AbouElkhair et al., (2014)**.

Soil sampling: Representative soil samples were collected from, the 30 cm top layer of the experimental field sieved through two mm screen and air-dried. The main physical and chemical properties of the soil among the two cultivated seasons were conducted as described by **Page et al., (1982)** and **Cottenie et al., (1982)**and recorded in **Table-1**.

Table-1: Physical and chemical analyses of cultivated soil among the two growing seasons.

Parameters		1st Season	2nd Season
Sand	(%)	40.4	39.7
Find sand	(%)	42.8	41.9
Silt	(%)	11.5	12.6
Clay	(%)	5.30	5.80
Textures		Sandy	Sandy
pH	(1:2.5)	8.04	8.14
E.C.	(dSm^{-1} at 25ºC)	0.52	0.52

SP (%)	27.0	28.0
Soluble cations (mmol/L)		
Ca^{2+}	1.50	1.40
Mg^{2+}	0.50	0.50
Na^{+}	0.22	0.21
K^{+}	2.95	2.92
Soluble anions (mmol/L)		
CO_3^{2-}	00.0	00.0
HCO_3^{-}	0.50	0.47
Cl^{-}	3.50	3.38
SO_4^{2-}	1.17	1.18
Nitrogen (%)	0.018	0.019
Total soluble-N (ppm)	97.00	97.50
Available-P (ppm)	08.38	8.950
Available-K (ppm)	95.00	94.90
DTPA-extract		
Fe (ppm)	1.22	1.52
Mn (ppm)	0.36	0.39
Zn (ppm)	0.12	0.13
Cu (ppm)	0.05	0.07

DTPA: Di-ethylene tri-amine penta acetic acid.

Organic fertilizer: Animal organic fertilizer (farm-yard manner) collected from Ismailia Research Station, Ismailia Governorate, Egypt, with the characteristics shown in **Table-2**was added to seven treatments in a trail to determine its role within the combination in improving the productivity of soybeans in sandy soils among the two growing seasons. Similar seven treatments without it were also used as control.

Table-2: Properties of farmyard manure conditioner used in the two experiment seasons.

Properties	First season	Second season
pH	7.35	7.28
E.C. (dS/m at 25ºC)	4.26	4.48
Organic-C (%)	15.25	16.84
Total N (%)	1.34	1.27
C/N ratio	11.83	13.26
Total-P (%)	0.64	0.95
Total-K (%)	1.35	1.30
Total soluble-N (ppm)	92.5	84.6
Available-P (ppm)	18.3	21.5
Available-K (ppm)	645.0	682.5
DTPA extractable (ppm):		
Fe	135.5	142.8
Mn	32.9	30.80
Zn	21.8	28.40
Cu	2.35	2.95
Total count of bacteria	$6.0X10^7$	$9.2X10^7$
Total count of bacteria	$5.4X10^6$	$2.6X10^6$
Total count of actinomycetes	$3.8X10^6$	$2.7X10^6$
Dehydrogenase activity (µg TPF/g)	108.4	113.8

Microbial total count: The total counts of microbes, *i.e.*, bacteria, fungi and actinomycetes in the cultivated soils treated with and without organic fertilizer were determined before and after cultivation following the protocol of **Clark (1965).**

Determination of NPK contents: Content of NPK in each of uncultivated and cultivated soils with or without organic fertilizer was determined before and after cultivation according to the method of **Attanandana et al., (1999)**. From each sample, 0.5 g was digested using mixture of sulfuric (H_2SO_4) and perchloric ($HClO_4$) acids (1:3, v:v) as described by **Cottenie et al., (1982)**. Nitrogen was determined by micro Keldahl, according to **Jackson (2005).** Phosphorus was determined by spectrophotometer using Ammonium Molybdate/Stannus Chloride method described by **Chapman and Pratt (1962)**. Potassium was determined by a flame photometer at wave length of 766.5 nmaccording to **Page et al., (1982).**

Dehydrogenase activity in soil: Soil activity was tested by determination of dehydrogenase activity (DHA) in cultivated soils with and without organic fertilizer before and after cultivation based on the protocol of **Stevenson (1959).**

Source and preparation of biofertilizer inoculants: Inoculants were prepared in Biofertilizer Unit; Soils, Water and Environment, Res. Inst., ARC, Giza, Egypt. Okaden inoculant contains *Bradyrhizobium japonicum* (ARC201 local isolate) was used for treating soybean seeds. Strain was grown on yeast extract mannitol broth (**Vincent and Humphrey 1970**), incubated at 28±C for five days until early log phase ($5x10^9$cfu/mL culture). SWERI inoculant contains *Bacillus megatherium* was also used after grown on nutrient broth (**Power and Johnson 2009**) under shaking condition (250 rpm) for 48 hr at 28±1ºC, till its populations reached of 5.5 $x10^9$cfu/mL culture. Vermiculite supplemented with 10% Irsh peat was packed in polyethylene bags (180 g carrier per bag), then sealed and sterilized by gamma irradiation ($5.0C10^6$rads).Afterwards, each culture of *Bradyrhizobium* and *Bacillus* was injected (120 mL) into the carrier to satisfy 60% of water holding capacity.

Actinomycete strain and preparation of its inoculants: An identified halotolerant actinomycete strain named as *Streptomyces luteogriseus*-8was obtained from Department of Agricultural Microbiology, ARC, Giza, Egypt. This strain was previously isolated from soil, Taif, KSA and completely identified by **Mohamed et al., (2013).** Inoculant of the applied *Streptomyces* strain was prepared by scraping the heavy spores from the surface of the growth of starch nitrate slant in the presence of 5 mL sterilized H_2O as described by **Osman et al., (2007)** by using starch nitrate agar medium **(Mohamed 1998).** The supernatants and pellets were then distributed in 50 mL Fisher tubes and kept at 4ºC until used.

Field experiments design: Two field experiments were carried out in two summer seasons of 2020 and 2021 at the Ismailia Research Station, Ismailia Governorate, Egypt.Super-phosphate (15.5% P_2O_5) and potassium sulphate (48% K_2O) were incorporatedinto soil before sowing at rats of 200 and 50 kg/fed, respectively.Field was divided into two main plots, the 1stplot wasuntreated with organic fertilizer and the 2ndsecond was treated with organic fertilizer at the rate of 2000kg/fed. Each plot was divided into 21 part, and the tested treatments were set up randomly in the subplots of 3x3.5 m with 50 cm apart between ridges (6 ridges/ subplot). A split plot design (RSPD) was used in this study with three replicates.

Sowing soybean seeds were cultivated as recommended practical and inoculated with each inoculant as treatment using Arabic gum solution (2.0%) as adhesive material seeds. Soybean seedlings were thinned out to two plants/hill at 21days from sowing. The other recommended agronomic practices for soybean cultivation were used just before sowing. It is worth to mention that the seven treatments as shown in **Table -3** were conducted in the presence and absences of organic fertilizer as mention above.

Table-3: Treatments of soybean seeds using a combination of different biofertilizers and low concentration of mineral fertilizer in the presence and absence of organic fertilizer.

Treatments		Description
Without organic fertilizer	**T01**	Soil+70 units of mineral N-fertilizer (Control)
	T02	Soil+20 units of mineral N-fertilizer + *Bacillus megatherium* (BM)
	T03	Soil+20 units of mineral N-fertilizer + *Streptomyces luteogriseus*
	T04	Soil+20 units of mineral N-fertilizer + *Bradyrhizobiumjaponicum*(BradyR)
	T05	Soil+20 units of mineral N-fertilizer + BradyR + (BM)
	T06	Soil+20 units of mineral N-fertilizer + BradyR + *Streptomyces luteogriseus*
	T07	Soil+20 units of mineral N-fertilizer + BradyR + (BM) + *Streptomyces luteogriseus*
With organic fertilizer	**T08**	Soil+70 units of mineral fertilizer (Control)
	T09	Soil+20 units of mineral fertilizer + (BM)
	T10	Soil+20 units of mineral fertilizer + *Streptomyces luteogriseus*
	T11	Soil+20 units of mineral N-fertilizer + *Bradyrhizobiumjaponicum*(BradyR)
	T12	Soil+20 units of mineral N-fertilizer + BradyR + (BM)
	T13	Soil+20 units of mineral N-fertilizer + BradyR + *Streptomyces luteogriseus*
	T14	Soil+20 units of mineral N-fertilizer + BradyR + (BM) + *Streptomyces luteogriseus*

It was used due to the recommendation of Ministry of Agriculture.

Samples each of contains five plants were randomly taken from each plot at 60 days after planting to determine nodule number and its dry weight as well as straw dry weight and its nitrogen contents according to **Page et al., (1982)**.

At harvest, 150 days post planting, plots were harvested and each of the following parameters, *i.e.*, plant height (cm), number of branches, number of pods per plant seed and straw yield were measured. Nitrogen in each of seed as well as straw weredetermined according to methods of **Page et al., (1982)**and**Pepe and Robert (1975)**. Crude protein percentage was calculatedby (N %X 6.25) based on the method of **Hames et al., (2008)**.Data were subjected to analysis of variance for each season according to **Gomez and Gomez (1984)**.

RESULTS AND DISCUSSION

The soybean (*Glycine max* L.) is among the most important legumes that feed a large number of people in the world. It is the most important oil crop as it contains about 18 to 22% cholesterol-free oil with 85% unsaturated fatty acids and 38 to 42% protein. Therefore, it is considered as one of the most important oil seeds worldwide (**Ali et al., 2009**).

The experimental results (**Table 4**) showed that the total numbers of microbial counts representing in numbers of bacteria, fungi and actinomycetes were 29.5x10^5, 6.5x10^3 and 1.9x10^4, respectively, which representing lower numbers compared to the treated soil. Data also show that the presence of organic fertilizer increased the number of microbial counts compared to organic fertilizer-free soil. This was obvious by the increasing in numbers of microbial total counts up to 31.45x10^5, 8.5x10^3 and 2.65x10^4in case of bacteria, fungi and actinomycetes (T08), respectively. The combination contains low dose of mineral fertilizer, biofertilizers (Okaden, SWERI and Actinomycetes)and organic fertilizer (T14) gave the highest numbers of microbial counts (47.00X10^5, 9.0X10^3 and 3.6X10^4,respectively) (**Table 4**).

Table-4: Microbial total counts in soil cultivated with biofertilizers-treated soybean seeds in the presence and absence of organic fertilizer among two seasons.

Treatments		Bacteria #x10^5			Fungi #x10^4			Actinomycetes #x10^4		
		Seasons			Seasons			Seasons		
		1st	2nd	Means	1st	2nd	Means	1st	2nd	Means
Without organic fertilizer	**T01**	28.8	30.2	29.50	0.6	0.7	0.65	1.6	2.2	1.90
	T02	39.3	35.4	37.35	0.7	0.9	0.80	1.9	2.2	2.05
	T03	28.9	32.4	30.65	0.7	0.8	0.75	2.8	3.2	3.00
	T04	26.7	34.7	30.70	0.8	0.9	0.85	2.7	2.9	2.80
	T05	37.1	39.3	38.20	0.7	0.8	0.75	2.0	2.8	2.40
	T06	33.3	33.6	33.45	0.7	0.8	0.75	2.6	3.1	2.85
	T07	39.4	40.8	40.10	0.8	0.9	0.85	2.5	2.9	2.70
With organic fertilizer	**T08**	30.4	32.5	31.45	0.8	0.9	0.85	1.9	3.4	2.65
	T09	33.3	36.7	35.00	0.8	1.2	1.00	2.1	2.8	2.45
	T10	28.8	35.4	32.10	0.7	0.9	0.80	3.0	3.6	3.30
	T11	37.1	39.8	38.45	0.8	0.9	0.85	2.8	3.0	2.90
	T12	41.8	43.4	42.60	0.9	1.1	1.00	2.9	3.2	3.05
	T13	39.7	40.6	40.15	0.7	1.0	0.85	3.2	3.5	3.35
	T14	45.6	48.4	47.00	0.7	1.1	0.90	3.4	3.8	3.60

The combined N and P application promoted the formation of a favorable soil aggregate structure and improved soil microbial activity, which accelerated fertilizer use, and enhanced the capacity of soil to maintain fertilizer supply (**Liu et al., 2020)**. They also showed that crop yield increased significantly when compared to the control conditions, with soybean and millet yields increasing by 82.5–560.1% and 55–490.8%, respectively. The combined application of N and P fertilizers increased soil water consumption, improved soil water use efficiency, and satisfied crop growth and development requirements. The results of **Liu et al. (2020)** provided a scientific basis for rational crop fertilization in semi-arid areas on the Loess Plateau.

The experimental results showed that the selected sandy soil of Ismailia which cultivated with soybean under different biofertilizer treatments in the presence of organic fertilizer, low N mineral, and an actinomycete strain as a biofertilizer agent was poor in NPK elements before plantings (667.6, 9.5 and 32.55 ppm, respectively; T01) compared to post cultivation (790.65, 11.45 and 37.20 ppm, respectively; T08) (**Table-5**). In other mean, the use of biological fertilizers (Okaden+SWERI+Actino) + 20 kg N/fed plus the organic fertilizer increased the amount of NPK (894.5, 14.4 and 43.10 ppm, respectively; T14) in cultivated soil compared to the soil-free organic fertilizer. At the same trend, soil activity recorded based on the rate of dehydrogenase enzyme before planting and reached 4.05 µg/g soil/24 hours was lower than that recorded in cultivated soil whatever fertilized with organic fertilizer (6.30 µg/g soil/24 hr, T14) or without (5.80 µg/g soil/24 hours, T07) (**Table-5)**.

Response of soybean growth to biofertilizers and organic fertilizer: Results in **Table-6** showed that no significant differences in soybean plant heights were recorded between inoculation with bradyrhizobia alone (T04 and T11) or plus SWERI (T05 and T12) in the absence (T04 and T05) or presence (T11 and T12) of organic fertilizer. On the other hand, significant differences in soybean plant heights were found when soybean seeds were inoculated with Okaden plus SWERI (T06 and T13) or inoculated with Okaden, SWERI and actinomycetes (T07 and T14) whatever in the presence of organic fertilizer (T13 and T14) or without (T06 and T07).

Both of nodules number and its dry weight of soybean plants cultivated under different fertilizer treatments in an open field experiment were presented in **Table-6**. The use of organic fertilizer plus biofertilizers (T14) enhanced the activity of soybean, and this was clear from the high number of nodules and its dry weight compared to the use of Rhizobia+20 kg N/fed (T04) among the two seasons.

Table-5: Available nitrogen, phosphorus & potassium and dehydrogenase activities in soil cultivated with biofertilizers-treated soybean seeds in the presence and absence of organic fertilizer among two seasons.

Treatments		Available nitrogen in soil (ppm)			Available phosphorus in soil (ppm)			Available potassium in soil (ppm)			Dehydrogenase activities µg TPF/g soil/24hr		
		Seasons			Seasons			Seasons			Seasons		
		1st	2nd	Means	1st	2nd	Means	1st	2nd	Means	1st	2nd	Means
Without organic fertilizer	T01	650.4	684.8	667.60	08.5	10.5	09.50	30.6	34.5	32.55	3.5	4.6	4.05
	T02	576.2	624.7	600.45	09.6	10.7	10.15	32.4	34.8	33.60	4.2	5.4	4.80
	T03	540.4	628.5	584.45	08.9	9.4	09.15	28.4	30.4	29.40	4.7	5.7	5.20
	T04	564.8	647.5	606.15	08.5	9.2	08.85	29.4	32.6	31.00	4.6	4.8	4.70
	T05	668.4	689.0	678.70	10.8	11.4	11.10	34.5	38.4	36.45	5.2	6.2	5.70
	T06	660.5	675.8	668.15	08.4	10.7	09.55	32.4	34.5	33.45	5.4	5.8	5.60
	T07	675.4	684.9	680.15	11.4	12.5	11.95	35.6	42.5	39.05	5.2	6.4	5.80
With organic fertilizer	T08	724.5	856.8	790.65	10.6	12.3	11.45	35.6	38.8	37.20	5.2	6.2	5.70
	T09	768.0	869.5	818.75	12.8	13.4	13.10	38.7	40.8	39.75	4.6	6.5	5.55
	T10	764.9	846.5	805.70	10.4	11.8	11.10	34.6	36.6	35.60	5.4	6.2	5.80
	T11	768.8	848.6	808.70	11.6	12.5	12.05	34.8	36.7	35.75	5.8	6.2	6.00
	T12	864.7	884.7	874.70	12.8	13.6	13.20	38.8	42.6	40.70	5.2	6.6	5.90
	T13	862.8	827.8	845.30	11.5	12.7	12.10	34.8	36.7	35.75	5.4	6.4	5.90
	T14	894.5	894.5	894.50	13.4	15.4	14.40	40.4	45.8	43.10	5.8	6.8	6.30

Table-6: Response of soybean plant (height and nodulation) to bio-fertilizer inoculation combined with organic fertilizer under sandy soil during two successive seasons.

Treatments		Plant height (cm/plant)			Nodules number (#/plant)			Nodules dry weight (mg/plant)		
		Seasons			Seasons			Seasons		
		1st	2nd	Means	1st	2nd	Means	1st	2nd	Means
Without organic fertilizer	T01	46.7fg	54.0hi	50.35E	0.0f	0.0f	0.00D	0.0e	0.0e	0.00D
	T02	44.3g	52.7i	48.50E	0.0f	0.0f	0.00D	0.0e	0.0e	0.00D
	T03	45.7fg	55.3ghi	50.50E	0.0f	0.0f	0.00D	0.0e	0.0e	0.00D
	T04	53.3de	65.0ef	59.15D	11.3e	16.0d	13.65C	147.2d	242.5c	194.85C
	T05	57.3cd	68.3de	62.80C	24.0b	27.7a	25.85A	292.7b	339.7a	316.2A
	T06	61.7bc	75.0bc	68.35B	20.3c	23.3bc	21.8B	164.4d	301.4b	232.9B
	T07	75.7a	86.7a	81.20A	25.3ab	25.7ab	25.5A	314.9ab	344.a	329.45A

With organic fertilizer	**T08**	50.3ef	59.3fgh	54.8D	0.0f	0.0f	0.00D	0.0e	0.0e	0.00D
	T09	47.7fg	57.0ghi	52.35D	0.0f	0.0f	0.00D	0.0e	0.0e	0.00D
	T10	50.3ef	61.3fg	55.8D	0.0f	0.0f	0.00D	0.0e	0.0e	0.00D
	T11	56.3cd	68.7de	62.5C	13.7e	19.7d	16.7C	147.2d	242.5c	194.85C
	T12	61.3bc	72.7cd	67.0C	29.0b	34.0a	31.5A	292.7b	339.7a	316.2A
	T13	66.7b	79.3b	73.0B	25.0c	29.7b	27.35B	164.4d	301.5b	232.95B
	T14	79.7a	86.3a	83.0A	30.7ab	31.7ab	31.2A	314.9ab	344.7a	329.8A

Means in a column not followed by the same letters are significantly different by LSD test (p >0.05)

Results in **Table-7** representing the soybean straw dry weights and strawnitrogen contentsof the fourteen treatments whatever organic fertilizer was not added (T01-T07) or added (T08-T14). Data show that the highest straw dry weights as well as straw nitrogen contents were recorded when Okaden, SWERI, actinomycetes, and 20 kg mineral nitrogen per Feddan were combined (T14). In other mean, biofertilizers in the presence of low N-fertilizer and organic fertilizer increased the straw dry weight from 33.33 g/plant(T01) to 38.20g/plant (T14) g/plant with a significant value.Similar significant differences were recorded in case of straw-nitrogen contents, i.e., increased from 837.3 mgN/plant (T01) to 1093.75 mgN/plant (T14).

Table-7: Response of soybean straw and nitrogen content to biofertilizer inoculationcombined with organic fertilizer under sandy soil during two successive seasons

Treatments		Straw dry weight (g/plant)			Straw N-content (mg/plant)		
		1st Season	2nd Season	Means	1st Season	2nd Season	Means
Without organic fertilizer	**T01**	30.83g	35.83e	33.33C	30.83g	35.83e	837.3D
	T02	24.63j	29.57g	27.1D	24.63j	29.57g	691.5E
	T03	23.10k	27.63h	25.365E	23.10k	27.63h	630.35F
	T04	31.70fg	37.47de	34.585C	31.70fg	37.47de	913.45C
	T05	32.70def	39.10cd	35.9B	32.70def	39.10cd	954.9B
	T06	32.07efg	39.53bc	35.8C	32.07efg	39.53bc	973.3C
	T07	33.40cdk	39.80bc	36.6A	33.40cdk	39.80bc	1038.35A
With organic fertilizer	**T08**	33.97bcd	38.53cd	36.25C	33.97bcd	38.53cd	948.30D
	T09	27.9h	31.37f	29.6D	27.9h	31.37f	765.10E
	T10	26.13i	28.93gh	27.53E	26.13i	28.93gh	697.10F
	T11	34.30bc	39.90bc	37.1B	34.30bc	39.90bc	1046.2C
	T12	35.33b	41.13ab	38.23A	35.33b	41.13ab	1079.15B
	T13	33.17cde	40.30abc	36.735B	33.17cde	40.30abc	1039.7B
	T14	34.40a	42.00a	38.2A	34.40a	42.00a	1093.75A

Means in a column not followed by the same letters are significantly different by LSD test (p >0.05)

Results of branches number and its pod contents of soybean plant cultivated under different fertilizer registered in treatments of open field experiment are reported in **Table-8**. Data show that the mean branches of soybean plants were ranged from 1.3 (T02) to 4.0 (T14) branches/plant. The averages of branches and pods were higher in T07 compared to the control (T01) in spite of absence of organic fertilizer, and this could be due to the use of mixture contains Okaden, SWERI and actinomycetes as biofertilizers plus low N-fertilizer (T07). While, the addition of organic fertilizer to such mixture as shown in T14 raised the mean branches up to 4.0 branches/plant(**Table-8**).Similar results were subsequently recorded regarding the number of pods per plant.

Regarding the weight of 100 seeds (g) of soybean plant under different fertilizer treatments in the same experiment, the average weight was ranged from 117.5 g to 150.65 g in the absence of organic fertilizer compared to present (122.0 g to 156.7 g).

Table-8: Effect of bio-fertilizer inoculation combined with organic fertilizer on branch number, pod number and weight 100 seed of soybean grown under sandy soil during two successive seasons.

Treatments		Branch number per plant			Pod number per plant			Weight 100 seed (g)		
		Seasons			Seasons			Seasons		
		1st	2nd	Means	1st	2nd	Means	1st	2nd	Means
Without organic fertilizer	T01	2.0d	1.7d	1.85CD	36.0c	41.3fg	38.65B	11.80f	11.74e	11.77D
	T02	1.3d	1.3d	1.3D	35.7c	41.0fg	38.05C	11.77f	11.73e	11.75D
	T03	2.0d	1.7d	1.85CD	36.3c	41.00g	38.65B	11.87f	11.87e	11.87D
	T04	2.7cd	2.7c	2.7BC	42.0ab	50.0d	46.0A	14.17de	14.27d	14.22C
	T05	2.7cd	2.7c	2.7BC	42.0ab	51.7bcd	46.85A	14.37de	14.73cd	14.55C
	T06	2.7cd	2.7c	2.7BC	42.3ab	52.3abc	47.3A	13.97e	14.47cd	14.22C
	T07	3.0bc	3.0bc	3.00A	44.7ab	54.0ab	49.35A	15.17abc	14.96bcd	15.065A
With organic fertilizer	T08	1.7d	1.7d	1.7D	42.0ab	39.3e	40.65B	12.13f	12.27e	12.20C
	T09	2.3cd	3.0bc	2.65B	39.7bc	45.0e	42.35B	12.33f	12.37e	12.35C
	T10	3.0bd	3.0bc	3.0B	40.0bc	44.3e	42.15B	12.33f	12.37e	12.35C
	T11	3.0bc	3.7ab	3.35A	44.3ab	50.7cd	47.5A	14.77bcd	15.07bcd	14.92B
	T12	3.0bc	3.7ab	3.35A	44.7ab	53.3abc	49.0A	14.70cd	15.13abc	14.915B
	T13	3.7ab	4.0a	3.85A	44.0ab	52.7abc	48.35A	14.37de	15.00bcd	14.685B
	T14	4.0a	4.0a	4.0A	46.0a	55.0a	50.5A	15.37ab	15.97a	15.67A

Means in a column not followed by the same letters are significantly different by LSD test (p >0.05)

Promising expected yields of each of seeds and straw (Ton/fed) of soybean plants cultivated under different fertilizers and bio-fertilizers conditions are illustrated in **Table (9)**. This was clear from increasing the yield seed up to 1.8400Ton/fed in the presence of organic fertilizer (T14) compared to 1.4565 Ton/fed when soybean was cultivated in the absence of organic fertilizer (T01). Significant differences between the straw yields in the presence and absence of organic fertilizer as well as bio-fertilizers were recorded (**Table 9**).

Effect of biofertilizer inoculation combined with organic fertilizer on crude protein in seed and straw of soybean treated with different fertilizers conditions among two successive seasons is shown in **Table-10.** Crude proteins of each of seeds and straw were affected by adding the organic fertilizer to the biofertilizers (Okaden, SWERI and actinomycetes) treatments, as their values were raised up to the highest value when organic fertilizer was combined to biofertilizers in the presence of low N-fertilizer (T14).

Table-9: Response of soybean seed yield and straw to biofertilizer inoculation combined with organic fertilizer under sandy soil during two successive seasons.

Treatments		Seed yield (Ton/fed)			Straw yield (Ton/fed)		
		1st Season	2nd Season	Means	1st Season	2nd Season	Means
Without organic fertilizer	T01	1.470de	1.443ef	1.4565B	1.757c	1.957ce	1.857D
	T02	1.140de	1.437ef	1.2885C	1.220f	1.587fg	1.4035E
	T03	1.110e	1.373f	1.2415C	1.147fg	1.403g	1.275E
	T04	1.500c	1.760cd	1.630AB	1.840cd	2.040cde	1.94CD
	T05	1.557bc	1.813bcd	1.6850A	1.977bc	2.150bcd	2.0635AB
	T06	1.533bc	1.867abc	1.700AB	1.837cd	2.237bc	2.037BC
	T07	1.623ab	1.970ab	1.7965A	2.047ab	2.307bc	2.177A
With organic fertilizer	T08	1.380d	1.620de	1.5000C	1.433ef	2.200bcd	1.8165C
	T09	1.267de	1.490ef	1.3785C	1.223f	1.813ef	1.518D
	T10	1.170de	1.427ef	1.2985C	1.220f	1.767ef	1.4935D
	T11	1.540bc	1.857abc	1.6985B	1.810cd	2.327b	2.0685BC
	T12	1.620ab	1.950abc	1.785AB	2.057bc	2.360b	2.2085BC
	T13	1.547bc	1.930abc	1.7385AB	2.033bc	2.390b	2.2115AB
	T14	1.627ab	2.053a	1.8400A	2.170ab	2.667a	2.4185A

Means in a column not followed by the same letters are significantly different by LSD test (p >0.05)

Table-10: Effect of biofertilizer inoculation combined with organic fertilizer on crude protein in seed and straw of soybean grown under sandy soil during two successive seasons.

Treatments		Seed crude protein (%)			Straw crude protein (%)		
		1st Season	2nd Season	Means	1st Season	2nd Season	Means
Without organic fertilizer	T01	14.79ef	16.67f	15.730C	9.48de	12.00ab	10.740C
	T02	13.96g	17.40e	15.680D	8.23f	9.96cd	9.095D
	T03	13.54g	16.63f	15.085D	8.02f	9.69d	8.855D
	T04	15.31cde	18.19de	16.75B	9.27e	11.50abcd	10.385C
	T05	15.73c	18.02e	16.875A	9.50cde	11.67abc	10.585B
	T06	15.42cd	18.85cd	17.135AB	9.69bcde	11.98ab	10.835B
	T07	15.83bc	19.58ab	17.705A	10.21ab	12.48ab	11.345A
With organic fertilizer	T08	14.90def	17.71e	16.305D	9.19e	13.33a	11.26A
	T09	14.69f	17.81e	16.250C	8.27f	11.56abcd	9.915B
	T10	14.69f	17.81e	16.25CD	8.13f	11.35bcd	9.740B
	T11	15.83bc	19.48abc	17.655B	10.00abc	12.81ab	11.405A
	T12	16.46a	19.23bc	17.845B	10.10abc	12.67ab	11.385A
	T13	16.35ab	19.85ab	18.100A	10.00abcd	12.40ab	11.200A
	T14	16.67a	20.10a	18.385A	10.42a	13.13ab	11.775A

Means in a column not followed by the same letters are significantly different by LSD test (p >0.05)

As overall view, this study was supported by the findings of **Kumar et al**., (**2017**) who mentioned that biofertilizers as PGPR stimulate root growth by producing some hormones and antimetabolites. Its effects can occur via local antagonism to soil-borne pathogens or by induction of systemic resistance against pathogens throughout the entire plant of these bacteria (*Bacillus*spp., *Pseudomonas* spp., *Bradyrhizobiumjaponicum* and *B. elkanii*) strains. PGPR improve plant growth directly by producing plant growth regulators such as auxins, gibberellins and cytokinins; by eliciting root metabolic activities and/or by supplying biologically fixed nitrogen. Consequently, germination, rootdevelopment, nutrient and water uptake are improved (**Kumar et al., 2017**).

Results of **Javid and Mahmoud (2010)** supportedthese findings of the currentstudy, as they reported that inoculation of soybean seeds with *Bradyrhizobiumjaponicum* was effective as it enhanced the nodulation status, plant growth, pod biomass and yield of soybean). Rhizobacteria, through nitrogen fixation, are able to convert gaseous nitrogen (N_2)toammonia (NH_3) making it an available nutriaent to the host plant which can support and enhance plant growth (**Kumari et al., 2019**). They reported that several microorganisms are commonly used as biofertilizers includingnitrogen-fixing soil bacteria (*Azotobacter*, *Rhizobium*), nitrogen-fixing cyanobacteria (*Anabaena*), phosphate-solubilizing bacteria (*Pseudomonas* sp.), and AM fungi.

Conclusion: Finally, *Bradyrhizobium japonicum* + *Bacillus megatherium*+*Streptomyces luteogriseus* +20 kg N/fed (T07) was effective in the presence of organic fertilizer compared to organic fertilizer-free soil or Uninoc.+70kg N/fed. The yield of each of seeds and straw reflect the importance of using the actinomycetes as bio-fertilizers. This could be supported by the amounts of NPK, N-percentage, protein content, numbers of branches and pods.

Recommendation: One can recommend with use of treatment T07 and T14 (*Bradyrhizobium japonicum* +*Bacillus megatherium*+*Streptomyces* plus activated dose of nitrogen) to improve soybean yield, especially in sandy soils, and to reduce the use of mineral fertilizers in soil.

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