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Effect of Bio-Fertilizer, Organic and Mineral Fertilizaters on Soybean Yield and Nutrients Uptake under Sandy Soil Conditions

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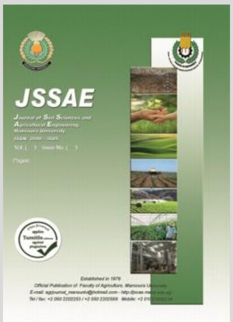

ABSTRACT

During the two consecutive summer seasons of 2015 and 2016 at Faculty of Agric.; Damietta University, Damietta Governorate, Egypt, two pot experiments were conducted to examine the effect of phosphorus fertilization, organic fertilization and inoculation with *Azotobacter* as well as their interactions on nutrient uptake and yield of soybean plants (*Glycine max* L.). The experiment was performed using split-split plot design with 18 treatments in three replicates, three treatments of organic manure in form of chicken manure (without, 8 and 12 $ton.fed^{-1}$) as the main plot, three rates of phosphorus fertilization (0, 36 and 54 $kgp_{2}o_{5}.fed^{-1}$) as subplot and all treatments were studied twice; once in the presence of *Azotobacter* strains inoculants and the other without it as sub-sub plot. Individual organic manure increased N, P and K uptake by the plant, yield and its component (100-seed, g/plant, seed yield, g/plant, pod length, cm and pods weight, g/plant) and chemical composition of seed (N, P, K, protein and total carbohydrates%). Obtained results declare that phosphorus fertilizer treatments at 54 kg $P_2O_5.fed^{-1}$ recorded the highest values of the mentioned parameters. Bio-fertilizers increased the traits especially in presence of *Azotobacter* comparing with the un-inoculated plant. So, the interaction between 12 $ton.fed^{-1}$ chicken manure and 54 $kg.fed^{-1}$ P_2O_5 in presence of *Azotobacter* was the best treatment to achieve the highest values of parameters under investigation during two seasons.

***Keywords**:* chicken manure, P-fertilization, *Azotobacter* and soybean plant

INTRODUCTION

Sandy soils are spread in Egypt as well as in the world which are dominated by sand particles containing enough sand over 85% , low clay particles content, lower nutrient content, very low in fertility, lower water holding capacity, lower cation exchange capacity and lower buffering capacity than clayey and loamy soils (Brady and Weil, 2013), weak structure or no structure, poor water retention properties, high permeability, highly sensitivity to compaction with many adverse consequences. Therefore, it is necessary to improve and maximize the productivity of these lands for sustainable agriculture by paying attention to organic, mineral and biological fertilization.

On the same time, soybean (*Glycine max* L. Merril), is the most important oil seed crop in the world, belongs to the family Fabaceae, under subfamily Faboideae provides vegetable protein for millions of people and ingredients for hundreds of chemical products. It has been classified more as an oil seed crop than as a pulse (Devi *et al.* 2013). Soybean is the cheapest protein source as compared to other protein sources such as egg, fish and meat. With high protein content of 40% on dry matter basis and is the considered second only to groundnut in terms of oil content of 20% among the food legumes (Ahiabor *et al.* 2014). Egypt imports large quantities of soybeans to provide vegetable protein and extract oils. To increase production and reduce imports, attention must be paid to increasing the cultivated area and paying attention to mineral and organic fertilization in addition to biological fertilization, especially in poor sandy soils.

Firstly, many researchers stated that organic inputs are an important source of nutrients; nutrients such as nitrogen, phosphorus, magnesium and calcium content are all released through mineralization (Fairhurst, 2012). Organic resources help the crop to respond better to the mineral fertilizer applied. It also helps in improving the soils capacity to store moisture, helps in regulating soil chemical and physical properties that affect nutrient storage and availability as well as root growth. Also organic inputs help in adding nutrients that are not contained in mineral fertilizers. They create a better rooting environment, improves the availability of phosphorus for plant uptake, ameliorates problems such as soil sodicity and helps in soil organic matter replenishment.

On the other hand, phosphorus is a major nutrient, especially for legumes. It is considered the second essentialnutrient for both microorganisms and plants. Its uptake and utilization by soybean is essential for ensuring proper nodule formation and improving yield and quality of the crop (Anon, 2004). Despite the considerable addition of phosphorus to soil, the amount of available for plant is usually low. Many researchers showed positive effect of phosphorus fertilization on legumes (Fouda, 2017). One of the problems in phosphorus fertilization is rapidly immobile and less available for plant use after application to the soil. Some factors affecting the availability of phosphorus include the pH of the soil and the availability of Ca, Al and Mn (Anderson and Magdoff 2005). Very high soil phosphate depressed seed protein and oil content, while yield would be low if available phosphorus was less than 30 kg P ha^{-1} (DAFF, 2010). The most important phosphorus sources in arable soils are chemical fertilizers, though 75 to 90 percent of the phosphorus is fixed with iron, calcium and aluminum in soil (Turan *et al.*, 2006).

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Besides, applications of chemical fertilizer for increase crop yield are also largely affecting environment, normal flora of soil and human health. For this reason, seed inoculation with nitrogen fixing microorganisms and other beneficial microorganisms as a bio-fertilizer, is an acceptable alternative to chemical fertilizer application. Bio-fertilizers increase soil fertility, while reducing the environmental impact of chemical fertilizers (Wu *et al.* 2005). The greatest proportion of biological nitrogen in agricultural soils originates from the activity of symbiotic and free-living nitrogen-fixing bacteria living in the association with plants. Therefore, bio-fertilization is the best alternative of chemical fertilizer is necessary . Azotobacter abundance in the soil is a good indicator of all toxicological and degradational changes in the soil (Cvijanović *et al.* 2011). Azotobacter is one of the alternatine methods to enhance the soil fertility which helps in synthesis of growth regulating substances like auxins, cytokinin and gibberellic acid (GA). In addition, it stimulates rhizospheric microbes, protects the plants from phytopathogens, improves nutrient uptake and ultimately boost up biological nitrogen fixation. The abundance of these bacteria in soil is related to many factors, mostly soil pH and fertility (Jnawali *et al.* 2015) . In addition, microbial activity in the soil also affects the availability of phosphorus. It was reported that through the process of carbohydrates oxidation by microorganisms, resulting organic acids that have a strong affinity with Al, Ca, Mg and Fe, it can release phosphorus which initially binds to these elements (Henri *et al,* 2008).

Generally , great efforts have been made to ensure more and better food production to bridge the food gap between production and consumption, so the balance between the comical fertilizer , organic fertilizers and bio fertilizers is the goal to achieve the highest yield , reduce production costs for farmers and increased profits for farmers.

Having all these facts in mind, the objective of this study was to examine the effects of organic fertilization, phosphorus fertilization on soybean inoculated with or without *Azotobacter* under sandy soil conditions on soybean plant growth, chemical content and seed yield.

MATERIALS AND METHODS

During the two consecutive summer seasons of 2015 and 2016 at Faculty of Agric.; Damietta University, Damietta Governorate, Egypt, pot experiments were conducted to examine the effect of organic fertilization, phosphorus fertilization and inoculation with *Azotobacter* as well as their interactions on nutrient uptake and yield of soybean plants (*Glycine max* L.) under sandy soil conditions.

The experiment was performed using split-split plot design with 18 treatments in three replicates, three treatments of organic manure in form of chicken manure (without, 8 and 12 ton.fed^{-1}) as the main plot, three rates of phosphorus fertilization (0, 36 and 54 kg.P_2O_5 fed^{-1}) as sub plot and all treatments were studied twice; once in the presence of *Azotobacter* strains inoculants and the other without it as sub-sub plot. Thus, the total number of treatments were fifty four pots for each season.

The soil texture. was sandy in texture. The soil sample was collected from the surface layer (0-15 cm) Qelabshowah City and analyzed for some physical and chemical properties as shown in Table (1)

Table 1. The physical and chemical properties of the soil used in 2015 and 2016 seasons.

Particle size distribution (%)			Textural class	EC, dSm^{-1*}	pH **	$CaCO_3$ (%)	O.M (%)	SP (%)
Sand	**Silt**	**Clay**						
87.3	3.5	9.2	**Sandy**	1.21	8.03	2.89	0.28	32
Available element, mg.kg^{-1}								
N	P	K	Fe	Mn	Cu	Pb	Ni	Cd
39.5	4.96	189	5.46	2.82	0.19	2.68	1.51	0.33

*** soil paste ext** **** 1:2.5 soil: water suspension**

54 polyethylene pot of 25 cm. in diameter and 30 cm. length were used. Each pot was filled with 10 kg air dried soil which was brought from the surface layer of a special farm in Qelabshowah city.

Soybean seeds (variety Giza 111) were mixed thoroughly with *Azotobacter* three hours before their sowing in the soil at a rate of 952 g inoculum/ha. The Arabic gum solution (16%) was used as an adhesive agent to fix the inoculum with soybean seeds (Badawi *et al.,* 2011).

Chicken manure (CH.M) as a source of organic manure was taken from a private farm near El-Mansoura city. Before sowing of soybean seeds; pots were mixed with CH.M at rate of (8 and 12 ton. fed^{-1}) i.e. (80 and 120 g/pot) and irrigated to the saturation percentage. Then, left for two weeks to avoid the damage on seeds and their roots resulted from the heat of decomposition. Some chemical properties of CH.M used are presented in Table (2).

Table 2. Some chemical properties of the chicken manure used during both seasons of 2015 and 2016.

%						Avail mgkg^{-1}		SP %	pH
O.M	**C**	**T.N**	**C/N**	**T.P**	**T.K**	**P**	**K**		**1: 5**
57.1	34.14	2.25	15.17	0.116	0.93	4.1	991	149	6.78

After that, on 25 April during both seasons 2015 and 2016, 10 seeds of soybean were sown in a small whole with equal distances in each pot. Two weeks later; soybean plants were thinned to the most five uniform plants per pot.

Urea and potassium sulfate were applied at doses of 42 kg fed^{-1} (46% N) and 50 kg fed^{-1}, respectively. The applied N was used as an activation dose for soybean and added after plants thinning. Potassium fertilizer was added before the second irrigation. The pots experiment was irrigated every 10 days and the irrigation was stopped 3 weeks before the harvest process. Phosphorus treatment fertilizers at the rates of 0, 36 and 54 kg. P_2O_5 fed^{-1} i.e. 0, 2.25 and 3.40 g.pot^{-1} for P were used, superphosphate was used at the first irrigation after soybean sowing.

Azotobacter (N fixing bacteria) was purchased from the bio-fertilizers Production Unit, Agricultural Microbiology Department at the Institute of Soils, Water and Environment Research, ARC, Giza, Egypt.

At harvesting stage (150 days after sowing) dry pods of each plant were harvested, at full maturity stage accounted;

as the following parameters were measured: Pod length (cm), pod weight (g),plant Weight of 100 seeds in (g). (seed index) and seed yield (g/plant).

Samples of seeds from each sample were oven dried at 70 °C tell the constant weight was reached. Then, dried parts were thoroughly ground and stored for chemical analysis of N, P and K % as well as its uptake (mg/plant), which calculated from the following equation:

$$\textbf{Nutrient uptake mg plant}^{-1} = nutrient\% \textbf{ X dry weight of plant } (\textbf{g}) \textbf{X 10}$$

According to (Jones *et al*. (1991), |(Peters *et al*. (2003), respectively as well as total carbohydrates% according to (Sadasivam and Manickam, 1996) and Crude protein was calculated by multiplying nitrogen percentage by the factor 6.25 according to (AOAC, 2000).

All data were statistically analyzed according to the technique of analysis variance (ANOVA) and the least significant difference (L.S.D) method was used to compare the difference between the means of treatment values to the methods described by (Gomez and Gomez, (1984). All statistical analyses were performed using analysis of variance technique using CoSTATE Computer Software.

RESULTS AND DISCUSSION

- N, P and K uptake (mg.plant^{-1}):

Data tabulated in Table (3) pointed out that increasing organic manure rates in form of chicken manure significantly increase N, P and K uptake mg.plant^{-1}. Addition with 12 ton.fed^{-1} recorded the highest value of N, P and K uptake mg.plant^{-1} during both seasons. On the other hand, control treatment adversely has significant effect on N, P and K uptake (mg.plant^{-1})as recorded in two seasons.

In respect to the phosphorus application effect, data in Table (3) showed that all used treatments of P-fertilization significantly increased N, P and K uptake (mg.plant^{-1})over the control. At the same time addition of 54 kg P_2O_5.fed^{-1} recorded the maximum significant value of N-uptake (785.32 & 978.44), P-uptake (46.14 & 57.88) and K-uptake (347.34 & 448.30 mg plant^{-1}), respectively during two seasons comparing with the untreated plants.

A significant effect was spotted on increasing the percentage of N, P and K uptake mg.plant^{-1} of soybean leaves with the addition of bio-fertilization during 2015 and 2016 seasons as shown in Table (3). The presence of *Azotobacter* obtained the highest percentage of N, P and K uptake mg.plant^{-1} as comparing with un-inoculated plants. Concerning the interaction effect, data in Table (3) a N, P and K uptake mg.plant^{-1} indicated that the significantly highest values of mentioned parameters resulted from the treatment of 12 ton.fed^{-1} + 54 kg P_2O_5 fed^{-1} + *Azotobacter* comparing with the untreated plant in two seasons.

Table 3. Effect of organic manure, phosphorus fertilization and *Azotobacter* as well as their interaction on N, P and K-uptake of soybean leaves during 2015 and 2016.

Treatments			N-uptake,	mg plant^{-1}	P-uptake,	mg plant^{-1}	K-uptake ,	mg plant^{-1}
			2015	2016	2015	2016	2015	2016
Organic fertilization								
Control			563.55	709.08	32.81	41.52	227.77	286.44
8 ton.fed^{-1} chicken manure			726.46	905.72	42.26	53.80	314.23	412.42
12 ton.fed^{-1} chicken manure			770.35	964.31	45.55	56.60	341.62	438.23
LSD $_{at\ 5\%}$			15.61	19.62	1.73	1.09	3.37	8.65
Phosphorus fertilization								
Without			523.04	654.52	30.44	38.22	206.66	277.67
36 kg. P_2O_5 fed^{-1}			751.99	946.15	44.04	55.82	329.62	411.12
54 kg. P_2O_5 fed^{-1}			785.32	978.44	46.14	57.88	347.34	448.30
LSD $_{at\ 5\%}$			16.21	16.90	0.93	0.71	4.68	9.35
Bio-fertilization								
Without			629.70	785.87	36.82	46.46	264.08	350.21
Azotobacter			743.87	933.53	43.59	54.82	325.00	407.85
LSD $_{at\ 5\%}$			5.20	7.52	0.41	0.80	6.81	4.33
Interaction effect								
Control	Without	Without	436.55	528.93	25.17	31.64	159.21	216.83
		Azoto.	460.13	575.34	26.55	33.92	178.35	230.64
	36 kg.fed^{-1} P_2O_5	Without	545.84	696.61	32.19	40.58	212.09	287.96
	Without	Azoto.	675.90	847.23	38.74	48.40	278.79	259.74
	54 kg.fed^{-1} P_2O_5	Without	565.80	721.85	33.77	41.93	234.24	328.89
		Azoto.	697.07	884.52	40.47	52.65	303.97	394.61
8 ton.fed^{-1} chicken manure	Without	Without	497.99	608.83	28.03	35.47	183.87	246.81
		Azoto.	600.54	759.30	35.76	44.35	249.82	345.39
	36 kg.fed^{-1} P_2O_5	Without	733.09	921.36	42.47	55.08	321.02	424.60
	Without	Azoto.	858.63	1065.42	49.77	64.12	389.90	498.99
	54 kg.fed^{-1} P_2O_5	Without	768.12	957.12	44.74	57.00	338.89	444.38
		Azoto.	900.35	1122.29	52.79	66.79	401.88	514.34
12 ton.fed^{-1} chicken manure	Without	Without	514.33	639.22	29.97	38.30	202.07	266.41
		Azoto.	628.69	815.48	37.16	45.65	266.64	359.96
	36 kg.fed^{-1} P_2O_5	Without	776.51	997.59	46.61	58.92	355.51	470.77
	Without	Azoto.	921.96	1148.69	54.46	67.80	420.42	524.63
	54 kg.fed^{-1} P_2O_5	Without	829.04	1001.36	48.44	59.20	369.85	465.25
		Azoto.	951.54	1183.50	56.65	69.70	435.25	542.34
LSD $_{at\ 5\%}$			15.59	22.56	1.22	2.41	20.43	12.98

- Yield and its components:

It was clear that the addition of chicken manure showed a remarkable increase not only in nutrients availability (macro and micronutrients) in the rizosphere, but also nutrients uptake by the plant and nutrients accumulated in the plant tissues. The obtained results might be attributed to that organic fertilizer improves different soil characteristics (Ayoola and Maknide, 2009; Oagile and Mufwanzala, 2010), which led to maintain fertility of the soil to be able to supply grown plants with nutrients requirement over the season. Soil applied chicken manure significantly increased soil living micro-organisms number, activity, and provided necessary nutrients in available forms in the root zone, consequently improved the absorption and accumulation of mineral contents in plant tissue in comparison to chemical fertilizers (Talaat *et al.,* 2015; Shaheen *et al.,* 2016). Chicken manure could be supplied the crops with adequate nutrients in available form easy for the plant to absorb, in particularly nitrogen, phosphorus and potassium. In this regard, (Guo and Song (2009) stated that amongst the organic fertilizers, chicken manure has the highest nitrogen, phosphorus and potassium contents. Total nitrogen and phosphorus contents in chicken manure were about 4 and 2%, respectively, (Gross *et al.,* 2008).

Further N, P and K uptake in leaves increased by increasing P-fertilization This is may be due to that P-fertilization plays a role for improving the growth of root system and, consequently enhancing the capacity of root to absorb more nutrients. These results are similar to those of (Darwesh *et al.* (2013); Fouda, (2017) and Soltan *et al*. (2018).

These improving effects by biofertilization may be attributed to the role played by N-fixing bacteria in secreting chelating substances; as organic acids which are important for solubilization of macro and micro elements from the organic manure sources. Similar results have been reported by El Sayed *et al.* (2015); Rahmayani *et al.* (2017) and Sánchez-Navarro, (2020)

Data of soybean yield and its components expressed in (100-seed (g), seed yield g/plant, pod length (cm) and pods weight g/plant) as affected by organic manure, P-fertilization, bio-fertilization and their interactions presented in Table (4).

Increasing organic manure rates (0, 8 and 12 ton.fed^{-1}) significantly increased parameters of yield during both seasons of the experiment as indicated in Table (4). The highest mean values of 100-seed (g), seed yield g/plant, pod length (cm) and pods weight g/plant realized with 12 ton.fed^{-1}. The rate of increase in traits compared with the control for 8 and 12 ton.fed^{-1} for 100 seeds recorded as (7.51, 9.82% in 2015 and 6.27, 8.34% in 2016), seed yield (7.07, 4.08 in 2015 and 6.22, 8.62% in 2016), pod length (10.51, 14.41 in 2015 and 11.30, 14.78% in 2016) and pod weight (18.51, 25.93 in 2015 and 19.01, 25.35% in 2016).

Table 4. Effect of organic manure, phosphorus fertilization and *Azotobacter* as well as their interaction on yield and its components of soybean during 2015 and 2016.

Treatments			100 seeds (g)		Seed yield gplant^{-1}		Pod length (cm)		Pods weight gplant^{-1}	
			1st	2nd	1st	2nd	1st	2nd	1st	2nd
Organic fertilization										
Control			16.21	17.39	46.23	48.52	3.33	3.45	1.35	1.42
8 ton.fed^{-1} chicken manure			17.18	18.48	49.50	51.54	3.68	3.84	1.60	1.69
12 ton.fed^{-1} chicken manure			17.51	18.84	50.50	52.70	3.81	3.96	1.70	1.78
LSD at 5%			0.06	0.09	0.21	0.09	0.06	0.04	0.05	0.04
Phosphorus fertilization										
Without (0)			15.98	17.11	45.39	47.56	3.23	3.34	1.31	1.36
36 kg. P_2O_5 fed^{-1}			17.38	18.71	50.09	52.31	3.75	3.92	1.65	1.73
54 kg. P_2O_5fed^{-1}			17.55	18.89	50.75	52.89	3.83	3.98	1.69	1.79
LSD at 5%			0.06	0.06	0.40	0.32	0.04	0.03	0.03	0.03
Bio-fertilization										
Without			16.61	17.84	47.58	49.80	3.48	3.61	1.47	1.53
Azotobacter			17.33	18.63	49.91	52.04	3.73	3.88	1.63	1.72
LSD at 5%			0.05	0.07	0.36	0.39	0.04	0.02	0.02	0.02
Interaction effect										
Org 0	Without	Without	15.43	16.34	43.40	45.33	3.02	3.14	1.17	1.23
		Azoto.	15.55	16.66	44.12	45.85	3.08	3.19	1.22	1.26
	36 kg. P_2O_5fed^{-1}	Without	16.11	17.34	45.93	48.75	3.28	3.42	1.34	1.38
	Without	Azoto.	16.85	18.12	48.33	50.75	3.55	3.69	1.49	1.58
	54 kg. P_2O_5 fed^{-1}	Without	16.27	17.55	46.44	49.24	3.38	3.47	1.38	1.43
		Azoto.	17.06	18.34	49.15	51.22	3.65	3.77	1.53	1.62
Org 1	Without	Without	15.66	16.84	44.76	46.49	3.13	3.26	1.29	1.32
		Azoto.	16.53	17.76	47.09	49.88	3.44	3.53	1.41	1.47
	36 kg. P_2O_5fed^{-1}	Without	17.24	18.56	49.76	51.79	3.73	3.87	1.58	1.67
	Without	Azoto.	18.04	19.43	52.18	54.20	3.96	4.18	1.83	1.92
	54 kg. P_2O_5 fed^{-1}	Without	17.45	18.74	50.36	52.20	3.74	3.92	1.65	1.75
		Azoto.	18.18	19.55	52.85	54.66	4.08	4.27	1.86	1.98
Org 2	Without	Without	15.95	17.12	45.32	47.72	3.22	3.32	1.32	1.35
		Azoto.	16.74	17.94	47.65	50.11	3.50	3.64	1.47	1.52
	36 kg. P_2O_5fed^{-1}	Without	17.62	18.95	50.79	52.91	3.87	4.04	1.73	1.80
	Without	Azoto.	18.44	19.86	53.54	55.45	4.13	4.32	1.93	2.04
	54 kg. P_2O_5 fed^{-1}	Without	17.76	19.13	51.47	53.76	3.93	4.11	1.78	1.87
		Azoto.	18.57	20.04	54.23	56.26	4.22	4.36	1.97	2.07
LSD at 5%			0.16	0.21	1.11	1.17	0.12	0.05	0.06	0.07

P- fertilization application treatments, data in Table (4) indicated that all used tested treatments significantly increased 100-seed (g), seed yield g/plant, pod length (cm) and pods weight g/plant over the control in the two seasons. Meanwhile, 54 kg.fed^{-1} was achieved the highest significant yield and its component in the two seasons in comparison with the other treatments. The control plant was given the lowest significant values in both seasons.

It was observed from the Table (4) that bio-fertilization significantly increased 100-seeds (g), seed yield g/plant, pod length (cm) and pods weight g/plant. The highest values were recorded in presence of *Azotobacter* in the two seasons.

In respect to interaction data in Table (4) ,it was indicated that organic manure at rate of 12 ton.fed^{-1} combined with 54 kg.fed^{-1} from P-fertilization and *Azotobacter* resulted in the highest significant 100-seed (18.57 & 20.04 g plant^{-1}), seed yield (54.23 & 56.26 g plant^{-1}), pod length (4.22 & 4.36 cm) and pods weight (1.97 & 2.07 g plant^{-1}). While the lowest significant 100-seed (g), seed yield g/plant, pod length (cm) and pod weight g/plant were resulted from the treatment of control as gave (15.43 & 16.34), (43.40 & 45.33), (3.02 & 3.14) and (1.17 & 1.23) for both seasons, respectively.

The enhancement of plant yield and yield attributes with the application of chicken manure could be explained mainly due to increase of nutrients availability over the time and nutrients uptake (Feleafel and Mirdad, 2014) and improve soil physical, chemical and biological characteristics (Alabadan *et al*., 2009). Besides, (Oustani *et al.* (2015) showed that the increase in yield could be attributed to the improvement of both soil moisture retention and the potentials of nutrient supply (with macro and micronutrients). A similar observation was found by (Shaheen *et al.* (2018); Patel and Tiwari (2018) and AL-Deen Al-Leela *et al.* (2019).

The increase in seed yield due to phosphorus application is attributed to the source and seed relationship. It appears that greater translocation of photosynthates from source to (seed) increased the seed yield. It might also be due to improvement in yield attributes which ultimately increased the seed yield as evident by the existence in a strong positive correlation between seed yield and pods per plant, seeds per pod and 100-seeds weight (Table 6). These findings clearly suggest the profound role of phosphorus fertilization in exploiting the inherent potential of vegetative and reproductive growth which ultimately resulted in increased productivity of soybean crops (Balai *et al.* 2017). These results are in line with that reported by Kuntyastuti and Suryantini, (2015), Shakori and Sharif (2016) and Alemayehu and Shumi, (2018).

The enhancing effect of using the biofertilizer *Azotobacter* might be attributed to supporting the growth of soybean plants with nitrogen in the soil and the role of nitrogen in increasing the leaf area which enhances the photosynthetic rate and ended with the increased formation of the carbohydrates and its translocation down to the tuber as described by (Al-Moshileh, 2001). Similarly results were obtained by Sabilu *et al.* (2015); Jafari *et al.* (2018); Hindersah *et al.* (2019)

- **Quality of soybean seeds:**

The obtained results in Tables (5 and 6) showed the effect of organic manure, P-fertilization, bio-fertilization and their interaction on quality parameters of soybean plant as (N, P, K concentration, protein and total carbohydrates) during both seasons of the experiments.

N, P and K concentration in seeds.

Data in Table (5) illustrated that treatments of 12 ton.fed^{-1} from chicken manure gave the highest significant N, P and K concentration in seeds as (5.15 & 5.37), (0.226 & 0.242) and (1.70 & 1.88), respectively during two seasons in comparison with other organic treatments. On the other side, the control achieved the lowest significant N, P and K concentration in seeds in the two seasons, respectively.

Regarding the effect of phosphorus application, data in Table (5) showed that all used treatments caused a significant increment over the control treatment. In the meantime, addition of 340 kg.fed^{-1} from P-fertilization achieved the significantly highest N, P and K concentration in seeds (5.17 & 5.42), (0.228 & 0.244) and(1.71 & 1.90) against control treatment (4.60 & 4.78), (0.180 & 0.194) and (1.29 & 1.48) in the two seasons, respectively.

As for the effect of bio-fertilization, data in Table (5), indicated that the addition of *Azotobecter* recorded the highest values of N, P and K concentration in seeds comparing with the untreated plants.

N, P and K concentration in soybean seeds were strongly influenced by the interaction treatments, data in the same Table indicated that the maximum significant nutrient concentration in seeds was recorded for the treatments of 12 ton.fed^{-1} combined with 340 kg.fed^{-1} in presence of *Azotobacter* comparing to the other ones. On the opposite, the minimum significant N, P and K concentration in soybean seeds resulted from untreated plants with any fertilizers during both seasons.

Table 5. Effect of organic manure, phosphorus fertilization and *Azotobacter* as well as their interaction on N, P and K% in soybean seeds during 2015 and 2016.

Treatments			N% 1st	N% 2nd	P% 1st	P% 2nd	K% 1st	K% 2nd
Organic fertilization								
Control			4.68	4.87	0.188	0.202	1.34	1.53
8 ton.fed^{-1} chicken manure			5.04	5.28	0.217	0.233	1.61	1.79
12 ton.fed^{-1} chicken manure			5.15	5.37	0.226	0.242	1.70	1.88
LSD at 5%			0.04	0.03	0.003	0.003	0.02	0.01
Phosphorus fertilization								
Without			4.60	4.78	0.180	0.194	1.29	1.46
36 kg. P_2O_5 fed^{-1}			5.10	5.32	0.222	0.239	1.65	1.84
54 kg. P_2O_5fed^{-1}			5.17	5.42	0.228	0.244	1.71	1.90
LSD at 5%			0.03	0.02	0.002	0.001	0.03	0.02
Bio-fertilization								
Without			4.84	5.03	0.200	0.215	1.47	1.65
Azotobacter			5.08	5.32	0.220	0.236	1.63	1.82
LSD at 5%			0.02	0.02	0.002	0.001	0.03	0.02
Interaction effect								
Control	Without	Without	4.37	4.56	0.161	0.169	1.12	1.28
		Azoto.	4.47	4.65	0.168	0.183	1.16	1.32
	36 kg. P_2O_5 fed^{-1}	Without	4.65	4.87	0.187	0.201	1.34	1.51
		Azoto.	4.92	5.09	0.206	0.225	1.43	1.72
	54 kg. P_2O_5fed^{-1}	Without	4.72	4.91	0.193	0.207	1.40	1.56
		Azoto.	4.98	5.18	0.211	0.229	1.57	1.79
8 ton.fed^{-1} chicken manure	Without	Without	4.55	4.74	0.176	0.189	1.23	1.39
		Azoto.	4.78	4.94	0.195	0.211	1.44	1.63
	36 kg. P_2O_5 fed^{-1}	Without	5.06	5.27	0.218	0.238	1.63	1.84
		Azoto.	5.32	5.63	0.241	0.258	1.83	2.01
	54 kg. P_2O_5fed^{-1}	Without	5.12	5.35	0.225	0.241	1.67	1.87
		Azoto.	5.41	5.73	0.247	0.261	1.88	2.04
12 ton.fed^{-1} chicken manure	Without	Without	4.61	4.78	0.180	0.194	1.28	1.47
		Azoto.	4.84	5.02	0.201	0.217	1.48	1.66
	36 kg. P_2O_5 fed^{-1}	Without	5.19	5.30	0.228	0.245	1.73	1.91
		Azoto.	5.47	5.78	0.253	0.268	1.93	2.08
	54 kg. P_2O_5fed^{-1}	Without	5.28	5.48	0.234	0.254	1.78	2.00
		Azoto.	5.53	5.88	0.258	0.272	1.98	2.14
LSD at 5%			0.05	0.06	0.005	0.004	0.08	0.06

As mentioned before, the addition of chicken manure showed a remarkable increase in seed nutrients. The obtained results might be attributed to that organic fertilizer improves different soil characteristics (Ayoola and Maknide, 2009; Oagile and Mufwanzala, 2010), which led to maintain fertility of the soil to be able to supply grown plants with nutrients requirement over the season. Soil applied chicken manure significantly increased soil living micro-organisms number and activity, and provided necessary nutrients in available forms in the root zone, consequently improved the absorption and accumulation of mineral contents in plant tissue in comparison with chemical fertilizers (Talaat *et al.,* 2015 and Shaheen *et al.,* 2016).

As for the increase in N, P and K in soybean seed as a result of increasing the levels of P-fertilization, consequently might be due to that phosphorus might have improved and developed good root system of plant and the capacity of root to absorb more N, P and K accordingly their contents increased by phosphorus applications for the soil, which reflected on N, P and K in seeds. Obtained results are confirmed with those reported by Darwesh *et al.* (2013); Fouda, (2017) and Soltan *et al.* (2018).

These improving effects by bio fertilization may be attributed to the role played by N-fixing bacteria in secreting chelating substances; as organic acids which are important for solubilization of macro and microelements from the organic manure sources. Similar results have been reported by El Sayed *et al.* (2015); Rahmayani *et al.* (2017) and Sánchez-Navarro, (2020).

- Protein and total carbohydrates% :

Data presented in Table (6) elucidate that both protein and total carbohydrates% increased due to increasing rates of chicken manure during both seasons. The protein and total carbohydrates% were significantly influenced by different organic treatments, the maximum protein% was (34.56 & 36.73) and total carbohydrates% was (36.27 & 38.48) noticed in 12 ton.fed^{-1} from chicken manure treatment, respectively in the two seasons which was statistically significant compared to different organic treatments.

Concerning the effect of phosphorus fertilization application, data at the same Table showed that protein and total carbohydrates% increased with all treatments comparing with the control. At the same time, 54 kg.fed^{-1} of phosphorus application recorded the maximum significant value of protein and total carbohydrates% comparing with the untreated plants during 2015 and 2016 without any significant with 36 kg.fed^{-1}.

Data depicted in Table (6) significantly increased protein and total carbohydrates% with inoculation with *Azotobacter* treatment. The values of such traits in increasing order were (31.75 & 33.26%) of protein and (35.02 & 36.78%) of total carbohydrates% due to the addition *of Azotobacter* during both seasons, respectively comparing with the un-inoculated plants.

Concerning the comparative and interactive effects, data in Table (6) illustrated that the combined treatments of organic manure in form of chicken manure, phosphorus fertilization and *Azotobacter*. The protein and total carbohydrates% contents in soybean seed were strongly influenced by integrated nutrient management. Between various treatments, integration 12 ton.fed^{-1} chicken manure , phosphorus fertilizers applied with 54 kg.fed^{-1} and presence of *Azotobacter* caused a significant increase in these traits in comparison with the other applications. While control without any fertilization gave a lower significant value in both seasons.

As for the effect of organic manure on quality of soybean seeds , similar conclusion was reported by Mahmoud and Ibrahim (2012) who reported that append organic manure increased carbohydrate content of turnip roots in clay loamy soil. In pea pods, Mahmoud *et al.* (2013) found that the chemical ingredients protein% significantly increased with increasing the level of organic manure up to 180 kg N/fed in sandy soil. Similarly results were found by Ali *et al* (2014) and Shaheen *et al.* (2018)

Table 6. Effect of organic manure, phosphorus fertilization and *Azotobacter* as well as their interaction on protein and carbohydrates in soybean seeds during 2015 and 2016.

Treatments			Total carbohydrates%		Protein %	
			1st	2nd	1st	2nd
Organic fertilization						
Control			33.92	35.27	29.28	30.46
8 ton.fed^{-1} chicken manure			34.95	36.67	31.50	32.98
12 ton.fed^{-1} chicken manure			35.25	37.08	32.20	33.57
LSD at 5%			0.61	0.95	0.24	0.17
Phosphorus fertilization						
Without			33.76	34.91	28.76	29.87
36 kg. P_2O_5fed^{-1}			35.11	36.90	31.89	33.27
54 kg. P_2O_5fed^{-1}			35.24	37.20	32.33	33.87
LSD at 5%			0.44	0.53	0.17	0.13
Bio-fertilization						
Without			34.39	35.89	30.24	31.42
Azotobacter			35.02	36.78	31.75	33.26
LSD at 5%			0.37	0.27	0.09	0.12
Interaction effect						
Control	Without	Without	33.33	34.25	27.33	28.48
		Azoto.	33.32	34.45	27.94	29.06
	36 kg. P_2O_5fed^{-1}	Without	33.83	35.09	29.08	30.42
		Azoto.	34.50	36.17	30.73	31.79
	54 kg. P_2O_5fed^{-1}	Without	33.91	35.20	29.48	30.67
		Azoto.	34.65	36.44	31.10	32.35
8 ton.fed^{-1} chicken manure	Without	Without	33.41	34.69	28.42	29.60
		Azoto.	34.33	35.38	29.88	30.88
	36 kg. P_2O_5fed^{-1}	Without	35.19	36.81	31.60	32.94
		Azoto.	35.65	37.86	33.27	35.21
	54 kg. P_2O_5fed^{-1}	Without	35.23	37.12	32.02	33.42
		Azoto.	35.86	38.15	33.81	35.83
12 ton.fed^{-1} chicken manure	Without kg.fed^{-1} P_2O_5	Without	33.76	34.80	28.79	29.85
		Azoto.	34.44	35.86	30.23	31.35
	36 kg. P_2O_5fed^{-1}	Without	35.36	37.19	32.44	33.15
		Azoto.	36.15	38.29	34.19	36.13
	54 kg. P_2O_5fed^{-1}	Without	35.51	37.87	32.98	34.23
		Azoto.	36.27	38.44	34.56	36.73
LSD at 5%			1.11	0.82	0.28	0.36

It could be related to increasing in nodulation then increasing content of N or protein in legumes as a result of phosphorous fertilization. Magani and Kochinda (2009) reported that crude protein content of cowpea seed was increased significantly with an increasing rate of P (0, 37.5, and 75 kg ha^{-1}). Kumar *et al.*, (2012) reported that ether extract and ash, crude fiber content was increased with each increment of P_2O_5 levels, in forage cowpea. Jha *et al.*, (2014) observed that the application of 80 kg P_2O_5 ha^{-1} recorded significantly higher crude fiber yield (6.1 q ha^{-1}). Also, Fouda (2017) resulted an increase in protein and carbohydrates due to the application of P-fertilization.

The crude protein increment in *Azotobacter* inoculated plants may be ascribed to the nitrogen fixed within the root nodules and translocated to the seeds. An increase in seeds protein content of other legumes as a response to *Azotobacter* inoculation has previously been reported by Rugheim and Abdelgani (2012) and Sabilu *et al.* (2015).

CONCLUSION

Finally, it could be concluded that the integration of biofertilizers, phosphorus and organic manure enhanced the yield attributes such as pod length, pod weight, weight of 100 seeds (seed index) and seed yield. The highest grain yield was recorded in the treatment of 54 kg. P_2O_5 fed^{-1} through phosphorus fertilizers coupled with chicken manure at rate of 12 $ton.fed^{-1}$ and *Azotobacter* biofertilizer. Similarly, the highest chemical content was also obtained from the integration of 54 $kg.fed^{-1}$ through phosphorus fertilizers coupled with chicken manure at rate of 12 $ton.fed^{-1}$ and *Azotobacter*. Thus the objective of maximizing yields as well as maintaining soil health and productivity can be furnished by a balanced use of inorganic fertilizers conjunctively with fertilization and organic manure.

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تأثير التسميد العضوى و الحيوي و المعدنى علي العناصرالغذائية الممتصة ومحصول نبات فول الصويا تحت ظروف الأراضى الرمليه

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تم اجراء تجربه أصص خلال موسمى الصيف المتتالين 2015 و 2016 في كليه الزراعه، جامعه دمياط ، محافظه دمياط، مصر لدراسه تأثير التسميد الفوسفاتى و العضوى و التلقيح ببكتريا الازوتوباكتر و تفاعلاتهم على امتصاص العناصر و انتاجيه نبات فول الصويا و جودتها. صممت تجربه في قطاعات منشقه متعامده من خلال 18 معامله في ثلاث مكررات كالتالى: ثلاث مستويات من التسميد العضوى (بدون، 8 و 12 طن/فدان) كقطاعات رئيسيه، ثلاث مستويات من التسميد الفوسفاتى (بدون، 36 و 54 كجم خامس أوكسيد الفوسفور/ فدان) كقطع تحت رئيسيه مع دراسه جميع المعاملات مرتين في وجود بكتريا الازوتوباكتر و مره أخرى بدون تلقيح كقطع منشقه و بالتالى اصبح عدد الاصص للتجربه 54 اصيص لكل موسم. أوضحت النتائج أن التأثير الفردى لاضافه السماد العضوى أدى الى زياده معنويه في كل من الصوره الممتصه للنيتروجين و الفوسفور والبوتاسيوم في النبات بالاضافه الى المحصول و مكوناته (وزن 100 حبه، وزن المحصول، طول القرن، ووزن القرون للنبات) كذلك جوده البذور (نسبه النيتروجين، الفوسفور، البوتاسيوم، البروتين و الكربوهيدرات الكليه). بالنسبه لتأثير التسميد الفوسفاتى سجلت اعلى القيم للصفات السابقه عند استخدام معدل 54 كجم خامس أوكسيد الفوسفور/ فدان. التسميد الحيوي في وجود الازوتوباكتر أدى الى زياده في جميع الصفات المدروسه مقارنه بالنباتات الغير معامله. الاضافات المشتركه بين 8 طن/فدان سماد دواجن مع 54 كجم خامس أوكسيد الفوسفور/ فدان في وجود بكتريا الازوتوباكتر سجل أعلى القيم للصفات السابقه خلال كلا الموسمين.