

Original Research Article

Assessment of the efficiency of nano, bio and organic fertilizers on soil chemical properties and soybean productivity in saline soil

Abstract

Two ~~filed~~-field experiments were conducted at the village of Khaled Ibn El-waleed, Sahl El-Hossinia Agricultural Research Station, El-Sharkia Governorate, Egypt, during the two summer seasons 2019 and 2020 to estimate the effect of NPK nano-fertilizers, bio-fertilizers and humic acid (in addition to different doses of mineral fertilizers) on soil chemical properties and productivity of soybean in saline soil. The treatments were NPK-chitosan, NPK-Ca, humic acid, biofertilizer and control (mineral NPK only). In the two seasons, the experiment design was a split plot in three replicates.

The results indicated a slight decrease in soil pH, while there was a significant decrease in EC values in all the studied treatments as compared to control. Also, the soil available macronutrients (nitrogen, phosphorous and potassium) and micronutrients (iron, manganese and zinc) were significantly increased after application of the studied treatments as compared to control. Also, there was a significant increase in the content of macro and micronutrients in soybean grains as affected by the studied treatments. This may be attributed to that addition of the used amendments like nano chitosan, humic acid and biofertilizers companied by decomposition of organic materials released acids that reduced soil pH which caused nutrients to be more soluble hence more available for plant uptake. The results indicated also a significant increase in the soybean yield parameters as plant height, weight of seeds and weight of pods in all treatments as compared to control. Also, the studied additions improved soybean quality parameters. There was also a significant increase in the soybean content of protein, oil and chlorophyll in all of the investigated treatments as compared to control.

Keywords: Nano chitosan, humic acid, bio fertilizer, saline soil, soybean.

Introduction

Soil salinity is one of the most critical problems that can cause land degradation (Taha et al. 2021). Plant growth is affected by high values of soil salts that causing osmotic stress, that affect spread of plant roots to soil solution, causing a negative effect on crop yield parameters (Dustgeer et al. 2021). There are several regions of the Nile Delta in Egypt suffer from soil salinity due to climate factors that increase salinization (Mohamed *et al.*, 2019 and Taha 2021). Different practices such as farm manure, soil amendments and biofertilizers can be applied for remediation of saline soils. These practices can improve soil properties and crop productivity in saline soils of the North Delta, (Ding *et al.*, 2020).

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Nitrogen, phosphorous and potassium were loaded into chitosan nanoparticles to produce NPK nano-fertilizer (Seleiman *et al.*, 2021). Nanofertilizers (NFs) as a new technology and a suitable substitution for traditional chemical fertilizer in agricultural practice (Seleiman *et al.*, 2020), it can prevent the soil and water pollution by gradual and controlled release of nutrients into the soil and subsequently on the plant (Sekhon, 2014 and Seleiman *et al.*, 2021). Nano NPK fertilizers can increase the uptake of N, P and K in the plants along with that can lead to an improvement of crop yield (Liu and Liao 2008 and Seleiman *et al.*, 2021). Nanofertilizers significantly increased the yield of fresh seeds, dry seed yield, number of seeds/bush, number of pods/bush, pod yield, total biomass and also increased the weight of 100 seeds of pea plants (Moosapoor *et al.* 2013).

Foliar fertilization has the ability to improve the efficiency and rapidity of utilization of a nutrient urgently required by the plant for maximum growth and yield. Foliar applications of nutrients also and other biostimulators can provide for a more rapid material utilization and permits the correction of observed deficiencies in less time than can be accomplished by soil applications (Kandil and Eman 2017).

Application of chitosan can increase crop yield because it stimulates plant growth, germination of seeds, and enhanced nutrients uptake (Nguyen *et al.* 2019). So, in the current study, we used chitosan nanoparticles (as a nanomaterial) for loading NPK to achieve the benefits of application of both NPK macronutrients and chitosan nanoparticles to crops.

Humic acid application on soybean significantly increased Fe, Mn and Zn uptake and reduced the influence of the salt stress on soybean plant (Renata *et al.* 2017). El-Shafey and Zein El-Dein (2016) studied the effect of soil and foliar addition of humic acid on soybean yield and growth parameters. They added it by rates 5 kg fed⁻¹ and 5 g L⁻¹ for soil and foliar application, respectively. They deduced a significant increase in all yield parameters as plant height, weight of pods/plant, weight of seeds/plant, weight of 100 seeds and weight of seeds (ton fed⁻¹) in the treated plots as compared to the untreated ones.

Biofertilizers are organic, bio-degradable as well as contain micro-organisms, provide nutrients viz., nitrogen, phosphorous, potassium, hormones like auxins, antibiotics, vitamins which make enriched environment in the zone of root rhizosphere. It reduces the use of chemical fertilizers, reduces environmental pollution and increases the validity of nutrients and easily absorbed (Kamlesh and Smritikana 2019).

Soybean (*Glycine max L.*) is the best crop for obtaining oil and protein. Its seeds have the highest protein content among leguminous crops (Taha *et al.*, 2020). Soybean protein is one of the most important sources of plant protein, because it contains a high ratio of the essential

amino acids (Taha *et al.*, 2020). Its oil is used either directly in the human consumption or indirectly in the many manufactured valuable materials. Indeed; soybean seeds have many uses such as, human food, animal feed. However, soybean plants foliage can be used as hay, pasture, cover and green manure crop. A high yield of soybean per unit area is the aim of agronomists and farmers under the limited area and water resources. This goal can be achieved by cultivating high yielding cultivars coupled with application of the best package from agricultural practices including optimum levels of several factors (Taha *et al.*, 2020). Soybean has good benefits for adult humans, it contains 40-50% proteins, 26-30% carbohydrates and 20-30% lipids (Gibbs *et al.*, 2004). It is affected to high extent by water salinity (Ayman *et al.*, 2015). In Egypt, the decrease in the productivity and area in the last twenty years due to competition with other strategic summer crops on the limited cultivated area and higher production costs with lower net income which related to marketing problems and the damage resulted from leaf feeding insects. So, increasing the production for such crop is a main goal. This could be achieved via cultivating high yielding cultivars combined with application appropriate cultural practices (El-Karamity *et al.*, 2015).

So the aim of the present study was to evaluate the effect of NPK as nanofertilizers that were carried on Ca^{2+} and chitosan, biofertilizers and humic acid in addition to different doses of NPK mineral fertilizers on some soil chemical properties and soybean productivity in saline soil.

Materials and methods

Two field experiments were conducted at Khaled Ibn El-waleed village, Sahl El-Hossinia Agricultural Research Station, El-Sharkia Governorate, Egypt, located at $31^{\circ} 8' 12.461''$ N latitude and $31^{\circ} 52' 15.496''$ E Longitude, during the two summer seasons 2019 and 2020, to estimate the effect of NPK nanofertilizers, biofertilizers and humic acid (in addition to different doses of mineral fertilizers) on some chemical properties of soil and soybean productivity under saline soil conditions. Soil samples from surface layer (0-30 cm) were air-dried, then ground and sieved to pass through a 2 mm sieve, kept and analyzed the physical and chemical soil properties before sowing of soybean and after soybean harvesting according to the methods described by Cottenie *et al.*, (1982), Klute (1986) and Page *et al.*, (1982). The main chemical and physical properties of soil samples before soybean planting were recorded in Table (1).

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Table (1): Some physical and chemical properties of soil before planting

Particle size distribution (%)				Texture	O.M (%)	CaCO ₃ (%)		
Coarse sand	Fine sand	Silt	Clay					
6.89	12.89	33.20	53.91	Clay	0.62	12.90		
pH (1:2.5)	EC (dS m ⁻¹)	Cations (meq L ⁻¹)				Anions (meq L ⁻¹)		
		Ca ²⁺	Mg ²⁺	Na ⁺	K ⁺	HCO ₃ ⁻	Cl ⁻	SO ₄ ²⁻
8.12	10.48	9.88	14.50	79.64	0.78	10.75	65.90	28.15
Macronutrients (mg kg ⁻¹)				Micronutrients (mg kg ⁻¹)				
N	P	K		Fe	Mn		Zn	
35.87	4.30	169.00		2.95	1.50		0.65	

The experiments were designed in a split plot design with three replicates in the two seasons. The main plots were the type of treatment (bio, humic acid and nano NPK), while the subplots were the rate of mineral NPK fertilizers). All farming processes were fulfilled in the experiments area before planting. The treatments were as follows:

The main plots were 5 treatments:

1. Control (without amendment).
2. Biofertilizers (5 L of the biofertilizer dissolved in 100 L water/fed.).
3. Humic acid (20 kg/fed.).
4. NPK nano fertilizers loaded on calcium (5 L of the prepared nano NPK dissolved in 200 L water/fed.).
5. NPK fertilizers loaded on nano chitosan (5 L of the prepared nano NPK dissolved in 200 L water/fed.).

The subplots were 4 mineral NPK fertilizers as follows:

1. 0 kg (N) + 0 kg (P) + 0 (K) kg/fed.
2. 10 kg (N) + 15 kg (P) + 15 (K) kg/fed.
3. 20 kg (N) + 25 kg (P) + 30 (K) kg/fed.
4. 40 kg (N) + 31 kg (P) + 60 (K) kg/fed.

The mineral fertilizers were as follows: Nitrogen as urea (46 % N) at rates (0, 10, 20 and 40 kg N fed⁻¹) was applied after 31, 45 and 65 days from planting. Phosphorous as calcium super phosphate (15.5 % P₂O₅) was added to the soil at rates (0, 15, 25 and 31 kg P₂O₅ fed⁻¹) before planting during soil tillage. Potassium as potassium sulphate (48 % K₂O) fertilizer was applied at rates (0, 15, 30 and 60 kg K₂O fed⁻¹) after 31 and 45 days from planting. Humic acid was applied at a rate 20 kg fed⁻¹ during the soil tillage before planting.

The biofertilizers were as follows: Radiobacter strain as a salt tolerant rhizobacteria (Salt Tolerant PGPR) was applied with urea for nitrogen fixing. *Bacillus megatherium* was applied with calcium superphosphate for dissolving phosphorous. *Bacillus circulans* was applied with

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potassium sulphate to enhance potassium availability. The bio-fertilizers were supplied from the Unit of Biofertilizer Production, Microbiology Department, Soils, Water and Environment Research Institute, Agricultural Research Center, Giza, Egypt.

Nanoparticles of NPK nanofertilizer were prepared by polymerization process of methacrylic acid (MAA) in chitosan solution via two steps according to the method of Hasaneen *et al.* (2014). Macronutrients (nitrogen, phosphorus and potassium) were loaded on polymethyl acrylic acid nanoparticles at rates 500, 60, 400 ppm, respectively (100% concentration stands for 500 ppm of nitrogen, 60 ppm of phosphorous and 400 ppm of potassium in both nano and normal NPK solutions and other concentrations were made from these stock solutions). The NPK nano-fertilizer was applied as a foliar application to soil and plant after 31, 45 and 65 days from planting.

Each unit area of the experiment was 10.8 m² (3.60 m x 3 m) with six ridges, the distance between ridges was 60 cm and 20 cm between hills, Plants were thinned to secure one plant for each hill after 30 days from sowing. Soybean (*Glycine max L.*) cultivar Giza 35 supplied by Field Crop Research Institute, ARC, was sown on the 25th of May 2019 and 2020. Other cultural practices for growing soybean were conducted as recommended.

Soybean varieties were harvested on 25 September 2019 and 2020 and the following growth and yield parameters were estimated: Plant height, number of pods/plant, weight of pods/plant (g), weight of seeds/plant (g), weight of 100 seeds (g), and weight of seeds (ton fed⁻¹).

Samples of the plant parts (grains and leaves) were oven dried then ground and digested using H₂SO₄ and HClO₄ acids mixture according to the method of Chapman and Pratt (1961). The grains contents of N, P, K, Fe, Mn and Zn were estimated in plant digestion using the methods described by Cottenie *et al.* (1982). Oil content in the grains was estimated using Soxhlet apparatus and petroleum ether as solvent according to method of A.O.A.C. (1990). Protein percentage of grains was calculated by multiplying the nitrogen percentage by the factor 6.25 as described by Hymowitz *et al.* (1972). Proline content was measured in the grains according to Bates *et al.*, (1973) and as described by Sofy *et al.* (2020). Chlorophyll was estimated in the leaves according to Saric *et al.* (1967).

The one-way analysis of variance (ANOVA) was carried out to determine the statistical significance of the treatment effects with Duncan grouping procedure at p = 0.05 by using SPSS program.

Results and discussions

Soil pH, EC and soil available macro and micronutrients:

Soil reaction (pH) is a very important factor that affects the chemical properties of soil. It is clear from the data shown in Table (2) that there were no statistical differences in pH values due to treatment with NPK-chitosan, humic acid and bio-fertilizers.

The slight decrease in values of soil pH may be due to the microorganisms activity that decomposed organic matter and led to high concentrations of released organic acids in the soil. Shaban and Omar (2006) deduced that the soil pH was affected by bio-fertilizer due to

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dehydrogenase activity of biofertilizer and formation of μ moles of H_2 in the rhizosphere of maize root media which react in the root zone to form hydrocarbon acid which cause a decrease in soil pH. El-Maaz and Ismail (2016) found that application of bio-fertilizer, humic acid and compost tea as soil or foliar applications caused a decrease in soil pH values.

Concerning soil salinity (EC in the normal soil should be less than 4 dS m^{-1}); results indicated that, in general all treatments significantly decreased soil salinity as compared to the control (Table 2). The lowest EC value (4.67 dS m^{-1}) exists in case of humic acid treatment. It was noticed that the mean EC values could be arranged in ascending order of: humic acid < NPK-chitosan < Bio-fertilizer < NPK-Ca < control. There were non significant differences between EC values concerning rates of mineral NPK fertilization. Also, the interaction between the added treatments and the mineral fertilizers was non significant.

The significant decrease in values of soil electrical conductivity was may be due to that addition of humic acid to the soil releases H^+ into the soil solution where its position was replaced by cation salt that led to a decrease in the salt concentrations in the soil solution, so that the EC values were reduced (Ali and Mindari 2016). Mohamed (2012) deduced a significance decrease in soil EC values after applying of humic acid. The humic acid application led to a decrease in values of soil EC and Na due to high supplies of Mg and Ca. Also, nano-fertilizers plus NPK mineral fertilizers led to activity of micro-organism to reduce salinity and simultaneously improve characterization of soil structure (increasing drainable porosity and aggregate stability) and consequently enhanced leaching process through growth of soybean (El-Sayed et al. 2020). Also, these results are in accordance with those of Sushila et al. (2017), they deduced that of biofertilizers application to salt affected soil decreased EC values because the bio-fertilizers led to activating microorganisms in soil and production of dehydrogenase enzyme in soil led to a decrease in the soil salinity as compared to control.

Regarding macronutrients availability, the results indicated that, in general, there were positive significant effects on nutrients availability in soil. Available nitrogen, phosphorous and potassium in soil were significantly increased by using all treatments. The results in this study are in agreement with some other studies on using nanofertilizer and chitosan nanoparticles. Nguyen et al. (2013) found that applying of chitosan nanoparticles had increased nutrients uptake by about 9.8-27.4 % nitrogen and 30-45 % potassium. Also Ha et al. (2019) found that application of the NPK chitosan nanofertilizers increased the content of nitrogen, phosphorous and potassium by about 17.04, 16.32 and 67.51 % in the leaves of treated plots as compared to control, respectively.

Concerning available micronutrients (Fe, Mn and Zn) in soil, it is clear from the data in Table (3) that there was a positive significant effect in all treatments as compared to control. The trend in increasing soil available content of iron, manganese and zinc was NPK-chitosan > NPK-Ca > humic acid > bio fertilizer > control. These results are in agreement with those

obtained by El-Sayed et al. (2020) who found significant increase in available micronutrients (Fe, Mn and Zn mg kg⁻¹) as affected by NPK nanofertilizers application.

Table (2): Soil pH, EC and available macronutrients in the soil after harvesting

Treatments	Rates of NPK (kg/fed)	pH (1:2.5)	EC (dS m ⁻¹)	Available macronutrients (mg kg ⁻¹)		
				N	P	K
Control	0+0+0	8.08	8.56	38.00	5.20	183.00
	10+15+15	8.06	7.45	39.55	5.84	185.36
	20+25+30	8.03	6.88	40.21	5.93	189.42
	40+31+60	8.01	6.17	40.96	6.02	193.41
	Mean	8.05	7.27 e	39.68 e	5.75 e	187.80 e
Bio-fertilizers	0+0+0	8.02	5.80	39.78	6.03	201.00
	10+15+15	8.00	5.63	40.39	6.12	205.00
	20+25+30	7.98	4.77	41.88	6.53	210.00
	40+31+60	7.95	4.32	42.18	6.70	216.00
	Mean	7.99	5.13 c	41.06 d	6.35 d	208.00 d
Humic acid	0+0+0	8.00	5.99	41.30	6.09	208.00
	10+15+15	7.97	4.73	42.40	6.15	215.00
	20+25+30	7.93	4.12	42.99	6.88	220.00
	40+31+60	7.92	3.85	43.20	7.02	226.00
	Mean	7.96	4.67 a	42.47 a	6.54 c	217.25 c
NPK Nano carrier Ca	0+0+0	8.00	6.12	40.88	6.13	209.00
	10+15+15	7.98	5.88	41.70	6.44	218.00
	20+25+30	7.95	5.30	42.75	6.89	226.00
	40+31+60	7.93	4.95	43.90	7.04	238.00
	Mean	7.97	5.56 d	42.31 b	6.63 b	222.75 a
NPK Nano carrier chitozan	0+0+0	8.00	5.90	40.89	6.23	208.00
	10+15+15	7.97	4.88	41.80	6.55	215.00
	20+25+30	7.94	4.21	42.25	6.90	228.00
	40+31+60	7.92	4.10	43.10	7.06	235.00
	Mean	7.96	4.77 b	42.01 c	6.69 a	221.50 b
L.S.D (0.05)	-	-	A= 0.020	A= 0.037	A= 0.008	A= 0.63
	-	-	M= ns	M= 0.031	M= 0.006	M= 0.56
	-	-	A*M= ns	A*M=0.071	A*M=0.011	A*M= 1.26

A= Amendments treatments

M= Mineral NPK treatments

Also, there were significant differences in macro and micronutrients concentrations in soil between the different rates of the applied mineral fertilizers. Also, the interaction between the added treatments and the applied mineral NPK fertilizers was significant.

The increase in values of soil available macro and micronutrients may be attributed to the decomposition of organic materials released acids that reduced soil pH which caused nutrients to be more soluble hence more available for plant uptake. These beneficial effects were also reported by El-Kouny (2007) who found that application of organic materials caused a substantial increase in total N, available P, K, Fe, Mn, Cu and Zn. These results are in

agreement with those of Bhardwaj et al (2014) who deduced that the availability of micronutrients (Mn, Fe and Zn) were positively affected by inoculation (PGPR) when compared to the un-inoculated treatments.

Also, nano NPK fertilizers, due to their high surface area to volume ratio, are more effective than most of the conventional fertilizers. Their nature could also allow slow release and promote efficient nutrient uptake by the crops (Al-Juthery and Sahar 2019).

There were also significant differences in soil available contents of macro and micronutrients between the different rates of mineral NPK fertilizers. Also, the nano particles of NPK fertilizers led to increase in soil nutrients because these nanoparticles enhance the availability and solubility of these nutrients (Lyarin and Aravinda 2019).

Table (3): Micronutrients contents in soil after harvesting

Treatments	Rates of NPK (kg fed ⁻¹)	Available micronutrients (mg kg ⁻¹)		
		Fe	Mn	Zn
Control	0+0+0	3.24	2.41	0.63
	10+15+15	3.59	2.53	0.65
	20+25+30	3.75	2.58	0.68
	40+31+60	3.98	2.62	0.69
	Mean	3.64 e	2.54 e	0.66 d
Bio-fertilizers	0+0+0	4.05	2.85	0.73
	10+15+15	4.08	2.98	0.75
	20+25+30	4.13	3.02	0.78
	40+31+60	4.23	3.07	0.82
	Mean	4.12 d	2.98 d	0.77 c
Humic acid	0+0+0	4.08	2.88	0.71
	10+15+15	4.16	3.04	0.75
	20+25+30	4.38	3.08	0.79
	40+31+60	4.77	3.13	0.83
	Mean	4.35 c	3.03 c	0.77 c
NPK Nano carrier Ca	0+0+0	4.09	2.95	0.73
	10+15+15	4.44	3.08	0.77
	20+25+30	4.70	3.15	0.80
	40+31+60	4.89	3.45	0.85
	Mean	4.53 b	3.18 b	0.79 b
NPK Nano carrier chitosan	0+0+0	4.22	3.05	0.75
	10+15+15	4.45	3.24	0.81
	20+25+30	4.70	3.55	0.86
	40+31+60	4.89	3.66	0.89
	Mean	4.57 a	3.38 a	0.83 a
	L.S.D (0.05)	A= 0.011	A= 0.008	A= 0.009
		M= 0.008	M= 0.006	M= 0.006
		A*M=0.023	A*M=0.017	A*M=0.015

A= Amendments treatments

M= Mineral NPK treatments

Macro and micronutrients contents in seeds of soybean:

It can be deduced from the data of macro and micronutrients contents in seeds of soybean (Table 4), that all of these contents were significantly increased in all treatments as compared to control. It is clear from the values of macronutrients, that the treatment NPK-chitosan was the best treatment in increasing the total contents of nitrogen, phosphorous and potassium, followed by NPK-Ca then humic acid and biofertilizer, while control (without amendment) recorded the least values. Also, the values of micronutrients (Fe, Mn and Zn) followed the same trend of macronutrients. Also, there were significant differences in the seeds contents of macro and micronutrients between the different rates of mineral NPK fertilizers. These results are in accordance with El-Sayed et al. (2020) who deduced increase in N, P, K, Fe, Mn and Zn contents in soybean seeds as affected by nanofertilizers application.

There were also significant differences in macro and micronutrients contents in soybean grains between the different rates of the applied mineral fertilizers. Also, the interaction between the added treatments and the applied mineral NPK fertilizers was significant.

Some studies elucidated that chitosan can affect metabolism and cause biological responses in the plants (Ghormade et al. 2011). These affects led to an increase in nutrients uptake in the leaves and other parts of the plant. Application of N nanofertilizer loaded on methacrylic acid and chitosan nanoparticles led to a higher N uptake as compared to free urea application (Ledezma-Delgadillo et al. 2016). Nitrogen content in the leaves of lettuce after using of urea nanofertilizer was the same as compared to free urea, but the amount of urea used in nanofertilizer was about 16 % only of urea rate used in the control. This is due to a continuous and slow release of NPK nanofertilizers as compared to normal chemical fertilizers.

The application of amendments in the current study increased macro and micronutrients concentrations in soybean seeds because these materials act as valuable soil amendments that offer a balanced nutritional release pattern to plants, providing nutrients in readily available form that can be easily taken up by plants, (Ferrerias et al., 2006).

The foliar application of NPK nano fertilizers causes immediate uptake of applied nutrients. Under problems of soil fixation, foliar fertilization is the most effective ways of fertilizer placement. And usually necessitates using smaller quantities of nutrients comparing with soil application. The most important use of foliar nutrition is the application of micronutrients in small amounts as well as macronutrients (e.g., nitrogen, phosphorus, or potassium) without causing any phytotoxicity (Rietra 2017). Also, the increase in the grains content of nutrients after foliar application of nano NPK fertilizers may be attributed to that foliar application improves the platform sustainable and novel nutrient delivery systems, which will exploit the nano porous surfaces of the plant parts on plant surfaces (Elemikie 2019).

The increase in concentrations of N, P, K, Fe, Cu, Zn and Mn in grains at foliar nutrition of nano NPK fertilizers (NCSF) are attributed to the role of these nutrients in stimulating plant growth. Also, the high content of macro and micronutrients in soybean grains was due to that foliar fertilization on plant and soil led to (1) a quick remediation for unexpected deficiencies, (2) for late supply of NPK nutrients during advanced growth stages, (3) as a preventive

measure against unsuspected (hidden) deficiencies, and (4) to overcome fixation of nutrients in soils (Stepien and Katarzyna 2016).

Furthermore, humic acids enhance nutrients uptake by plants, because it affects the permeability of root membranes (Türkmen et al., 2004). These results are in agreement with those recorded by Mesut et al., (2010) who found that humic substances act in a very similar way to growth hormones. The positive effects of the applied materials on increasing macro- and micro-nutrients availability and their uptake may be attributed to one or more of the following: a) high initial nutrients content of such materials, b) reduction of soil pH values resulting from the applied materials, c) slow mineralization of organic matter and long rate release of the nutrients and d) converting unavailable soil phosphate into available forms due to humic acids application, (Bhattacharjee et al., 2001 and Angelova et al., 2013). Also, Abdel Aziz et al. (2018) found a significance increase in phosphorous and potassium content in wheat grains after application of NPK fertilizers loaded on nano chitosan compared to the control (untreated).

Table (4): Macro and micronutrients contents in seeds of soybean

Treatment s	Rates of NPK (mg kg ⁻¹)	Macronutrients (%)			Micronutrients (mg kg ⁻¹)		
		N	P	K	Fe	Mn	Zn
Control	0+0+0	3.10	0.28	2.25	53.69	42.16	16.85
	10+15+15	3.25	0.32	2.34	57.52	43.69	17.52
	20+25+30	3.46	0.36	2.48	59.63	44.10	18.00
	40+31+60	3.69	0.39	2.55	62.94	44.80	18.55
	Mean	3.38 d	0.34 e	2.41 d	58.45 e	43.69 e	17.73 e
Bio-fertilizer	0+0+0	3.75	0.38	2.59	64.55	46.20	22.49
	10+15+15	3.89	0.44	2.63	66.00	48.10	23.55
	20+25+30	3.99	0.47	2.66	69.33	48.85	23.90
	40+31+60	4.12	0.52	2.75	74.20	52.18	24.15
	Mean	3.94 c	0.45 d	2.66 c	68.52 d	48.83 d	23.52 d
Humic acid	0+0+0	3.77	0.42	2.55	68.50	51.99	23.68
	10+15+15	3.85	0.46	2.64	74.77	54.38	25.77
	20+25+30	4.15	0.55	2.74	78.40	56.84	27.44
	40+31+60	4.25	0.58	2.76	84.00	59.65	27.98
	Mean	4.01 c	0.50 c	2.67 c	76.42 c	55.72 c	26.22 c
NPK Nano carrier Ca	0+0+0	3.95	0.44	2.62	72.30	53.90	25.33
	10+15+15	4.08	0.52	2.75	76.39	56.39	27.52
	20+25+30	4.22	0.58	2.78	79.00	58.88	29.80
	40+31+60	4.35	0.59	2.82	86.00	62.25	32.10
	Mean	4.15 b	0.53 b	2.74 b	78.42 b	57.86 b	28.69 b
NPK Nano carrier chitosan	0+0+0	4.09	0.52	2.66	77.30	55.90	29.00
	10+15+15	4.36	0.62	2.77	82.19	59.33	33.04
	20+25+30	4.62	0.66	2.81	85.97	65.10	36.98
	40+31+60	4.77	0.69	2.84	89.20	70.90	37.65
	Mean	4.46 a	0.62 a	2.77 a	83.67 a	62.81 a	34.17 a
	L.S.D	A= 0.104	A= 0.008	A= 0.014	A= 0.161	A= 0.031	A= 0.042

(0.05)	M= 0.093	M= 0.006	M= 0.008	M= 0.144	M= 0.025	M= 0.040
	A*M= 0.209	A*M= 0.014	A*M= 0.023	A*M= 0.323	A*M= 0.059	A*M= 0.085

A= Amendments treatments

M= Mineral NPK treatments

Effect of the used treatments on growth and yield parameters of soybean:

It is clear from the data of growth and yield parameters as shown in Table (5), that the used treatments significantly increased all of the growth parameters as compared to control. The best treatments affected these parameters were NPK-chitosan and NPK-Ca followed by humic acid and bio fertilizer. Concerning plant height, the best treatments were NPK-Ca and NPK-chitosan, followed by bio fertilizer and humic acid then control with means 60.27, 57.80, 55.61, 54.88 and 45.32 cm, respectively. While for weight of pods/plant and weight of seeds/plant, the trend was NPK-chitosan > NPK-Ca > bio fertilizer > humic acid > control with mean values 64.52 , 64.23 , 54.16 , 47.84 and 28.52 g for weight of pods/plant, and 30.35, 28.99, 25.78, 22.49 and 14.81 g for weight of seeds/plant, respectively. NPK-Ca and NPK-chitosan treatments recorded the highest values of weight of 100 seeds with mean values 12.94 and 12.17 g, respectively. While the highest values of seeds yield were found in the treatments bio fertilizer and NPK-Ca with mean values 0.782 and 0.772 ton fed⁻¹, respectively. Also, there were significant differences in all growth parameters between the different rates of mineral NPK fertilizers. These results are in agreement with those obtained by El-Shafey and Zein El-Dein (2016) who deduced a significant increase in all yield parameters of soybean as plant height, weight of pods/plant, weight of seeds/plant, weight of 100 seeds and weight of seeds (ton fed⁻¹) in humic acid treated plots as compared to the untreated ones. Similar results were obtained by Ha et al. (2019) who deduced an increase in plant height (of coffee) due to using NPK loaded on nano chitosan. Also Nguyen et al. 2019 deduced that chitosan increased crop yield parameters because it stimulates seed germination, growth of plants and enhanced nutrients uptake.

It can be deduced from the data in Table (6) that there were significant differences in the growth and yield parameters between the different rates of mineral NPK fertilizers. Also, the interaction between the added treatments and the applied mineral NPK fertilizers was significant.

El-Metwally et al. (2018) reported that the nano-fertilizers had significant increases effect on plant height, no. of branches , no. of pods/plant , fresh weight of straw/plant, dry weight of grains/plant and 100 grains. These results may be due to nano-fertilizers increase of nutrients to the plant which increase pigments formation, photosynthesis rate and dry material production.

The data in Table (6) show that there was a significant increase in values of protein, proline, oil and chlorophyll in soybean as compared to control. For protein and chlorophyll the trend was NPK-chitosan > NPK-Ca > humic acid > bio fertilizer > control with mean values 27.88 , 25.94 , 25.03 , 24.61 and 21.10 % for protein content, and 5.19 , 5.01 , 4.91 , 4.89 and 3.82 mg/g (f.w.) for chlorophyll, respectively. While for oil content, the trend was NPK-Ca >

NPK-chitosan > bio fertilizer > humic acid > control with mean values 23.88, 23.78, 23.21, 23.15 and 21.41 % for oil content, respectively.

The increase in growth parameters as plant height and total chlorophyll was attributed to the increase in concentrations of N, P, K, Fe, Cu, Zn and Mn in soybean plant due to foliar application of nano NPK fertilizers due to the role of these nutrients in stimulating plant growth. These essential elements are required for optimum growth of the plant to complete its life cycle (Ali 2012). Macronutrients nutrients are N, P and K enhance the photosynthesis and thylakoid and the development of chloroplasts (Abdulahdy 2018). It also plays a role in the transfer of energy within the plant, and in many enzymatic activities and photosynthesis as well as respiration and synthesis of proteins therefore has a key role in plant growth (Ali 2012).

The results are near to those of Ha et al. (2019) who found that the NPK loaded on nano chitosan increased content of chlorophyll in the leaves of by about 14.7-35.3 % and 10.7-25.1 %, respectively, as compared to control. This increase may be attributed to the increase of the nutrients uptake of the nanofertilizer. Liu and Lal (2014) deduced that chitosan induced improvement of chloroplast gene in plants that lead to an increase in the leaves content of chlorophyll.

Table (5): Soybean productivity as affected by the studied treatments

Amendments Treatments	Rates of NPK (kg fed ⁻¹)	Plant height (cm)	No. of Pods/plant	Weight of pods/plant (g)	Weight of seeds/plant (g)	Weight of 100 seeds (g)	Weight of seeds (ton fed ⁻¹)
Control (Mineral)	0+0+0	38.52	22.89	12.60	9.10	5.88	0.238
	10+15+15	45.62	52.90	22.40	13.84	6.33	0.587
	20+25+30	47.24	56.67	33.96	16.90	7.12	0.665
	40+31+60	49.88	70.88	45.10	19.39	7.83	0.734
	Mean	45.32 e	50.84 e	28.52 e	14.81 e	6.79 e	0.556 e
Bio fertilizers	0+0+0	45.19	35.00	26.46	16.70	8.07	0.589
	10+15+15	55.32	69.00	55.49	26.87	7.50	0.793
	20+25+30	59.80	80.00	59.45	28.01	10.98	0.834
	40+31+60	62.14	95.00	75.23	31.55	6.98	0.912
	Mean	55.61 c	69.75 c	54.16 c	25.78 c	8.38 d	0.782 a
Humic acid	0+0+0	48.95	32.00	25.71	14.20	8.68	0.569
	10+15+15	53.24	66.00	43.00	17.93	9.70	0.695
	20+25+30	56.88	76.00	55.20	27.54	11.50	0.769
	40+31+60	60.46	90.00	67.43	30.30	10.76	0.815
	Mean	54.88 d	66.00 d	47.84 d	22.49 d	10.16 c	0.712 d
NPK Nano carrier Ca	0+0+0	52.41	40.00	32.56	15.62	11.91	0.579
	10+15+15	59.35	84.00	64.87	29.20	12.76	0.790
	20+25+30	62.41	89.00	73.80	35.30	14.20	0.825
	40+31+60	66.89	98.00	85.70	35.83	12.88	0.892
	Mean	60.27 a	77.75 a	64.23 b	28.99 b	12.94 a	0.772 b
NPK Nano carrier chitosan	0+0+0	50.45	39.00	32.70	20.66	12.30	0.570
	10+15+15	57.95	80.00	60.96	28.65	14.12	0.783
	20+25+30	60.33	85.00	75.43	34.38	13.16	0.815
	40+31+60	62.48	95.00	89.00	37.69	9.10	0.885
	Mean	57.80 b	74.75 b	64.52 a	30.35 a	12.17 b	0.763 c
L.S.D (0.05)		A= 0.014	A= 0.71	A= 0.32	A= 0.045	A= 0.011	A= 0.06
		M= 0.011	M= 0.64	M= 0.29	M= 0.042	M= 0.008	M= 0.04

		A*M=0.028	A*M=1.42	A*M= 0.65	A*M=0.093	A*M=0.023	A*M=0.11
A= Amendments treatments		M= Mineral NPK treatments					

Table (6): Soybean quality parameters as affected by the studied treatments

Treatments	Rates of NPK (kg fed ⁻¹)	Protein (%)	Proline (mg g ⁻¹ f.w.)	Oil (%)	Chlorophyll (mg g ⁻¹ f.w.)
Control	0+0+0	19.38	56.38	20.58	3.25
	10+15+15	20.31	42.20	21.33	3.86
	20+25+30	21.63	39.20	21.78	3.95
	40+31+60	23.06	35.12	21.95	4.20
	Mean	21.10 e	43.23 a	21.41 e	3.82 e
Bio-fertilizers	0+0+0	23.44	49.63	22.36	4.66
	10+15+15	24.31	33.25	22.95	4.85
	20+25+30	24.94	27.13	23.65	4.98
	40+31+60	25.75	20.14	23.89	5.07
	Mean	24.61 d	32.54 d	23.21 c	4.89 d
Humic acid	0+0+0	23.56	51.63	22.85	4.75
	10+15+15	24.06	35.77	22.96	4.88
	20+25+30	25.94	29.81	23.15	4.99
	40+31+60	26.56	22.59	23.65	5.03
	Mean	25.03 c	34.95 b	23.15 d	4.91 c
NPK Nano carrier Ca	0+0+0	24.69	44.69	22.89	4.82
	10+15+15	25.50	36.25	23.85	4.99
	20+25+30	26.38	27.23	24.13	5.09
	40+31+60	27.19	20.63	24.65	5.14
	Mean	25.94 b	32.20 e	23.88 a	5.01 b
NPK Nano carrier chitozan	0+0+0	25.56	46.98	22.44	5.01
	10+15+15	27.25	38.95	23.91	5.12
	20+25+30	28.88	30.14	24.13	5.28
	40+31+60	29.81	22.18	24.63	5.36
	Mean	27.88 a	34.56 c	23.78 b	5.19 a
L.S.D (0.05)		A= 0.034	A= 0.036	A= 0.023	A= 0.017
		M= 0.031	M= 0.030	M= 0.019	M= 0.014
		A*M=0.068	A*M=0.071	A*M=0.045	A*M=0.028

A= Amendments treatments

M= Mineral NPK treatments

Conclusion

Application of NPK nanofertilizers, biofertilizers and humic acid led to improvement in soil chemical properties and soybean productivity under saline soil conditions. Available macronutrients (N, P and K) and micronutrients (Fe, Mn and Zn) in soil were significantly increased by using all treatments due to the decomposition of organic materials released acids that reduced soil pH which caused nutrients to be more soluble hence more available for plant uptake. Chitosan increased crop yield parameters because it stimulates growth of plants, seed germination, and enhanced nutrients uptake. Also, application of nano NPK fertilizers led to increase in all growth parameters of soybean due to its slow release of nutrients that make these nutrients available to the plant more time than traditional mineral fertilization.

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