



Positive Role of Applied Chitosan as a Supplement Fertilizer on Okra Plants

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ABSTRACT

This experiment was conducted under modified greenhouse (net house) conditions at the Central Laboratory for Agricultural Climate (CLAC), Agricultural Research Center (ARC), to investigate the effect of using chitosan as a supplement fertilizer. Seeds of okra (*Abelmoschus esculentus* cv Balady) were sown on 15th February from each season in 2020 and 2021. Two factors were tested (i) applied method of chitosan (spray and adding to soil), and (ii) concentration of chitosan such as (100, 150, 200, 250 and 300 ppm) with fourth replicates designed in a randomized complete block. Results reflected the positive role of using a high concentration of chitosan on the growth, yield and quality of okra plants. The greatest values of all tested parameters i.e., vegetative growth (plant height, number of leaves and fresh and dry weights of leaves), chemical contents of leaves (N, P and K plus chlorophyll reading) and yield and its components (number of fruits/plant, average fruit weight, early and total yield, total protein, phosphorus and potassium) were obtained with 80% recommended doses from "N" chemical fertilizer + chitosan adding to soil 250 ppm "T10" and 80% recommended doses from "N" chemical fertilizer + chitosan spray 300 ppm "T6" treatments rather than grower treatment and reduced content of dietary fiber in okra fruit. While stem diameter was not affected by applied two tested factors.

INTRODUCTION

Okra (*Abelmoschus esculentus*) is an important vegetable crop with high demand and high economic value. Okra is an important and high-yielding vegetable of the mallow family grown in many countries. Okra, also known as 'Lady's cinquefoil' or 'Bendi' in Malaysia, is popular for its health benefits such as high fiber content, antioxidants, vitamin C, minerals, potassium and calcium. It is also important as a medicinal plant for plasma replacement in tropical and subtropical countries (Kumar et al., 2013; Sorapong, 2012). Popular for its short production time and ease of cultivation, okra is a versatile crop as fresh leaves, flowers, buds, pods, stems and seeds provide multiple uses. Additionally, the medicinal properties

of okra have been recognized (Gemede et al., 2015). In addition, okra mucus can be used as a blood volume-increasing agent or plasma substitute for medical purposes. Okra mucilage binds cholesterol, toxins in bile acids are excreted by the liver, and most parts of okra are edible and used as food (Gemede et al. 2015; Maramag, 2013).

Chitosan is a biopolymer, a chitin derivative, and a compound that is completely harmless to the environment. That is mean it's very safe and environmentally friend. Moreover, this compound is characterized by unique properties such as bioactivity and biocompatibility (Dias et al., 2013). Its derivative, chitosan, is therefore described as a linear, semi-crystalline polysaccharide composed of glucosamine

(C₆H₁₃NO₅) and N-acetylglucosamine linked by glycosidic β-linkages (1-4), with free amino groups. It differs from chitin polymers by the presence of in the polymer; we distinguish the second carbon atom of the D-glucose unit, not the acetamide group (Agbodjato et al. 2021). Also, literature results show that the use of chitosan in plants increases yield (Mondal et al., 2012), decreases transpiration (Dzung et al. 2011), and induces many metabolic changes to reduce plant viral load. It has been shown to increase tolerance to bacterial and fungal infections (Al-Hetar et al. 2011). In addition, chitosan-treated plants may be less susceptible to stress caused by adverse conditions such as drought, salinity, cold or hot temperatures (Lizarraga-Pauli et al. 2011; Jabeen and Ahmad, 2013; Pongprayoon et al. 2013). Chitosan stimulates important plant processes at all levels of biological organization, from single cells and tissues, through physiological and biochemical processes, to molecular changes associated with gene expression (Limpanavech et al. 2008; Hadwiger, 2013; Nguyen Van et al. 2013). Chitosan refers to a group of commercially available copolymers rather than a unique compound. In addition, treatment with chitosan makes plants more resistant to various soil and foliar pathogens and induces root-knot formation (Hamel and Beaudoin, 2010), making chitosan a useful tool for agricultural sustainability (Iriti and Varoni, 2015; Pichyangkura and Chadchawanb, 2015). Moreover, Kah et al. (2013) found that chitosan increases the absorption of bioactive compounds, allowing plants to absorb nutrients more effectively.

On other hand, the main goal of modern agriculture is to produce sufficient quantities of good quality food to meet the growing world population with low environmental impact (FAO, 2013). Agricultural production is severely affected by many pests and diseases, which can lead to huge losses. Chemical fertilizers and pesticides have been used over the past 100 years to combat these problems and increase yields. Large-scale development of these products has greatly increased productivity, but has also led to biodiversity loss and degradation of natural and agricultural systems. Furthermore, residue accumulation has caused environmental pollution and public health problems with the emergence of resistant pests (Sun et al. 2012). Therefore, alternative methods are needed to address these issues and reduce the environmental impact of activities without compromising agricultural productivity and achieving economic returns. Recently, chitosan-based materials have been used to create nanoparticles that can

efficiently supply chemicals and nutrients to plants (Kah et al., 2013). Indeed, chitosan is readily absorbed by the epidermis of leaves and stems, prolonging the contact time and facilitating the absorption of bioactive molecules.

Furthermore, fertilizer requirements are important in early growth to improve okra productivity and quality. Currently, chemical fertilizers such as NPK (nitrogen, phosphorus, and potassium) are widely used in agricultural fields, including vegetable cultivation, because they can achieve high productivity in a short period of time. However, it is very expensive and causes nutrient imbalance and soil acidification (Akande et al. 2010). Furthermore, overuse of chemicals used to fertilize plants can lead to the accumulation of minerals and nutrients that are not readily available for plant consumption, ultimately leading to soil contamination and toxicity (Savci, 2012). Therefore, increasing crop production depends on improving soil fertility to ensure food for all in the current global food security scenario (Godfray et al. 2010). Therefore, it is imperative to develop various eco-friendly methods to improve soil fertility and increase agricultural production.

This investigation aims to evaluate the effect of using two ways for adding chitosan as a supplement fertilizer on the growth, yield, and quality of okra plants.

MATERIAL AND METHODS

This investigation was conducted at Central Laboratory for Agricultural Climate (CLAC), Agricultural Research Center (ARC), Giza, Egypt, under a modified greenhouse (net house). Through two summer seasons 2020 and 2021 to evaluate using chitosan as a supplement fertilizer on growth, yield and quality of okra plants.

Experimental layout

A modified greenhouse covered by a white net was 40m long, 8m wide and 5.25m height. This house was divided into 5 beds (1m wide and 40m long). Seeds of okra (*Abelmoschus esculentus* cv Balady) were sown on 15th February from each season by the spacing of 0.5m between plants inside the raw and separated 0.50m between beds. A drip irrigation system was placed in this experiment. Recommended doses from chemical fertilizer which should be added "200 Kg

ammonium nitrate (33.5% NH₄NO₃), 100 Kg potassium sulphate (48% K₂O) and 150 Kg calcium super phosphate (15.5 % P₂O₅)/fad." was applied (as 100%) only with grower treatment (control), while, application chitosan treatments as supplement fertilizer were applied by 80% from nitrogen chemical fertilizer recommended doses. Fertilizer treatments were divided into three equal doses; the first was added after 30 days from the sowing date, the second at the flowering stage and the third after 30 days from the second does.

Treatments

Two factors were tested as follows compared to the grower treatment (as control):

(A) The applied method of chitosan (spray and adding to soil), and

(B) The concentration of chitosan such as (100, 150, 200, 250 and 300 ppm).

Treatments were arranged as follows:

1. 100% recommended doses from chemical fertilizer (control treatment) "T1",
2. 80% recommended doses from "N" chemical fertilizer + chitosan spray 100 ppm "T2",
3. 80% recommended doses from "N" chemical fertilizer + chitosan spray 150 ppm "T3",
4. 80% recommended doses from "N" chemical fertilizer + chitosan spray 200 ppm "T4",
5. 80% recommended doses from "N" chemical fertilizer + chitosan spray 250 ppm "T5",
6. 80% recommended doses from "N" chemical fertilizer + chitosan spray 300 ppm "T6",
7. 80% recommended doses from "N" chemical fertilizer + chitosan adding to soil 100 ppm "T7",
8. 80% recommended doses from "N" chemical fertilizer + chitosan adding to soil 150 ppm "T8",
9. 80% recommended doses from "N" chemical fertilizer + chitosan adding to soil 200 ppm "T9",
10. 80% recommended doses from "N" chemical fertilizer + chitosan adding to soil 250 ppm "T10", and
11. 80% recommended doses from "N" chemical fertilizer + chitosan adding to soil 300 ppm "T11".

Data recorded

(1) Vegetative growth parameters

Plant height, number of leaves, fresh and dry weights of leaves and stem diameter were measured at med of the season from fifth okra plants as random samples.

(2) Chemical contents of leaves

Content percentages from N, P and K in leaves were recorded plus chlorophyll reading at med of season. Nitrogen was determined in leaves by the distillation in a Macro-Kjeldahle according to (FAO, 2008). Phosphorus was colorimetrically determined in leaves in the acid digest using ascorbic acid and ammonium molybdate as described by FAO (2008). Potassium was estimated in leaves photometrically as described by FAO, 2008. When, chlorophyll reading was measured in leaves by using a digital chlorophyll meter (model Minolta chlorophyll meter SPAD-501).

(3) Yield and its components

A number of fruits/plant, average fruit weight (from 10 fruits), and early and total yield (per plant) were measured at the harvest stage. Additionally, total protein and dietary fiber were determined according to A.O.A.C. (2005). Phosphorus and potassium concentration was determined according to FAO (2008).

Experimental design and data analysis

This experiment was designed in randomized complete block with fourth replications and obtained data were statistically analyzed using the analysis of variance method. Duncan's multiple range tests at a 5% level of probability were used to compare the means of the treatments (SAS, 2005).

RESULTS

Vegetative growth parameters

Data in Table (1) reflected the effect of applied chitosan as a supplement fertilizer on the vegetative growth of okra plants at tested two grown seasons 2020 and 2021.

Generally, indicated that, the application of okra plants with a high concentration of chitosan (spray or adding to soil) enhanced all tested vegetative growth parameters such as (plant height, number of leaves, fresh and dry weights of leaves) through two grown seasons compared to grower treatment. Contrary, the stem diameter parameter was not affected by the applied two tested factors at all two growing seasons. The greatest values of those parameters were obtained with applied treatments T10 (80% recommended doses from "N" chemical fertilizer +

chitosan adding to soil 250 ppm) and T6 (80% recommended doses from "N" chemical fertilizer + chitosan spray 300 ppm) more than other treatments without any significant difference. When application of okra plants by 80% of recommended doses from

"N" chemical fertilizer + chitosan spray 100 ppm "T2" reduced all those parameters followed by 80% of recommended doses from "N" chemical fertilizer + chitosan spray 150 ppm "T3", respectively.

Table (1): Effect of using chitosan as supplement fertilizer by spraying or adding to soil on vegetative growth parameters i.e., plant height (cm), number of leaves, fresh and dry weights of leaves (g) and stem diameter (cm) of okra plants through 2020 and 2021 seasons.

Treatments	plant height	number of leaves	Fresh weight of leaves	Dry weights of leave	stem diameter
T ₁	60.24D	16.25E	78.68D	16.20G	1.40A
T ₂	52.63G	12.92J	64.94H	13.88K	1.34A
T ₃	56.76EF	13.77I	68.65G	14.35J	1.36A
T ₄	61.54D	15.55F	79.35D	16.66F	1.41A
T ₅	64.33C	17.66D	80.55C	18.83E	1.44A
T ₆	72.80A	24.96A	93.85A	23.54B	1.51A
T ₇	54.35FG	14.25H	72.77F	14.99I	1.37A
T ₈	58.85DE	14.86G	75.83E	15.53H	1.39A
T ₉	65.42C	19.68C	82.66C	19.68D	1.46A
T ₁₀	73.95A	25.22A	95.45A	24.15A	1.54A
T ₁₁	68.53B	22.12B	88.67B	21.75C	1.49A
	Second Season				
T ₁	58.43D	15.93E	75.53D	15.55G	1.39A
T ₂	51.05G	12.66J	62.34H	13.32K	1.33A
T ₃	55.06EF	13.49I	65.90G	13.78J	1.35A
T ₄	59.69D	15.24F	76.18D	15.99F	1.40A
T ₅	62.44C	17.31D	78.29C	18.08E	1.43A
T ₆	70.62A	24.46A	90.10A	22.60B	1.49A
T ₇	52.72FG	13.97H	69.86F	14.39I	1.36A
T ₈	57.08DE	14.56G	72.80E	14.91H	1.38A
T ₉	63.46C	19.29C	79.35C	18.89D	1.45A
T ₁₀	71.73A	24.72A	91.61A	23.18A	1.52A
T ₁₁	66.47B	21.68B	85.12B	20.88C	1.48A

Chemical Contents of Leaves

Illustrated data in Table (2) showed the effect of applied chitosan (spray or adding to soil) on the chemical contents of leaves (N, P, K and chlorophyll reading). The greatest contents in leaves from those parameters were observed with application treatments 80% recommended doses from "N" chemical fertilizer + chitosan adding to soil 250 ppm "T10" and 80% recommended doses from "N" chemical fertilizer + chitosan spray 300 ppm "T6", respectively, without any significant differences compared to other treatments. Although, the application of 80% recommended doses from "N"

chemical fertilizer + chitosan spray 100 ppm "T2" led to reduced it.

Yield and its components

Presented data in Tables (3 and 4) indicated the effect of using chitosan as supplement fertilizer yield and its components such as (number of fruits/plant, average fruit weight fruits), early yield, total yield and fruit quality i.e., (total protein, dietary fiber, phosphorus and potassium).

Both tables observed that applying a high concentration of chitosan increased yield and

enhanced fruit quality more than the grower treatment (control). The highest yield and quality were obtained with okra plants applied by 80% recommended doses from "N" chemical fertilizer + chitosan adding to soil 250 ppm "T10" and 80% recommended doses from "N" chemical fertilizer + chitosan spray 300 ppm "T6" treatments, without any significant differences. When, applied 80% recommended doses from "N" chemical fertilizer +

chitosan spray 100 ppm "T2" treatment reduced values of all tested yield and its components parameters. Furthermore, control treatment (100% recommended doses from chemical fertilizer (control treatment) "T1") replaced sixth rank after 80% recommended doses from "N" chemical fertilizer + chitosan spray 200 ppm "T4", without any significant differences.

Table (2): Effect of using chitosan as supplement fertilizer by spraying or adding to soil on chemical contents of leaves chlorophyll reading (SPAD), N, P and K (%) of okra plants through 2020 and 2021 seasons.

Treatments	Chlorophyll reading	N	P	K
T ₁	40.75E	1.65D	0.39D	1.68D
T ₂	31.20I	1.43G	0.31G	1.52G
T ₃	33.34H	1.49F	0.33F	1.57F
T ₄	41.95E	1.67D	0.40D	1.70D
T ₅	45.87D	1.73C	0.42C	1.74C
T ₆	56.53A	1.88A	0.47A	1.85A
T ₇	35.42G	1.53F	0.34F	1.59F
T ₈	39.65F	1.59E	0.37E	1.64E
T ₉	49.84C	1.79B	0.44B	1.78B
T ₁₀	57.76A	1.92A	0.48A	1.87A
T ₁₁	53.27B	1.82B	0.45B	1.81B
Second Season				
T ₁	39.12E	1.58D	0.37D	1.61D
T ₂	29.95I	1.37G	0.30G	1.46G
T ₃	32.01H	1.43F	0.32F	1.51F
T ₄	40.27E	1.60D	0.38D	1.63D
T ₅	44.04D	1.66C	0.40C	1.67C
T ₆	54.27A	1.80A	0.45A	1.78A
T ₇	34.00G	1.47F	0.33F	1.53F
T ₈	38.06F	1.53E	0.36E	1.57E
T ₉	47.85C	1.72B	0.42B	1.71B
T ₁₀	55.45A	1.84A	0.46A	1.80A
T ₁₁	51.14B	1.75B	0.43B	1.74B

DISCUSSION

In this study obtained that using chitosan as a supplement fertilizer in high concentration had the best impact on improving and increasing all evaluated parameters of okra crop i.e., (plant height, number of leaves, fresh and dry weights of leaves, chlorophyll reading in leaves, content percentage from N, P and K in leaves, number of fruits/plant, average fruit weight fruits, early yield/plant, total yield/plant, total protein, dietary fiber, phosphorus and potassium).

This increase may be due to the use of chitosan which increases the activity of key enzymes of nitrogen metabolism (nitrate synthesis, glutamine synthetase, proteases), improves nitrogen transport in functional leaves, and promotes plant growth (Chibu and Shibayama, 2003; Shehata et al., 2012). Also, Hafez et al., 2019 found that chitosan application played a positive role in promoting functional leaf nitrogen transport, which promoted plant growth. Furthermore, Mondal and Malek et al., 2012 stated that vegetative growth parameters of okra increased with increasing chitosan application concentration. On the other hand, chitosan facilitated plant growth

by treating the plants with necessary mineral elements, which the plants could not supply in sufficient supply, probably due to soil problems or the supply of certain necessary amino compounds to the plants (Chibu and Shibayama, 2003). In addition, plants grew better due to improved root growth and

greater root spread in the soil (Zubaidi and Zainab, 2016).

Table (3): Effect of using chitosan as supplement fertilizer by spraying or adding to soil on number of fruits/plant, average fruit weight (g), early yield/plant (g) and total yield/plant (Kg) of okra plants through 2020 and 2021 seasons.

Treatments	Number of fruits	Average fruit weight	Early yield/plant	Total yield/plant
T ₁	60.78E	3.05E	188.65E	2.850E
T ₂	50.00I	2.32H	110.38I	2.150I
T ₃	52.64H	2.54G	122.24H	2.335H
T ₄	61.28E	3.08E	192.50E	2.995E
T ₅	64.58D	3.32D	215.12D	3.162D
T ₆	74.92A	3.86A	298.85A	3.724A
T ₇	55.15G	2.58G	131.28G	2.510G
T ₈	57.23F	2.83F	162.47F	2.680F
T ₉	68.42C	3.55C	235.34C	3.388C
T ₁₀	75.76A	3.98A	300.79A	3.853A
T ₁₁	72.55B	3.62B	275.29B	3.558B
Second season				
T ₁	57.74E	2.93E	182.991E	2.793E
T ₂	47.50I	2.23H	107.069I	2.107I
T ₃	50.01H	2.44G	118.573H	2.288H
T ₄	58.22E	2.96E	186.725E	2.935E
T ₅	61.35D	3.19D	208.666D	3.099D
T ₆	71.17A	3.71A	289.885A	3.650A
T ₇	52.39G	2.48G	127.342G	2.460G
T ₈	54.37F	2.72F	157.596F	2.626F
T ₉	65.00C	3.41C	228.280C	3.320C
T ₁₀	71.97A	3.82A	291.766A	3.776A
T ₁₁	68.92B	3.48B	267.031B	3.487B

Chitosan has various functional groups such as hydroxyl groups and amine groups, and because it binds to metal ions by chemisorption or physical adsorption, it has a high adsorption capacity for various metal ions. Chitosan works with metallic elements because it fits the basic natural properties of multiple cations. Chitosan stimulates the activity of key enzymes in nitrogen metabolism and improves nitrogen transfer to leaves. This stimulates leaf function in growth and development. In addition, chitosan is a polysaccharide that is very important for plant defense and yield increase in plant nutrition, especially in horticulture. It causes a doubling of photosynthesis. The action of chitosan molecules

varies from cell to cell and depends on their physiochemistry. It leads to increased root mass, flowering and final production (Abdel-Mawgoud et al., 2010; Monirul et al., 2018; Al-Hassani and Majid, 2019). Furthermore, chitosan had a positive effect on root effectiveness and nutrient uptake, leading to increased photosynthetic efficiency and carbohydrate and sugar production, resulting in increased internode growth and length and food accumulation led to an increase in stem diameter (Monirul et al., 2018).

Also, this result is harmony with Mondal et al., 2013, who, indicated that photosynthesis and chlorophyll in cowpea plants are increased by chitosan treatment,

which also influences the increase in biomass to water content by reducing transpiration (Bittelli et al., 2001). In addition, chitosan played an important role in plant nutrition and also had positive effects on growth rate, plant properties, and increasing metallic elements (Mondal and Malek, 2012; Mondal et al., 2013). Similarly, Guan et al., 2009 have shown that, this increases the availability and uptake of water and essential nutrients by modulating cellular osmotic pressure, and the release of harmful free radicals by increasing antioxidants and enzymatic activity. We

believe this is due to a decrease in accumulation. Malerba and Cerana, 2016 reported that chitosan application enhances leaf chemistry, promotes water and nutrient uptake by vigorous roots, combats oxidative damage by reactive oxygen species (ROS), and enhances antioxidant enzymes. Furthermore, found that it can activate both defense systems and photosynthetic enzymes. Photosynthesis and biosynthesis of essential organic molecules are improved, increasing the accumulation of assimilates.

Table (4): Effect of using chitosan as supplement fertilizer by spraying or adding to soil on total protein (%), dietary fiber (%), phosphorus (%) and potassium (%) of okra fruits through 2020 and 2021 seasons.

Treatments	Total protein	Dietary fiber	Phosphorus	Potassium
T ₁	3.58E	2.18A	0.68E	2.42E
T ₂	3.24I	1.55B	0.52I	2.04I
T ₃	3.33H	1.52C	0.57H	2.13H
T ₄	3.62E	1.37F	0.70E	2.44E
T ₅	3.83D	1.34G	0.74D	2.53D
T ₆	4.54A	1.24I	0.84A	2.82A
T ₇	3.44G	1.46D	0.60G	2.22G
T ₈	3.52F	1.41E	0.64F	2.33F
T ₉	4.22C	1.30H	0.78C	2.64C
T ₁₀	4.57A	1.25I	0.86A	2.88A
T ₁₁	4.35B	1.29H	0.82B	2.73B
Second season				
T ₁	3.47E	2.09A	0.65E	2.40E
T ₂	3.14I	1.49B	0.50I	2.02I
T ₃	3.23H	1.46C	0.55H	2.11H
T ₄	3.51E	1.32F	0.67E	2.42E
T ₅	3.72D	1.29G	0.71D	2.50D
T ₆	4.40A	1.19I	0.81A	2.79A
T ₇	3.34G	1.40D	0.58G	2.20G
T ₈	3.41F	1.35E	0.61F	2.31F
T ₉	4.09C	1.25H	0.75C	2.61C
T ₁₀	4.43A	1.20I	0.83A	2.85A
T ₁₁	4.22B	1.24H	0.79B	2.70B

Furthermore, the enhancement at yield and its components parameters attribute to chitosan-containing plants were the best in most traits of vegetative growth and had a high proportion of mineral matter, which allowed the plants to build carbohydrate matter and high carbonation, reflected in an increase in average fruit weight (Al-Hassani and Majid, 2019). Application of chitosan increased all vegetative growth traits and yields and their constituents (Hafez et al., 2019). The important effects of chitosan on yield and its composition are

likely due to the fact that chitosan has mimetic effects on physiological processes, improving nitrogen transport in functioning leaves and improving vegetative growth and development (Gornik et al., 2008).

The vast impact of chitosan is probably because of that chitosan is a brand new plant boom promoter which include GA3 that can be have impact at the plant boom and yield (El-Bassiony et al., 2014). On the other hand, for the impact of chitosan on macro and

micro elements (N, P, K, Fe, Zn, Cu and B), chitosan may be used as a remedy for mineral factors infected soil (Sheikha and Al-Malki, 2011). Also, the main position of chitosan in ameliorating plant roots potential to uptake water and vital elements and use them correctly inside plant in promoting of antioxidant enzyme sports, prevention of reactive oxygen species (ROS), activation of photosynthesis and biosynthesis of carbohydrates, proteins and different natural compounds that wanted for distinct plant metabolic sports and manufacturing of greater assimilates which translocated to end result in result of K and P mode of action, and there for the marketable end result yield and first-rate increased (El-saady, 2016). Moreover, the effective effect of chitosan on chemical additives leaves in phrases of photosynthetic pigments and NPK elements can be ascribed to the chitosan-mediated enhance root device performance to soak up greater to be had water and nutrients that wanted for critical physiological sports which include photosynthesis and biosynthesis of critical assimilates which ameliorate leaves chemical first-rate (Malekpoor et al., 2016).

In addition, chitosan increases the uptake and availability of water and essential nutrients by regulating intracellular osmotic pressure, promoting plant growth. Over the past decade, the signaling mechanisms of chitosan and its derivatives that regulate plant growth and developmental processes have been studied. Initial results show that chitosan helps activate the hydrolytic enzymes required to degrade and mobilize stored food materials such as starch and protein. Chitosan promotes root cell division by activating plant hormones such as auxin and cytokinin, further increasing nutrient intake. Furthermore, the increased yield can be attributed to chitosan's plant growth-promoting activity may be directly related to its effects on plant physiological mechanisms such as nutrient uptake, cell division, cell elongation, enzyme activation, and protein synthesis (Amin et al., 2007). In addition, Chissan improved the photosynthetic index by improving stomatal function and chlorophyll content, and also significantly increased crop yield. Polycationic chitosan increases the osmotic pressure of stomatal cells, resulting in increased stomatal opening and CO₂ uptake (Chakraborty et al., 2020).

On other hand, soil-applied chitosan also significantly enhanced seedling growth and induced early flowering in many ornamental plants (Pichyangkura and Chadchawan, 2015). Similarly, Xu and Mou (2018)

found that applying chitosan to soil increased lettuce leaf number, area, fresh weight, dry weight, and chlorophyll index. Suppression of plant diseases, insects and nematodes, increased biomass and beneficial microbial activity, high nitrogen and calcium content, improved soil physical structure and nutrient availability, direct stimulation of plant growth the synergistic effect of many factors, may play a role derived from chitosan as a soil conditioner.

Furthermore, addition of chitosan alters rhizosphere conditions, shifting the microbial balance in favor of beneficial organisms and against plant pathogens (Sharp, 2013). Chitosan provides a carbon source for soil microorganisms, promotes the conversion of organic matter to inorganic matter, and helps roots to absorb more nutrients from the soil (Xu and Mou, 2018).

Chitosan and all other chitin derivatives have a high nitrogen content of 6% to 9%. Plants can access nitrogen in chitin through microbial degradation and release of inorganic nitrogen, or by direct uptake of monomers as organic nitrogen (Roberts and Jones, 2012). Chitosan can be used to add organic matter to soil without increasing the carbon-to-nitrogen ratio. In addition to nitrogen, chitosan also contains large amounts of calcium minerals that provide structural strength to crustacean exoskeletons (Boßelmann et al., 2007). Although chitosan contains nitrogen and calcium, its beneficial effects on plant growth and yield are not due to that nutrient alone, and some studies have shown that the control plots treated with mineral fertilizers have been shown to contain chitosan nutrients was balanced.

Chitosan significantly enhanced the seedling growth of several plants compared to conventional mineral fertilizers. Due to its cationic properties, chitosan is also suitable as a vehicle to provide additional essential nutrients (Sharp, 2013). Hydroxyl and amino functional groups on deacetylated chitosan allow the formation of coordination compounds with ions such as copper, zinc, iron, but not alkali metals (e.g. potassium) or alkaline earth metals (e.g. calcium or magnesium) is not possible (Ramírez et al., 2010). For this reason, chitosan is a sustainable alternative to synthetic chelators such as ethylenediaminetetra acetic acid, which are routinely used to supply iron and other nutrients to overcome their low solubility in calcareous/neutral soils. It has become a viable alternative (Strawn et al., 2019). Due to its high molecular weight and porous structure, chitosan

forms a gel that absorbs large amounts of water and enhances the water-holding capacity of soil (Jamnongkan and Kaewpirom, 2010).

Application of chitosan to soil increased levels of nitrogen, phosphorus, potassium, total sugars, soluble protein and total amino acids (Farouk et al., 2011). Chitosan application to soil has been reported to increase chlorophyll content in leaves of many crops (Farouk et al., 2011; Sheikha and Al-Malki, 2011). As a bio-stimulant, chitosan may also enhance the fluorescence of chlorophyll and enhance the photosynthetic rate.

Furthermore, the use of chitosan as a bio-stimulator in plant development can increase leaf and shoot size. Chitosan has been found to exert molecular effects on flowers, directly affecting growth and physiological parameters (Salachna and Zawadzińska, 2014).

CONCLUSION

From this investigation indicated that application chitosan by high concentration started by 200 ppm have a positive role for increasing and enhancing all tested parameters of okra plants such as (vegetative growth, chemical contents in leaves, yield and fruits quality. On other hand, the suitable concentration depending on method applies of chitosan. The greatest values of almost i.e., vegetative growth, chemical contents in leaves, yield and best fruits quality parameters were obtained with applied 80% recommended doses from chemical fertilizer + chitosan adding to soil 250 ppm "T10" and 80% recommended doses from chemical fertilizer + chitosan spray 300 ppm "T6" treatments, without any significant differences followed by 80% recommended doses from chemical fertilizer + chitosan adding to soil 300 ppm "T11", 80% recommended doses from chemical fertilizer + chitosan adding to soil 200 ppm "T9", 80% recommended doses from chemical fertilizer + chitosan spray 250 ppm "T5" and 80% recommended doses from chemical fertilizer + chitosan spray 200 ppm "T4" treatments, respectively. More that, control treatment (100% Recommended doses from chemical fertilizer (control treatment) "T1") placed the six place.

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