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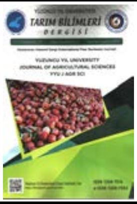

Research Article (Araştırma Makalesi)

The Effects of Magnetic Compounds on Growth and Yield of Cucumber under Greenhouse Conditions

Mohammad AHMADI[1], Sahebali BOLANDNAZAR[2], Jaber PANAHANDEH[3], Seyedeh Somayyeh SHAFIEI MASOULEH[*4]

[1,2,3]Department of Horticultural Sciences, Faculty of Agricultural Sciences, University of Tabriz, Tabriz, Iran
[4]Department of Genetics and Breeding, Ornamental Plants Research Center (OPRC), Horticultural Sciences Research Institute (HSRI), Agricultural Research, Education and Extension Organization (AREEO), Mahallat, Iran
[1]https://orcid.org/0000-0002-5476-604x [2]https://orcid.org/0000-0001-9396-7373 [3]https://orcid.org/0000-0001-7788-5300
[4]http://orcid.org/0000-0001-5455-0970
*Corresponding author e-mail: shafiee.masouleh@areeo.ac.ir; shafyii@gmail.com

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Cucumis sativus,
Growth,
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Yield

Abstract: This study was performed to examine the effects of different concentrations of magnetic nano-chitosan (MNC) and nano-Fe_3O_4 (NF) on the growth and yield of cucumber (*Cucumis sativus* L. cv. 'Negin'). The sizes of nano-particles were in the range of 20 nm to 50 nm, and their concentrations ranged between 10 to 100 mg L^{-1}. The experiment was designed with nine treatments of MNC (10, 25, 50 and 100 mg L^{-1}), NF (10, 25, 50 and 100 mg L^{-1}) and one control treatment (iron chelate; IC: 60 mg L^{-1}). The results showed that the foliar application of MNC and NF could improve yield and plant growth in cucumber, as well as iron chelate. The results of the present study suggest that the foliar application of nano-fertilizers similar to iron chelate could improve the plant growth and yield of the greenhouse cucumber. As regards toxicity of iron chelates, especially in high doses in the soil, the application of MNC may be safer in sustainable agriculture and even could be better than NF, because it has the combined effects of magnetism, chitosan and being nano-size.

Manyetik Bileşiklerin Sera Koşullarında Hıyarın Büyüme ve Verimine Etkileri

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Anahtar kelimeler

Cucumis sativus,
Büyüme,
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Öz: Bu çalışma, farklı konsantrasyonlarda manyetik nano-kitosan (MNK) ve nano-Fe_3O_4 (NF)'nin hıyarın (*Cucumis sativus* L. cv.' Negin') büyüme ve verimi üzerindeki etkilerini incelemek için yürütülmüştür. Nano parçacıkların boyutları, 20 nm ile 50 nm aralığında ve konsantrasyonları 10 ile 100 mg/L arasında yer almıştır. Denemede, MNK (10, 25, 50 ve 100 mg/L), NF (10, 25, 50 ve 100 mg/L) uygulamaları ve bir kontrol uygulaması (demir şelat; IC: 60 mg/L) dahil toplam dokuz uygulama yer almıştır. Sonuçlar, demir şelatta olduğu gibi MNK ve NF'nin yapraktan uygulanmasının, hıyarda verim ve bitki büyümesini iyileştirebileceğini göstermiştir. Mevcut çalışmanın sonuçları, demir şelata benzer şekilde nano gübrelerin yapraklara uygulanmasının, sera hıyarının bitki büyümesini ve verimini artırabileceğini önermektedir. Özellikle topraktaki yüksek dozlarda demir şelat toksisitesi ile ilgili olarak, MNK'nin uygulanması, sürdürülebilir tarımda daha güvenli ve hatta manyetizma, kitosan ve nano boyutlu olmanın birleşik etkilerine sahip olduğu için NF'den daha iyi olabilecektir.

1. Introduction

Cucumber is one of the main vegetables in Iran that is predominantly cultivated in plastic greenhouses also it constitutes the fourth most important vegetable crop after tomatoes, cabbage, and onion in Asia (Eifediyi and Remison, 2010). Correspondingly, the increasing world population requires an equal increase in food production. Therefore, to add to existing food production levels, it is necessary to employ a range of diverse technologies in agriculture.

The fertilizers have an important role in increasing food production, and particularly the foliar application of micronutrients has a positive effect in increasing fruit yield and quality. Researchers have indicated that foliar spraying of macro and micronutrients have been used as a means to improve the yield and quality of fruits (Kumar et al., 2014). The fertilizers derived from nanotechnology in agriculture have garnered excessive attention and have a fast-growing field. Interestingly, nanotechnology has been used in different agricultural fields such as production, packaging, and transport of agricultural products (De Rosa et al., 2010; Baruah and Dutta, 2009; Srilatha, 2011). Nano-fertilizers simply dissolve in solution and release the nutrient(s) as soluble ions. Furthermore, the use of nano-fertilizers leads to increased efficiency of the elements and improved growth, with improved fruit set and yield. This potential contribution of nano-fertilizers in improving growth and yield is derived from greater absorbance and high reactivity.

Iron is an essential element for plants that plays a critical role in oxidizing and reducing systems. In addition, iron is a basic element in the synthesis of chlorophyll, which is essential for the maintenance of chloroplast structure and function (Sawan et al., 2001). Iron deficiency is a major problem in plants that result in the yellowing of leaves, poor growth, and reduced yield. During iron deficiency, chlorophyll production in leaves decreases and this is the primary cause of leaf chlorosis. Correspondingly, in proper concentrations, magnetic fluids play a positive effect on photosynthesis capacity, and similarly, iron ions in the structure of magnetic fluids play a significant role in plant metabolism (Răcuciu and Creangă, 2007; Shafiee-Masouleh et al., 2014).

Chitosan is one of the biodegradable compounds, which is a deacetylated form of chitin. It exists abundantly as a natural polymer that is derived from crustaceous shells such as crabs and shrimps. Moreover, chitosan is biocompatible and non-toxic compound and used in the formulation of slow-release fertilizers and late-degradable (Wu and Liu, 2008). Several studies have evidenced foliar-applied chitosan promoted growth and yield of various crops such as bean plants (Sharifa, 2013), cabbage (Hirano, 1988), radish (Farouk et al., 2008), and cucumber (Ghoname *et al*., 2010). Similarly, *in vitro* use of chitosan showed an increase in photosynthesis in *Vitis vinifera* L. (Barka et al., 2004). Shafiee-Masouleh et al. (2014) used the magnetic nano-chitosan and carboxymethyl chitosan in various concentrations with a nutrient solution of lily during yearling bulblets and forcing sized bulbs production, and they proposed 15 mg L^{-1} magnetic nano-chitosan (the highest assayed concentration) for increasing size of bulblets and bulbs. In another study, Hatamzadeh and Shafiei-Masouleh (2019) applied both compounds during forcing of lily bulbs to produce cut flowers and reported that both the compounds at 10 and 15 mg L^{-1} had the highest effects of the soluble carbohydrates and amylases of flower buds without toxicity and for morphological effects needed to use higher concentrations. In a study on artichoke, between three levels of chitosan (0, 150, or 300 ppm), the highest concentration caused the more growth such as head yield (Elsharkawy and Ghoneim, 2019). They explained the reason on the effect, the direct and indirect role of chitosan on plant physiology. Xu and Mou (2018) reported the highest photosynthesis rate and gas exchanges with highest concentration of chitosan. Therefore, chitosan effects on plant growth can be well interpreted as before mentioned.

Up to now, the effects of foliar application of chitosan as nano-magnetic compound and nano-Fe_3O_4 were not studied in the soil culture of cucumber in the greenhouse. Therefore, this study aimed to determine the effects of different concentrations of magnetic nano- chitosan and nano-Fe_3O_4 as a foliar spray on the growth and yield of cucumber (*Cucumis sativus* L. cv. 'Negin') compared to a usual iron chelate that is applied in farms and greenhouses.

2. Material and Methods

2.1. Plant materials

The study was carried out at the Agricultural Research Center of Yazd (31° 49' 9.65" N, 54° 21' 52.30" E) , Iran. Cucumber (cv. Negin) plants were planted under a glass greenhouse. Plants were grown with natural light conditions, approximate day/night temperatures of 26/16ºC, and 60% relative humidity during the experiment. The study was conducted based on a completely randomized design with totally nine treatments, including magnetic nano-chitosan (MNC: 10, 25, 50, and 100 mg L^{-1}), nano-Fe_3O_4 (NF: 10, 25, 50, and 100mg L^{-1}) and one control treatment (IC: 60 mg L^{-1}) and three replications. Seeds were sown in 48-cell seed germination trays filled with peat and perlite (V:V; 50:50). Then, each seedling was transferred to a pot whose size was 55 ×35 × 20 cm. Some of the physicochemical properties of the soils are shown in Table 1. The N-P-K fertilizers were applied in 4 g L^{-1} (20-20-20; vegetative phase) and 5 g L $^{-1}$ (15-5-30; reproductive phase). For the control plants, 6% chelated iron was used.

Table 1. Physical and chemical characteristics of the soil.

Organic matter (%)	pH	Bulk density (g cm^{-3})	Sand (%)	Silt (%)	Clay (%)	EC (ds m^{-1})	Field capacity (%)	Extractable DTPA-Fe (mg Kg^{-1})
1.06	8.1	1.2	52	34.5	13.5	1.7	24	4.9

2.2. Preparation of magnetic nano-chitosan and nano-Fe_3O_4 and foliar application

The preparation of the nano-particles of Fe_3O_4 was implemented according to Kang et al. (1996). Briefly, concentrated HCl (37%) and deionized water (30 mL) were deoxygenated by N_2 for 45 mins. $FeCl_3$ (5.4 g) and $FeCl_2.4H_2O$ (3.25 g) were added and dissolved completely. This solution was dripped to NaOH (1.5 M, 250 mL) within one hour at the deoxygenated conditions with N_2, and the end of the reaction, the black precipitate was formed. The product was washed with distilled water and dispersed in deionized water and kept at 4 °C up to drying at 55°C. To analyze the size and morphology of nano-Fe_3O_4, SEM (Scanning Electron Microscopy, XL-30, Philips, Eindhoven, USA) was used (Figure 1a). For the preparation of magnetic nano-chitosan, Fe_3O_4 by carboxymethyl chitosan (see its preparation in Shafiee-Masouleh et al. (2014)) was encapsulated according to Chang and Chen (2005). The size and morphology of the nanoparticles were analyzed with SEM (Figure 1b). The mentioned concentrations of each compound were separately sprayed on the plant leaves and fruits with a commercial hand-held sprayer. Sprays were repeated at weekly intervals, starting from two weeks after transplanting. The plants were sprayed enough to cover all of leaves.

a b

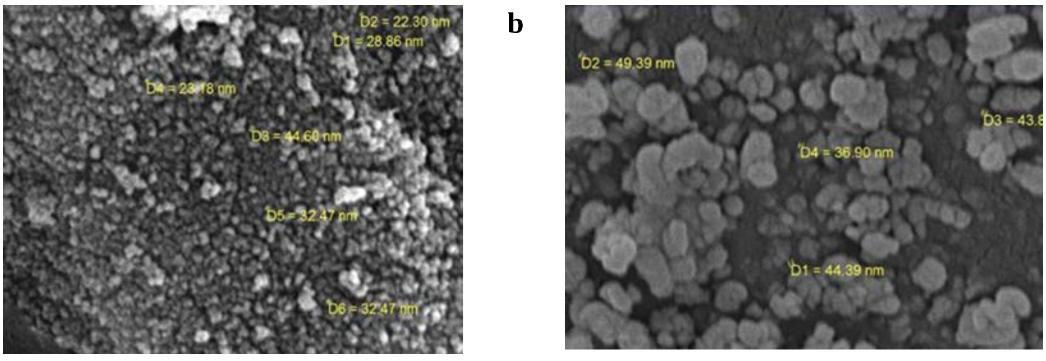

Figure 1. SEM images of nanoparticles; size and morphology of a) nano-Fe_3O_4 and b) magnetic nano-chitosan.

2.3. Measurement of plant physical characteristics

The dry weight, after drying in an oven at 70°C for 48 h, was measured for the shoot (leaves and stem). The leaf area of the plant was recorded with a leaf area meter (Model AM 100-002). The chlorophyll content was measured with the SPAD (Spad-502 plus, Konica Minolta, Tokyo, Japan). The plant height was recorded on the day of the harvest with a measuring tape.

2.4. Measurement of fruit physical and chemical characteristics

A sample of 10 mature fruits was taken to measure physical quality parameters such as diameter (cm), length (cm), and fruit shape (L/D). Fruit juice was extracted to analyze the total soluble solids and titratable acidity (Xie et al., 2011). The total soluble solids were measured using a hand refractometer at room temperature (Zhu et al., 2017).

2.5. Statistical analysis

The variance analysis was conducted using SAS software 9.1 and the means were compared with Duncan's multiple range test at the levels of $p \leq 0.01$ or $p \leq 0.05$, according to the variance analysis.

3. Results

3.1. Growth and yield

Variance analysis of different concentrations of MNC and NF with control treatment (60 mg L^{-1} iron chelate) had significant effects ($p \leq 0.01$) on the yield and shoot dry weight and also significant effect ($p \leq 0.05$) on the leaf area, chlorophyll content and fresh weight (Table 2). The effects of magnetic nano-chitosan and nano-Fe_3O_4 foliar application on yield and growth parameters are reported in Table 3. The application of MNC significantly influenced on growth characters (leaf area, shoot fresh and dry weights). The highest shoot fresh weight (565.60 g) was recorded by foliar application of MNC at a rate of 50 mg $L^{-1}$and the lowest shoot fresh weight (413 g) was found in treatment with 100mg L^{-1} MNC and there were significant differences ($p < 0.05$) between the two treatments. The application of MNC significantly improved the shoot dry weight, and the maximum shoot dry weight was observed in MNC50 that was non-significant with IC. The minimum shoot dry weight was observed in MNC10. The highest leaf area (645.00 cm^2 $plant^{-1}$) was recorded by foliar spraying of MNC at the concentration of 25 mg L^{-1}. However, the lowest leaf area (395.00 cm^2 $plant^{-1}$) was observed in NF100.

Table 2. Variance analysis of yield and growth parameters of cucumber (*Cucumis sativus* L. cv. 'Negin') under the application of magnetic nano-chitosan and nano-Fe_3O_4.

Source of variation	df	Yield	Leaf area	Plant height	Shoot fresh weight	Shoot dry weight	Chlorophyll content (SPAD)
Treatments	8	0.238**	23219*	0.26ns	7221.3*	9.93**	50.08*
Error		0.064	8782.7	0.30	2726.7	2.32	17.77
CV(%)		6.540	16.8	15.64	10.3	10.01	10.51

* and ** indicate significant differences at 5 and 1% probability levels, respectively.

The effect of foliar application with different concentrations showed that the highest and the lowest yields were obtained from plant sprayed with MNC50 and NF10 (4.35 and 3.40 kg $plant^{-1}$), respectively. The results showed the application of MNC in some characteristics was not significant compared to other treatments, but it seems that 50 mg L^{-1} NMC is more effective compared to the other treatments. Chlorophyll content was significantly affected by MNC and NF treatments. At the end of the growth period, the relative amount of chlorophyll increased up to 46.00when plants were treated with 100 mgL^{-1} MNC, while chlorophyll content was lowest (35.00) for plants that received 10 mgL^{-1} NF.

Table 3. Effects of magnetic nano-chitosan and nano-Fe_3O_4 on yield and growth parameters.

Treatments	Yield (kg $plant^{-1}$)	Leaf area (cm^2 $plant^{-1}$)	Plant height (m)	Shoot fresh weight (g)	Shoot dry weight (g)	Chlorophyll content (SPAD)
IC	4.15 ab	610.00 ab	3.38 ab	523.60 ab	17.53 ab	37.50 bc
NF10	3.40 c	600.00 ab	3.96 a	524.40 ab	13.66 c	35.00 c
NF25	3.80 abc	560.00 abc	3.70 ab	512.20 ab	14.20 bc	36.00 c
NF50	3.85 abc	470.00 abc	3.60 ab	483.10 ab	15.86 ab	44.50 ab
NF100	3.50 bc	395.00 c	3.10 b	470.00 ab	13.63 c	39.90 abc
MNC10	3.65 bc	450.00 bc	3.60 ab	467.00 ab	12.73 c	35.80 c
MNC25	3.90 abc	645.00 a	3.20 b	562.00 a	16.13 ab	38.00 bc
MNC50	4.35 a	620.00 a	3.90 a	565.60 a	18.00 a	41.00 abc
MNC100	3.65 c	555.00 abc	3.33 ab	413.00 b	15.23 abc	46.00 a

Mean separation by a differ letter in column are different based on Duncan’s multiple range test ($p < 0.05$).

3.2. Fruit quality

Variance analysis of fruit parameters under the effects of NF and MNC foliar application are presented in Tables 4 and in Table 5. It seems that more concentrations in both compounds should need to achieve the significant effects on parameters of fruit quality.

Table 4. Variance analysis of cucumber fruit parameters under the effect of treatment with magnetic nano-chitosan and nano-Fe_3O_4.

Source of variation	df	TSS	TA	Fruit diameter	Fruit length	Fruit shape (L/D)
Treatments	8	0.10^{ns}	0.01^{ns}	0.049^{ns}	0.57^{ns}	0.12^{ns}
Error		0.19	0.06	0.064	1.87	0.48
CV (%)		13.2	27.3	8.32	8.38	12.88

^{ns} non- significant.

Table 5. Effect of magnetic nano-chitosan and nano-Fe_3O_4 on fruit parameters.

Treatments	TSS (%)	TA (%)	Fruit diameter (cm)	Fruit length (cm)	Fruit shape (L/D)
IC	3.23 a	0.90 a	3.30 a	16.43 a	5.02 a
NF10	3.2 a	0.95 a	2.97 a	16.06 a	5.44 a
NF25	3.53 a	1.02 a	3.10 a	16.66 a	5.46 a
NF50	3.33 a	0.91 a	3.03 a	15.33 a	5.06 a
NF100	3.2 a	1.01 a	2.86 a	16.25 a	5.68 a
MNC10	3.1 a	0.93 a	3.00 a	15.86 a	5.29 a
MNC25	3.66 a	1.00 a	3.16 a	17.03 a	5.43 a
MNC50	3.13 a	0.82 a	3.03 a	15.66 a	5.18 a
MNC100	3.16 a	1.06 a	2.95 a	15.83 a	5.39 a

Mean separation by a differ letter in column are different based on Duncan’s multiple range test ($p < 0.05$).

4. Discussion and Conclusion

In the present study, the obtained data revealed that foliar sprays with MNC have similar effects with control treatment in terms of growth and yield of cucumber. The highest values of yield, the fresh and dry weights of shoots were recorded by foliar spraying of MNC at a concentration of 50 mg L^{-1}.

Improving the growth and productivity of various crops by application of chitosan was recorded by several studies such as on cabbage (Hirano, 1988), *Lilium* (Shafiee-Masouleh et al., 2014), grape (Gornik et al., 2008), sweet basil (Kim, 2005), strawberry (Abd El Mawgoud et al., 2010) and sweet pepper plants (Ghoname et al., 2010). According to Shafiee-Masouleh et al. (2014), it seems that chitosan has a role to increase the photosynthesis by promoting the nutrient element uptake, and then this would increase growth and yield of the plant. In a study, it was reported that the vegetative growth and leaf chlorophyll content of tomato improved by the application of chitosan, as well as goal manure and farmyard manure (El Tantawy, 2009). He stated that the effect of chitosan is due to increasing immunity system, photosynthesis pigments and photosynthesis rate, as well as promoting growth. Significant improvement has been observed by the application of different concentrations of chitosan in growth rates and yield of Chinese cabbage (Chandrkrachang et al., 2003). The positive influence of chitosan treatment in the yield was observed in soybean sprouts (Lee et al., 2005). They observed the highest growth of sprouts (hypocotyl and root length) in the seeds that treated with high molecular weight of chitosan as soaking, compared to low and medium molecular weight of chitosan and the control. They said that high molecular weight of chitosan increased the respiratory rate of seeds compared to other treatments. Therefore, being high molecular weight of chitosan cannot be explained that have effects on growth, but more respiratory rate increased the use of seed storage. Therefore, this is not in contrast with our results, which magnetic nano-chitosan with low molecular weight and in nano-size particles. Shafiee-Masouleh et al. (2014) reported that magnetic chitosan increased growth and development of bulb and yearling bulblets in *Lilium*. They found that treatment by 15 mg L^{-1} MNC compared to other concentrations, including 0, 1, 2.5, 5 and 10 mg L^{-1} in both of the experimented compounds (MNC and carboxymethyl chitosan (CMC)) and the control, has a positive and significant effects on fresh and dry weights of shoots and basal roots, diameter and fresh weight of bulblets. Mondal et al. (2012) reported that yield attributes and fruit yield increased significantly with increasing concentration of chitosan up to 100 ppm (the concentrations of 0, 50, 75, 100 and 125 ppm were investigated) in okra. Peyvendi et al. (2011) reported that foliar spray with nano-chelated iron (1, 3 or 5 kg ha^{-1}) compared to iron chelated fertilizer (1.5, 4.5 or 7.5 kg ha^{-1}) increased the mean dry and fresh weights of stems, roots, and leaves in levels less than 5 kg ha^{-1}. It was similar to the highest iron chelated fertilizer (7.5 kg ha^{-1}). Application of iron into the soil or on the foliage as chelate or sulfates improved the vegetative growth, yield, and fruit quality of various crops (Ma and Hong, 1998). It seems that some positive effects of nano-fertilizers in improving growth and yield of various crops are based on its ability for greater uptake by roots and high reactivity.

To conclude, according to the results, the significant effects of magnetic treatments towards the control are not evident in vegetative quality and high yield and also there is no significant difference for changing the biochemical structure of plants (quality parameters of fruits). In other words, organic and environment-friendly compounds had similar effects to iron chelate (control treatment), and in some cases it is more obvious to be towards the more effects. According to previous our results (Shafiee-Masouleh et al., 2014; Hatamzadeh and Shafiei-Masouleh, 2019), we achieved the best results at high concentrations of magnetic nano-chitosan, It seems that better results may achieve if higher concentrations of magnetic nano-chitosan, however, it needs to assay for a safe recommendation. Furthermore, it may be explained that the safe structure of magnetic nano-chitosan in making the nutritional and biochemical balance of the plant makes it an alternative to iron chelates and inorganic iron fertilizers in the near future. There are reports about toxicity of the chelated iron when apply in the soil and foliar applications in high doses (Siebach et al., 2015; Ren et al., 2018), but there is not any report about magnetic nano-compounds when apply in safe conditions, i.e. without magnetic fields. However, both of the use type, including in the soil or as a foliar application of magnetic nano-chitosan have indicated a safe range of application of nano-materials, and is the promising for sustainable agriculture. It could be concluded that MNC, as well as IC, is more effective than NF for yield and quantitative characteristics of cucumber under the experimental conditions. This could be due to the combined effects of MNC on supplying the iron source by magnetic core (magnetite) and increasing another element uptake for the plant by chitosan. This role of chitosan when uses as root application can be interpreted for its chelating role, but to know the cause of its effect in foliar application for increase in the growth (mechanism), more studies are needed. Furthermore, MNC was a suitable compound compared to NF. This research showed that nano-fertilizer could play an important role in the plant growth and yield of cucumber. Because both chitosan and magnetite are environment-friendly and low-cost compounds and for their synergic effects into one compound could be used as supplementary fertilizer to promote the efficiencies of other fertilizers.

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