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Research Article
Growth, Yield and Biochemical Changes of Soybean Plant in Response to Iron and Magnesium Oxide Nanoparticles

[1]Amany Abd El-Mohsen Ramadan, [1]Hala Mohamed Safwat El-Bassiouny, [2]Bakry Ahmad Bakry, [1]Maha Mohamed Shater Abdallah and [3]Magda Aly Mahmoud El-Enany

[1]Department of Botany, Agricultural and Biological Research Division, National Research Centre, 33 El Bohouth Street, P.O. Box 12622, Dokki, Giza, Egypt
[2]Department of Field Crops Research, Agricultural and Biological Research Division, National Research Centre, 33 El Bohouth Street, P.O. Box 12622, Dokki, Giza, Egypt
[3]Department of Cytology and Genetics, Genetic Engineering and Biotechnology Research Division, National Research Centre, 33 El Bohouth Street, P.O. Box 12622, Dokki, Giza, Egypt

Abstract

Background and Objective: New technology in agriculture focused on the use of nano fertilizers. Mineral oxides nanoparticles used in industries of fertilizers and therefore studying their impact on different plants were of concern. Minerals in forms of nanoparticles found to have the ability to enhance growth and yield of plants and helps on overcoming the adverse effects of soil conditions. **Materials and Methods:** Different foliar concentrations of Iron-oxide and magnesium-oxide nanoparticles materials (0, 10, 20, 30 and 40 ppm) were sprayed twice on soybean at 30 and 45 days after sowing. Plant samples were taken after 60 days from sowing for growth and biochemical parameters measurement. **Results:** There are improvements in growth parameters of soybean plants due to foliar spraying of both tested materials. The two tested nanoparticle materials increased significantly some biochemical constituents, photosynthetic pigments, compatible solutes, endogenous growth regulators and phenol yield and its attributes. Catalase content reached its maximum value with 30 ppm treatment concentrations of both tested materials. The values of seed components (protein and carbohydrate percentages) reflected the positive effect of using nanoparticles of tested materials, but such difference was not significant in case of oil (%). The SDS-PAGE banding patterns showed that tested nanoparticle materials induce slight differences between treatments. **Conclusion:** Both tested materials may act as nano-fertilizer sources that help the plant to overcome the adverse effects of such soil shortage in nutrients necessary for plant growth and development and accordingly its yield and quality.

Key words: Soybean, protein patterns, yield, iron oxide nanoparticles, magnesium oxide nanoparticles

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Corresponding Author: Amany Abd El-Mohsen Ramadan, Department of Botany, Agricultural and Biological Research Division, National Research Centre, 33 El Bohouth Street, P.O. Box 12622, Dokki, Giza, Egypt

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

In the third world countries, the increase of population and their needs results in the need for great efficient agricultural production system to meet their needs of food. Since agriculture considers the main crucial activity of the economy of such countries. However, changing environmental conditions introduce a challenge to the agriculture production, so that developing any agricultural system of the country is a must to meet its population needs.

Soybean (*Glycine max* L. Merr.) belongs to Fabaceae family and is an annual crop. Soybean has many important roles in human and animal nutrition due to it contains useful compounds, e.g. unsaturated fatty acids, protein, mineral salts as well as plant secondary metabolites such as isoflavin. Soybean grains are used for different purposes so it consumed at large scale all over the world. One of these uses is the production of soybean oil for human consumption. It is also used mainly in forms of feed, flour, soap, cosmetics, resins, paints, solvents and biodiesel[1]. Optimum quantity and quality of soybean seeds depend upon many critical important factors such as weed control and plant nutrition[2].

Nanotechnology is a new emerged technology during the last decade and promises to help agriculture and can lead to a new revolution[3]. Nanotechnology has been found to solve many agricultural problems with considerable advancement as compared to conventional agricultural means. Roco *et al.*[4] defined nanoparticles (Nano-scale particles = NSPs) as atomic or molecular aggregates with at least one dimension between 1 and 100 nm. This dimension can extremely change material physical-chemical properties compared to the bulk material[5]. As reviewed and discussed by Sheykhbaglou *et al.*[6], using of nanoparticles and Nano-powders can be useful in producing controlled or delayed releasing fertilizers. This feature may be because of nanoparticles characteristic of high reactivity. This reactivity resulted from its more specific surface area, more density of reactive areas, or increased reactivity of these areas on the particle surfaces. This take place throughout simplifying the absorption of fertilizers and pesticides that produced in nano scale.

From biological point of view, Singh *et al.*[7] stated that nanoparticles (NPs) are very attractive materials since it is very suitable in sensing and detection of biological structures and systems. Due to the unique physicochemical properties of NPs it has the potential to boost the plant metabolism[8]. Currently, scientists tried to minimize nutrient losses in fertilization and to increase the crop yield quantity and quality. This can be achieved through the exploitation of new applications of nanotechnology and nanomaterials. The advantages of nano-fertilizers or nano-encapsulated nutrients might be release of nutrients on demand, control the release of chemical fertilizers that regulate plant growth and enhance plant activity[9,10]. So that, agricultural application of NPs became an interesting area for minimizing the use of chemical fertilizers and improves growth and yield of crops[11-13]. Several predicted pathways for nanoparticle association and uptake in plants are exist. So, Tamilselvi *et al.*[14] reported that the key point to apply the nanotechnology to agriculture is analyzing the level of penetration and transportation mechanism of NPs in plant growth.

Various studies were carried out to understand the effect of nanoparticles on the growth of plants. For example, Iron as one of the essential elements for plant growth which plays a vital role in the photosynthetic reactions. Malakouti and Tehrani[15] reported that Iron activates several enzymes and contributes in RNA synthesis and improves the performance of photosystems. Soybean plant is sensitive to iron deficiency and efficiency of iron consumption differed among different soybean genotypes. Ghasemi *et al.*[16] stated that application of iron in low-iron soils can increase grain yield of soybean. Iron compounds can be used as foliar on leaves and as seed coating[17].

Magnesium is an alkaline earth metal essential to all living cells. Its ionic state play an important role in the formation of biological polyphosphate compound like ATP, DNA and RNA. Mg is also crucial ion at the center of photosynthetic pigment, chlorophyll and thus a common additive to fertilizers. It has been documented that the shape and size of nanocrystalline magnesium oxide particles provide them with high specific surface and reactivity, because of the high concentration of edge/corner sites and structural defects on their surface[18]. This feature may be reflected on the availability of Mg to plants thus affect the processes responses for plant yield quality and quantity.

Jhansi *et al.*[19] found that the smaller size (15 nm) MgO NPs have been enhanced the seed germination and growth parameters of peanut (*Arachis hypogaea* L.) as compared with remaining sizes (20, 18.5, 18 and 16.5) of MgO NPs and control. They also added that the MgO NPs caused an increase in photosynthesis in peanut which enhances the ability to absorb and utilize light. However, this NPs stimulated antioxidant systems and hastened the germination and growth of plants[20]. Raliya *et al.*[21] found that the application of biologically synthesized MgO nanoparticle demonstrate greater improvement in shoot-root growth and chlorophyll photosynthetic pigment of cluster bean (*Cyamopsis tetragonoloba*).

Hieng *et al.*[22] reported that one of the forms of plant response to stress and adaptation to environmental conditions is protein variation. George and Ghareeb[23] and Haiba *et al.*[24] used electrophoretic techniques of protein as a tool to estimate the possible mutagenic potentialities.

This experiment was conducted to investigate the effects of foliar spraying soybean plants with nano-iron or magnesium oxide particles on overcoming the adverse conditions of sandy soil and avoiding their negative effects on crop yield quantity and quality.

MATERIALS AND METHODS

A field trial was carried out at the Experimental Station of National Research Centre, Al-Nubaria district, El-Beheira Governorate, Egypt, during two successive summer of 2017 and 2018 seasons. The experiment aimed to study the effect of iron oxide and magnesium oxide nano-fertilizer form on growth, yield and its components as well as seed quality of soybean under sandy soil conditions. Soil of the experimental site was sandy soil. Mechanical, chemical and nutritional analysis of the experimental soil is reported in Table 1 according to Chapman and Pratt[25].

The experimental designed of this work in split-plot design with 3 replications. Where FeO and MgO nanoparticles materials occupied the main plots and their concentrations foliar applications at the rates of (0.0, 10, 20, 30 and 40 ppm) were allocated at random in sub plots. Soybean (*Glycine max* L.) Merr.) Var. Giza-83 seeds were obtained from Agricultural Research Centre, Giza, Egypt and inoculated just before sowing with the specific rhizobium bacteria inoculants at the rate of (920 g ha^{-1}). Soybean seeds were sown in the first week of May in both seasons in rows 3.5 m long with distance between rows of 60 cm apart. Plot area was 10.5 m^2 (3.0 m width$\times$3.5 m length). The normal cultural practices were applied as recommended in the district. The seeding rate was 120 kg ha^{-1}. Pre-sowing, 360 kg ha^{-1} of calcium super-phosphate (15.5% P_2O_5) were used. Nitrogen was applied after emergence in the form of ammonium nitrate 33.5% at the rate of 180 kg ha^{-1} in 5 equal doses. Potassium sulfate (48% K_2O) was added at two equal doses of 144 kg ha^{-1}. Irrigation was carried out using the new drip irrigation system where water was added every 5 days for 2 h. Foliar application of different concentrations of FeO and MgO nanoparticles materials (0, 10, 20, 30 and 40 ppm) were carried out twice at 30 and 45 days after sowing. Plant samples were taken after 60 days from sowing for measurements of growth characters and some biochemical parameters.

Growth measurements: Samples were taken after 60 days from sowing, the morphological traits measured were shoot length (cm), number of branches and leaves/plant, root length (cm), fresh and dry weight of shoot (g/plant) and fresh and dry weight of root (g/plant).

Biochemical analysis: Some biochemical tests carried out including photosynthetic pigments (chlorophyll a, b, carotenoids and total pigments) in fresh leaves using the method of Lichtenthaler and Buschmann[26]. Indole acetic acid content were extracted and analyzed by the method of Larsen *et al.*[27]. Phenolic content was measured as described by Danil and George[28]. Free amino acids (FAA) was extracted according to Vartanian *et al.*[29] and determined with the ninhydrin reagent method[30]. Proline content was extracted and calculated according to Bates *et al.*[31]. Total soluble sugars (TSS) were extracted according to Prud'homme *et al.*[32] and assayed according to Yemm and Willis[33]. Enzyme extracts were prepared according to method of Chen and Wang[34]. Catalase (CAT, EC 1.11.1.6) and super oxide dismutase (SOD, EC 1.12.1.1) activity was calculated by nitro-blue-

Table 1: Some physical and chemical characteristics of the experimental soil

Season	Constant depth (cm)	Coarse sand (%)	Fine sand (%)	Silt (%)	Clay (%)	Texture class
Physical characteristics						
2017	00-30	40.7	44.6	10.7	4.0	Sandy
	30-60	38.2	43.0	13.8	5.0	Sandy
2018	00-30	38.7	42.6	13.7	5.0	Sandy
	30-60	36.5	38.1	17.8	7.6	Sandy

	Constant depth (cm)	pH	EC (dS m^{-1})	SP	Anions (meq L^{-1}) CO_3^{2-}	HCO_3^-	Cl	SO_4^{2-}	Cations (meq L^{-1}) Ca^{++}	Mg^{++}	Na^+	K^+	$CaCo_3$ (%)	Organic matter (%)
Chemical characteristics														
2017	00-30	7.84	11.76	32	-	0.50	8.40	1.11	1.80	0.90	7.10	0.20	1.00	0.40
	30-60	7.89	5.79	27	-	0.60	8.00	1.40	2.10	1.50	6.20	0.20	6.00	0.07
2018	00-30	7.95	1.59	23	-	0.32	12.70	1.98	4.00	1.80	9.00	0.20	1.90	0.38
	30-60	7.85	1.81	25	-	0.45	15.40	2.15	5.60	2.00	10.20	0.20	1.30	0.32

tetrazolium reduction method[34]. Peroxidase (POX, EC 1.11.1.7) activity was evaluated according to Kumar and Khan[35]. The level of membrane damage was estimated by measuring malondialdehyde (MDA) according to Hodges *et al.*[36]. The oil content of soybean seeds was determined according to the procedure reported by AOAC[37]. Determination of total carbohydrates was carried out according to Herbert *et al.*[38]. Total protein concentration was determined according to the method described by Bradford[39].

Electrophoretic protein profile of soybean leaves was analyzed according to sodium dodecyl-sulphate polyacrylamide gel electrophoresis (SDS-PAGE) technique according to the method described by Hanna *et al.*[40].

Yield measurements: With signs of full maturity stage appearance yield and its attributes (plant height (cm), root lengths (cm), number of branches/plant, number of pods/plant, biological, straw and seeds yields/plant (g), pods yield/plant (g), 100-seed weight (g) and seed yield (t ha^{-1}).

Statistical analysis: The data were statistically analyzed on split plot design according to Snedecor and Cochran[41]. Combined analysis of the two growing seasons was carried out. Means were compared by using least significant difference (LSD) at 5% levels of probability.

RESULTS

Changes in growth parameters: Data of growth parameters in terms of shoot length, number of branches and leaves/plant, root length, shoot fresh weight, shoot dry weight; root fresh weigh and root dry weight are presented in Table 2. The presented values showed clearly that FeO or MgO NPs at different concentrations significantly ($p<0.05$) improved all the studied parameters compared to the control plants. In general, the use of FeO NPs at the rate of 30 ppm significantly ($p<0.05$) improved most growth parameters of soybean plants The resulted values indicated that the foliar spraying of MgO NPs at the rate of 20 ppm on soybean grown in sandy soils was more effective in improving growth parameters, except for number of branches and leaves/plant. The highest ($p<0.05$) values of soybean growth parameters were recorded with applying 30 ppm FeO NPs.

Changes in photosynthetic pigments: Data in Fig. 1a-d presented for the effect of foliar spraying of FeO or MgO NPs on photosynthetic pigments of soybean plant grown in sandy soils. The obtained results showed that both treatments with FeO or MgO NPs increased significantly ($p<0.05$) chl a, chl b, carotenoids as well as total pigments compared to those recorded for the control treatment (Fig. 1a-d). In the meantime, MgO NPs showed to be superior than FeO NPs in improving photosynthetic pigments the treated plants. The highest ($p<0.05$) values of the measured photosynthetic pigments were obtained with foliar spraying of both FeO and MgO NPs at the rate of 30 ppm compared to the control and other concentrations of treated plants.

Changes in some compatible solutes: Figure 2a-d presents for the resulted changes in some compatible solutes [total soluble sugar (TSS), proline (Pro), free amino acids (FAA) and total soluble protein (TSP)] of soybean plants grown under sandy soil conditions when foliar sprayed with NPs of FeO or MgO.

The obtained results indicated that foliar spraying of soybean with both FeO and MgO NPs increased significantly ($p<0.05$) all the measured compatible solutes traits compared to the control plants. In general, FeO NPs foliar spraying treatments on soybean plants proved to be superior in increasing compatible solutes values compared to that

Table 2: Effect of different concentrations of iron or magnesium oxide nanoparticles on growth parameters of soybean plants grown in sandy soil (at 60 days from sowing) (mean of 2 seasons)

Materials	Concentration (ppm)	Shoot length (cm)	Number of branches	Number of leaves	Shoot fresh weight (g)	Shoot dry weight (g)	Root length (cm)	Root fresh weight (g)	Root dry weight (g)
FeO NPs	0	40.33±0.58	0.00±0.00	11.00±0.58	17.15±0.40	6.52±0.08	15.00±1.15	1.24±0.05	0.75±0.02
	10	44.33±1.20	2.66±0.58	16.66±0.67	24.23±0.63	8.35±0.11	21.67±0.88	1.78±0.07	0.88±0.03
	20	52.66±1.53	3.34±1.00	24.33±0.36	34.50±1.22	9.42±0.06	25.33±0.43	2.49±0.12	1.87±0.04
	30	58.34±0.58	3.67±1.20	32.35±0.89	39.34±1.41	18.03±0.22	27.57±0.53	3.61±0.05	2.92±0.06
	40	54.33±0.88	1.68±0.88	16.68±0.33	30.82±1.46	12.58±0.42	23.47±0.49	3.46±0.04	2.53±0.10
MgO NPs	10	50.68±1.00	0.00±0.00	20.00±1.25	27.98±0.38	9.98±0.03	23.33±0.54	2.91±0.03	2.13±0.01
	20	53.66±1.23	3.56±0.52	32.00±1.15	39.57±1.78	20.68±0.11	27.98±0.85	3.41±0.05	2.73±0.12
	30	51.00±1.58	3.69±0.68	42.34±1.67	35.73±0.83	11.90±0.28	24.31±0.93	3.15±0.05	1.49±0.02
	40	50.00±1.15	3.48±0.43	37.62±0.33	31.31±1.14	8.65±0.15	21.72±0.69	2.43±0.08	1.06±0.03
LSD 5%		1.22	1.65	10.74	7.94	4.57	4.45	0.87	0.82

Each value represents the mean±standard error (n = 3)

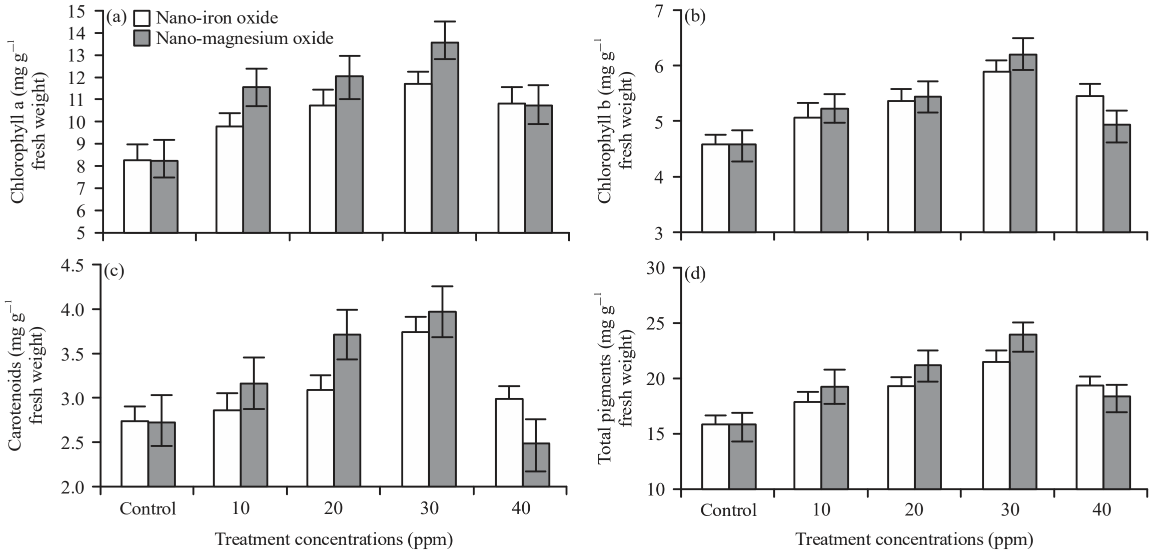

Fig. 1(a-d): Changes in photosynthetic pigments (a) Chlorophyll a, LSD at 5% 0.43, (b) Chlorophyll b, LSD at 5% 0.26, (c) Carotenoids, LSD at 5% 0.26 and (d) Total pigments, LSD at 5% 0.63, contents of soybean plants grown in sandy soil when treated with iron or magnesium oxide nanoparticles (at 60 days from sowing) (mean of two seasons)
Each value represents the mean±standard error (n = 3)

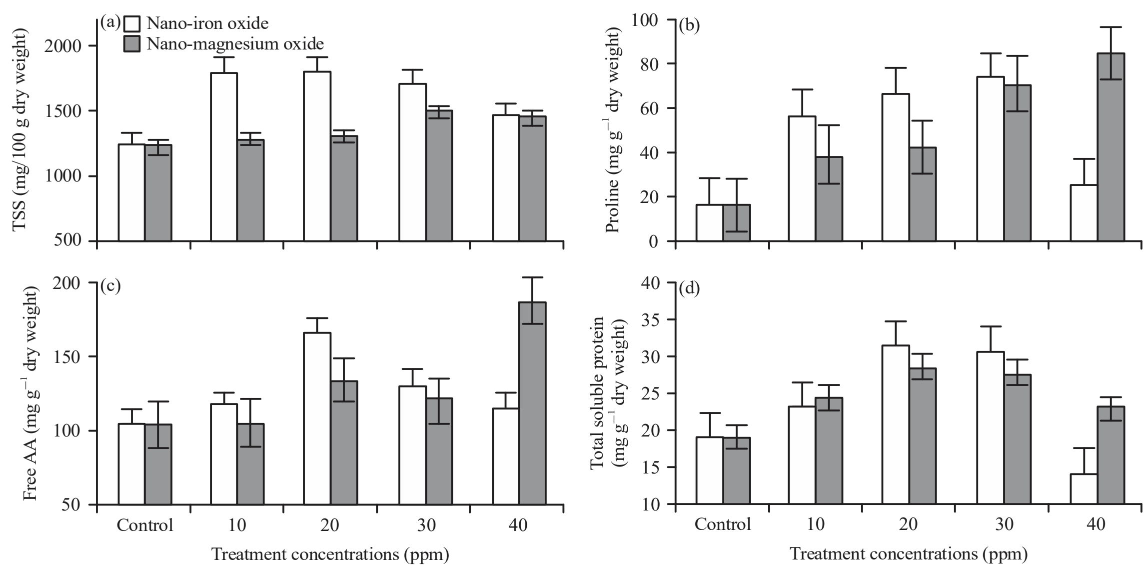

Fig. 2(a-d): Changes in some compatible solutes (a) Total soluble sugars, LSD at 5% 36.54, (b) Proline, LSD at 5% 4.86, (c) Free AA, LSD at 5% 10.43 and (d) Total soluble protein, LSD at 5% 0.85 of soybean plants treated with iron and magnesium oxide nanoparticles grown in sandy soil (at 60 days from sowing) (mean of two seasons)
Each value represents the mean±standard error (n = 3)

recorded from control or MgO NPs treated plants. It is obvious that the most pronounced increase in TSS, FAA and TSP were observed in plant sprayed with 20 ppm FeO NPs. Meanwhile, proline recoded the highest value at 30 ppm treatment.

Regarding the effect of MgO, it is clear from Fig. 2a-d that the most pronounced increase in proline and FAA were observed in plants sprayed with the highest concentration of MgO NPs (40 ppm), but TSS recorded the highest value at 30 ppm treatment.

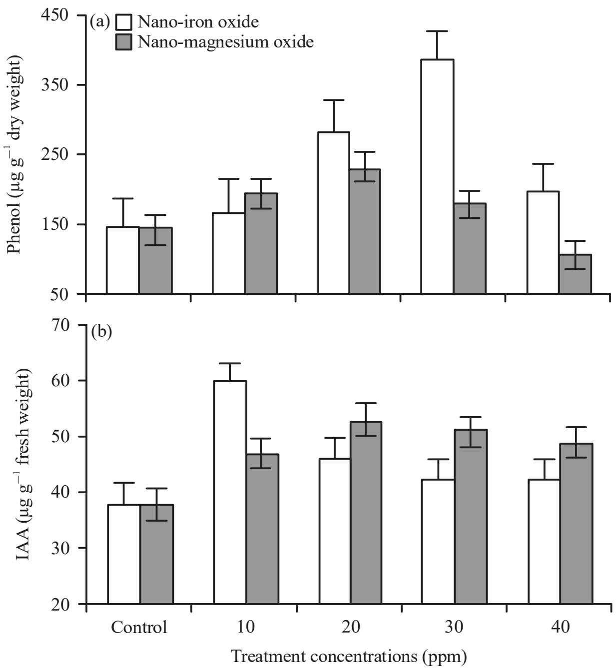

Fig. 3(a-b): Effect of iron and magnesium oxide nanoparticles treatment on (a) Phenol content (µg g^{-1} dry weight), LSD at 5% 18.62 and (b) IAA content (µg g^{-1} fresh weight), LSD at 5% 1.21 of soybean plants grown in sandy soil (at 60 days from sowing) (mean of two seasons)

Each value represents the mean±standard error (n = 3)

Phenol and indole acetic acid contents: Figure 3a illustrated that foliar spraying with NPs of FeO or MgO on soybean plant generally increased ($p<0.05$) phenol concentrations in treated plants than the control ones. Generally, the FeO NPs treatment recorded higher ($p<0.05$) values of phenol than that obtained from MgO NPs application. The highest value of phenol (382.44 µg g^{-1} dry wt.) was obtained with applying 30 ppm FeO NPs. While the highest phenol content (229.49 µg g^{-1} dry wt.) recorded with MgO NPs treatment at the rate of 20 ppm which showed decreased trend with increasing concentration treatments. It is of interest to note that the highest concentration (40 ppm) of FeO NPs improved phenol content by about 35.87% of soybean plants, while MgO NPs at the same level decreased it by about 25.47%.

In general, both FeO or MgO NPs application increased ($p<0.05$) IAA content of soybean plants in the present work. The obtained results in Fig. 3b reported that MgO NPs foliar application on soybean plants at high concentrations (20, 30 and 40 ppm) proved to be better in increasing IAA content than FeO NPs which showed a reduction trend. The lowest concentration (10 ppm) of FeO NPs produced the highest IAA value (59.44 µg g^{-1} dry wt.), while the highest value (52.88 µg g^{-1} dry wt.) obtained from MgO NPs application when 20 ppm concentration was used with significant ($p<0.05$) differences between them.

Lipid peroxidation: Figure 4a illustrated that foliar spraying with NPs of FeO and MgO on soybean plant generally decreased ($p<0.05$) lipid peroxidation in treated plants than the control ones. This observed gradual reduction ($p<0.05$) in MDA values recorded with increasing the concentrations of both FeO and MgO NPs sprayed plants.

Antioxidant enzymes activity: The obtained values of SOD, POX and CAT are presented in Fig. 4b-d. The lowest ($p<0.05$) concentration of SOD and POX of soybean plants showed remarkably by about, 59.09 and 51.18% in plants treated with 40 ppm FeO NPs and by about 57.91 and 51.85% in the plants treated with 40 mg L^{-1} MgO NPs than that recorded with the control treatment. Low concentrations of both treatment materials produced intermediate values of these enzymes among that of the control plants and those treated with high concentrations. While CAT content followed the reveres trend, since it increased in the treated plants by about 46.97% using 30 ppm FeO NPs treatment and 44.35% when 30 ppm MgO NPs was applied compared to the untreated plants.

Changes in protein electrophoretic patterns: An electrophoretic separation of total soluble proteins in leaves of unsprayed and sprayed soybean plants with different concentrations of FeO or MgO NPs, using SDS-PAGE were depicted in Fig. 5 and Table 3. A total number of 14 polypeptides of soybean leaves were showed heterogeneity among control and the treated plant with FeO or MgO nanoparticles with molecular weights (MWs) ranged from 10-238 kDa. Regarding the effect of FeO NPs, lanes from 2-5 as compared with control plants (lane, 1), the data clearly demonstrated that there is appearance for a new polypeptide bands (de novo synthesis) at molecular weights 171 and 32 kDa in response to 10 and 20 ppm. Also, 120, 140 and 32 kDa in response to 30 ppm and 51 kDa in response to 40 ppm. On the other hand, there is a disappearance in other bands at molecular weights of 190 kDa in response to 10 and 20 ppm, 124 kDa in response to 30 and 40 ppm and 42 kDa in response to 40 ppm.

Results in Fig. 5 and Table 3 indicate that treatment of soybean plants with different concentrations of MgO NPs induced changes in profile of protein pattern in the plant leaves. It is obvious that at the lowest conc. there is no change in bands number (6 bands similar to the control one). On the

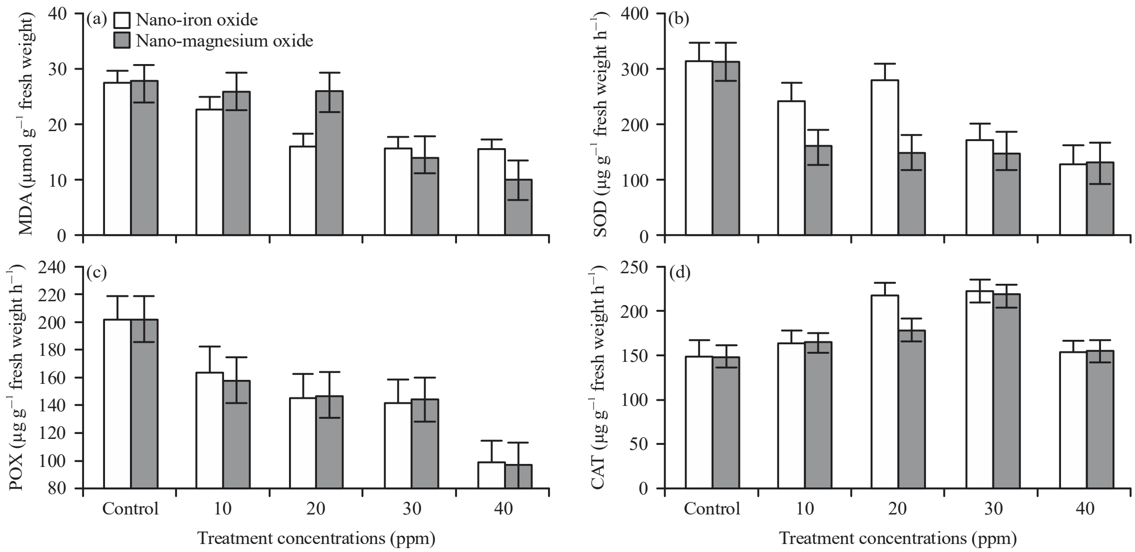

Fig. 4(a-d): Effect of iron and magnesium oxide nanoparticles treatment on (a) Malondialdehyde (MDA) content, LSD at 5% 3.26, (b) Superoxide dismutase, LSD at 5% 16.48, (c) Peroxidase, LSD at 5% 11.48 and (d) Catalase activities, LSD at 5% 3.02 of soybean plants grown in sandy soil (at 60 days from sowing) (mean of two seasons)

Each value represents the mean±standard error (n = 3)

Table 3: Analysis of total soluble proteins (SDS-PAGE) in leaves of soybean plants sprayed with different concentrations of iron and magnesium oxide nanoparticles grown in sandy soil showing band number and molecular weight (Mwt)

			FeO NPs (ppm)				MgO NPs (ppm)			
Band No.	MWt (kDa)	Control	10	20	30	40	10	20	30	40
1	238	+	+	+	+	+	+	+	+	+
2	190	+	-	-	+	+	+	+	-	-
3	171	-	+	+	-	-	-	-	+	-
4	150	-	-	-	-	-	-	+	+	+
5	140	-	-	-	+	-	-	-	-	-
6	124	+	+	+	-	-	+	-	+	+
7	120	-	-	-	+	-	-	+	-	-
8	70	+	+	+	+	+	+	+	+	+
9	51	-	-	-	-	+	-	+	+	+
10	42	+	+	+	+	-	+	+	+	+
11	32	-	+	+	+	-	-	-	-	-
12	30	-	-	-	-	-	-	-	-	+
13	26	-	-	-	-	-	-	-	+	-
14	10	+	+	+	+	+	+	+	+	+
Total number of bands = 14		6	7	7	8	5	6	8	9	8

+: Presence of band, -: Absence of band

other hand, there are increases in profile of protein pattern in response to 20 and 40 ppm MgO NPs recorded 8 bands (two bands more than control). In response to 30 ppm MgO NPs recorded 9 bands (3 bands more than control). It is clear from data that the appearance of new polypeptides bands at molecular weights 150 and 51 kDa in response to 20, 30 and 40 ppm. Moreover, 26 and 171 kDa in response to 30 and 30 kDa in response to 40 ppm as compared with control.

Changes in yield and yield components: Yield and yield attributes in forms of shoot and root lengths, number of branches and pods/plant, shoot and root weights, pods weight/plant, 100-seeds weight and seed yield are listed in Table 4. Generally, the studied yield traits found to be increased ($p<0.05$) significantly due to treatment with different concentrations of both FeO and MgO NPs. The resulted values indicated that the foliar spraying of FeO NPs at the rate of 30 ppm on soybean was more effective in

Table 4: Effect of different concentrations of iron and magnesium oxide nanoparticles on yield and yield components of soybean plants grown in sandy soil (mean of two seasons)

Materials	Concentration (ppm)	Shoot length (cm)	Root length (cm)	Number of branches	Shoot weight (g)	Number of pods/plant	Pods weight/ plant (g)	Root weight (g)	100-seeds weight (g)	Seed yield (t ha^{-1})
Iron oxide	0	44.33±1.67	22.33±1.03	4.33±0.43	23.94±0.60	76.00±1.53	11.69±0.81	2.50±0.23	10.49±0.10	1.822±0.03
NPs	10	51.67±1.53	34.67±1.12	6.33±0.19	48.83±0.29	107.00±1.15	23.50±0.71	4.18±0.08	12.93±0.03	2.522±0.03
	20	55.33±0.67	52.67±1.45	8.67±0.12	60.73±0.78	112.00±1.15	34.23±0.80	5.67±0.04	14.33±0.11	2.928±0.04
	30	72.67±1.33	36.00±1.58	7.67±0.79	74.29±0.64	129.00±0.58	36.37±0.31	6.48±0.07	15.94±0.04	3.391±0.04
	40	44.00±1.00	27.67±0.98	8.00±0.58	51.10±0.11	96.67±0.88	26.66±0.72	4.61±0.06	12.63±0.07	2.436±0.01
Magnesium	10	57.00±1.26	25.00±0.67	6.00±0.10	64.42±0.56	95.67±1.20	23.51±0.74	2.56±0.03	13.74±0.07	2.758±0.03
oxide NPs	20	68.00±1.20	38.67±1.33	6.67±0.03	85.32±2.35	110.33±0.33	37.22±0.43	3.49±0.04	14.36±0.26	2.935±0.05
	30	65.00±1.4 3	27.67±0.67	6.33±0.88	47.04±0.62	87.33±0.88	23.58±0.79	2.75±0.15	13.36±0.05	2.647±0.01
	40	51.33±1.55	18.00±1.00	5.00±0.09	27.23±0.62	78.00±1.00	14.57±0.20	2.26±0.11	12.72±0.14	2.465±0.02
LSD at 5%		2.32	2.63	1.11	2.81	2.68	1.20	0.31	0.35	0.054

Each value represents the mean±standard error (n = 3)

Table 5: Effect of different concentrations of iron or magnesium oxide nanoparticles on protein, carbohydrate and oil percentages in the seeds of soybean plants grown in sandy soil (mean of two seasons)

Materials	Concentration (ppm)	Protein (%)	Carbohydrate (%)	Oil (%)
FeO NPs	0	27.63±0.26	46.75±0.79	20.64±0.18
	10	32.26±1.29	48.50±0.37	21.50±0.79
	20	33.03±1.23	55.37±0.68	22.81±0.32
	30	33.33±1.01	60.45±1.30	25.62± 0.66
	40	30.39±0.63	54.32±0.85	22.61±0.80
MgO NPs	10	27.51±1.63	54.46±0.31	21.72±0.82
	20	28.29±0.26	68.32±0.62	25.48±0.76
	30	34.20±2.00	61.15±0.54	23.61±0.37
	40	29.41±3.49	54.30±0.62	21.38±0.77
LSD at 5%		2.44	2.24	1.78

Each value represents the mean±standard error (n = 3)

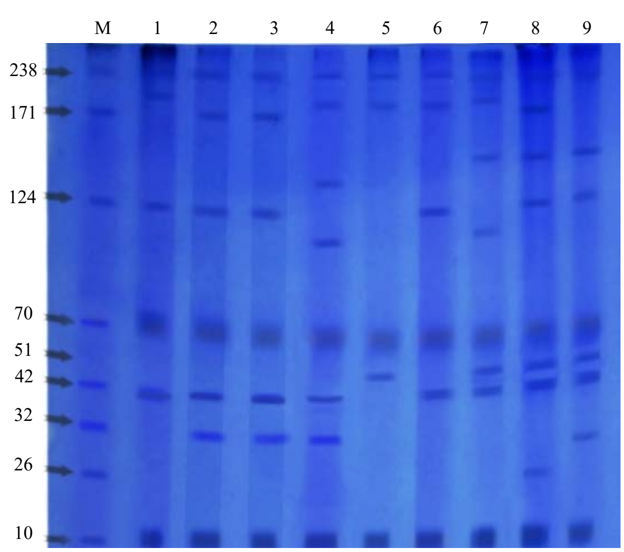

Fig. 5: Electrograph of soluble protein pattern by one-dimensional SDS-PAGE showing the change of protein bands (marked by arrowheads) in response to different concentrations of iron and magnesium oxide nanoparticles

Lane M: Marker, Lane 1: Control, Lane 2: 10 ppm FeO, Lane 3: 20 ppm FeO , Lane 4: 30 ppm FeO, Lane 5: 40 ppm FeO, Lane 6: 10 ppm MgO, Lane 7: 20 ppm MgO, Lane 8: 30 ppm MgO, Lane 9: 40 ppm MgO

improving yield component characteristics, except for root length (cm) and number of branches which showed their increase with 20 ppm treatment. The concentration of 20 ppm MgO NPs produced the highest values of yield parameters compared to the other three concentrations and control plants as well. Seed yield (t ha^{-1}) as the most important parameter in this crop increased significantly ($p<0.05$) by about 86.11% with FeO NPs foliar spraying at the rate of 30 ppm, while it increased by about 61.09% with MgO NPs foliar spraying at the rate of 20 ppm compared to the control plants.

Nutritive value of the seeds yield: In general, the use of foliar spraying for FeO or MgO NPs increased ($p<0.05$) significantly protein (%), carbohydrate (%) and oil (%) of the yielded seeds of treated soybean plants. It seems from the data in Table 5 that 30 ppm FeO NPs foliar spraying was the most effective ($p<0.05$) in increasing protein, carbohydrates and oil percentages of the yielded seeds compared to that of the control plants. Meanwhile, carbohydrate and oil (%) resulted from seeds of plants treated with 20 ppm MgO NPs foliar spray were increased than the other three concentrations (10, 30 and 40 ppm), while protein (%) increased with using 30 ppm MgO NPs solution.

DISCUSSION

It is well evident that plants did not meet their water or nutrient needs in sandy soils due to its poor mechanical structure and chemical composition that hindered plant growth and productivity. Leakage of water and nutrients needed by growing plants can be overcome throughout foliar spraying with NPs of nutrients as fertilizers[9] and may help in overcoming such problems.

The significant increase in growth parameters of soybean plants (Table 2) in the present study are in harmony with results obtained by Dhoke *et al.*[42]. They obtained similar trend with foliar spraying of ZnO, FeO and Zn, Fe, Cu-oxide NPs on mung seedlings. The increases in growth parameters due to treatment with both FeO and MgO NPs may be due to the increases in photosynthetic pigments (Fig. 1a-d) and endogenous promoters especially IAA (Fig. 3b) which in turn alleviate the harmful effect of water deficiency. Iron improves the performance of photosystems via activating several enzymes and contributes in RNA synthesis thus increase the biomass accumulation[43]. The enhancement in root length, root growth, chlorophyll content due to FeO NPs are in harmony with Li *et al.*[44] on watermelon, Ren *et al.*[45] on Chinese mung bean, Alidoust and Isoda[46] on soybean and Alidoust and Isoda[47] on rice.

The obtained values of photosynthetic pigments came on line with Raliya *et al.*[21] who reported increased pigments content with application of MgO nanoparticle in cluster bean plant leaves. They explained that the better response of MgO nanoparticle might be owing to its essentiality and acting as central metal atom for chlorophyll structure. In this regard, Jayarambabu *et al.*[48] found that synthesized MgO NPs treatment on maize plants increased its photosynthetic pigments content.

Both FeO and MgO NPs application significantly increased TSS, proline, FAA, TSP (Fig. 2a-d), phenol (Fig. 3a) and IAA (Fig. 3b). Increasing the compatible solutes (TSS, proline and FAA) improves plant cells tolerance to the growth in sandy soil via increasing osmotic pressure in the cytoplasm as well as relative water contents essential for plant growth. In this concern, Liu *et al.*[49] recorded that low concentrations of nano-ferric oxide caused increasing in soluble sugar and protein of peanut plants. The increase in phenol content (Fig. 3a) are in agreement with findings of Sadak[50] on fenugreek plants using Ag NPs.

The treated soybean plants with FeO and MgO NPs (Fig. 4a-c) showed a gradual decrease in MDA, SOD and POX compared with the control plants. Meanwhile, CAT content (Fig. 4d) significantly increased in response to all nanoparticle treatment. The obtained results are in harmony with those recorded by Ren *et al.*[45] on Chinese mung bean plants treated with γ-Fe_2O_3 NPs and decreased CAT, POX, SOD and malondialdehyde (MDA). On the other hand, Iannone *et al.*[51] on wheat plant found that Fe_3O_4 nanoparticles increased the enzymatic antioxidant defense system (CAT, GPOX, APOX, SOD) on both the root and the aerial part respect to the controls. The increased production of the antioxidant molecule under the influence of nanoparticles confirms the regulation of antioxidant system as a response to nanoparticles interaction with plant[52-54].

Regarding the effect of FeO NPs, it induced slight changes in the protein profile (Fig. 5 and Table 3) with an appearance in some polypeptide bands in their respective storage protein profiles in the leaves and other polypeptides disappeared. In this concern, Siva and Benita[55] found that ginger plants exposed to iron oxide nanoparticles respond at molecular level with slight increase in the amount of protein and there is expression of some bands due to increase in the total protein amount which was not present in control. Moreover, the changes in protein electrophoretic patterns of soybean leaves has been postulated by Elsadany *et al.*[56] who found that Si NPs on two soybean cultivars induced appearance and disappearance of some polypeptide protein bands. It is obvious from the obtained data that treating soybean plants with MgO NPs cause appearance the polypeptide protein band with molecular weight of 51 kDa. In this connection, Abedi *et al.*[57] reported that the band with a molecular weight of 51 kDa band may be related to Rubisco activase enzyme. Such observation in their study and the present study can be explained by findings of Spreitzer and Salvucci[58] who stated that the mentioned enzyme (Ribulose-1, 5-bisphosphate carboxylase activase), in a process that requires ATP hydrolysis; it is a key enzyme initiates both photosynthetic and photorespiratory carbon metabolism by facilitating the dissociation of tightly bound sugar-phosphates from Rubisco. Moreover, the appearance of polypeptide with molecular weight 26 kDa have an osmo-protection function in response to 30 ppm MgO NPs. In support to this observation of Shukry[59], El-Bassiouny and Sadak[60] and El-Bassiouny *et al.*[61] mentioned that protein with molecular weight of 26 KDa seems to be osmotin expressed under salinity stress in flax and wheat plants, respectively.

The significant increase in yield and yield attributes (Table 4) due to treatment with both FeO and MgO NPs has been postulated by many investigators. In this concern, Liang *et al.*[62] reported that the carbon nano particles treatments increased leaf area of tobacco plants at the maturity stage compared with conventional fertilizer. Also, Razzaq *et al.*[63] induced the highly favorable effects of Ag NPs on number of grains/spike, 100-grain weight and grain yield per pot in wheat. Raliya *et al.*[64] on *Solanum lycopersicum* showed a better growth of the plant, increase in fruit yield and chlorophyll concentration due to TiO_2 NPs foliar treatment. The increases in yield and its components might be attributed to the increases in growth parameters, photosynthetic pigments and IAA content of treated soybean plants. Several studies showed that nanoparticles play an important role in improving photosynthetic quantum efficiency and chlorophyll content[65] and increased water and fertilizer use efficiency[66].

In general, the use of foliar spraying for FeO or MgO NPs increased significantly protein (%), carbohydrate (%) and oil (%) (Table 5) of the yielded seeds. The increase in the content of protein and carbohydrate due to nanoparticles treatments were in consistence with findings of Salama *et al.*[67] on *Phaseolus vulgaris* and Krishnaraj *et al.*[68] on *Bacopa monnieri*.

On the light of the above-mentioned findings, many morphological and physiological changes in plants had been detected and led to conclude that the effectiveness of NPs varies depending on dose concentration. Thus, the present results open the field for future work to find out the effect of different chemical composition, particle size, surface area covering, shape and reactivity of NPs on different plant performance under different stress conditions.

CONCLUSION

Foliar application of soybean plants grown in sandy soil with FeO NPs or MgO NPs may act as nano fertilizer sources that help the plant to overcome the adverse effects of such soil shortage in nutrients necessary for plant growth and development and accordingly its yield and quality. Some positive changes in polypeptide protein banding pattern by the appearance of bands took place due to NPs treatment.

SIGNIFICANCE STATEMENT

This study discovers that the possible roles of nano-particles increase the horizontal expansion of plant cultivation in the sandy soils. The application of nano-particles became an area of interest for minimizing the use of chemical fertilizers, improves yield quantitatively and qualitatively to produce eco-friendly plants. Nano-particles can protect the plant from the harmful effect of sandy soil like, low water availability, high irradiances, temperature fluctuations and nutrients deprivation.

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