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Iron oxide nanoparticles effect on growth, physiological traits and nutritional contents of *Moringa oleifera* grown in saline environment

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Abstract

Background: Using of nanoparticles in various aspects of life including agriculture, medicine and industry is very crucial. One of the important source for Fe nutrition in plants is Iron oxide nanoparticles (Fe_3O_4NPs) due to its efficiency in releasing under different pH range. Thus, in the Model Farm of National Research Centre Egypt at El Tour South Sinai, a field experiment was carried out, to study the effect of different concentration of Fe_3O_4NPs (0, 20, 40, 60 ppm) on the physiological parameters and the nutritive value of Moringa under saline condition.

Results: The obtained results indicate that foliar spraying of Fe_3O_4NPs significantly promote growth (plant height, branches leaves number per plant, leaf area, stem diameter and biomass). Foliar treatment also increased photosynthetic pigments (*chlo*.a, *chlo* b, *chlo* a/b and carotenoids) and indole acetic acid (IAA) contents comparing with control. Hydrogen peroxide and lipid peroxidation contents of *Moringa oleifera* leaves were decreased significantly as compared with control plant. The maximum activities of antioxidant enzymes Peroxidase (POX), poly phenol oxidase (PPO), super oxide dismutase (SOD) and nitrate reductase (NR) were observed in plants treated with 40 ppm. Different concentrations of Fe_3O_4NPs increased significantly crude protein, crude fiber and ash percentages as well as, some nutrient contents of moringa leaves (N, P, K and K/Na) compared with untreated control plants, meanwhile decreased Na contents.

Conclusion: Treatment of *Moringa oleifera* plant with Fe_3O_4NPs at different concentrations greatly decrease the harmful effect of salinity on growth by its promotive role on different studied biochemical and physiological aspects.

Keywords: Fe_3O_4 NPs, Growth, *Moringa oleifera*, Physiological parameters, Saline environment

Background

Moringaa oleifera is referred to "miracle vegetable". It belongs to the Moringaceae family. It is a perennial herb and it is a common ingredient in functional foods and pharmaceuticals. It is used as a nutritional additive because of its high contents of essential amino acid, some vitamins such as vitamin A, B, C and E, various minerals, some bioactive secondary metabolites such flavonoids, glucoinolate and phenolics (Anwar et al. 2007; Ezzo et al. 2018). Furthermore, *Moringa oleifera* seeds are regarded as oil plant because they have 62–75% oleic acid of 80% unsaturated fatty acids. It is also a powerful antioxidant thus; it is used in food products, cosmetics and lubricants (Zheng et al. 2017). *Moringa oleifera* can be used in water purification (Eman 2014; Mekonnen 2016).

Salinity remains the biggest negative environmental challenge, because elevated levels of sodium chloride are commonly found in many farm zones. Hence, the great restriction facing agriculture in arid and semiarid zones due to increased salt levels both in soil and irrigation water, which hinder growth and productivity of various crops. One of the main causes of salt over accumulation

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in soil is irrigation with poor water. Salinity stress cause both osmotic and ionic stresses on plant cells. Osmotic stress due to the excessive level of salt in soil, which hampered plant´s ability to absorb and retain water (Omisun et al. 2018). Ionic stress even, is produced by an ionic imbalance, which alters the proportion of potassium (K^+) to sodium (Na^+). The rise in of Na^+ can affect chlorophyll formation either by suppressing the biosynthesis of chlorophyll or speeding up its destruction, causing a decline in the rate of photosynthesis (Karimi et al. 2011). Furthermore, reactive oxygen species (ROS) contents also increased by increased Na^+ content, causing harmful effect, such as protein destruction, DNA mutation, and lipid peroxidation (Munns et al. 2006; Farhangi-Abriz and Ghassemi-Golezani 2018).

Both soil salinity and global water scarcity cause reduction in agricultural productivity. Salinity in the soil reduces the availability and competitive absorption of nutrients and in turn affect on crop (Silva et al. 2008). To sustain soil and turgor water absorption, the plant´s internal water capacity must be lower than of the soil. This impact is induced by an increase in osmotic pressure caused by soil solute uptake. This involves an increase in osmotic, either by soil solutes absorption or synthesis of suitable solutes (Tester and Davenport 2003). Increased levels of these solutes resulted in more water taking up from the soil and a decline in water level in plant cell (Nanjo et al. 1999). Survival of plants under such stress environment depends on the ability of plants to perceive the stimulus, generate and transmits signals and instigates physiological alterations thus maintain metabolic processes accordingly (Dolatabadian and Saleh 2009). Adipate with salinity is the only way to provide alimentary agriculture production in saline setting (Al-Rawahy et al. 2011). Therefore, an appropriate arrangement to minimize the harmful effects of salinity must be found (Alharby et al. 2016).

Nanotechnology a rapidly developing field recently, has had an important role on people´s life and has improved quality of life through a variety of fields (Tripathi et al. 2017). Nanoparticles have essential characters such as increase surface-to-volume ratio, thermal, optical and electrical characters, in addition to physical, chemical and biological characters in terms of absorption and activity (Rastogi et al. 2017). It is a procedure for producing, manipulating and deploying nanomaterials into a device (Baruah and Dutta 2009). This technique enables nanoparticles (NPs) with at least one dimension in the order of 100 nm or less (Auffan et al. 2009). Therefore, nanomaterials hold great promise regarding their application in agriculture in terms of plant nutrition and protection (Cossins 2014) due to their size-dependent qualities, high surface-to-volume ratio, and unique optical properties (Dimetry and Hany, 2016). It also modifies material at the nuclear, atomic, atomic or macromolecular level to create nanoscale objects with novel properties due to their small size.

Iron (Fe) is an important microelement affected on different physiological and biochemical processes and ranks the fourth abundant elements regarding to its value, however its quantity is poor or inadequate for plant needing (Askary et al. 2016, 2017). Iron is essential to normal plant growth, development and play important roles in enzyme reactions, photosynthesis, improves the performance of photosystems, DNA transcription, RNA synthesis and auxin activity (Sheykhbaglou et al. 2018). Since Fe is often present in the insoluble Fe^{3+} form, particularly in increased pH and aerobic soil, such soils are iron-deficient (Rui et al. 2016). Using of nanoparticles to treat iron deficiency is one alternative due to poor solubility of minerals containing iron, as well as, improve plant tolerance to different abiotic stress (Askary et al. 2017). Iron nanoparticles reacts at a molecular level inside plant cells have the ability to improve nutrient uptake (Hasan et al. 2011). Several reports about remedy the harmfully effects of salinity by magnetic iron on the other plants are available, El-Sayed (2014) postulated that magnetic water enhanced growth, yield and water content of broad bean plants grown in saline soil. Moreover, chlorophyll a, chlorophyll b, carotenoids, total carbohydrates, protein, total amino acids, proline, total indoles, phenols, P, K, Na and Ca contents were also improved in all parts of plant under salt stress by treatment with magnetic iron. Shahrekizada et al. (2015) investigated the effect of Nano-Fe_3O_4-EDTA fertilizer on aerial organ biomass, number of leaves, plant height, chlorophyll content as well as elemental quantities of sunflower plants and observed stimulatory effects of Nano-Fe_3O_4-EDTA fertilizer.

Magnetite nanoparticles (MNPs) have some advantages in biocompatibility, biodegradability (Yew et al. 2016) and easily encapsulated. It has been indicated also that MNPs has characterized physiochemical and super paramagnetism that affect biomass and biochemical properties and in turn plant (Abou El-Nasr et al. 2015).

Therefore, the aim of this research is to study the effect of magnetite nanoparticles (MNPs) treatments on growth, photosynthetic pigments content, some physiological aspects as well as crude content of *Moringa oleifera* L.

Methods

A field experiment was conducted in the Model Farm of National Research centre, El Tour, South Sinai to study the impact of Fe_3O_4 nanoparticles on growth and some phsiochemical parameter of Moringa. Plants were transplanted in El Tour, South Sinai at 18th Sep 2019

Table 1 Water analysis of Abo Kalam well, El Tour. South Sina

pH	7.49
EC dS^{-1}	8.7
Soluble cations (Meq/L)	
K^+	0.5
Na^+	69.2
Mg^{++}	11.9
Ca^{++}	21.6
Soluble anions (Meq/L)	
SO_4^-	26.6
Cl^-	74.2
HCO_3^-	2.4
CO^-	–

Table 2 Chemical and mechanical analysis of the soil

Depth	00–30 cm	30–60 cm
Soil texture	Sandy soil	Sandy soil
pH	8.1	8.4
EC dS^{-1}	15.1	4.52
Soluble cations (Meq/L)		
K^+	0.4	0.24
Na^+	112.0	27.0
Mg^{++}	28.8	5.5
Ca^{++}	60.5	12.5
Soluble anions (Meq/L)		
$SO4^-$	61.0	10.64
Cl^-	139.0	31.0
HCO_3^-	2.7	3.6
CO^-	–	–

grown under drip irrigation system with saline water (EC 8.7 dSm^{-1}), water analysis of Abo Kalam Well are presented in Table 1.

Experiment was laid out in completely randomized block (0.5 × 1.5 m distance between plants) i.e. 5600 plants /fed., the mechanical and chemical analysis of the soil was carried out by using the standard method described by Klute (1986) Table 2. Each plant was fertilized with 20 g calcium superphosphate (15.5% P_2O_5) and 10 g potassium sulphate (48.0% K_2O) and 20 g urea (46.5% N) mixed with 300 g green manures. Foliar spraying with Fe_3O_4 nanoparticles (control, 20, 40 and 60 ppm) was applied 45 days from transplantation and 30 days later.

After 210 days from transplantation four replicates of representative samples from each treatment was taken to record plant height (cm), stem diameter (cm), number of branches and leaves per plant, leaf area (cm^2) and total biomass (dry wt.gm/plant).

Measurements: Photosynthetic pigments content were determined as mg/gm fresh weight (chlorophyll a, chlorophyll b, carotenoids and total pigments according to Lichtenthaler and Buschmann (2001). Indole acetic acid (IAA) content was extracted and analysed by the method of Gusmiaty et al. (2019). Proline and free amino acid content were extracted according to the method described by Vartainan et al. (1992). Proline was estimated according to Bates et al. (1973). Free amino acid was determined with the ninhydrin reagent method (Yemm and Cocking 1955). The procedure for the extraction of TSS from stem, leaf, and pod wall was described by Ciha and Brun (1978). TSS was quantified using a modified phenolsulfuric acid assay (Zhang 1993). The hydrogen peroxide (H_2O_2) level was colorimetrically measured as described by Jana and Choudhuri (1981). Lipid peroxidation was determined by estimating the malondialdehyde content following the method of Heath and Packer (1968). For enzyme determination: The method used for extracting the enzyme was that of (Mukherjee and Choudhuri 1983). Peroxidase (POX, EC 1.11.1.7) activity assayed according to the method described by Bergmeyer (1974). Polyphenol oxidase (PPO, EC 1.10.3.1) activity assayed using the method of Kar and Mishra (1976). Superoxide dismutase (SOD, EC 1.12.1.1) activity measured according to the method of Dhindsa et al. (1981). The activity of nitrate reductase (NR, EC 1.7.1.1) was measured according to Jaworski (1971). The contents of sodium and potassium were determined in the digested material using Jenway flame photometer as described by Eppendrof and Hing(1970). K/Na ratio was also calculated for each treatment. Crude protein was calculated by multiplying nitrogen contents by 5.75. Crude fiber and ash were determined by standard analytical methods after (AOAC 2010). Nitrogen was determined with micro Kjeldhal's apparatus according to the method described by AOAC (2010). Crude protein was calculated by multiplying nitrogen contents by 5.75. Analysis of variance (ANOVA) for completely randomized block and LSD at 5% to compare mean were used by M-STAT-C statistical analysis program (MSTAT 1988).

Results

Effect of Fe_3O_4 nanoparticles on some growth parameter and total biomass of *Moringa oleifera* L.

The obtained results as presented in Table 3 show that, plant height (cm.) increased gradually and significantly with increasing concentration of Fe_3O_4 NPs. The highest increase in plant height obtained with 60 ppm Fe_3O_4 NPs, while the least plant height recorded in control plant (0 Fe_3O_4). The same table also showed that, the highest number of branches and leaves/plant were recorded

Table 3 Effect of Fe_3O_4 nanoparticles treatments on some growth parameter of *Moringa oleifera* L.

Fe_3O_4 NPs (ppm)	Length of the plant (cm)	No. of branch/ plant	No. of leaves/ plant	Leaf area (cm^2)	Stem diameter (cm)	Total biomass (g dry wt /plant)
0	84.49	22.60	26.78	35.23	1.91	58.37
20	88.44	23.10	27.88	36.69	2.17	66.52
40	91.10	23.14	32.23	39.60	2.32	74.38
60	92.28	22.21	28.62	37.66	2.07	68.47
LSD 5%	7.28	NS	NS	3.98	0.14	4.30

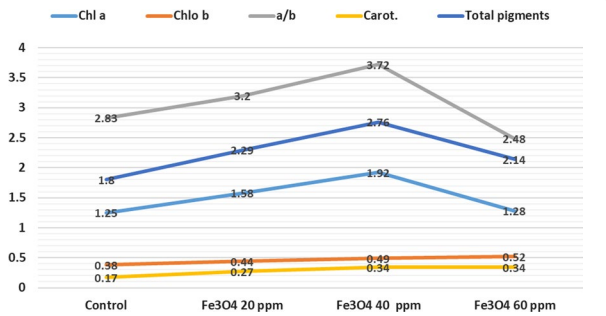

Fig. 1 Effect of foliar application with Fe_3O_4NPs on photosynthetic pigments (mg/g 100 fresh wt) content of *Moringa oleifera* L. (LSD 5% chl *a* = 0.13, chl *b* = 0.03, chl a/b = 0.36, carot = 0.02 and total pig = 0.27)

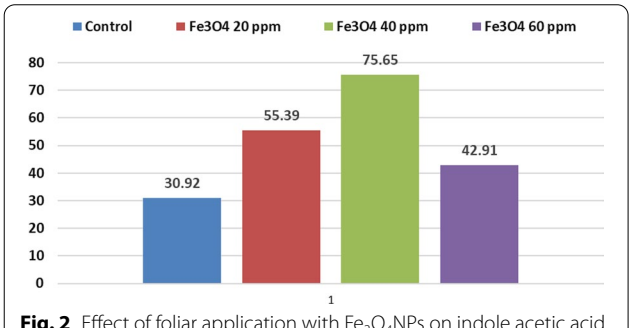

Fig. 2 Effect of foliar application with Fe_3O_4NPs on indole acetic acid content (µ g/g fresh Wt) of *Moringa oleifera* L (LSD 5% = 5.33)

with 40 ppm of Fe_3O_4 NPs without significant differences between other treatments.

Moreover, foliar application of Fe_3O_4 NPs significantly increased leaf area (cm^2), stem diameter (cm) and total biomass (g) dry wt/plant. Furthermore, the highest increases of leaf area (39.60 cm^2), stem diameter (2.32 cm) and total biomass (74.38 g) dry wt/plant were recorded in plants sprayed with 40 ppm Fe_3O_4 NPs. However, increasing concentration of Fe_3O_4 NPs up to 60 ppm caused progressive reduction in these growth parameters compared with Fe_3O_4 NPs at 40 ppm except plant height. Foliar application with Fe_3O_4NPs of moringa plants gave superiority in inducing the highest degree of adaptation under saline habitat, which resulted in significant increases in most studied growth parameters and dry matter accumulation.

Effect of foliar application with Fe_3O_4NPs on photosynthetic pigments content of *Moringa oleifera* L.

As indicated in Fig. 1 it is clearly show that different concentrations of Fe_3O_4NPs, significantly increased photosynthetic pigments as compared with untreated control. It is evident that foliar application with Fe_3O_4NPs at 40 ppm has the most advantageous effect on photosynthetic pigments content as compared to other treatments.

Effect of foliar application with Fe_3O_4 NPs on indole acetic acid content of *Moringa oleifera* L.

The effect of foliar treatment of different concentrations of Fe_3O_4NPs on indole acetic acid (IAA) contents of *Moringa oleifera* L. grown under saline condition is presented in Figure 2. Foliar treatment with different concentrations of Fe_3O_4 nanoparticles induced significant increases in IAA content of *Moringa oleifera* L. as compared with untreated control plant. It is clear that, increasing concentration of Fe_3O_4 nanoparticles significantly increased IAA content up to Fe_3O_4 40 ppm which caused the highest increase in IAA content as compared with other treatments, then treatment with 60 ppm decreased IAA content but still greater than control plants.

Effect of foliar application with Fe_3O_4NPs on osmoprotectants (proline, free amino acids and total soluble sugars) of *Moringa oleifera* L.

The obtained data presented in Table 4 show that, different concentrations of Fe_3O_4 nanoparticles foliar treatment caused significant increases in the content of proline, free amino acids and total soluble sugars of *Moringa oleifera* L. plant grown under saline conditions as compared with controls. The data also show that the largest record of the studied osmolyte resulted by the foliar application of 40 ppm Fe_3O_4NPs treatment.

Table 4 Effect of foliar application with Fe_3O_4NPs on osmoprotectants (proline, free amino acids and total soluble sugars) of *Moringa oleifera* L.

Fe_3O_4 NPs (ppm)	Proline	Free amino acids	TSS
0	24.52	349.41	89.55
20	32.88	468.54	124.74
40	37.01	527.39	142.13
60	31.79	452.94	120.13
LSD 5%	1.68	21.03	7.56

Table 5 Effect of foliar application with Fe_3O_4NPs on antioxidant enzymes POX, PPO, SOD (U/min/g FW) and NR (nM NO_2/g FW/h) of *Moringa oleifera* L.

Fe_3O_4 NPs (ppm)	POX	PPO	SOD	NR
0	373.758	23.793	24.524	394.278
20	501.189	32.153	32.884	521.709
40	564.143	36.283	37.014	584.663
60	484.498	31.058	31.789	505.018
LSD 5%	28.235	2.015	2.018	29.321

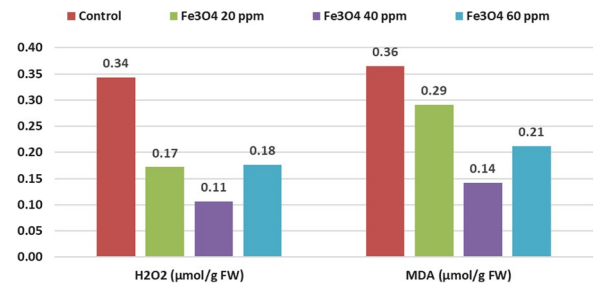

Fig. 3 Effect of foliar application with Fe_3O_4NPs on hydrogen perxide and lipid peroxidation of *Moringa oleifera* L. (LSD 5%: H_2O_2 = 0.012 & MDA = 0.008)

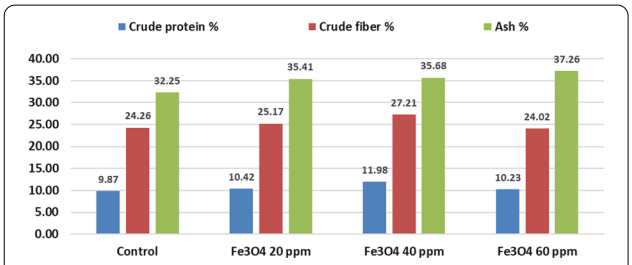

Fig. 4 Effect of foliar application with Fe_3O_4NPs on nutritional values of *Moringa oleifera* L (LSD 5%: crude protein = 0.56 & crude fiber = 1.21 & Ash = 1.87)

Effect of foliar application with Fe_3O_4NPs on hydrogen perxide and lipid peroxidation of *Moringa oleifera* L.

The effect of foliar treatment with different concentrations of Fe_3O_4 NPs on H_2O_2 and MDA contents of *Moringa oleifera* L. plant grown under saline condition are presented in (Fig. 3). Data revealed that the contents of H_2O_2 and MDA were decreased with increasing Fe_3O_4 NPs levels up to 40 Fe_3O_4 NPs then increased with 60 ppm treatment but still lower than control plant as compared with untreated control plant.

Effect of foliar application with Fe_3O_4NPs on Antioxidant enzymes (POX, PPO, SOD and NR) of *Moringa oleifera* L.

Data presented in Table 5 show that, foliar treatment of Fe_3O_4 NPs caused significant increases in antioxidant enzymes Peroxidase (POX), poly phenol oxidase (PPO), super oxide dismutase (SOD) and nitrate reductase (NR)) of *Moringa oleifera* L. leaves as compared with untreated plants (control). Maximum increases were obtained by treatment of *Moringa oleifera* L. plants with 40 ppm of Fe_3O_4 nanoparticles (Table 5).

Effect of foliar application with Fe_3O_4NPs on nutritional values of *Moringa oleifera* L.

Data in Fig. 4 shows the effect of different concentrations of foliar application of treatments on nutritional values of *Moringa oleifera* L. Foliar application

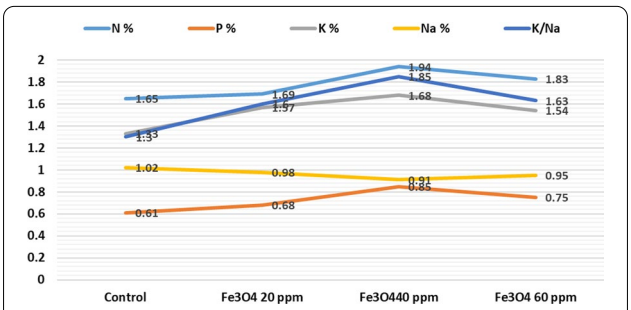

Fig. 5 Effect of foliar application with Fe_3O_4NPs on nutrients content of *Moringa oleifera* L. (LSD 5%: N % = 0.09, P % = 0.06, K % = 0.10, Na % = NS, K/Na = K/Na)

with Fe_3O_4NPs different concentrations (0, 20, 40 and 60 ppm) caused significant increases in nutritional values in comparison to control plants. It is clear from Fig. 4 that, foliar application with 40 ppm Fe_3O_4 NPs recorded the highest values for the content of crude protein, crude fiber and ash in plant leaves.

Effect of foliar application with Fe_3O_4NPs on nutrients content of *Moringa oleifera* L.

Data in Fig. 5 showed that, foliar application with Fe_3O_4 NPs different concentrations, significantly and

gradually increased contents of N, P, K, Na and K/Na values as compared with control treatment. However, plants sprayed with 40 ppm Fe_3O_4 recorded the highest values of N, P and K as well as K/Na. On the contrary, the lowest Na content was recorded under same treatment. The increase of N, P and K in this experiment could be due to the promotional effect of Fe_3O_4 NPs on growth, physiological and biochemical processes of *Moringa oleifera* L.

Discussion

Effect of Fe_3O_4 nanoparticles on some growth parameter and total biomass of *Moringa oleifera* L.

Foliar application with Fe_3O_4NPs of moringa plants gave superiority in inducing the highest degree of adaptation under saline habitat, which resulted in significant increases in most studied growth parameters and dry matter accumulation. These results confirmed by the finding of Abdel-Fattah (2014) he declared that application of magnetite, 3 times at the rate of 4 g/pot improved vegetative growth of *Jacaranda acutifolia seedlings* grown in salinized soil up to 4000 ppm. Also, Shankramma et al. (2016) stated that magnetic iron (III) oxide nanoparticles (Fe_3O_4 NPs) increased growth parameters of tomato plant. In addition, Sanati et al. (2018) confirmed our results on *Capsicum annuum* plant. This enhanced role of magnetic iron (III) oxide nanoparticles (Fe_3O_4 NPs) might be ascribed to the role of magnetic iron in enhancing of N, P and K uptake which stimulate plant growth rather than the harmful effect of Na and Cl which inhibit plant growth (Sodium and chloride ions separate when salts are dissolved in water. The dissolved sodium and chloride ions, in high concentrations, can displace other mineral nutrients in the soil. Plants then absorb chlorine and sodium instead of needed plant nutrients such as potassium and phosphorus, leading to deficiencies). It induces cell metabolism and mitosis of meristematic cells (Askary et al. 2017).

Effect of foliar application with Fe_3O_4NPs on photosynthetic pigments content of *Moringa oleifera* L.

The effect of foliar spraying with Fe_3O_4NPs on photosynthetic pigments of moringa plants is shown in Fig. 1. Data clearly indicated that, different treatments of Fe_3O_4NPs, significantly increased photosynthetic pigments constituents (Chl a, Chl b, carotenoids and total pigments) as compared with untreated control plants. Our results were concurrent with those of Siddiqui et al. (2015), Bastani et al. (2018), Sheykhbaglou et al. (2018) on different plant species. Moreover, Ahmed et al. (2016) concluded that the leaf content of chlorophyll a, b and carotenoids was gradually decreased with increasing salinity level, but was progressively increased as the rate of Fe_3O_4 was increased. The enhancing effect of nano Fe_3O_4 treatment could be attributed to the role of iron in the synthesis of protochlorophyllide from magnesium protoporfyrin complex. It is also participate in the synthsis of aminoleyulinic acid and as coenzyme (Mengel 1991). Furthermore, iron is important in plant growth and development for a wide range of biochemical processes from photosynthesis to respiration. Fe is essential for maintaining the chloroplast structure and function, for biosynthesis of Fe-S clusters and chlorophyll (Chl), and is involved in the electron transport systems (Broadley et al. 2012; Briat et al. 2015; Mai and Bauer 2016).

Effect of foliar application with Fe_3O_4 NPs on indole acetic acid content of *Moringa oleifera* L.

The effect of foliar treatment of different concentrations of Fe_3O_4NPs on indole acetic acid (IAA) contents of *Moringa oleifera* L. grown under saline condition is presented in (Fig. 2). Foliar treatment with different concentrations of Fe_3O_4 nanoparticles induced significant increases in IAA content of *Moringa oleifera* L. as compared with untreated control plant. Similar results were obtained by El-Sayed (2014) who confirmed the positive role of magnetic water on increasing total indole content of broad bean plants grown under saline soil. The increased levels of endogenous IAA contents of Moringa olefera L plant in response to Fe_3O_4NPs might be due to its promotive role in IAA biosynthesis via activiation of enzymes and/or decreasing IAA degradation.

Effect of foliar application with Fe_3O_4NPs on osmoprotectants (proline, free amino acids and total soluble sugars) of *Moringa oleifera* L.

The obtained data presented in Table 4 show that, different concentrations of Fe_3O_4 nanoparticles foliar treatment under saline condition caused significant increases in the content of proline, free amino acids and total soluble sugars of *Moringa oleifera* L. plants conditions as compared with controls. Our result are supported by the findings of Soliman et al. (2015) they stated that, ZnO and Fe_3O_4 nanoparticles induced proline synthesis, and improved tolerance to abiotic stress. Proline, free amino acids and total soluble sugars contents of plant cells play an important role in osmotic adjustment under stress thus protecting macromolecules structure and membranes of cell. Moreover, stabilization of components of sub cellular and free radical and cellular redox buffering induced by proline (Kaur and Asthir 2015) (Table 6).

Table 6 Effect of foliar application with Fe_3O_4NPs on proline, free amino acids and total soluble sugars (mg/100 g dry wt) of *Moringa oleifera* L

Fe_3O_4 NPs (ppm)	Proline	Free amino acids	TSS
0	24.52	349.41	89.55
20	32.88	468.54	124.74
40	37.01	527.39	142.13
60	31.79	452.94	120.13
LSD 5%	1.68	21.03	7.56

Effect of foliar application with Fe_3O_4NPs on hydrogen perxide and lipid peroxidation of *Moringa oleifera* L.

The effect of foliar treatment with different concentrations of Fe_3O_4 NPs on H_2O_2 and MDA contents of *Moringa oleifera* L. plant grown under saline condition are presented in Figure 3. Data revealed that the contents of H_2O_2 and MDA were decreased with increasing Fe_3O_4 NPs levels up to 40 Fe_3O_4 NPs then increased with 60 ppm treatment but still lower than control plant as compared with untreated control plant. Malonyldialdehyde (MDA) is known as marker to assume the degree of peroxidation of lipids and injure of plasmalemma and membranes of organelle resulted from the damage induced by ROS due to environmental stresses (Ozkur et al. 2009). In this regard, Mirzaee et al. (2013) reported that there is modification in matrix of lipid of the membrane of the plasma induced by salinity, they also added that salinity induce changes in the physical organization of the membrane. Moreover, saline stress resulted in obvious increase in the levels of MDA and H_2O_2 in different plant species. Such reduction in MDA contents could be resulted from the effect of Fe_3O_5 in reducing the harmful effect of salinity on the built of membranes cell and decreasing the formation of greatly free radicals. H_2O_2 is considered a signal molecule in plants subjected to different abiotic stresses. These decreases in response to different treatments were reported earlier by Salama et al. (2009) they reported that the contents of H_2O_2 increased in iron-deficient flax leaves. Nanoparticles Fe_3O_4 could increase the POD activity (Table 5), which can decrease H_2O_2 accumulation and maintain cell membrane integrity (Alexandre et al. 2017). This results as described previously by Elstner and Osswald (1994) may be due to the effect of iron on many enzymes to act correctly especially on the active site of catalase and supermutase which induce wiping of reactive oxygen species.

Effect of foliar application with Fe_3O_4NPs on Antioxidant enzymes (POX, PPO, SOD and NR) of *Moringa oleifera* L.

Data presented in Table 5 show that, foliar treatment of Fe_3O_4 NPs caused significant increases in antioxidant enzymes Peroxidase (POX), poly phenol oxidase (PPO), super oxide dismutase (SOD) and nitrate reductase (NR)) of *Moringa oleifera* L. leaves as compared with untreated plants (control).Maximum increases were obtained by treatment of *Moringa oleifera* L. plants with 40 ppm Fe_3O_4NPs nanoparticles (Table 5). Iron is a cofactor for a large numbers of enzymes that catalyze several biochemical processes within the plant. Recently, Rui et al. (2016) and Alexandre et al. (2017) confirmed the induced effect of Fe_3O_4 nanoparticles on antioxidant enzymes activities of peanut and wheat plants under stress conditions. Under salinity stress, antioxidant enzymes as well as antioxidant compounds has the ability to protect the plant from harmful effects of stress. Thus, these antioxidant compounds is the very important to safe the plant cell from the harmful effect of Salama et al. (2009). Soliman et al. (2015) reported that, the enhancing effect of the activities of these enzymes could be attributed to the improving role of Fe_3O_4 NPs as an important element for plant which acts as a metal constituents of different enzymes as well as, a functional structure or cofactor for protein biosynthesis (Marschner 1995).

Effect of foliar application with Fe_3O_4NPs on nutritional values of *Moringa oleifera* L.

Data in Figure 4 shows the effect of different concentrations of foliar application of treatments on nutritional values of *Moringa oleifera* L. Foliar application with Fe_3O_4NPs different concentrations (0, 20, 40 and 60 ppm) caused significant increases in nutritional values in comparison to control plants. The same findings reported by Soliman et al. (2015) who found that spraying plants of Moringa with Hoagland containing ZnO and Fe_3O_4 NP significant increase crude protein level. These increases of nutritional values of moringa leaves could be attributed to the effect of Fe_3O_4NPs on growth, development as it play important roles in enzyme reactions, photosynthesis, improves the performance of photosystems, DNA transcription, RNA synthesis and auxin activity (Sheykhbaglou et al. 2010).

Effect of foliar application with Fe_3O_4NPs on nutrients content of *Moringa oleifera* L.

Data in Fig. 5 showed that, foliar application with Fe_3O_4 NPs different concentrations, significantly and gradually increased contents of N, P, K, Na and K/Na values as compared with control treatment. However, plants sprayed with 40 ppm Fe_3O_4 recorded the highest values of N, P and K as well as K/Na. On the contrary, the lowest Na content was recorded under same treatment. The increase of N, P and K in this experiment could be due to the promotional effect of Fe_3O_4 NPs on growth, physiological and biochemical processes of *Moringa oleifera* L.

Magnetite Nanoparticles (Fe_3O_4) proved to have unique physicochemical properties and super paramagnetism that boosted over all plant metabolisms that affected by biomass and biochemical properties (Abou El-Nasr et al. 2015). In this concern, Soliman et al. (2015) concluded that spraying plants of Moringa with Hoadland containing ZnO and Fe_3O_4 NP resulted in obvious decrease in Na Cl and increase in N, P and K. Moreover, these results related with the findings of Ahmed et al. (2016) they reported that N, P and K was gradually decreased with increasing salinity level, but was progressively increased as the rate of Fe_3O_4 was increased, they added Na content was in descending order, but that of Cl and proline was gradually increased with increasing Fe_3O_4 dose.

Conclusion

The results showed that, foliar application with 40 ppm Fe_3O_4 nanoparticles enhanced all studied growth characters as well as photosynthetic pigments content and crude protein as well as the physiological aspects of the Moringa. We can recommend that Fe_3O_4 nanoparticles can play important role to promote the growth of plants especially in salt affected environment by regulating physiological and biochemical processes in response to salt induced stress. High and low, concentration of Fe_3O_4 nanoparticles was less effective.

Abbreviations

Fe_3O_4 NPs: Iron oxide nanoparticles; *chlo*.a: Chlorophyll a; *chlo* b: Chlorophyll b; *chlo* a/b: Chlorophyll a/Chlorophyll b; IAA: Indole acetic acid; POX: Peroxidase; PPO: Polyphenol oxidase; SOD: Super oxide dismutase; NR: Nitrate reductase; N: Nitrogen; P: Phosphorus; K: Potassium; K/Na: Potassium sodium ratio; ROS: Reactive oxygen species; DNA: Deoxy ribonucleic acid; RNA: Ribo nucleic acid; MNPs: Magnetite nanoparticles; TSS: Total soluble sugars.

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Authors' contributions

MMT, MHM and ATT designed and farming plants, statistical analysis and wrote and reviewed the manuscript. MShS, designed and performed the experiment, responsible of all the physiological and biochemical analysis and also wrote and reviewed the manuscript. All authors read and approved the final manuscript.

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Availability of data and materials

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Declarations

Ethics approval and consent to participate

Not applicable.

Consent for publication

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Competing interests

The authors declare that they have no competing interests.

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