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Effect of Treating the Black Seed Plan, *Nigella sativa* L with Nano-iron Oxide and Vitamin C on Some Physiological Traits and Yield

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Abstract. *In this study we sought to determine the effects of spraying nano-iron oxide and vitamin C on certain physiological traits, yield components, and volatile oil content of black seed plant (Nigella sativa) during the 2023 season, November 5. Concentrations of nano-iron oxide (0, 40, 80, 120 mg L^{-1}) and vitamin C (0, 50, 100 mg L^{-1}) were applied as foliar sprays to plants cultivated in plastic pots using a completely randomized design (CRD) in experiment. Results indicate that the treatment of 120 mg L^{-1} nano-iron oxide together with 100 mg L^{-1} vitamin C produced a highly significant improvement on most of the selected traits. The treatment proved better in yield components of plant such as number of capsules per plant (34.50), number of seeds per capsule (48.17), 1000 seed weight (3.99 g) and volatile oil percent (2.09%).*

Keywords. *Iron nanoparticles, Vitamin C, Nigella sativa.*

INTRODUCTION

The natural products of medicinal and aromatic plants have gained global appeal due to their therapeutic potential against many diseases that affect humans. The medical importance of medicinal plants has dynamic roles in promoting disease prevention and their use in proportion to all prevailing prevention strategies. They are an important source of phytochemical components with Medicinal properties [1].

Nigella sativa is a medicinal plant widely used throughout the world and belongs to the Ranunculaceae family. It is very popular in many traditional medicine systems. Nigella seeds have been widely used in treating various diseases and ailments. Studies have indicated the importance The medicinal benefits of this plant are in treating many inflammatory diseases, rheumatism, and diabetes, as well as improving liver and kidney functions and increasing the activity of immune cells within the human body [2]

The medicinal properties of *Nigella sativa* seeds, specifically antioxidant, antiinflammatory, immunomodulatory, anticancer, neuroprotective, antimicrobial, antihypertensive, antidiabetic and cardioprotective effects, have long been recognized. They are also known for protective parts in liver and kidney health. *Nigella sativa* are still considered of therapeutic importance, particulary its seed especially its essential oil whose constituent includes many bioactive compounds. Volatile oil of the plant consists of phenols, including thymol, thymohydrochinone, thymochinone, and trans -anethole, and alkaloids nigellicine, nigellimine, nigellidine. The seeds are also rich in glycosides, tannins, amino acids, vitamins, minerals etc. Moreover, they consist of 20% protein and 30 to 35% fixed oil which play an important role in their pharmacological effect and therapeutic benefit [3].

The foliar feeding technique is a commonly used method, especially with the development of sprinkler irrigation techniques, which facilitated the addition of nutrients with irrigation water. Nanotechnology has provided many new and convenient products and applications, and the pharmaceutical, agricultural and food industries are investing heavily in nanotechnology research. Nanoparticles are more useful in increasing crop production, as nanoparticles of interest are manufactured directly from mineral salts and minerals [4]. There has also been great progress in developing nanoparticle systems for use as fertilizers, herbicides, insecticides, and high-quality catalysts. Nanoparticles are characterized by dimensions ranging between 1 and 100 nanometers [5].

Vitamin C (ascorbic acid) is considered an antioxidant, and its effect on plants is similar to the effect of growth regulators. This has encouraged its increased use in the agricultural field. At the same time, it is considered a powerful compound for removing toxicity, through rapid interaction with free radicals and ozone. It has been noted that vitamin It plays an important and major role in physiological processes within the plant, such as cell division and growth, signal transmission, and biosynthesis [6]. It also regulates oxidation and reduction processes and is considered a cofactor for enzymes in building hormones and an antagonist of reactive oxygen compounds that are formed during photosynthesis and the building of oxalate compounds and other compounds. Tartate also regulates the response to biological and environmental stress and regulates flowering time.

MATERIALS AND METHODS

The experiment was performed in the agricultural nursery in Salah al-Din Governorate, Iraq, during the 2023 growing season began on November 5. Plastic pots of 30 cm in diameter, 40 cm in height and 20 kg in weight capacity, were used. Mixed soil substrate used for the pots and black seed (*Nigella sativa*) seeds taken from agricultural offices were planted. During the experiment standard agricultural practices were practiced.

A factorial experiment was designed using a completely randomized design (CRD) with two factors:

1. Nano-Iron Oxide Application (F) : Two foliar treatments at four concentrations (0, 40, 80 and 120 mgL^{-1} distilled water).
2. Vitamin C Application (C) : At three applying concentrations (0, 50 and 100 mg L^{-1} distilled water).

Two applications of foliar sprays were made with one month of interval between the two. The gap between spraying nano-iron oxide and vitamin C was three days. Each pot contained one plant. Each replicate consisted of 12 treatment combinations arising from the interaction of the two factors, with the treatments replicated three times.

The following parameters were evaluated :

1. Chlorophyll Content (SPAD) : Chlorophyll content was measured by using SPAD device. We took three readings from the leaves of each plant, and recorded the average value.
2. Plant Dry Weight (g) : Three plants were dried in an electric oven at 65–70°C until they reached a constant weight. Average dry weight of the plant was then determined.
3. Number of Capsules per Plant : Randomly five plants from the middle rows of each experimental unit were selected. Counted the number of capsules per plant and averaged.
4. Number of Seeds per Capsule : The number of seeds per capsule was recorded in five plants randomly selected from the middle rows in each experimental unit. Then we calculated the average.
5. Weight of 1000 Seeds (g) : 1000 seeds from five randomly selected plants were mixed, and weighed using a precision balance.
6. Percentage of Volatile Oil (%) : Were the weight of extracted oil divided by the weight of sample (1 g of seeds) and multiplied by 100 following the equation:
7. Oil Percentage (%)=(Weight of Extracted Oil (g)
   Weight of Sample (1 g))×100 [7].

The data were analyzed using the SAS (2001) program. Significant differences among treatment means were determined using Duncan's Multiple Range Test at a probability level of 0.05, irrespective of the calculated F-value.

RESULTS

As shown in Table 1, a small treatment of nano-iron oxide sprayed combined with vitamin C added to plastic cups produced a significant positive effect on the chlorophyll content of the leaves. The lowest mean chlorophyll content was recorded at control treatment (without spraying) (50.30 SPAD) while the highest mean chlorophyll content was recorded at 60.63 SPAD. The results also indicate synergistic effect of the nano iron oxide and vitamin C on increasing photosynthetic efficiency.

Table 1. Effect Spraying Iron Oxide Nanoparticles and Vitamin C on Chlorophyll Content(SPAD).

Fe / C	0	40	80	120	Vit.Mean
0	50.30j	52.70i	54.22g	58.10d	53.83c
50	53.50h	56.60e	58.09d	65.00b	58.29b
100	55.50f	58.70d	61.22c	67.10a	60.63a

Fe Mean	53.1d	56.00c	57.84b	63.4a

From the result presented in Table 2, the interaction treatment by applying foliar spray with combination of nano-iron oxide and vitamin C increased the average dry weight of shoot per plant. The control treatment (without spraying) showed the lowest average (1.01 g plant-1) and the highest average dry weight (2.71 g plant⁻1). The combined efficacy of nano-iron oxide and vitamin C in increasing shoot biomass is highlighted.

Table 2. Effect Spraying Iron Oxide Nanoparticles and Vitamin C on Plant Dry Weight(gm).

Fe / C	0	40	80	120	Vit.Mean
0	1.01jk	1.10j	1.22i	1.80eh	1.28c
50	1.20i	1.44g	1.61f	1.99e	1.56b
100	1.95d	2.69c	3.00b	3.20a	2.71a
Fe Mean	1.38d	1.74c	1.94b	2.33a	

Table 3 illustrates that the interaction treatment of spraying with nano-iron oxide and vitamin C significantly enhanced the average number of capsules per plant. The highest recorded value was 31.15 capsules plant^{-1}, whereas the control treatment (without spraying) exhibited the lowest average of 14.34 capsules plant^{-1}. These findings underscore the synergistic effect of nano-iron oxide and vitamin C in promoting reproductive development in the black seed plant.

Table 3. Effect Spraying Iron Oxide Nanoparticles and Vitamin C on Number of Capsules Plant^{-1}

Fe / C	0	40	80	120	Vit.Mean
0	14.34k	16.00j	19.90i	22.20h	18.11c
50	22.00h	24.60fg	25.10f	26.90e	24.65b
100	28.00d	30.10c	32.00b	34.50a	31.15a
Fe Mean	21.44d	23.56c	25.66b	27.86a	

From Table (4), it is noted that the interaction treatment, spraying with nano-iron oxide and spraying with vitamin C, had a positive significant effect on the average trait number of seeds, plant-1, as the highest value in the average trait reached 45.77 seeds, plant-1, compared to the treatment without spraying, which gave the lowest average. It reached 22.50 plant seeds-1.

Table 4. Effect Spraying Iron Oxide Nanoparticles and Vitamin C on Number of Seeds Cupsuls^{-1}.

Fe / C	0	40	80	120	Vit.Mean
0	22.50i	29.9h	35.88g	40.44e	32.10c
50	38.11f	42.1d	42.80d	46.60b	42.40b
100	44.14c	44.80c	46.00b	48.17a	45.77a
Fe Mean	34.91d	38.93c	41.56 b	45.07a	

The results in Table (5) indicate that the interaction treatment, spraying with nano-iron oxide and spraying with vitamin C, had a positive significant effect on the average trait of the average seed weight of 1000 grams, as the highest value in the average trait reached 3.37 grams compared to the treatment without spraying, which gave the lowest average of 3.37 grams. 2.29 gm.

Table 5. Effect Spraying Iron Oxide Nanoparticles and Vitamin C on Weight 1000 Seed(gm).

Fe / C	0	40	80	120	Vit.Mean
0	2.00h	2.27g	2.29f	2.51de	2.26c
50	2.28g	2.28g	2.41f	2.66d	2.40b
100	2.61d	3.01c	3.90b	3.99a	3.37
Fe Mean	2.29d	2.52b	2.86c	3.05a	

It is clear from the results of Table (6) that the interaction treatment of spraying with nano-iron oxide and spraying with vitamin C had a positive significant effect on the average characteristic of the percentage of volatile oil, as the highest value in the average characteristic reached 209% compared to the treatment without spraying, which gave the lowest average of 1.25. %.

Table 6. Effect Spraying Iron Oxide Nanparticles and Vitamin C on Volatile Oil Percentage(%).

Fe / C	0	40	80	120	Vit.Mean
0	1.25h	1.36g	1.49f	1.55e	1.41c

50	1.48f	1.50f	1.56e	1.78d	1.58b
100	1.74d	2.00e	2.21b	2.44a	2.09a
Fe Mean	1.49d	1.62c	1.75b	1.92a	

Spraying with nano-iron oxide has led to an increase in the characteristics of chlorophyll and the dry weight of the plant. Iron may have a role in protein formation by contributing to the activity of many enzymes, in particular those responsible for building proteins, as well as building chlorophyll through the ability of iron ions to gain and lose electrons, and this helps in The effectiveness of the enzymes that are involved in the processes of oxidation and reduction within the plant that occur within the processes of respiration and photosynthesis, and to the effect of the iron element and its effectiveness in these characteristics, as it is included in the synthesis of flavoprotein and has an effective role in the processes of biological oxidation and its need the plant is involved in the process of division and increasing the length of the cells, and it is also involved in the synthesis of some pigments. This increase or superiority in vegetative characteristics has been reflected positively on the rest of the characteristics of the dry weight of the plant and the characteristics of the yield components [8]. The element iron is concentrated in a high concentration in the chloroplasts of the plant. The percentage reaches 90% of its weight, and this indicates the importance of iron in the process Photosynthesis increases the characteristics of the yield [9]. Adding nano-element iron to the plant leads to an increase in vegetative characteristics, and increasing its concentration leads to an increase in the activity of physiological processes and an increase in the number of chloroplasts, and this would lead to an increase in the content of the chlorophyll pigment. The kidneys inside the plant. The reason for the increase in total chlorophyll pigments may also be attributed to the role of the iron element in the representation of nucleic acids in the plastids. This increases the efficiency of photosynthesis, as it increases the manufactured carbohydrates, which in turn leads to an increase in vegetative growth

The significant effect of some traits as a result of spraying with vitamin C and the synergistic effect of the vitamin with nano-iron oxide may be attributed to the fact that vitamin C plays an important role in some physiological processes in the plant, namely cell division and growth, signal transmission, and vital metabolism and regulates oxidation and reduction processes and is a cofactor. It contains enzymes in the construction of hormones and the construction of oxalate and tartarate [10]. It is involved in the electron transfer system and protects chloroplasts from oxidation. The role of ascorbic acid in encouraging the photosynthesis process and its impact on growth characteristics.

CONCLUSION

Spraying with nano-iron oxide plays a crucial role in enhancing chlorophyll content and plant dry weight by contributing to protein formation, enzyme activity, and chlorophyll synthesis. The redox properties of iron improve enzyme efficiency in respiration and photosynthesis, while its high concentration in chloroplasts highlights its significance in boosting photosynthetic efficiency and overall plant yield. Additionally, the synergistic effect of vitamin C with nano-iron oxide supports cell division, growth, signal transmission, and metabolism while regulating oxidation-reduction processes and protecting chloroplasts from oxidative damage. This combination effectively enhances vegetative growth, physiological activity, and yield characteristics in plants.

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