



Physiological Effects of Nanoparticles Prepared from Banana Peel Extract and Nickel Oxide on Strawberry Plants

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ARTICLE INFO

Article history:

Received 12 March 2024

Revised 19 May 2024

Accepted 29 May 2024

Published online 01 July 2024

ABSTRACT

Nanoparticles have unique physical and chemical properties due to their small size, allowing them to exhibit unexpected optical, chemical, and physical characteristics. The present study aimed to investigate the physiological effects of nanoparticles biologically synthesized from banana peel extract and nickel oxide on the strawberry plant (*Fragaria ananassa*). Banana peel extract was prepared and subjected to qualitative phytochemical screening. Nickel oxide nanoparticles (NiONPs) were biologically synthesized by combining banana peel extract with a nickel oxide precursor and capping agent. Various techniques, including Fourier-transform infrared spectroscopy (FTIR), ultraviolet-visible (UV-Vis) spectroscopy, scanning electron microscopy (SEM), and X-ray diffraction (XRD) analyses, were employed to characterize the synthesized NiONPs. Different concentrations (10, 20, and 30% designated as N1, N2, and N3, respectively) of NiONPs were made, and their effects on strawberry plants were evaluated. The results of the characterization confirmed the production of NiONPs. Among the different concentrations, N2 demonstrated the most favorable results for seedling leaf length, leaf width, and the number of unripe fruits. However, N2 outperformed in the number of ripe fruits, number of leaves, and number of flowers. Notably, N1 had the highest concentration of DNA (134.7 ng/ μ L), while N2 had the best DNA purity. Furthermore, the N3 had the highest chlorophyll content. The findings of the study revealed that novel nanoparticles could be effectively produced from banana peel extract and nickel oxide. The optimal concentration of 20% NiONPs produced the best strawberry plants, while the highest chlorophyll content in strawberry leaves was obtained with the 30% concentration.

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Keywords: Banana peels, Chlorophyll, DNA, Genotype, Nickel oxide, Strawberry plant.

Introduction

The banana is a tropical fruit that is grown in more than 122 countries. With a total yield of 56.4 million metric tons of fruit, its cultivated area of 3.8 million hectares ranked fourth in terms of output, after milk, rice, and corn. Banana peels have been utilized in several industrial operations recently, including the generation of biofuel and the identification of potassium-40 isotope traces in naturally occurring potassium. Banana plants are used in several medicinal applications. Young leaves are used as poultices for burns and other skin conditions. The astringent ashes of the unripe peel and the leaves are used to treat dysentery, diarrhea, and malignant ulcers. Bronchitis, dysentery, and ulcers are treated with banana flowers. Furthermore, the cooked flowers of bananas are administered to diabetic patients.

Nanoparticles (NP) are microscopic particles with a diameter usually ranging from 1 to 100 nm that are found in nature and are studied in a variety of scientific fields, including chemistry, physics, and biology.

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Citation: Yousif AM, Ahmed SH, Lafta SN, Al-Newani HRH. Physiological Effects of Nanoparticles Prepared from Banana Peel Extract and Nickel Oxide on Strawberry Plants. Trop J Nat Prod Res. 2024; 8(6): 7483-7488. <https://doi.org/10.26538/tjnpr/v8i6.22>

Official Journal of Natural Product Research Group, Faculty of Pharmacy, University of Benin, Benin City, Nigeria

They have essential roles in the composition of industrial products, such as paints, metals, ceramics, plastics, and magnetic objects, and contribute significantly to air pollution. Nanoparticles exhibit diverse shapes, such as nanorods, nanospheres, nanorods, nanoflowers, nanoreefs, nanostars, nanowhiskers, nanoboxes, and nanofibers. Frequently, nanoparticles develop coatings of other materials that are different from the material of the particle and the surrounding medium. When these coverings are present in only a few particles, they can significantly change the properties of the particles.¹ Nanoparticles have unique physical and chemical properties due to their small size, which enables them to exhibit unexpected physical, optical, chemical, and other properties. Nevertheless, they find extensive applications in modern medicine, serving as contrast agents in imaging and carriers for delivering genes and drugs into tumors. The primary goal of utilizing nanoparticles for drug delivery is to either enhance the delivery of the drug to target cells or mitigate the adverse effect of the drug on other organs. Furthermore, they can perform both functions. It is crucial for nanoparticles used in this situation to be able to travel great distances while evading the defenses of the body. To accomplish this, nanoparticles are designed to adhere to cell membranes, penetrate specific cells within the body or tumors, and traverse through cells. Because they are so small, nanoparticles have quite different physicochemical characteristics compared to their superior material counterparts. They are widely used in many different disciplines, including medicine.^{2,3}

Nickel nanoparticles have unique properties because of their quantum effects, small size, and high surface area. These characteristics enable a multitude of applications across multiple disciplines. One of the

notable applications of nickel nanoparticles is that they play a catalytic role. Nickel nanoparticles are applied as catalysts in a variety of chemical reactions owing to their reactivity and wide surface area. They find applications in hydrogenation, carbon-carbon coupling reactions, and more.⁴ Another application of nickel nanoparticles is their function in electronics. Nickel nanoparticles are used in electronic devices for their unique magnetic properties. They can be employed in data storage, magneto-optical devices, and sensors.⁵ Nickel nanoparticles can also function in the detection of biomolecules in biosensor applications. Their large surface area promotes enhanced binding of biomolecules, leading to improved sensitivity.⁶ Another use of nickel nanoparticles is in medicine. They can be used in drug delivery and medical imaging. Also, they are employed to improve the efficiency of drug delivery systems and enhance contrast in medical imaging techniques.⁷ Nickel nanoparticles are used in cancer therapy due to their ability to generate heat under an alternating magnetic field (hyperthermia). This property can be utilized for targeted cancer treatment.⁸

Because of its potential uses in several fields, such as environmental remediation, medicine, and agriculture, banana peel extract has gained attention recently. These nanoparticles are often synthesized using green and sustainable methods, making them environmentally friendly.^{9,10} Ibrahim reported that silver nanoparticles showed significant antibacterial activity against a range of bacterial and yeast infections.¹¹ Both the minimal bactericidal and inhibitory concentrations were determined by the study. Interestingly, the produced nanoparticles displayed a synergistic effect when combined with the levofloxacin antibiotic, resulting in increased antimicrobial activity by 1.16 to 1.32 times.¹¹ According to Rigopoulos *et al.*,⁹ the environmentally friendly production of silver nanoparticles using banana peels holds great promise for nanoparticle synthesis, and there is a clear possibility for optimization using statistical experimental design. Future research endeavors examining the potential effects of diverse synthesis factors on the yield and characteristics of nanoparticles may enhance their valuable biological attributes, hence facilitating prosperous implementations in diverse fields beyond the food sector.

The present investigation was conducted to examine the physiological impacts of nanoparticles biologically synthesized from banana peel extract and nickel oxide on strawberry plants.

Materials and Methods

Preparation of banana peel extract

Banana peels were obtained in January (2024) by separating them from the banana fruit. It was recommended to use bananas that are noticeably ripe or in an "oxidized" state. Afterward, the banana peels were washed and dried. The dried peels were finely ground into powder. Then, 3 g of the dried powder was combined with 100 mL of distilled water and stirred for 10 minutes at room temperature. Following this, the mixture was filtered, resulting in the extraction of water from the banana peels.¹²

Qualitative phytochemical analysis of banana peel extract

By mixing 1 mL of the plant extract with 2 mL of Benedict's reagent in a test tube, the glycoside concentration of the strawberry extract was found. After giving the solution a good shake, it was submerged in a bath of hot water for five minutes. After allowing the tube to cool, a red residue was seen, which indicated the presence of glycosides. A test tube with 0.5 mL of the extract was filled with a few drops of 1% FeCl₃ solution; the appearance of a bluish-green colour indicated the presence of tannins. A 1% ferric chloride solution was used to measure the presence of phenols. The plant extract in a watch glass containing the phenolic compounds changed colour from blue to green when the solution was introduced. The detection solution was made by mixing 10 mL of 50% ethyl alcohol with 10 mL of a 50% potassium hydroxide (KOH) solution to test for flavonoids. After mixing an equal proportion of the solution and the plant extract, flavonoids were visible due to the appearance of a yellow color.¹³

Two methods were used to detect the presence of saponins in the plant extract. In the first method, an aqueous solution of the dried banana

powder was prepared. A test tube containing the solution was filled and given a thorough shaking. The presence of saponins was indicated by the long-lasting, thick foam that appeared. In the second saponin detection procedure, 5 ml of the plant extract was mixed with 1-3 mL of 1% mercuric chloride solution (HgCl₂); There was the presence of saponins because a white precipitate formed.¹³ To find out if there were any resins present, 10 mL of each extract was taken, and 20 mL of distilled water acidified with 4% hydrochloric acid (HCl) was added. The appearance of turbidity suggested the presence of resins. Additionally, two techniques were used to detect alkaloids. A few drops of Marquis reagent were combined with an aliquot of 1 mL of the extract. The presence of alkaloids was indicated by the colour changing to a gritty gray. By adding Mayer's reagent to 1 mL of the extract in the second procedure, alkaloids were detected by the production of a white precipitate.¹³

Synthesis of nickel oxide nanoparticles

The synthesis of nickel nanoparticles (NiONPs) was made according to Ehiowemwenguan *et al.*¹⁴ The nickel precursor was dissolved in deionized water to create a clear solution. The concentration of the precursor solution is influenced by the desired nanoparticle concentration and size. The stabilizing agent was added to the solution, which helps control the particle size and prevents aggregation. Common stabilizing agents include polyvinyl pyrrolidone (PVP) or sodium dodecyl sulfate (SDS). The solution was stirred to ensure a thorough mixing of the precursor and stabilizing agent. A separate solution of the reducing agent was prepared in deionized water. The reducing agent was added to the nickel precursor solution to initiate nanoparticle formation. The reducing agent solution was gradually added to the nickel precursor solution under constant stirring, which led to the formation of nickel oxide nanoparticles. The stirring was continued for a specified period to allow for complete nanoparticle formation. The reaction time and temperature may vary depending on the specific synthesis conditions. After the reaction was complete, the resulting nanoparticle suspension was centrifuged or filtered to separate the nanoparticles from the solution. The nanoparticles were washed with deionized water and a suitable solvent to remove any residual reactants or byproducts. Finally, the nickel oxide nanoparticles were dried under a vacuum or at a controlled temperature.

Preparation of nickel-banana peel nanoparticles

After the completion of both the extraction of banana peels and the synthesis of nickel oxide nanoparticles, the next step involved utilizing the banana peel extract as a reducing and capping agent for the synthesis of nickel oxide nanoparticles. The extract from banana peels was added to a suspension of nickel oxide nanoparticles, allowing the reaction to occur under the right circumstances (such as stirring and temperature). This procedure enabled the bioactive compounds in the extract to reduce the amount of nickel on the nanoparticle surface.

Characterization of nickel oxide nanoparticles

The synthesized nickel oxide nanoparticles were characterized using the Fourier transform infrared spectroscopy (FTIR),¹⁵ ultraviolet-visible (UV-Vis) spectroscopy,¹⁶ scanning electron microscopy (SEM), and X-ray diffraction (XRD) analyses.

DNA extraction from the strawberry leaves

To extract DNA from the strawberry plants, 50 mg of fresh leaves were weighed, crushed, and transferred to 1.5 mL Eppendorf tubes. Then, 600 μ L of cetyltrimethylammonium bromide (CTAB) buffer solution was added. The tubes were placed in a water bath at 65°C for 30 minutes. After adding an aliquot of 3 μ L of RNase, the mixture was incubated for 60 minutes at 37°C. Afterward, 400 μ L of a solution containing isoamyl alcohol: chloroform solution (1:24) was added. The resulting solution was mixed thoroughly and centrifuged at 12,000 rpm for 15 minutes. The upper layer was collected and transferred to a new sterile tube. Six hundred microliters of isopropanol were added and centrifuged at 12,000 rpm for 10 minutes. The supernatant was removed, and the precipitated DNA was washed with 600 μ L of 75% alcohol and centrifuged at 7,500 rpm for 5

minutes. The supernatant was discarded, and the DNA was air-dried for 30 minutes before being dissolved in 50 μ L of TE buffer. Using the nanodrop spectrophotometer, the concentration of the isolated DNA was measured.

Statistical analysis

The statistical software for the social sciences (SPSS) was used to perform a statistical analysis of the data. The data were subjected to analysis of variance (ANOVA) and the values are presented as mean \pm standard error of the mean (SEM).

Results and Discussion

Qualitative phytochemical content of the aqueous banana peel extract

The qualitative phytochemical screening of the hot aqueous extract of banana peel showed positive results for glycosides, alkaloids, and phenols, but did not contain flavonoids and resins. This is consistent with the findings previously reported by Ehiowemwenguan *et al.*¹⁴

Characteristics of nickel oxide nanoparticles

Fourier-transform infrared spectroscopy (FTIR) revealed the presence of two broad bands at 3,340 and 1,635 cm^{-1} as shown in Figure (1). These bands are attributed to the presence of water, with the first band corresponding to the stretching vibration of hydroxyl groups in water and the second band associated with the bending vibration of water molecules. These peaks resulted from moisture absorption caused by the large surface area of the material. Furthermore, the measurement revealed the existence of fundamental bands of nickel oxide at 632 and 597 cm^{-1} , which are attributed to the stretching vibrations of the Ni-O and O-Ni-O bonds.¹⁵ These results are consistent with those of Hong *et al.*,¹⁷ where the N-O coordination bond appeared below 1,000 cm^{-1} and precisely at 550 cm^{-1} , while the N-O bond appeared at 470 cm^{-1} in the findings of Shi *et al.*¹⁸ The UV-Vis spectroscopy is a pivotal technique for identifying nanomaterials since these materials frequently have unique colours and, hence, unique peaks in this measurement. It can be effectively used in characterizing nanomaterials because of their unique colours and ability to produce distinct peaks during analysis. In the present study, a measurement was made, and the results confirmed the existence of a peak at 214 nm, which is the manganese oxide electron transition band.¹⁶ In a recent study, a peak at 214 nm was observed, confirming the presence of the electron transition band for nickel oxide. This shifts the peak lower than the one reported by Hong *et al.*,¹⁷ because the spectra measurement was carried out at 300 $^{\circ}$ C, while the current study was carried out at room temperature (RT).

Scanning electron microscopy (SEM) was employed for microscopic analysis to determine the nanosize and shape of nickel oxide nanoparticles, as illustrated in Figure 3. Additionally, the examination revealed that these particles did not meet the criteria for quantum size, thereby increasing their safety for potential biological applications. Four main peaks were recorded for the X-ray diffraction pattern of nickel oxide nanoparticles, which were at 38.100, 44.200, 64.450 and 77.350 degrees. These peaks correspond to the crystallographic planes of 111, 200, 220, and 222, respectively. Moreover, the particle size of nickel oxide nanoparticles was calculated using the Scherrer equation, and the results showed that the particle size was within the range of 18.72-21.79 nm with an average of 21.73 nm as shown in Figure 4 and Table (1). According to Figure 4, the results of the SEM of the nano-scale dimensions and shape of nickel oxide nanoparticles showed the presence of irregular geometric forms, primarily spherical. Importantly, the analysis indicated that the particles did not fall within the quantum size range, enhancing their safety for biological applications. The spectra in Figure (4) highlighted distinct peaks

corresponding to Ni and O, unveiling the chemical composition of the prepared nanoparticles. The profile of the produced nanoparticles confirmed that oxygen and nickel were present in the substance as described.¹⁸

Influence of the nano-nickel extract on the phenotypic characteristics of the strawberry seedlings

In strawberry seedlings, phenotypic attributes refer to the visible physical features of the organism. Among these characteristics are the leaf shape, varying shades of green in leaf color, a smooth texture with a slightly shiny or waxy appearance, and a fibrous and shallow root system. Furthermore, strawberry seedlings are often small and compact in size, growing to a height of between a few inches and a foot.¹⁹ It is crucial to highlight that phenotypic traits may differ across various strawberry varieties, and the expression of these traits can be influenced by environmental factors²⁰. Table (3) illustrates the effects of the nickel oxide nanoparticles on the phenotypic characteristics of the strawberry seedlings. The best ripe fruits were found in N1 (10% nickel oxide nanoparticles treatment) and N2 (20% nickel oxide nanoparticles treatment), which showed the same value (6 \pm 1.41), while the best seeding length, number of leaves, and number of flowers were observed in N3 (30% nickel oxide nanoparticles treatment) with values of 16 \pm 5.66, 25 \pm 14.14, and 4 \pm 2.83, respectively. However, the results did not show any significant changes between the variables.

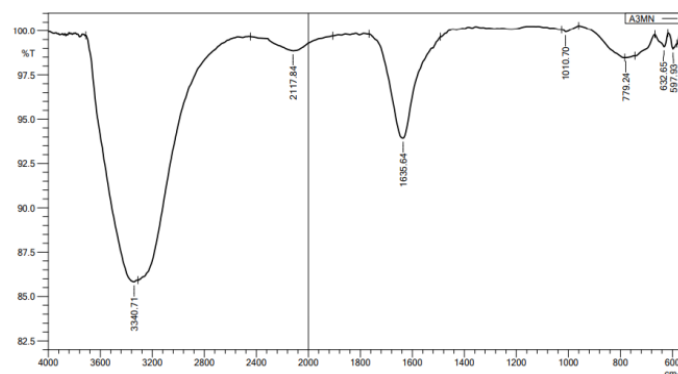


Figure 1: Characterization of nickel oxide nanoparticles with Fourier-transform infrared spectroscopy (FTIR).

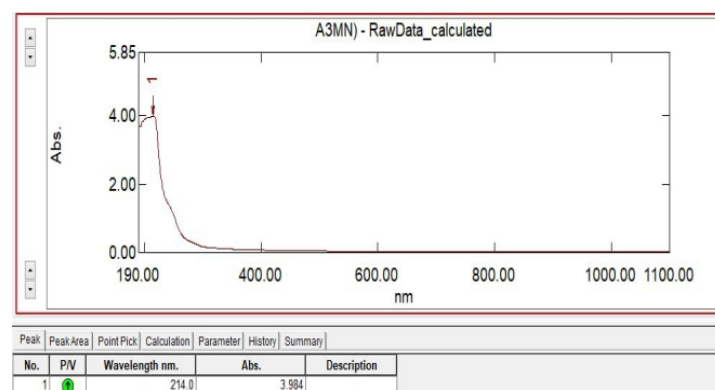


Figure 2: Characterization of nickel oxide nanoparticles with ultraviolet-visible (UV-Vis) spectroscopy.

Table 1: X-ray diffraction data of nickel oxide nanoparticles

Pos. [$^{\circ}$ 2Th.]	Height [cts]	FWHM [$^{\circ}$ 2Th.]	h k l	Diameter (nm)	Average diameter (nm)
38.100	6225.8	0.46369	111	18.95	21.73
44.200	4141.8	0.32638	200	27.46	

64.450	1673.0	0.45037	220	21.79
77.350	1453.2	0.56828	222	18.72

Influence of banana peel extract on strawberry genotype

There is not enough information available to determine how olive leaf extract affects strawberry genotypes. It probably depends on several variables, including exposure length, genotype of the strawberry plant, application technique, and extract concentration. Although olive leaf extract contains a variety of bioactive components, including flavonoids, antioxidants, and phenolic compounds, which may have an impact on plant development and gene expression regulation, further research is needed to determine the precise impact on the strawberry genotype.^{21,22} Plant's genetic composition mainly determines its genotypic traits, although environmental elements and specific treatments might impact gene expression. They usually do not alter the genotype directly. Gene expression changes may result from exposure to certain substances or from environmental stressors. Table (4) shows how the strawberry genotype was affected by banana peel extract. The optimal DNA concentration was observed in N1 (311.2 ng/ μ L), while the highest DNA purity was found in N2 (A_{260}/A_{280} ratio of 1.81).

The effect of banana peel extract on the quantity of chlorophyll in strawberry leaves

There is not much scientific investigation into how olive extract affects the amounts of chlorophyll in strawberry leaves. However, some general insights into how plant extracts might affect chlorophyll content in leaves have been reported.²³ Plants use chlorophyll, a pigment essential to photosynthesis, to convert light energy into chemical energy. Changes in photosynthetic activity and general plant health can be indicated by variations in chlorophyll levels.²⁴ Although olive leaf extract contains bioactive compounds with potential effects on plants, including chlorophyll content, the specific result depends on factors like extract concentration, application method, strawberry variety, and environmental conditions.^{23,24} Table (5), however, illustrates how the extract affects the amount of chlorophyll in the leaves of strawberry seedlings. Chlorophyll measurements utilizing soil plant analysis development (SPAD) showed that N2, N3, and biofertilizers significantly altered chlorophyll contents. These findings are consistent with those reported by Du *et al.*²⁵

Conclusion

Novel nanoparticles were prepared from nickel salt and banana peel extract. FTIR, UV-Vis spectroscopy, and XRD analysis were among the various physicochemical techniques effectively used to characterize the novel nanoparticles. The effects of the new nanoparticles on strawberry seedlings were evaluated. The findings of the research revealed that the best strawberry seedlings were observed in the 20% nano-nickel nanoparticles. The best impact of the extract on the genotype of strawberries was found with N2. With 30% nano-nickel nanoparticles, strawberry leaves had the highest chlorophyll content.

Conflict of Interest

The authors declare no conflict of interest.

Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

Table 2: Quantitative phytochemical constituents of the banana peels in hot water

Phytochemical	Quantity
Tannins	+
Glycosides	+
Phenols	+
Resins	-
Flavonoids	-
Alkaloids	+

+: Present in high amount; ++: Present in higher amount; -: Absent

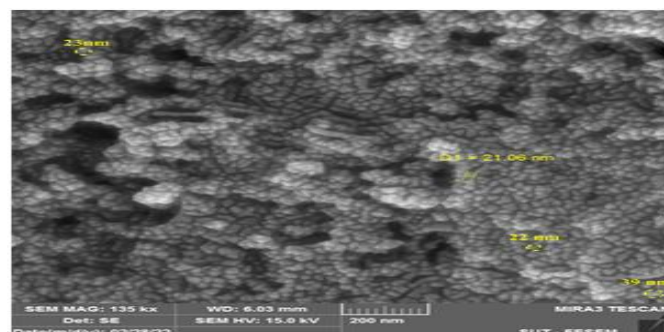


Figure 3: Characterization of nickel oxide nanoparticles with scanning electron microscopy (SEM).

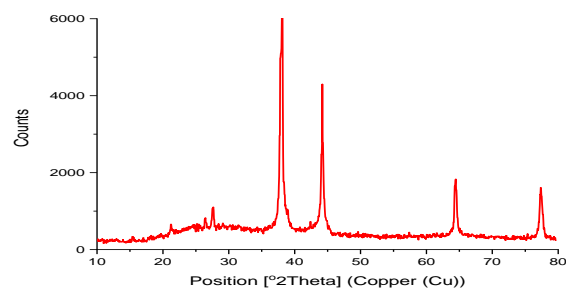


Figure 4: Characterization of nickel oxide nanoparticles with X-ray diffraction (XRD) analysis.

Table 3: The effect of nano-nickel nanoparticles on some phenotypic traits of strawberry seedlings

Treatment	Indicator	Seedling length	No. of ripe fruits	Leaf No.	Flower No.
N1	Mean \pm SD	11 \pm 2.83	6 \pm 1.41	21 \pm 1.41	2 \pm 0
N2	Mean \pm SD	14.5 \pm 9.19	6 \pm 1.41	18 \pm 11.31	3.5 \pm 3.54
N3	Mean \pm SD	16 \pm 5.66	4.5 \pm 0.71	25 \pm 14.14	4 \pm 2.83
F	Mean \pm SD	14.25 \pm 1.06	4.5 \pm 0.71	16 \pm 7.07	4.5 \pm 0.71

Control	Mean ± SD	19.25±6.72	5.5±2.12	23±2.83	3.5±0.71
LSD	Sig.	0.724 NS	0.677 NS	0.839 NS	0.798 NS

Nano-nickel concentrations of 10% (N1); 20% (N2); 30% (N3); Biofertilizer (F).

Table 4: The effect of the nano-nickel extract on the genotype of strawberries

Genotype	DNA concentration (ng/μL)	DNA purity (A ₂₆₀ /A ₂₈₀)
N1	311.2	1.75
N2	91.1	1.81
N3	86.1	1.64
Control	34.1	1.16
F	582.6	1.78

Nano-nickel concentrations of 10% (N1); 20% (N2); 30% (N3); Biofertilizer (F).

Table 5: The effect of the nano-nickel extract on chlorophyll content of strawberry leaves

Treatment	Mean ± SD	LSD
N1	47.4 ± 2.95	0.440 NS
N2	56.9 ± 6.41	0.012*
N3	59.73 ± 8.69	0.004*
F	54.93 ± 2.41	0.027*
Control	44 ± 1.73	

Nano-nickel concentrations of 10% (N1); 20% (N2); 30% (N3); Biofertilizer (F); SD: Standard deviation; LSD: Least significant difference; The chlorophyll content was measured using the Soil Plant Analysis Development (SPAD).

Acknowledgments

The authorsthankmembers of the staff of Mustansiriyah University (<https://www.uomustansiriyah.edu.iq>) for their assistance in completing this research.

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