Middle East Journal of Agriculture Research
Volume: 13 | Issue: 01| Jan. – Mar.| 2024

EISSN: 2706-7955 ISSN: 2077-4605
DOI: 10.36632/mejar/2024.13.1.4
Journal homepage: www.curresweb.com
Pages: 102-113

Phosphorus Nanoparticles Foliar Application as an Effective Strategy for Improving the Vegetative Growth, Biochemical Traits, and Essential Oil of Peppermint Plants

Tamer M. Abd El-Razik[1], Bakry A. Bakry [2] and Mervat Sh. Sadak[3]

[1]Department of Medicinal and Aromatic Plants Research, Pharmaceutical and Drug Industries Research Institute, National Research Centre, Dokki, Giza, Egypt.
[2]Department of Field Crops Research, Agricultural and Biological Research Institute, National Research Centre, Dokki, Giza, Egypt.
[3]Department of Botany, Agricultural and Biological Research Institute, National Research Centre, Dokki, Giza, Egypt.

Received: 20 Nov. 2023 **Accepted:** 15 Jan. 2024 **Published:** 30 Jan. 2024

ABSTRACT
Nanomaterials have been developed for various applications due to their unique features, and anew interest is now growing in their use in the agricultural sector as nano-fertilizers. Among the developed nanomaterials, nano-phosphorus fertilizer. (*Mentha longifolia* L.), which is an abundant source of essential oils (EO) and phenolic acids, is well known for its medicinal significance. The present research aimed to evaluate the impact of various concentrations of phosphorus nanoparticles (200, 400 and 600 ppm), on growth, Essential oil (Eos), and phenolic compounds of mint (*M. longifolia* L) plant. The results demonstrated that the simultaneous application of Phosphorus nanoparticles (P-NPs) from 200 to 600 ppm increased all studied traits as compared with control in both cuts. The foliar application with P-NPs at rate of (600 ppm) significantly increased the herb fresh and dry weight, essential oil and oil yield/plant, chlorophyll (a and b), carotenoids and total pigments contents of mint leaves in both cuts, compared to the control. The highest values of Carbohydrates % (51.83 and 53.358), Phenol (346.52 and 356.710), Flavonoids (241.89 and 249.000) and DPPH % (57.58 and 59.274) on leaves in both cuts respectively, were recorded with the foliar application treatment of P-NPs at rate of (600 ppm). The highest significant values of N% (3.22 and 3.28), P % (0.58 and 0.61 %), and K % (3.24 and 3.26) on leaves in both cuts respectively, were recorded with the foliar application treatments of P-NPs at rate of (600 ppm). The P-NPs at rate of (600 ppm) treatment gave the maximum values on the contents of Eucalyptol (15.83), Isopulegone (1.85) and Pulegone (34.54) on the leaves of (*M. longifolia* L.) herbs. While, l-Menthone (23.17) recorded the highest value with P-NPs at rate of (200 ppm).

Keywords: Mint, Nanotechnology, Phosphorous nanoparticles, Growth, Biochemical, essential oil

1. Introduction
Mint plants (*Mentha* spp.) Common names for peppermint include mint, spearmint, American mint, brandy mint, lamb mint, or lam mint. Peppermint is a member of the Lamiaceae family of herbs. Worldwide, mint is grown for its taste, aroma, medical properties, and potential uses in pharmaceuticals. One of the most often manufactured and used essential oils is peppermint oil (Brown, 1995; Bisset, 1995; EL-Gohary *et al*., 2014). Mint has long been used in medicine. Its applications include carminative, anti-inflammatory, antispasmodic, antiemetic, diaphoretic, analgesic, stimulant, emmenagogue, and anticatharrhal. Mint is also used in cuisine, herbal tea preparations, and confectioneries. Additionally, it is used to treat liver issues, anorexia, pneumonia, flatulence, nausea, and ulcerative colitis. Mint essential oils are commonly applied externally to treat neuralgia, myalgia, headaches, migraines, and for antipruritic, astringent, rubefacient, antibacterial, and antimicrobial reasons (Bisset, 1995, Lorenzi and Matos, 2002, and EL-Gohary *et al*., 2014). As per Singh et al. (2020).

Corresponding Author: Tamer M. Abdel-Razik, Department of Medicinal and Aromatic Plants Research, Pharmaceutical and Drug Industries Research Institute, National Research Centre, Dokki, Cairo, Egypt. E-mail: tamernrc81@gmail.com

Around the world, mint oils are manufactured, one of the most significant mint products is essential oil, which is utilized extensively in the food and beverage sector, medicine, relaxation, and fragrance industries. Fever, cough, and stomach issues can all be treated with mint essential oils and extracts (Gulluce *et al*., 2007; Tucker 2007 and Tarasevi *et al.,* 2021). Some species of mints have antioxidant and anti-cancer properties because they include phytosterols, phenolic compounds, and unsaturated fatty acids (Hussain *et al.*, 2010; Nazem *et al*., 2019 and Benabdallah *et al*., 2016).

Development of the agriculture sector can only be achieved by increasing the efficiency of resource use with minimal damage to the agricultural environment through the effective use of modern technologies (Gaikwad *et al.,* 2017). Among these fields, Nano technology has the ability to revolutionize agricultural systems, environmental engineering, biomedicine, security and safety, energy conversion, and many other fields. (Manjunatha *et al*., 2016 and Veronica *et al*., 2015). In addition, Nano technology is a fertile field of interdisciplinary research. The potential uses and benefits of nanotechnology are very promising.

Nanotechnology has enormous potential because of its applications in fields such as agriculture and the food system, which can improve people's quality of life. Ditta (2012) reported that, around the world Nanotechnology has become the future of any nation. Nanotechnology is considered the most important technological advancement in the recent period and is being applied in all industries due to its potential applications (Cicek and Nadaroglu, 2015). "Nanotechnology" This term is defined as a branch of science that deals with understanding and controlling matter at dimensions of about 1-100 nm (Dhewa, 2015, Singh *et al*., 2015 and Cicek and Nadaroglu, 2015).

The heavy use of fertilizers in previous years has a negative impact on the quality of the soil and harms the environment. Nanoparticles accelerate the germination process of seeds, boost agricultural yield, stimulate the production of chlorophyll, and improve the plant's ability to absorb elements. Nano-fertilizer is one of the most important applications of nanotechnology, which promote plant's ability to absorb nutrients, promote nutrient utilization efficiency, reduced soil toxicity, decreased the negative effects of overdosing and minimized the frequency of treatment (Ditta, 2012). Nano fertilizers can supply one or more nutrients to plants and promote their growth and yield, or they can enhance the performance of conventional fertilizers (Liu and Lal, 2015).

Crop quality and agricultural productivity both depend on plant nutrition. Therefore, improving plant nutrition may result in higher-yielding, higher-quality crops. In all living things, phosphorus (P) is a component of phospholipids, nucleic acids, and high-energy molecules like adenosine triphosphate (ATP). P is a crucial element for plant growth and reproduction and is a major component of fertilizers needed to support contemporary agriculture.

Concerning the importance of Phosphorus element to crop growth, Phosphorus plays a main role in promoting nodulation of roots in leguminous crops, enhancing the process of nitrogen fixation and increasing nutrient-use efficiency (Ogola *et al*., 2012 and Gitari and Mureithi 2003). Phosphorus nanofertilizers can play a vital role in making phosphorus and other nutrients more available to crops and thus reducing misuse of chemical fertilizers. (Selvakumar *et al.,* 2009). The efficiency of nutrients can also be improved by increasing the contact of fertilizers with plant surfaces, which leads to increased absorption by increasing the contact of fertilizers with plant surfaces, which can be achieved by reducing the particle size, which leads to an increase in the number of particles each time. Unit weight and specific surface area (Brady and Weil 2008).

During foliar application, nanoparticles (NPs) are deposited directly inside plants through nano sized pores of plasmodesmata, which usually carry ions between cells, or the stomata in leaves encouraging NPs uptake and transport into the plant leaf system. This leads to a higher nutrient use efficiency and consequent reduction in the use of P and waste of fertilizer (Mittal *et al*., 2020). Phosphorus (P) is one of the macro-nutrients needed for plant growth and productivity. Only 10%−20% of the popular conventional P fertilizers are absorbed by plants from soil (Kopittke *et al*., 2019), while the remaining 80%−90% are rapidly converted to low-availability or fixed forms to insoluble inorganic compounds, or are precipitated with Fe/Al and Ca minerals (Folle *et al.*, 1995). Consequently, excessive P applications are usually required, which may lead to the soil being saturated with P and to reduced sorption ability, as well as to increased P losses (Mozaffari & Sims, 1994). Thus, Nano-P could represent a more efficient and environmentally friendly innovation to replace conventional fertilizers. Thus, this study aimed to maximize the vegetative growth, herb, some physiological traits and essential

EISSN: 2706-7955 ISSN: 2077-4605 DOI: 10.36632/mejar/2024.13.1.4

oil yield of mint plant as affected by foliar application of phosphorus nanoparticles (NPs) as a source of P fertilizer compared to the use of NPK normal fertilizer.

2. Material and Methods

2.1. Site description and soil properties

In two successive seasons, 2021 and 2022, a pot trail investigation studies were conducted in the natural environment of the National Research Center's (NRC) greenhouse in Dokki, Giza, Egypt. Seedlings of Mentha, (*M. longifolia* L.) produced (12 cm in length and 5 cm in root length; derived from three-node rhizomes). On February 15 of each season, the seedlings were transplanted into pots (30 cm in diameter and 50 cm in depth). Every pot held three seedlings and was placed in direct sunlight. Ten kg of air-dried soil were inside the pots. Jackson (1973) and Cottenie *et al*. (1982) were consulted in order to determine the mechanical and chemical parameters of the soil. where the physical composition of the sandy soil was as follows: 81.50% sand, 13.23% silt, 5.2% clay, and 0.45% organic matter (OM). pH was 7.92, E. C. (mmohs/cm) was 0.72, total nitrogen was 0.1 percent, accessible phosphorus was 1.9 mg/100-grams, potassium was 21.1 mg/100 gram, and iron was 20.8 ppm according to the findings of the soil chemical study.

2.2. Experimental design, plant materials, and treatment details

The fertilizer treatments used in the field experiment were: control (distilled water only), NPK, different doses (200, 400 and 600 ppm) of Phosphorus Nanoparticles fertilizer (P-NPs). Based on soil testing and mint nutrient requirements, nitrogen (N), phosphorus (P) and potassium (K) were applied as ammonium nitrate (33.5% N) was added at an amount of 476 kg/ha. (0.021g/ pot), calcium superphosphate (15.5% P2O5) was added at an amount of 357 kg/ha. (0.015 g/pot) and potassium sulfate (48.5% K2O) was added at an amount of 238 kg/ha. (0.009 g/pot), P-NPs which was used in this study were supplied from Sigma-Aldrich Co. with the purity, 98 wt. %.

The plants were sprayed after one month from transplanting with P-NPs at two equal portions at 30, 60 days from transplanting in the first cut and 120, 150 days in the second cut.

The experimental design was complete randomized block design (RCBD) with four replications. Five pots were included in each replicate, with each pot sowing three plants. The Fertilizers treatments studies of this experiment as following:

T1: Control
T2: NPK
T3: NK + P-NPs at rate of (200 ppm)
T4: NK + P-NPs at rate of (400 ppm)
T5: NK + P-NPs at rate of (600 ppm)

Samples were taken for growth characters and chemical constituent's determinations after 90 and 180 days from transplanting in the first and second cut respectively. The following traits were studied. The height of plant (cm), branches number /plants, fresh and dry weights of herb / plant (g), essential oil percentage of the air fresh herb and essential oil yield per plant (g).

2.3. Chemical Determinations:

2.3.1. Photosynthetic pigment:

Photosynthetic pigments: The approaches of Li and Chen (2015) were used to estimate and quantify the levels of carotenoids, chlorophyll a, and chlorophyll b. using a mortar and pestle, the fresh tissue was mashed with 80% acetone. Concentrations of photosynthetic pigment are measured in mg g^{-1} fresh weight (FW).

2.3.2. Determination of phenol contents:

Following IAA extraction, 0.5 mL of the extract was added to 0.5 mL of Folin, shaken, and let to stand for three minutes. After adding one milliliter of saturated sodium carbonate to each tube, the distilled water was agitated and left to stand for sixty minutes. According to Gonzalez *et al*. (2003), the optical density was measured at a wave length of 725 nm using a spectrophotometer

2.3.3. Determination of Flavonoids contents:

Flavonoid content of crude extract was determined by the method of Chang *et al.* (2002).

2.3.4. Determination of antioxidant activity (DPPH):

A plant sample with a defined weight was homogenized with methanol and subsequently filtered. Gyamfi *et al.* (2002) utilized the filtrate to measure the amount of free radical scavenging activity using the 2,2,-diphenyl-2-picryl-hydrazyl (DPPH) technique at an optical density of 517 nm. The following formula was used to determine the antioxidant activity (%): The Sample (517 nm) / The Control (517 nm)×100.

2.3.5. Determination of total carbohydrate:

The measurement of total carbohydrates was done in accordance with Albalasmeh *et al* (2013). 10 mL of sulphuric acid (1N) was added to a test tube containing a known mass (0.2–0.5 g) of dry tissue. After sealing, the tube was kept in a 100°C oven for the whole night. After filtering the mixture into a 100 mL measuring flask, distilled water was added to bring it up to the proper level.

2.3.6. Essential oil (EO) isolation:

Using a Clevenger-type equipment and hydro-distillation for three hours, the complete plants (100 g) of the various mint fresh samples plants were used to extract the essential oil (EO) (Clevenger, 1928). Anhydrous sodium sulfate was used to separate and dry the resulting oily layer. Until additional examination, all of the EOs was stored in airtight, sealed glass vials that were wrapped with aluminum foil and kept at 4°C.

2.3.7. Gas chromatography-mass spectrometry (GC-MS):

The samples of all treatments were analyzed using an Agilent 8890 GC System gas chromatography system, which was connected to an Agilent 5977B GC/MSD mass spectrometer and fitted with an HP-5MS fused silica capillary column (30 m, 0.25 mm i.d., 0.25 mm film thickness). The temperature of the oven was kept at 50 °C at first, then programmed to rise to 200 °C at a rate of 5 °C/min then to rise to 280 °C at a rate of 10 °C/min. Finally, it was held at 280 °C for 7 minutes. The carrier gas, helium, was employed at a flow rate of 1.0 mL/min. A split ratio of 1:50 was used to inject 1 µL of the dissolved essential oil (20 µL essential oil / mL diethyl ether) into the gas chromatograph. The injection temperature was 230 °C. Mass spectra were acquired at 70 eV in the electron impact mode (EI) scanning a range of 39 to 500 amu in m/z. By comparing the isolated peaks with information from the mass spectra library (National Institute of Standard and Technology, NIST), the isolated peaks were found.

2.4. Statistical analysis:

The field experiment data was statistically analyzed using the MSTAT-C software program (Freed *et al.,* 1989) and the experiment in complete randomized block design (RCBD) (Gomez and Gomez 1984). The combined ANOVA analysis of the two seasons was performed after completing Bartlett's homogeneity test (Steel *et al*., 1997). The test of least significant differences (LSD) was utilized to identify any noteworthy distinctions between the means of the treatments that were evaluated (Steel *et al*., 1997).

3. Results and Discussion

3.1. Growth and Essential oil characteristics

Results presented in Table (1) show the vegetative growth characteristics, essential oil and oil yield of mint plants responded significantly to all fertilizer treatments compared with the control in both cuts. The addition of NPK, or any of the three P-NPs foliar treatments, resulted in taller plants with higher fresh and dry herb weight (g plant^{-1}) compared to the control. Maximum plant heights were resulted from plants fertilized with NPK and plants sprayed with P-NPs at 600 ppm, in the first and second cuts, where NPK produced the tallest plants with no significant differences detected between P-NPs at 400 ppm and 600 ppm. Also, the data in Table (1) indicated that the four fertilizer treatments increased significantly the herb fresh and dry weight in both cuts, compared to the control. The maximum values were reached by P-NPs at 600 ppm followed by the application at 400 ppm, followed by the application at 200 ppm, followed by the application at NPK. Additionally, the data also, indicate the foliar application with P-NPs at 600 ppm significantly increased essential oil and oil yield/plant in both cuts, compared to the other treatments.

Table 1. Growth and Essential oil characteristics of mint plant treated with foliar application of Phosphorus nanoparticles **(**P-NPs) fertilizers in the first and second cut (combined of two seasons).

Treatments	Plant height (cm)	No. of branches/ plant	Herb fresh weight (g)	Herb dry weight (g)	Essential oil (%)	Essential oil yield (g/plant)
			1st cut			
Control	42.40	4.33	85.91	24.55	0.053	0.055
NPK	55.00	5.67	102.19	29.20	0.083	0.082
200 ppm	47.20	5.67	103.40	30.96	0.100	0.103
400 ppm	53.20	6.00	108.35	29.54	0.103	0.112
600 ppm	55.00	8.00	117.26	33.50	0.123	0.149
LSD 5%	3.50	1.50	18.60	5.31	0.014	0.026
			2nd cut			
Control	46.64	5.33	94.50	26.25	0.073	0.069
NPK	60.50	6.00	112.41	31.22	0.095	0.107
200 ppm	51.92	6.67	119.19	33.11	0.110	0.136
400 ppm	58.52	7.00	113.74	31.59	0.114	0.145
600 ppm	60.50	8.33	128.99	35.83	0.139	0.190
LSD 5%	3.69	1.38	20.46	5.68	0.011	0.009

P-NPs at doses of 200, 400 and 600 ppm.

Because of their small particle size, ability to pass through cells, and large surface area that controls release kinetics to specific sites, P-NPs has a positive effect on mint plant growth. This makes them an effective delivery method for enhancing plant uptake of P, whether through foliar fertilizer or root absorption. Research indicates that P-NPs with a steady and slow release seems to be a more accessible, efficient, and eco-friendly source of P than conventional fertilizers (Sabry *et al.,* 2023; Bindraban *et al*., 2015; Montalvo *et al*., 2015; Fellet *et al.,* 2021; Szameitat *et al.,* 2021). Phosphorus nano-fertilizer foliar treatment occasion a significant increase in plant height, shoot fresh and dry weight, root circumference, total chlorophyl and P uptake of peanut plant under sandy soil (Bakry *et al.*, 2022).

A further explanation might be that increased gibberellin hormone in plants due to addition of P nutrition (Maghsoodi *et al.,* 2020). Nanofertilizers have been shown to improve plant growth over normal NPK application. One explanation for this would be that P is a necessary part of the ATP molecule, which is in charge of storing and transmitting energy. The essential metabolic activities that govern photosynthesis, respiration, protein synthesis, glycolysis, nutrient translocation, and transport inside plant cells depend on this. Furthermore, according to Elsayed *et al*. (2022) these mechanisms have the capacity to pass on genetic traits from one generation to the next.

3.2. Changes in Photosynthetic pigments

Data in Table (2) showed chlorophyll (*Chlo* a and *Chlo* b), carotenoids and total pigments contents of mint leaves were significantly increased by Nano-Phosphorus fertilizers treatments from 200 ppm to 600 ppm as compared with control plants Table (2). Meanwhile, Nano-Phosphorus foliar treatment of mint plants with various concentrations significantly increased photosynthetic pigments as compared to untreated plants. The maximum significant ($P < 0.05$) increment percentage in various photosynthetic pigments compared with control, NPK and P-NPs at rates of (200, 400 and 600 ppm) increased *Chlo* a by 28.90%, 35.32%, 54.01%, 56.56, and 28.92%, 35.40%, 54.03%, 56.63% in both cut respectively, *Chlo* b by 41.46%, 54.03%, 67.54%, 58.91%, and 41.28%, 53.91%, 67.44%, 58.72% in both cut respectively, Carotenoids by 38.14%, 62.16%, 87.39%, 71.17%, and 38.10%, 61.90%, 87.30%, 71.11% in both cut respectively, and total pigments 33.92%, 45.09%, 63.53%, 59.69%, and 33.95%, 44.99%, 63.24%, 59.56% in both cut respectively. These obtained results are agreed with the recent finding by (Bakry et al., 2022) discovered that, in comparison to the control treatment, the

EISSN: 2706-7955 ISSN: 2077-4605 *DOI: 10.36632/mejar/2024.13.1.4*

application of 200 ppm of phosphorus nano-fertilizer, either once at 30 Days After Sowing (DAS) or twice at 30 and 60 DAS, significantly increased the pod, seed, and oil yield of the peanut plant.

Nanoparticles accelerate the germination process of seeds, boost agricultural yield, stimulate the production of chlorophyll, and improve the plant's ability to absorb elements. Nano-fertilizer is one of the most important applications of nanotechnology, which promote plant's ability to absorb nutrients, promote nutrient utilization efficiency, reduced soil toxicity, decreased the negative effects of overdosing and minimized the frequency of treatment (Ditta, 2012). Phosphorus nano-fertilizer caused a significant increase in plant height, shoot fresh and dry weight, root circumference, total chlorophyll and P uptake of peanut plant under sandy soil (Bakry *et al*., 2022).

Table 2: Photosynthetic pigments, chlorophyll (*Chlo* a and *Chlo* b), carotenoids and total pigments characteristics of mint plant leaves treated with foliar application of Phosphorus nanoparticles (P-NPs) fertilizers in the first and second cut (combined of two seasons).

Treatments	Chlorophyll a (mg g^{-1} fresh wt.)	Chlorophyll b (mg g^{-1} fresh wt.)	Carotenoids (mg g^{-1} fresh wt.)	Total pigments (mg g^{-1} fresh wt.)
		1st cut		
Control	1.059	0.533	0.333	1.925
NPK	1.365	0.754	0.460	2.578
200 ppm	1.433	0.821	0.540	2.793
400 ppm	1.631	0.893	0.624	3.148
600 ppm	1.658	0.847	0.570	3.074
LSD 5%	0.059	0.022	0.026	0.062
		2nd cut		
Control	1.079	0.562	0.315	1.956
NPK	1.391	0.794	0.435	2.620
200 ppm	1.461	0.865	0.510	2.836
400 ppm	1.662	0.941	0.590	3.193
600 ppm	1.690	0.892	0.539	3.121
LSD 5%	0.060	0.023	0.025	0.062

P-NPs at doses of 200, 400 and 600 ppm.

3.3. Changes in Carbohydrates % and antioxidants components.

All application treatments of NPK and P-NPs at rates of (200, 400 and 600 ppm) resulted in a significant increase of (Carbohydrates %, Phenol, Flavonoids, and DPPH %) in both cut compared with untreated treatment as shown in Table (3). The highest values of Carbohydrates % (51.83 and 53.358), Phenol (346.52 and 356.710 mg per 100 g fresh wt.), Flavonoids (241.89 and 249.000 mg per 100 g fresh wt.) and DPPH % (57.58 and 59.274) on leaves in both cut respectively, were recorded with the foliar application treatment of P-NPs at rate of (600 ppm), followed by P-NPs at rate of (400 ppm), followed by P-NPs at rate of (200 ppm), compared with untreated treatment as shown in Table (3). The results obtained are consistent with a recent study that found that when applied externally, Nano-HAP enhances the development and biomass production of rosemary plants in comparison to NPK (Elsayed *et al.,* 2022). Easy penetration and translocation in plant leaves may be the cause of the foliar spray of Nano-HAP's effects on agricultural productivity and yield efficiency (El-Saadony *et al*., 2021). Thus, the use of NFs or slow-release compounds has been suggested as a way to increase P NPs nutrient use efficiency (NUE) in a range of agricultural products. Not only does a biosafe nano fertilizer that contains P contribute to a high NUE, but it also significantly increases fresh and dry biomass, multiplies fruit production, and improves fruit quality (Patra *et al*., 2013).

Table 3: Carbohydrates %, and antioxidants characteristics of (Phenol, Flavonoids, and DPPH %) on mint plant leaves treated with foliar application of Phosphorus nanoparticles (P-NPs) fertilizers in the first and second cut (combined of two seasons).

Treatments	Carbohydrates % (mg per 100 g fresh wt.)	Phenol (mg per 100 g fresh wt.)	Flavonoids (mg per 100 g fresh wt.)	DPPH %
		1st cut		
Control	47.19	289.79	157.51	39.36
NPK	48.34	314.16	200.22	44.13
200 ppm	49.65	324.99	202.30	48.26
400 ppm	50.90	333.51	227.50	51.61
600 ppm	51.83	346.52	241.89	57.58
LSD 5%	0.63	5.56	1.82	1.09
		2nd cut		
Control	48.581	298.317	162.147	40.519
NPK	49.766	323.398	206.104	45.423
200 ppm	51.112	334.550	208.253	49.678
400 ppm	52.393	343.318	234.196	53.128
600 ppm	53.358	356.710	249.000	59.274
LSD 5%	0.645	5.72	1.876	1.126

P-NPs at doses of 200, 400 and 600 ppm.

3.4. Changes in minerals contents on mint plant leaves:

All application treatments of NPK and P-NPs at rates of (200, 400 and 600 ppm) resulted in a significant increase in N, P, and K contents on mint plant leaves compared with the control in both cut as shown in Table (4). The highest significant values of N% (3.22 and 3.28), P % (0.58 and 0.61 %), and K % (3.24 and 3.26) on leaves in both cuts respectively, were recorded with the foliar application treatments of P-NPs at rate of (600 ppm), followed by P-NPs at rate of (400 ppm), followed by P-NPs at rate of (200 ppm), compared with untreated treatment as shown in Table (3). It was evident that applying phosphorus nano fertilizer topically improved the chemical composition of leaves, possibly as a result of phosphorus's synergistic effects. Use nano-fertilizer in addition to traditional fertilizer to improve crop nutrient uptake and achieve ideal mint development. The obtained results agree with the recent finding obtained by (Bakry *et al*., 2022) they found that the application of Phosphorus nano-fertilizer in two doses of 200 ppm resulted in the greatest levels of N (4.01%), P (0.42%), K (2.3%), Na (2.7%), and Fe (120.7 ppm), Mn (154.3 ppm) and Cu (6.3 ppm), Mn (154.3 ppm), and 120.7 ppm) in leaves when compared to the other treatments.

Table 4: Minerals (N, P, and K) contents on mint plant leaves treated with foliar application of Phosphorus nanoparticles (P-NPs) fertilizers in the first and second cut (combined of two seasons).

Treatments	N (%)	P (%)	K (%)
		1st cut	
Control	1.44	0.23	2.27
NPK	2.87	0.37	2.54
200 ppm	2.93	0.47	2.74
400 ppm	3.17	0.55	3.11
600 ppm	3.22	0.58	3.24
LSD 5%	0.10	0.03	0.11
		2nd cut	
Control	1.52	0.28	2.53
NPK	2.83	0.40	2.65
200 ppm	2.91	0.49	3.02
400 ppm	3.22	0.56	3.19
600 ppm	3.28	0.61	3.26
LSD 5%	0.11	0.04	0.1

P-NPs at doses of 200, 400 and 600 ppm.

3.5. Essential oil composition

A total of 19 compounds were identified in the essential oils extracted from the leaves of (*M. longifolia* L.) herbs (Table 5). The identified oil compounds represented 98.62 - 100% of the total oil compositions. Data in Table (5) represented the obtained compounds the leaves of (*M. longifolia*) herb application treatments of NPK and Nano-P at rates of (200, 400 and 600 ppm) comparing with control treatment. From the same table, the P-NPs at rate of (600 ppm) treatment gave the maximum values on the contents of Eucalyptol (15.83), Isopulegone (1.85) and Pulegone (34.54) on the leaves of (*M. longifolia* L.) herbs. While, NPK treatment gave the maximum values of Sabinene (2.33), β-Pinene (3.04), β-Myrcene (1.90), α-Terpineol (1.92), Estragole (2.15), Anethole (0.74), Piperitenone (1.86), β-Caryophyllene (1. 47) and Germacrene D (0.33) concentration on the leaves of (*M. longifolia*) herbs compare with the other treatments. On the other hand, with increasing P NPs the contents of p-Cymene, D-Limonene and p-Menthan-3-one decreased which the control treatment gave the highest values of (0.41, 4.44 and 18.44) respectably. l-Menthone (23.17) recorded the highest value with P-NPs at rate of (200 ppm). Additionally, potassium and phosphorus are required for the growth of plants that produce essential oils as well as for the production of those oils (Prasad *et al*., 2011).

Table 5: Essential oil composition on mint plant leaves treated with foliar application of Phosphorus nanoparticles (P-NPs) fertilizers in the first cut during second season.

Peak	RT	Component name	T1	T2	T3	T4	T5
1	6.297	α-Pinene	0.93	1.74	2.66	1.48	0.65
2	7.269	Sabinene	0.92	2.33	1.92	1.83	0.71
3	7.361	β-Pinene	1.48	3.04	2.75	2.59	1.18
4	7.681	β-Myrcene	1.06	1.9	1.45	1.61	0.69
5	8.603	p-Cymene	0.41	0.00	0.00	0.00	0.36
6	8.717	D-Limonene	4.44	3.85	2.31	3.08	4.01
7	8.814	Eucalyptol	14.98	15.06	12.85	14.19	15.83
8	12.276	l-Menthone	21.86	17.36	23.17	20.82	17.75
9	12.574	p-Menthan-3-one	18.44	14.89	16.91	15.94	15.43
10	12.746	(-)-Menthol	0.00	0.54	0.82	1.07	0.54
11	12.843	Isopulegone	1.64	1.65	1.83	1.63	1.85
12	13.26	α-Terpineol	0.61	1.92	0.87	1.46	1.69
13	13.449	Estragole	1.34	2.15	0.55	1.65	1.37
14	14.702	Pulegone	29.04	28.16	28.44	28.36	34.54
15	15.023	Piperitone	0.89	1.01	1.31	1.2	0.98
16	15.847	Anethole	0.00	0.74	0.00	0.5	0.00
17	17.369	Piperitenone	0.88	1.86	1.8	1.6	1.69
18	19.412	.β-Caryophyllene	1.1	1.47	0.36	1.0	0.73
19	20.95	Germacrene D	0.00	0.33	0.00	0.00	0.00

Pearson correlation coefficient (PCC) heat map matrix, with significance levels

Fig. (1) Represents the correlation coefficients between various traits and variables related to the mint plants. Correlation coefficients range from -1 to 1, with -1 indicating a strong negative correlation, 1 indicating a strong positive correlation, and 0 indicating no correlation. The summary of the correlations was, plant height has positive correlations with all other variables, such as number of branches plant, herb fresh and dry weight, Chlorophyll a, Chlorophyll b, carotenoids, total pigments, phenol, Carbohydrates % and DPPH%, Essential oil (%), phenol, Flavonoids, N%, P% and K%. This suggests that plant height has some influence on these traits. Number of branches plant^{-1} it is has positive correlations with several variables, including herb fresh and dry weight, Essential oil (%), Chlorophyll a, Chlorophyll b, Flavonoids, Carbohydrates %, and others. It may play a role in the growth and development of the mint plant. Herb fresh weight has strong positive correlations with herb dry weight, Essential oil (%), Chlorophyll a, Chlorophyll b, carotenoids, total pigments, phenol, Flavonoids, Carbohydrates %, and DPPH %, Essential oil yield and several other variables. This suggests that Herb fresh weight is closely related to these variables. Herb dry weight also shows strong positive correlations with Essential oil (%), Essential oil yield, Chlorophyll a, Chlorophyll b, carotenoids, total pigments, phenol, Flavonoids, and DPPH %, and several other variables, indicating a strong relationship with these traits. Essential oil (%) has positive correlations with several other variables, including Chlorophyll a, Chlorophyll b, carotenoids, total pigments, phenol, Flavonoids, Carbohydrates %, and DPPH %, and others, the Essential oil (%) appears to be related to with several variables, including Chlorophyll a, Chlorophyll b, carotenoids, total pigments, phenol, Flavonoids, DPPH %, Essential and oil yield these means that the Essential oil (%), may be influenced by these traits.

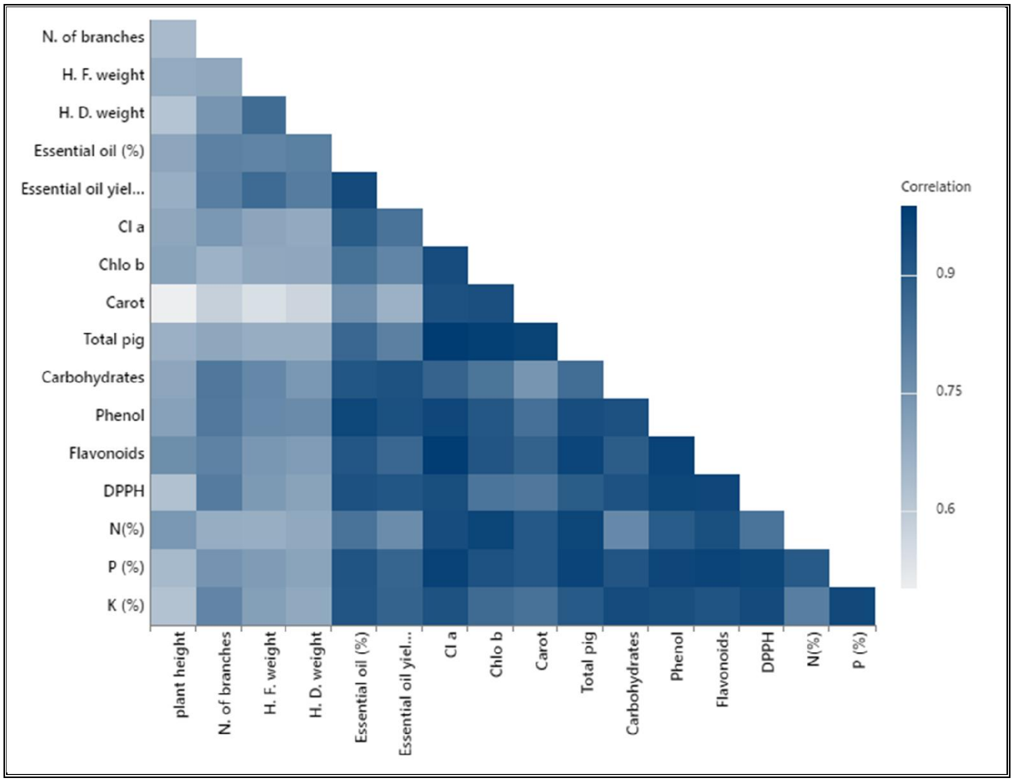

Fig. 1: Correlogram analysis of all investigated traits of plant height, N. of branches, H. F. weight, H. D. weight, Essential oil (%), Essential oil yield (g/plant), Chlo a, Chlo b, Carot, Total pigment, Carbohydrates %, Phenol, Flavonoids, DPPH %, N (%), P (%) and K (%) as affected by foliar application treatments of NPK and P-NPs at rates of (200, 400 and 600 ppm) comparing with control.

4. Conclusion

From the result it be concluded that, the treatment P-NPs at rate of (600 ppm) significantly increased growth characters, essential oil %, total pigments contents, carbohydrates %, Phenol, Flavonoids and DPPH % of mint leaves in both cuts compared to the other treatments, the treatment P-NPs at rate of (600 ppm) increased Pulegone content by 22.66 % compare to NPK treatment, which it is the main component of essential oil.

References

Albalasmeh, A.A., A.A. Berhe, and T.A. Ghezzehei, 2013. A new method for rapid determination of carbohydrate and total carbon concentrations using UV spectrophotometry, Carbohydrate Polymers. 97(2): 253-261.https://doi.org/10.1016/j.carbpol.2013.04.072

Bakry, B.A., S.S. El-Nwehy, R.R. Afify, O.M. Ibrahim and M.E. Abd El-Aziz, 2022. Preparation, characterization and evaluation of nano Phosphorus foliar application on peanut production under sandy soil. Asian J. Plant Sci., 21: 423-431.

Benabdallah, A., C. Rahmoune, M. Boumendjel, O. Aissi, and C. Messaoud, 2016. Total phenolic content and antioxidant activity of six wild *Mentha* species (Lamiaceae) from northeast of Algeria. Asian Pac. J. Trop. Biomed., 6:760–766.

Bindraban, P.S., C. Dimkpa, L. Nagarajan, A. Roy and R. Rabbinge, 2015. Revisiting fertilizers and fertilisation strategies for improved nutrient uptake by plants. Biology and Fertility of Soils, 51(8): 897-911. doi:10.1007/s00374-015- 1039-7

Bisset, N.G., 1995. Herbal Drugs. Medpharm Scientific Publishers: Stuttgart, Germany.

Brady, N.C. and R.R. Weil, 2008. The Nature and Properties of Soils. 14th Edn., Prentice Hall, Inc., New Jersey, ISBN: 13-978-0-13-227938-3.

Brown, D., 1995. Encyclopaedia of Herbs and Their Uses. Dorling Kindersley: London, UK.

Chang, C., M. Yang, H. Wen, and J. Chern, 2002. Estimation of total flavonoid content in propolis by two complementary colorimetric methods, J. Food Drug Anal. 10: 178–182.

Cicek, S. and H. Nadaroglu, 2015. The use of nanotechnology in the agriculture. Advances in Nano research, 3(4): 207–223.

Clevenger, J.F., 1928. Apparatus for the determination of volatile oil. J. Am. Pharm. Assoc., 17: 345-349.

Cottenie, A., M. Verloo, L. Kiekens, G. Velghe, and R. Camerlynck, 1982. Chemical Analysis of Plant and Soil. Laboratory of Analytical and Agrochemistry, State Univ., Ghent, Belgium.

Dhewa, T., 2015. Nanotechnology applications in agriculture: an update. Octa Journal of Environmental Research, 3(2): 204-211.

Ditta, A., 2012. How helpful is nanotechnology in agriculture Advances in Natural Sciences: Nanoscience and Nanotechnology., 3:10.

EL-Gohary, A.E., S.E. El-Sherbeny, G. M.E.M. Ghazal, K.A. Khalid, and M.S. Hussein, 2014. Evaluation of essential oil and monoterpenes of peppermint *(Mentha piperita* L.) under humic acid with foliar nutrition. J. Mater. Environ. Sci. 5 (6):1885-1890.

El-Saadony, M.T., A.M. Saad, A.A. Najjar, S.O. Alzahrani, F.M. Alkhatib, E. Selem, S.M. Desoky, S.S. Fouda, A.M. El-Tahan, and M.A.A. Hassan, 2021. The use of biological selenium nanoparticles in controlling *Triticum aestivum* L. crown root and rot diseases induced by *Fusarium* species and improve yield under drought and heat stress. Saudi J. Biol. Sci., 28:4461–4471.

Elsayed, A.A.A., A. El-Gohary, Z.K. Taha, H.M. Farag, M.S. Hussein and K. AbouAitah, 2022. Hydroxyapatite nanoparticles as novel nano-fertilizer for production of rosemary plants. Scientia Horticulturae, 295, 110851.doi:https://doi.org/10.1016/j.sci enta.2021.110851

Fellet, G., L. Pilotto, L. Marchiol and E. Braidot, 2021. Tools for nano-enabled agriculture: fertilizers based on calcium phosphate, silicon, and chitosan nanostructures. Agronomy, 11(6). doi:10.3390/agronomy11061239

Folle, F., J.W. Shuford, R.W. Taylor, A.A. Mehadi, and W. Tadesse, 1995. Effect of sludge treatment, heavy metals, phosphate rate, and ph on soil phosphorus. Communications in Soil Science and Plant Analysis, 26(9-10): 1369-1381.

Freed, R., S.P. Einensmith, S. Gutez, D. Reicosky, V.W. Smail, and P. Wolberg, 1989. User's Guide to MSTAT-C Analysis of agronomic research experiments. Michigan State University, East Lansing, U.S.A.

Gaikwad, G.S., S.C. Vilhekar, P.N. Mane, and E.R. Vaidya, 2017. Impact of organic manures and hydrophilic polymer hydrogel on conservation of moisture and sunflower production under rainfed condition. Advance Research Journal of Crop Improvement, 8(1): 31-35.

Gitari, J.N. and J.G. Mureithi, 2003. Effect of Phosphorus fertilization on legume nodule formation and biomass production in Mount Kenya region. East Afr. Agric. For. J., 69: 183-187.

Gomez, K.A. and A.A. Gomez, 1984. Statistical Procedure for Agricultural Research. 2nd Edition, Willey, Hoboken, 28-192.

Gonzalez, M., B. Guzman, R. Rudkyk, E. Romano, and M.A.A. Molina, 2003. Spectrophotometric determination of phenolic compounds in propolis. Latin American Journal of Pharmacy, 3: 243-248. Available from: http:// www.latamjpharm.org/trabajos

Gulluce, M., F. Shain, M. Sokmen, H. Ozer, D. Daferera, A. Sokmen, M. Polissiou, A. Adiguel, and H. Ozcan, 2007. Antimicrobial and antioxidant properties of the essential oils and methanol extract from *Mentha longifolia* L. spp. longifolia. Food Chem., 103: 1449–1456.

Gyamfi, M.A., M. Yonamine, and Y. Aniya, 2002. Free radical scavenging action of medicinal herbs from Ghana Thonningia sanguine on experimentally induced liver injuries. Gen Pharmacol. 32:661–7.

Hussain, A.I., F. Anwar, P.S. Nigam, M. Ashraf, A.H. Gilani, 2010. Seasonal variation in content, chemical composition and antimicrobial and cytotoxic activities of essential oils from four *Mentha* species. J. Sci. Food Agric., 90: 1827–1836.

Jackson, M.L., 1973. Soil Chemical Analysis. Prentice Hall of India Pvt., Ltd., M.97, Connght Citrus, New Delhi.

Kopittke, P.M., E. Lombi, P. Wang, J.K. Schjoerring and S. Husted, 2019. Nanomaterials as fertilizers for improving plant mineral nutrition and environmental outcomes. Environmental Science: Nano, 6(12): 3513-3524. doi:10.1039/C9EN00971J

Li, Y. and M. Chen, 2015. Novel chlorophylls and new directions in photosynthesis research. Funct. Plant Biol., 42: 493-501.

Liu, R. and R. Lal, 2015. Potentials of engineered nanoparticles as fertilizers for increasing agronomic productions. Science of The Total Environment, 514:131-139.

Lorenzi, H. and F.J.A. Matos, 2002. Plantas medicinais no Brasil: Nativas e exóticas cultivadas, Instituto Plantarum, Nova Odessa.

Maghsoodi, M.R., L. Ghodszad and B. Asgari Lajayer, 2020. Dilemma of hydroxyapatite nanoparticles as phosphorus fertilizer: Potentials, challenges and effects on plants. Environmental Technology & Innovation, 19, 100869. doi:https://doi.org/10.1016/j.eti.2020.10 0869

Manjunatha, S.B., D.P. Biradar, and Y.R. Aladakatti, 2016. Nanotechnology and its applications in agriculture: A review. J. Farm. Sci., 29(1): 1-13.

Mittal, D., G. Kaur, P. Singh, K. Yadav and S.A. Ali, 2020. Nanoparticle-Based Sustainable Agriculture and Food Science: Recent Advances and Future Outlook. Frontiers in Nanotechnology, 2.

Montalvo, D., M.J. McLaughlin and F. Degryse, 2015. Efficacy of Hydroxyapatite Nanoparticles as Phosphorus Fertilizer in Andisols and Oxisols. Soil Science Society of America Journal, 79(2): 551- 558. doi:10.2136/sssaj2014.09.0373

Mozaffari, M. and J.T. Sims, 1994. Phosphorus Availability and Sorption in an Atlantic Coastal Plain Watershed Dominated by Animal-Based Agriculture. Soil Science, 157(2).

Nazem, V., R.M. Sabzalian, G. Saeidi, and M. Rahimmalek, 2019. Essential oil yield and composition and secondary metabolites in selfand open-pollinated populations of mint (*Mentha* spp.). Ind. Crops Prod., 130:332–340.

Ogola, A.H., G.D. Odhiambo, J.R. Okalebo and F.N. Muyekho, 2012. Influence of Phosphorus on selected desmodium growth and nodulation parameters. ARPN J. Agric. Biol. Sci., 7: 294-301.

Patra, P., S.R. Choudhury, S. Mandal, A. Basu and A. Goswami, 2013. Effect sulfur and ZnO nanoparticles on stress physiology and plant (*Vigna radiata*) nutrition. In: Advanced Nanomaterials and Nanotechnology, Giri P.K., D.K. Goswami and A. Perumal (Eds.)., Springer Berlin Heidelberg, Germany, ISBN-13: 978-3-642-34216-5, 301-309.

Prasad varx, P.V., S.R. Pisipati, and M. Ivana, 2011. Independent and combined effects of high temperature and drought stress during grain filling on plant yield and chloroplast EF-Tu expression in spring wheat. Journal of agronomy and crop science, 197(6):430 – 441.

Sabry, R.M., A.A.A. Elsayed, R.A. El-Ziat, Z.K. Taha, H.M. Farag and K. AbouAitah, 2023. Hydroxyapatite nanoparticles as effective phosphorus nano-fertilizer on Italian parsley plants. Scientific Journal of Agricultural Sciences, 5 (1): 20-36. 10.21608/sjas.2023.185609.1278

Selvakumar, G., M. Lenin, P. Thamizhiniyan and T. Ravimycin, 2009. Response of biofertilizers on the growth and yield of blackgram (*Vigna mungo* L.). Recent Res. Sci. Technol., 1: 169-175.

Singh, P.P., P. Pandey, D. Singh, S. Singh, M.S. Khan, and M. Semwal, 2020. '*Mentha* Mitra'—an android app based advisory digital tool for menthol mint farmers. Ind. Crops Prod. 144, 112047. https://doi.org/10.1016/j.indcrop.2019.112047.

Singh, S., B.K. Singh, S.M. Yadav, and A.K. Gupta, 2015. Applications of nanotechnology in agricultural and their role in disease management. Research Journal of Nano science and Nanotechnology, 5(1): 1-5.

Steel, R.G.D., J.H. Torrie, and D.A. Dickey, 1997. Principles and procedures of statistics. A biometrical approach, 3rd edn. McGraw Hill Book Co. Inc, New York.

Szameitat, A.E., A. Sharma, F. Minutello, A. Pinna, M. Er-Rafik, T.H. Hansen and S. Husted, 2021. Unravelling the interactions between nanohydroxyapatite and the roots of phosphorus deficient barley plants. Environmental Science: Nano, 8(2): 444-459. doi:10.1039/D0EN00974A

Tarasevi˘ciene, Ž., A. Veli˘cka, and A. Paulauskiene, 2021. Impact of Foliar Application of Amino Acids on Total Phenols, Phenolic Acids Content of Different Mints Varieties under the Field Condition. Plants, 10: 599. https://doi.org/10.3390/plants10030599

Tucker, A.O., 2007. *Mentha*: Economic Uses. In Mint, the Genus *Mentha*; Lawrence, B.M., Ed., CRC Press: Boca Raton, FL, USA, 519–522.

Veronica, N., T. Guru, R. Thatikunta, and S.N. Reddy, 2015. Role of Nano fertilizers in agricultural farming. International Journal of Environmental Science and Technology, 1(1): 1-3.