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THE ROLE OF MELATONIN AND MAGNESIUM ON SOME CHEMICAL PROPERTIES OF SPINACH PLANTS (*SPINACEA OLERACEA* L.)

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Abstract:
On a farm in the Um Al-Akf area, the experiment was carried out in the winter of 2023–2024 to investigate the effects of spraying spinach plants with melatonin at three different concentrations (0, 50, and 100 mg L–1) and magnesium at three different concentrations (0, 100, and 200 mg L–1). The complete randomized block design (C.R.B.D.) was used to conduct a factorial experiment with three replicates, and the averages were compared at the 0.05 probability level. The most important results of the experiment are: Spraying spinach plants with melatonin had a noteworthy outcome on all plant Qualities
On a farm in the Um Al-Akf area, the experiment was carried out in the winter of 2023–2024 to investigate the effects of spraying spinach plants with melatonin at three different concentrations (0, 50, and 100 mg L–1) and magnesium at three different concentrations (0, 100, and 200 mg L–1). The complete randomized block design (C.R.B.D.) was used to conduct a factorial experiment with three replicates, and the averages were compared at the 0.05 probability level. The most important results of the experiment are: Spraying spinach plants with melatonin had a noteworthy outcome on all plant Qualities

Keywords: Melatonin, magnesium, chlorophyll, carbohydrates, vitamin C.

INTRODUCTION

Spinach (Spinacea oleracea L.) is a crop that is leafy green and a member of the Chenopodacea family. Asia is said to have been its initial habitat., specifically Iran, from where it moved to China before Christ and then to European countries at the beginning of the twelfth century. The cultivated area in the world is estimated at about 980,000 hectares (FAO, 2011), In contrast, the Central Statistical Organization - Ministry of Planning estimates that the cultivated area in Iraq is approximately 540 hectares, with an annual production rate of roughly 3234 tons.

Spinach is a leafy green crop abundant in phenolic and carotenoids, two types of antioxidant chemicals, flavonoids, and some vitamins such as vitamin C, in addition to the plant containing many nutritional elements such as iron, magnesium, potassium, and calcium (Gill et al., 1999). Melatonin is a plant hormone whose scientific name is (MelatoninN-acetyl-5-methoxy-tryptamine), and it has the molecular weight of 232.278 g mol-1 and the chemical formula $C_{13}H_{16}N_2O_2$.It is a white powder that dissolves in ethyl alcohol. Almost all living things include the amino acid tryptophan, which serves as the catalyst for its manufacture (Hardeland et al., 2011). This hormone, which controls both human and animal biological rhythms, is secreted by the pineal gland in the brain of the human. Melatonin is one of the compounds that have been recently used as it works to shield the plant from the impacts of oxidative stress brought on by salt stress, and to improve plants' tolerance to environmental challenges before attempting to lessen their detrimental effects (Altaf et al., 2020), given that a number of scientists have hypothesized that it might have certain effects akin to those of the auxin indole-3-acetic acid (IAA) (Kolár and et al.). Melatonin is thought to play a significant role in controlling a variety of physiological processes in higher plants, including growth, promoting the development of a strong root system, carbon representation, preserving the stability of chlorophyll, and internal water balance (Hernandez et al., 2015; Antoniou et al., 2017), as Wang et al. (2016) added melatonin to cucumber plants Curcumas sativa L. decreased the impact of salt stress and raised the levels of total chlorophyll content, photosystem II (PSII) maximum quantum efficiency, and net carbon assimilation rates. Mousse and Al-Jamal (2016) found that treating spinach plants with melatonin increased the fresh and the plant's dry weight, increased the number of leaves, and increased their total chlorophyll and carbohydrate content. Due to its unique role in the metabolism of carbon, the synthesis of food, and numerous other physiological processes, magnesium is considered one of the necessary nutrients for plants (Al-Lami, 1999). It is also a mineral that is essential to the makeup of the chlorophyll molecule. It has another role in carbohydrate metabolism because it is a catalyst for many enzymes. This element is also necessary to obtain energy from (ATP) as it links the enzyme protein with the phosphate group of (ATP) and it also plays a role in the process of protein formation as it stabilizes the structure of ribosomes that completes the process of protein construction (Abu Dahi, 1997). Hussein and Rabie (2009) found that treating pea plants *Pisum sativum* L. Using 10 g L-1 of magnesium sulfate resulted in a notable boost in plant growth, which in turn raised the plant's dry weight. Al-Jubouri (2016) also showed that adding magnesium to basil plants *Ocimum basilicum* L. In the form of magnesium chloride at the levels of 25 mg L-1 produced a notable impact on the amount of vital nutrients N, P, and K in the leaves and also raised the plant's dry weight in comparison to plants that were not treated.

THE OBJECTIVE OF THE STUDY

To investigate how giving spinach plants melatonin and magnesium can boost plant growth and, in turn, increase the plant's output of leaves.

MATERIALS AND WORKFLOW TECHNIQUES

The experiment was conducted on a farm in the Umm Al-Akf neighborhood of the city of Samawah / Muthanna Governorate during the winter of 2023–2024. The field soil is defined as a medium-fertile soil with a certain amount of salts. A few of the chemical and physical characteristics of the irrigation water and field soil are shown in Table 1.

Table 1: lists the physical and chemical characteristics of irrigation water and soil samples in pots.

Method used	Type of analysis	Measuring unit	
Page *et al. (1982)*	ECe	ds m^{-1}	4.5
	TDS	g L^{-1}	3.9
	pH	----	7.36
	Ready nitrogen	mg kg^{-1}	330
	Ready phosphorous	mg kg^{-1}	340
	Ready potassium	mg kg^{-1}	205
	Organic matter	%	0.59
	Clay ratio	%	26.47
	Silt ratio	%	65.45
	Sand ratio	%	8.08
	Soil tissue	%	Silty clay
Irrigation water properties			
Page et al. (1982)	ECe	ds m^{-1}	2.4
	TDS	g L^{-1}	0.8
	pH	----	7.2

Two parameters were examined in this experiment: the first involved foliar spraying spinach plants with melatonin at three different concentrations (0, 50, and 100 mg L-1); the second involved spraying plants with magnesium (magnesium chloride) at three different doses (0, 100, and 200 mg L-1). Only distilled water was sprayed on the comparative treatment. In accordance with Al-Rawi and Khalaf Allah (2000), the treatments were allocated in the experiment using a Randomized Complete Block Design (R.C.B.D.) with three replicates, resulting in nine treatments overall and 27 experimental units with 16 plants each. Twice, the plants were sprayed with the above concentrations; the first application occurred twenty days after the emergence of the third true leaf, and the second application occurred fifteen days following the first. Six plants were randomly taken from each experimental unit and the characteristics of the basic nutrients nitrogen, phosphorus and potassium, the leaf content of vitamin C, chlorophiyll and carbohydraties, the leaf content of total soluble solids and the dry weight of leaves were measured .

RESULTS AND DISCUSSION:

The effects of magnesium spraying and melatonin on the amount of vital elements (N, P, K) and vitamin C in spinach leaves are displayed in Table 2. It has been shown that when melatonin was sprayed at both doses, the amount of nitrogen, phosphorus, potassium, and vitamin C in the leaves rose, with the content increasing when treated with the greater concentration compared to untreated plants. The aforementioned table also shows that applying a magnesium spray significantly increased the amount of the aforementioned features in the leaves, with the greater concentration achieving the highest values when compared to untreated plants.

In comparison to the control treatment plants that were sprayed with distilled water alone, it is observed that there is a significant increase in the case of the interaction between the two factors, and the best results were obtained from spraying with melatonin at a concentration of (100 mg L-1) and magnesium at a concentration of (200 mg L-1).

Table 2: Impact of melatonin and magnesium spraying and their combination on the nutritional content of spinach leaves

Melatonin mg L^{-1}	Mg mg L^{-1}	N %	P %	K %	Vit. C mg $100g^{-1}$ ww
0	0	2.09	0.197	1.62	23.11
	100	2.37	0.233	1.82	25.72
	200	2.93	0.277	2.10	29.72
50	0	2.64	0.234	1.81	28.11
	100	2.98	0.292	2.13	33.97
	200	3.35	0.332	2.65	35.10
100	0	3.22	0.309	2.44	35.27
	100	3.88	0.370	3.05	39.38
	200	4.53	0.439	3.50	44.17
L. S. D.		0.12	0.017	0.14	1.47
Effect Melatonin	0	2.47	0.236	1.85	26.18
	50	2.99	0.286	2.20	32.39
	100	3.88	0.373	2.99	39.61
L. S. D.		0.07	0.010	0.08	0.85
Effect Mg	0	2.65	0.246	1.96	28.83
	100	3.08	0.298	2.33	33.02
	200	3.60	0.349	2.75	36.33
L. S. D.		0.07	0.010	0.08	0.85

Table 3 illustrates the impact of melatonin and magnesium spraying, as well as their combined effect, on the chlorophyll content of spinach leaves, total dissolved solids, soluble carbohydrates, and the plant's overall dry weight. It is shown that melatonin spraying significantly increased the amount of the aforementioned features in the leaves, with the concentration (100 mg L-1) being superior to that of the untreated plants. Magnesium spraying has a noteworthy impact as well; all of the features in the above table showed a substantial increase with the superiority of the treatment at the concentration (200 mg L-1) over untreated plants. When the two factors interact, the above traits increase significantly. The best results were obtained when plants were treated with melatonin at a concentration of 100 mg L-1 and magnesium at a concentration of 200 mg L-1, as compared to untreated plants.

Table 3: shows how magnesium and melatonin spraying affect the amount of chlorophyll in spinach leaves as well as the total soluble solids, soluble carbohydrates, and total dry weight of the plant.

Melatonin mg L^{-1}	Mg mg L^{-1}	Chlorophyll mg $100g^{-1}$ ww	TSS	Carbohydrates mg g-1	D. W.
0	0	25.74	2.73	19.40	4.46
	100	29.43	3.31	24.14	5.17
	200	32.89	3.81	29.22	6.33
50	0	29.20	3.38	25.00	5.39
	100	31.20	3.79	30.32	6.92
	200	33.39	4.26	34.29	7.87
100	0	34.95	4.04	29.17	7.72
	100	39.50	4.79	36.43	9.11
	200	44.80	5.43	43.96	10.81
L. S. D.		1.02	0.13	1.02	0.31
Effect Melatonin	0	29.35	3.29	24.25	5.32
	50	31.26	3.81	29.87	6.73
	100	39.75	4.75	36.52	9.21
L. S. D.		0.59	0.08	0.59	0.18
Effect Mg	0	29.96	3.38	24.52	5.86
	100	33.38	3.96	30.30	7.07
	200	37.02	4.50	35.82	8.34
L. S. D		0.59	0.08	0.59	0.18

The above tables show the significant effect of treating spinach plants with melatonin and magnesium on all the traits under study. We can attribute the reason to the positive role of both substances, as melatonin acts as an antioxidant and can effectively contribute to protecting DNA and also protects against damage to cell membranes. It also contributes to inhibiting peroxide reactions by removing free radicals formed by exposure to stress (Küçükyumuk, 2021). The reason may also be due to the fact that the external addition of melatonin works to reduce the pH between plant cells and activate enzymes responsible for the flexibility of cell walls, which in turn works to elongate cells, thus its work is similar to the work of auxin (IAA) (Yang et al., 2021), in addition to the fact that melatonin stimulates an increase in the biosynthesis of hormones and internal growth stimulants such as metabolites, auxins and enzymes responsible for removing free radicals in plants (Shafi et al., 2021), It causes a rise in the density of the root system and hence a rise in the vegetative system. It was also found that melatonin reduced the decomposition of the green pigment (chlorophyll) under environmental stresses by reducing the gene expression of the enzyme (Pheophorbide-a-oxygenase (PAO)), which contributes to the decomposition of chlorophyll (Liu et al., 2022). The significant effect of magnesium treatment may be due to its role in activating many enzymes associated with carbohydrate metabolism such as glued kinase or Hexokinase, etc., and may also be due to its useful part in the synthesis of nucleic acids (Deafen and Wesam, 1998), as well as its association with nitrogen atoms of the Porphyrin compound in the composition of the chlorophyll molecule. It also plays an important role in the release of electrons as a result of exposure to sunlight, and this is the basic step in the process of carbon metabolism in plants (Abdul, 1988). It is also necessary in activating the enzyme necessary to fix the carbon dioxide molecule in the Calvin cycle in the dark reactions of C3 plants in the process of food production (Marschaer, 1995).

CONCLUSIONS

1. All of the characteristics under investigation significantly increased when melatonin spraying at a concentration of 100 mg L-1.
2. A 200 mg L-1 magnesium spray produced a statistically significant improvement in every feature that was being studied.
3. All of the characteristics under investigation significantly increased after magnesium and melatonin sprays at concentrations of 200 mg L-1 and 100 mg L-1, respectively.

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