Effect of cobalt salts on some physiological indicators in the leaves of (*Lactuca sativa* L.) grown hydroponically

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Abstract

This study aimed to study the effect of adding two types of Cobalt salts, namely Cobalt sulfate (CoSO4.7H2O) and Cobalt nitrate Co(NO3)2.6H2O , on some vital indicators of the shoots of lettuce plants, which are leaf area, fresh weight, dry weight, leaf moisture content, and Chlorophyll concentration. Hydroponics technology was used in this experiment.

The concentrations added to both salts were 10, 30, and 50 mg L-1 . Adding Cobalt salts in high concentrations had toxic and negative effects on the plant's vegetative system.

There was a significant decrease in the leaf area of the plant, which gradually decreased with increasing concentration of Cobalt salts. At the same time, the same effect occurred on the wet and dry weight of the plant leaves. With minimal effects on the percentage of moisture content of the paper.

An effect was also observed on the total chlorophyll content of plant leaves, as the concentration of 50 mg L-1 had a significant effect in reducing the total chlorophyll content.

Key words : Hydroponic , Lettuce , Chlorophyll , leaf area , fresh weight , dray weight , moisture content

Introduction

Agriculture in the present day faces a variety of challenges such as a rise in population, a decrease in arable land, and rapid urbanization. Hence, there has been a shift from conventional cultivation towards soilless cultivation (20) .With the decline in the cost of hydroponic equipment and electricity, hydroponics may become a viable option for small cities and common crops (5). However, due to the current high energy requirements and costs associated with hydroponics, it is currently only feasible for big cities and high-value crops (23). Therefore, lettuce is grown using hydroponics due to its short growth cycle and high yield (6.(

Heavy metals (HVs) available for plant uptake are those present as soluble components in the soil solution, or those dissolved by root exudates. Plants need certain heavy metals for their growth and maintenance, and excess amounts of these metals can become toxic to plants. Since metals cannot be broken down, when concentrations within a plant exceed optimal levels, they negatively affect the plant directly and indirectly. These toxic effects (direct and indirect) lead to reduced plant growth, which ultimately leads to plant death. The effect of heavy metal toxicity on plant growth and development varies according to the specific heavy metal (2 .(

Cobalt is considered a plant micronutrient because it is essential for the growth of many higher and lower plants. (7). But, it also negatively affects plant growth and

ISSN 2072-3857

productivity at elevated levels. The damage to plant cells and cell membranes due to excessive amounts of Cobalt is irreversible, and this phenomenon disrupts water and nutrient uptake, thus impairing growth (3.(

Lettuce (Lactuca sativa L.), Belonging to the Compositae family, is one of the most significant vegetable crops globally. It is typically cultivated in open fields and greenhouses, with simple hydroponic systems often utilized for lettuce production (14). It is one of the most consumed leafy plants around the world and is available throughout the year. Lettuce is also an important source of phytochemicals(25). Lettuce is a widely cultivated vegetable. In addition, some studies have revealed that lettuce has high yields and good quality when grown in a soilless system (15.(

The leaf area index is a dimensionless variable and was first defined as the total one-sided area of photosynthetic tissue per unit of ground surface area (24).The leaf area index is a key variable for characterizing crop growth conditions and estimating crop productivity (9 .(

The importance of chlorophyll lies in the fact that it is a photoreceptor that plants cannot do without in the process of photosynthesis. Chlorophyll is also considered a physiological indicator that is widely used in botany research because of its close connection to the process of photosynthesis in plants (1) . Therefore, the study of this subject is crucial as it plays a significant role in the exchange of energy between the biosphere and the atmosphere, as well as in the functioning of terrestrial ecosystems. (4. (

Material and Methods

One-month-old lettuce seedlings were transferred to seven plastic pipes (PVC), each two meters long and four inches in diameter. The pipes have ten holes with a diameter of three inches, which contain perforated plastic cups filled with a perlite medium to support the seedlings. The tubes are designed using the nutrient membrane method, as shown in Figure 1. The seedlings were Provided with the Hoagland nutrient solution, which was updated every two weeks Until the end of the Experiment period. After transferring the seedlings to the hydroponic system, they were treated with two solutions of Cobalt salts two weeks later: Cobalt sulfate CoSO4.7H2O and Cobalt nitrate Co(NO3)2.H2O, at concentrations of 10, 30, and 50 mg L-1 for each salt with Hoagland's solution to treatments. in addition to a control treatment prepared with only Hoagland's solution. After a month of adding Cobalt salts, the plants were harvested to conduct laboratory tests

ISSN 2072-3857

Figure 1: description of the Experiment Designleaves area measurement

To measure the leaf area of the plant, the fourth lower leaf of each replicate used in the experiment for each treatment was drawn on a sheet of paper. The papers were fixed on the table with an adhesive, and a digital Planimeter (figure 2) was used to measure the area (10).

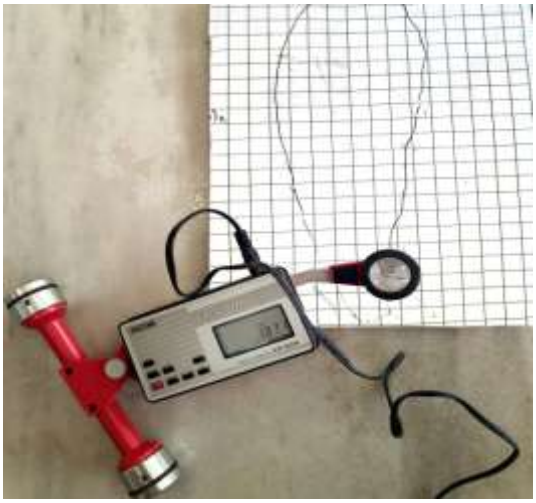

figure 2: digital Planimeter used in to experience Measure the leaf area

calculate the fresh weight , dry weight, and moisture content

To calculate the fresh weight, one leaf was taken from each replicate, five replicates for each sample, and its mass was calculated using a sensitive balance.

After that, these leaves were placed in an electric oven for two days at a temperature of70 ° C to calculate the dry weight after weighing the dry leaf completely, as well as calculating the moisture content According to the equation mentioned by (17.(

Chlorophyll a and b, and total chlorophylls

The content of chlorophyll a, b, total chlorophyll, and carotenoids was estimated based on the method (11). where 0.25 g of fresh lettuce leaves were taken and placed in 80% acetone for a week until all pigments were extracted from the plant leaf. The absorbance was then read at wavelengths of 645, 663 nm .

Statistical analysis

ISSN 2072-3857

The data of 5 replication for each treatment was analyzed using SPSS (version 26, SPSS Inc. Chicago, Illinois, USA). Differences were compared by One-way ANOVA, at $p \leq 0.05$ using Duncan's multiple-range test. The value of $p < 0.05$ was considered statistically significant.

Results

leaves area measurement

According to the table(1) , it was observed that adding Cobalt salts led to significant differences in the area of plant leaves. The addition caused a clear decrease in leaf area with increasing concentrations of both Cobalt sulfate and Cobalt nitrate. The lowest leaf area was recorded at a concentration of 50 mg L-1, where the lowest leaf area recorded was 89.4 cm2 for treatment with Cobalt sulfate. It is noteworthy that the control sample had the highest leaf area,

with an average of 215.9 cm2

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Table (1): The effect of Cobalt salts on the leaf area (cm2) of lettuce plants grown using hydroponic technology The results are average of five replicates. ($P \leq 0.05$).

Concentration of Cobalt salts (mg L^{-1}) / Parameters		Leaf area (cm2)
Hoagland's solution only	0	215.9 d
Hoagland's solution + Cobalt sulfate $CoSO_4.7H_2O$	10	193.9 c
	30	135.9 b
	50	89.4 a
Hoagland's solution + Cobalt nitrate $Co(NO_3)_2.6H_2O$	10	204.2 c
	30	135.0 b
	50	93.9 a

Different letters indicate significance at a significance level of $p \leq 0.05$.

fresh weight , dry weight, and moisture content

It was noted in Table No. (2) that treating plant with Cobalt salts resulted in significant differences in their fresh and dry weights. The study found that there was a gradual decrease in the fresh weight of the shoot as the concentration of Cobalt salts increased. The lowest fresh weight was recorded when a concentration of 50 mg L-1 of Cobalt sulfate was used, resulting in a weight of 3.95 grams compared to the control treatment, which resulted in a weight of 9.94 g.

It was also noted that there was a significant difference in dry weight in the same table when treated with Cobalt salts, as the lowest average dry weight was when treated with Cobalt salts at a concentration of 50 mg L-1, Which was estimated at 0.25 g compared to the control treatment, which resulted in a weight of 0.5 g.

ISSN 2072-3857

In the same table, the study found slight significant changes in the percentage of moisture content of plant leaves. The lowest significant decrease was recorded when using a concentration of 50 mg L-1 of Cobalt sulfate, which resulted in a moisture content of 93.56% compared to the highest percentage for the control sample, which amounted to 95.23%.

Table (2): The effect of Cobalt salts on the water content of the shoots of lettuce plants grown using hydroponic technology The results are average of five replicates. (P ≤ 0.05).

Concentration of Cobalt salts (mgL^{-1}) / Parameters		Fresh weight	Dray weight	Percentage of moisture content
Hoagland's solution only	0	9.94 d	0.50 c	95.23 bc
Hoagland's solution + Cobalt sulfate $CoSO_4.7H_2O$	10	6.24 b	0.38 abc	93.99 ab
	30	5.42 b	0.34 ab	93.70 a
	50	3.95 a	0.25 a	93.56 a
Hoagland's solution + Cobalt nitrate $Co(NO_3)_2.6H_2O$	10	8.00 c	0.42 bc	94.78 abc
	30	6.42 b	0.30 ab	95.38 c
	50	4.62 ab	.25 a0	94.75 abc

Different letters indicate significance at a significance level of p≤0.05.

Chlorophyll a and b, and total chlorophylls

A slight difference in the reduction of chlorophyll a and chlorophyll b content was observed in Table No.(3) . The treatment with Cobalt sulfate at a concentration of 50 mg L-1 showed the lowest significant decrease in the concentration of both chlorophyll a and b. In this treatment, the concentration of chlorophyll A was 1.48 mg g-1, and for chlorophyll b, it was 1.54 mg g-1. Thus, the same treatment showed the lowest significant decrease in the total chlorophyll concentration, which was 3.01 mg g-1. It is worth noting that the maximum value of total chlorophyll was observed in the treatment with Cobalt sulfate at a concentration of 30 mg g-1, where the amount of chlorophyll was 5.49 mg g-1

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ISSN 2072-3857

Table (3): The effect of Cobalt salts on the chlorophyll concentration of lettuce plants grown using hydroponic technology The results are average of five replicates. ($P \leq 0.05$).

Concentration of Cobalt salts (mgL^{-1}) / Parameters		Chlorophyll a	Chlorophyll b	Total chlorophyll
Hoagland's solution only	**0**	2.04 **b**	2.88 **d**	4.92 **e**
Hoagland's solution + Cobalt sulfate $CoSO_4.7H_2O$	**10**	2.07 **b**	2.47 **c**	4.54 **cd**
	30	2.06 **b**	3.44 **e**	5.49 **f**
	50	1.48 **a**	1.54 **a**	3.01 **a**
Hoagland's solution + Cobalt nitrate $Co(NO_3)_2.6H_2O$	**10**	2.56 **c**	2.19 **b**	4.74 **de**
	30	1.78 **ab**	2.22 **b**	4.00 **b**
	50	1.98 **b**	2.52 **c**	4.50 **c**

Different letters indicate significance at a significance level of $p \leq 0.05$.

Discussion

The results indicated a significant decrease in the leaf area of lettuce plants, as the leaf area decreased as the concentration of Cobalt salts increased. The accumulation of Cobalt absorbed by the roots increases significantly in plant leaves, and this leads to toxic effects In these plant leaves (13) and (8). Excessive amounts of heavy metal negatively affect plant growth, cause yield loss, and have undesirable effects on crops (16 . (

There was a significant decrease in biomass, as it was found that increasing the concentration of Cobalt salts leads to a decrease in the fresh and dry weight of the plant, as was expected in some studies of the enhancing effect of low doses of Cobalt on both the fresh and dry weights of the plant.(19) and (22) . (18) found that high levels of Cobalt addition inhibited cucumber plant growth This also affected the moisture content of the plant, which had varying effects and may be related to heavy metal toxicity.

The total chlorophyll concentration also decreased with increasing concentrations of Cobalt due to the negative effect of Cobalt, as mentioned by (2) and (21). Other studies indicated that Cobalt in high concentrations inhibits photosynthesis in secondary leaves more severely than Primary leaves (12.(

Conclusion

Based on the results obtained, it was found that adding high concentrations of Cobalt to these crop has negative effects on plants, as it causes a decrease in leaf area, fresh and dry weight of the plant, as well as chlorophyll content, with the Cobalt concentration increasing beyond the required limit. This in turn affects the quality of the crop. Therefore, the amount of cobalt to be added must be carefully determined based on the type of plant crop to avoid exceeding the required limit.

ISSN 2072-3857

It is important to note that there was no difference in the physiological effect between the Cobalt salts used.

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ISSN 2072-3857