Mitigating Tipburn True Foliar Calcium Application in Indoor Hydroponicly Grown Mini Cos Lettuce

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Abstract. Hydroponic systems provide a highly advantageous environment for cultivating green leafy vegetables. Among these advantages, there is a significant acceleration in harvest compared to conventional farming methods, the attainment of high yields, enhanced leaf coloring, increased nutritional content, and improved sweetness, all of which are crucial factors. However, a particular issue known as tip burn is encountered when cultivating, especially Yedikule (Romaine) type lettuce in hydroponic systems. This study investigates various calcium dosages applied via foliar spray to mitigate tip burn on Thespian and Suntred lettuce varieties (green and red Romaine mini types). Different calcium dosages were employed for both lettuce varieties. This experiment examined various parameters for both lettuce types, including plant height, root length, stem diameter, plant age, dry weight, root dry and fresh weight, macro and micronutrient content, dry matter, and total yield. The results indicate that combining foliar calcium and adequate ventilation effectively mitigates tipburn. This research emphasizes the significance of such a combined approach in preventing tip burn in hydroponically grown lettuce, specifically Thespian and Suntred varieties. Keywords:

Keywords: Lactuca sativa, spraying doses, calcium, yield,

1 Introduction

Agricultural areas and water resources have become increasingly limited in our world. This is due to the unconscious use of fertilizers and pesticides with traditional agricultural methods, which are mixed with groundwater and make water resources unusable. Decreased arable land, urbanization increase, water scarcity, and climate change have pressured agricultural producers (1,2). For this reason, soilless agriculture, shown as an alternative to traditional agriculture, has come to the agenda. The need for production is increasing daily due to the increasing population; however, production areas are decreasing with population growth. As a result, efforts are made to obtain the highest yield from the unit area. Hydroponic systems produce potted ornamental plants, seedlings, and transplants and increase plant metabolites in fruits, vegetables, and medicinal and aromatic plants. Hydroponic production technology affects plant growth, yield, and overall plant quality, increasing plants' cumulative benefits (3,4). Hydroponic culture is a modern cultivation technology that has developed rapidly in the last 30-40 years, mainly applied in greenhouses. Most hydroponic crops are grown in high-tech greenhouse structures with fully automatic climate control features(2,3). Both in the world and our country, vegetable cultivation is carried out in 92% of the soilless farms, and the rest are used for ornamental plants (5).

Tipburn is a necrosis of the margins of young developing leaves on the inside of vegetable crops, especially head-forming vegetables such as lettuce (Lactuca sativa), white cabbage (Brassica oleracea L. var. capitata), Chinese cabbage (B). This physiological disorder can cause severe economic losses. Susceptibility to tip blight is genetically determined but influenced by the environment (6,7,8,9). Ca2+ deficiency is generally recognized as the leading cause of tip burn disorder, and the term calcium deficiency is sometimes used synonymously with tip burn.

Lettuce (Lactuca sativa) is an annual or semi-annual cool-climate vegetable with broad green leaves from the daisy family (Asteraceae). In 2019, world lettuce production was 29,134.65 million kilograms (10). In Turkey, 540,569 tonnes of lettuce was produced in 2022 (11). According to the figures of the Food and Agriculture Organisation of the United Nations (FAO), 55.98% of the world lettuce production in 2019 was produced by China, 12.66% by the USA, 4.33% by India, 3.47% by Spain and 2.6% by Italy. In light of these data, China, the USA, India,

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Spain, and Italy are the largest lettuce-producing countries in the world. In the world where the average yield is 2.21 kg, Belgium is the most productive country in lettuce production with 4.58 kg per m2, followed by the USA with 3.51 kg per m2, Germany with 3.11 kg, the Netherlands with 3.07 kg and Iran with 3.05

Different locations tend to favor different types of lettuce. Northern Europe tends to consume Butterhead lettuce in the U.K. and stem-type lettuce in China and Egypt. In the late 20th century, icebergs became more popular. Sixty-five percent of lettuce is produced and consumed in America. Root lettuce was first cultivated and developed in China. Lettuce production methods, including price, increased in the 20th century. Agricultural production has vastly increased with the use of fertilizers and pesticides, but with organic production, the products of small-scale producers have become more preferred and trendy. In the early 21st century, packaged lettuce began in the markets. Especially in the markets where processed lettuces were not accepted in the previous years, lettuce with longer shelf life was offered to the consumers using a packaging system (13).

Lettuce can be grown all year round in regions with mild climates without harsh winters. Lettuce, which is partially resistant to cold, needs humid weather conditions. The most effective climatic factors limiting lettuce production are temperature and day length. While lettuce grows well in environments with an average temperature of 15-18°C, a maximum temperature of 27-30°C, and a minimum temperature of 2-4°C are considered suitable for the plant to maintain its growth (14, 15, 16, 17).

In calcium-poor soils, low yields are obtained, and the protein ratio in the product is significantly reduced. Calcium deficiency in plants slows down the growth of meristem tissues. The development of shoot tip buds and growth tips of roots stops, and thus, the development of the plant stops. Young leaves are deformed. Black and brown necrosis occurs on the leaf margins. Leaf tips become dry or brittle (easily broken), and the leaf eventually withers and dies (18, 19, 20, 21).

The study aimed to eliminate or reduce the tip blight due to calcium deficiency, which is an economic obstacle in lettuce cultivation in hydroponic cultivation in indoor environments in 2 different mini romain types, green and red lettuce.

2 Materials and Methods

The study was conducted in a climate room of Çukurova University, Faculty of Agriculture, Department of Horticulture, where many studies were conducted. This experiment was carried out during the autumn and winter growing periods of 2022 and 2023. Plants were harvested 51 days after transfer to the hydroponic system. Illumination was set to 16 hours day and 8 hours night. The temperature was fixed at 21 degrees C. This study applied foliar calcium to Romain-type Thespian and Suntred lettuce varieties to prevent tip burn formation, and the optimum dose was investigated . In the Thespian lettuce variety, control, 600 ppm, 800 ppm, and 1000 ppm CaO were foliar sprayed once a week to determine the ideal dose with 72 plants. In the Suntred lettuce variety, control, 200 ppm, 400 ppm, and 800 ppm CaO were foliar sprayed once a week to determine the ideal dose with 72 plants. Stock solutions were used for fertilization, and nutrient solutions were changed every four days. This dose difference was determined as a result of our preliminary experiments.

Total yield, plant height, root length, plant stem diameter, plant circumference, plant width, plant fresh weight, root fresh weight, number of leaves, leaf area, green parts dry weight, nutrient analyses (P, K, Mg, Ca, Cu, Mn, Fe, Zn) were performed. Plant and root height, plant circumference, and plant width were measured with a meter with a sensitivity of 1 cm, stem diameter was measured with a caliper in mm, and plant and root fresh weights were measured with a balance with a sensitivity of 0.1 and leaf area was measured with Image-J. The number of leaves was obtained by counting the whole plant by hand. Nutrient analysis was carried out by FS 220 Atomic Absorption

Spectroscopy (Jones, 1972). Nitrogen analysis was done by Khejdahl (1883) method. Phosphorus analysis was done colorimetrically using the vanadomolybdophosphoric yellow color method described in Lott et al. (1956).

Fig. 1. - Thespian and Suntred lettuce varieties grown in water culture in climate chamber.

3 Results and Discussion

Thespian

Table 1 Effects of CaO treatments at different ppm on plant height, plant width, plant circumference, and stem diameter of Thespian lettuce cultivar

Apps	Plant Height (cm)	Plant Width (cm)	Plant Circumference (cm)	Body Diameter (cm)
1	22,51	32,3	38,0	14,6
2	23,51	31,0	41,0	14,4
3	24,19	33,58	40,6	13,6
4	24,07	34,52	38,3	13,7
5	N.S.	N.S.	N.S.	N.S.

1: Control; 2: 200 ppm CaO; 3:400 ppm CaO; 4:800 ppm CaO; 5; LSD

When the plant height parameter was evaluated, it was observed that the highest plant height was 23.5 cm due to 800 ppm CaO application. In the control group, this value was 22.7 cm. Regarding plant width, the highest value obtained with 1000 ppm CaO application was 34.0 cm, while the lowest value was recorded as 31.0 cm with 600 ppm CaO application. In plant circumference measurements, the highest value of 41.0 cm was reached with 600 ppm CaO application. In the control group, this value was 38.0 cm. In the stem diameter evaluation, the highest value of 14.6 cm was obtained in the control treatment.

In comparison, the lowest value of 13.6 cm was reached due to 800 ppm CaO application. When the results were analyzed, it was determined that calcium application affected only plant width. It was statistically calculated that foliar calcium applications did not affect plant height, circumference, or stem diameter in lettuce plants.

Table 2. Number of leaves, root fresh weight, root length, Leaf fresh weight of Thespian lettuce cultivar

Apps	Number of Leaves (pcs/plant)	Root Fresh Weight (g)	Root Lenght (cm)	Leaf Fresh Weight (g/plant)
1	35,3	17,7	36,5 a	157,0 a
2	37,4	18,3	31,8 ab	133,1 ab
3	36,7	17,6	33,8 ab	148,5 ab
4	35,3	15	28,4 b	124,8 b
	N.S	N.S.	6,9	24,644

According to the evaluations on root length and leaf fresh weight parameters (Table 2) In root length measurements, the highest value of 36.5 cm was obtained in control plants. On the other hand, the lowest root length value was observed in 1000 ppm CaO treatment with 28.4 cm. In the leaf fresh weight evaluation, the maximum value was reached with 157.0 g obtained as a result of 1000 ppm CaO application, while this value was recorded at the lowest level with 124.8 g in Control plants.These results reveal the potential effects of different CaO concentrations on the Thespian lettuce cultivar's root length and fresh leaf weight. Foliar application of calcium increased plant fresh weight.

Table 3. Macro element values of Thespian lettuce cultivar

Apps	Cu (ppm)	Mn (ppm)	Fe (ppm)	Zn (ppm)
1	13,0 a	150,0 a	93,4	131
2	15,2 a	202,4 a	119,4	148,6
3	13,0 a	241,2 a	123,2	137,2
4	14,6 a	206,6 a	108,2	133,2
5	N.S	N.Sç.	N.S	N.S.

The effects of different concentrations of CaO applications on the mineral content of the Thespian lettuce cultivar were investigated. According to the results obtained:

Regarding potassium (K) content, the highest value was 2.8 in the control treatment. On the other hand, K content was recorded at the lowest level with 2.2 in 1000 ppm CaO application. In evaluating magnesium (Mg) content, the maximum Mg content was reached with 0.4 value obtained with 1000 ppm CaO application. On the other hand, due to 600 ppm CaO application, this value was determined at the lowest level of 0.2. No significant change was observed as a result of statistical analysis. In terms of calcium (Ca) content, the highest Ca content was reached with 0.5 value obtained as a result of 1000 ppm CaO application, while this value was determined at the lowest level as 0.2 in the control group. As expected, the 1000 ppm application obtained the highest calcium content. In the phosphorus (P) content evaluation, the maximum P content was determined at 0.48 due to the Control application. In 800 ppm CaO application, this value was recorded at the lowest level with 0.40. No significant change was observed as a result of statistical analysis.

These results indicate that different CaO concentrations caused significant changes in some macro element contents of the Thespian lettuce cultivar.

Table 4 . Micro element values of Thespian lettuce cultivar

Apps	K (%)	Mg (%)	Ca (%)	P (%)
1	2,8 a	0,3	0,2 c	0,4 a
2	2,6 ab	0,2	0,4 b	0,4 ab
3	2,4 ab	0,3	0,4 ab	0,4 b
4	2,2 b	0,4	0,5 a	0,4 ab
5	0,595	N.S.	0,163	0,047

The effects of CaO applications at different concentrations on some microelements were analyzed in the Thespian lettuce cultivar. According to the results of this analysis, the treatments did not cause a statistically significant difference in the control plants.

Suntred

Table 5 Plant height, plant width, plant circumference, and stem diameter values of Suntred red mini lettuce cultivar

Appns	Plant Height (cm)	Plant Width (cm)	Plant Surroundings (cm)	Body Diameter (mm)
1	18,9 b	30,1 b	36,1 c	12,4 b
2	19,6 b	30,8 b	37.7 bc	13.1 ab
3	20,7 b	31,8 b	40,3 b	14,2 a
4	23,8 a	35,4 a	44,2 a	14,5 a
5	1,822	3,098	3,504	1,693

The effect of CaO applications at different concentrations on the morphological characteristics of the Suntred red mini lettuce cultivar was investigated. In plant height evaluation, the highest plant height, 23.8 cm, was obtained in 800 ppm CaO application. In control plants, this value was the lowest, with 18.9 cm. The maximum value was reached in plant width measurements with 35.4 cm due to 800 ppm CaO application. All other treatments gave lower results and were in the same statistical group. In the plant circumference evaluation, the 800 ppm treatment showed the highest value with 44,2 cm.
On the other hand, the lowest plant circumference value was obtained at 36,1 cm in control plants. In stem diameter measurements, the maximum value of 14.5 cm was reached with 800 ppm CaO application. The lowest value of 12.4 cm was recorded in control plants.
These findings indicate that different CaO concentrations can cause significant changes in the morphological characteristics of the Suntred red mini lettuce cultivar.

Table 6 Number of leaves, root fresh weight, root length, and leaf fresh weight values of Suntred red mini lettuce cultivar

Apps.	Number of leaves (pcs/plant)	Root Fresh Weight(g)	Root Length (cm)	Leaf FreshWeight (g/plant)
1	29,7 b	14,9 b	32,6	128,8 a
2	29,7 b	17,8 b	33,7	143,0 b
3	31,1 b	18.2 ab	32,6	148,8 b
4	38,2 a	22,3 a	30,6	202,3 a
5	5,578	4,161	S.D.	30,812

In evaluating the number of leaves, the highest number was reached with the value of 38.2 obtained from 800 ppm CaO application. This value was the lowest, with 29.7, due to control plants and 200 ppm CaO application. Regarding root fresh weight, the maximum root fresh weight was reached at 22.3 g due to 800 ppm CaO treatment. This value was recorded at the lowest level with 14,9 g in 400 ppm CaO treatment. Leaf fresh weight showed the highest value with 202,3 g in 800 ppm CaO treatment. In control plants, this value was determined at the lowest level with 128,8 g. CaO application increased the plant weight.
These data indicate that different CaO concentrations may significantly affect the physiological characteristics of the Suntred red mini lettuce cultivar.

Table 7 Macro element values of Suntred lettuce cultivar

Apps.	K(%)	Mg (%)	Ca(%)	P(%)
1	2,8 b	0,3	2,79 b	0,4
2	2,8 b	0,3	2,84 b	0,4
3	3.1 ab	0,5	3.14 ab	0,4
4	3,6 a	0,4	3,62 a	0,5
5	0,478	S.D.	0,48	S.D.

When potassium (K) content was evaluated, the highest concentration reached 3.6% due to 800 ppm CaO application. In both 200 ppm CaO application and control plants, this value was determined at the lowest level, with 2.8%. In terms of magnesium (Mg) concentration, the maximum Mg concentration was reached at 0.5% due to 400 ppm CaO application. As a result of 200 ppm Ca application and control plants, this value was recorded at the lowest level at 0.3%. In the calcium (Ca) content evaluation, the highest concentration of 3.62% was obtained due to 800 ppm CaO application. This value was determined at the lowest level in control plants, with 2.79%. As a result of phosphorus (P) analysis in the results obtained, the treatments did not statistically differ from the control plants. These findings suggest that different CaO concentrations may cause significant changes in the

mineral content of Suntred red mini lettuce cv. Suntred.

Table-8 Micro element values of Suntred lettuce cultivar

Apps.	Cu(ppm)	Mn(ppm)	Fe(ppm)	Zn(ppm)
1	10,8 b	123,0 b	49,2 b	129,2
2	11,6 b	139,0 b	82,6 a	130,2
3	12,2 b	146,4 b	69.4 ab	130,2
4	18,2 a	215,6 a	89,8 a	126,8
5	0,478	S.D.	0,48	S.D.

Regarding copper (Cu) content, the highest Cu concentration was reached at 18.2 due to 800 ppm CaO application. Control plants showed this value at the lowest level, with 10.8. In evaluating manganese (Mn) concentration, the maximum Mn concentration was recorded at 215.6 due to 800 ppm CaO application. This value was determined at the lowest level, with 123.0 in the control application. When the iron (Fe) content was analyzed, the 800 ppm CaO application had the highest Fe concentration, with a value of 89.8 ppm.

In comparison, the control application had the lowest concentration, with a value of 49.2 ppm. Regarding zinc (Zn) concentration, both 200 ppm CaO application and 400 ppm CaO application resulted in the highest Zn concentration with a value of 130.2. 800 ppm CaO application showed this value at the lowest level with 126,8. However, no statistically significant difference existed between the treatments and control plants.
These results indicate that different CaO concentrations may have specific effects on the microelement content of lettuce cultivars.

Corriveau et al. 2012, in the study of testing irrigation, day/night foliar spraying, foliar calcium, and growth inhibitor as cultural practices to reduce tip blight in lettuce, a control group with no foliar application was established as a control group, a water spraying treatment and foliar calcium application were applied to the leaves as a control. 360mg/L calcium chloride was applied twice a week. Calcium application decreased the number of leaves leaf area with tip burn and increased the calcium content. Biomass, dry weight, leaf area, and leaf number did not change (25). When these results are compared with our study, some are compatible, but some are incompatible. Biomass weight increased as the amount of foliar sprayed calcium increased in our study.

In lettuce, Samarakoon et al. 2020, CaCl2 was sprayed foliar against high temperatures and tip blight problem in summer. Leaves were sprayed with 0, 200, 400 and 800 mg/L calcium. Foliar spray application at 400 or 800 mg-L -1 Ca twice a week provided better control of tip blight, as symptoms of tip blight were minimal, and the effect on yield was minimal compared to reducing E.C. Fresh weight decreased with increasing doses of CaCl2. However, it decreased tip blight (26). Jun Gu Lee et al. 2013, investigated the effects of air temperature and airflow rate control on the occurrence of tip blight in leaf lettuce in a closed-type factory system and experimented with daytime temperatures at 18, 22, and 25^0 C and horizontal airflow rates of 0.28, 0.55, 1.04 ms^{-1}. It was found that temperature did not affect lettuce, whereas constant horizontal airflow above 0.28 ms^{-1} effectively reduced the tip blight symptom. This study demonstrated that applying constant horizontal airflow across the growing beds was more effective than air temperature control in reducing tip blight symptoms in a closed plant factory system (27).

Kubota et al. 2022, which investigated technological methods to eliminate tip blight for lettuce under vertical farming conditions, suggested effective mitigation measures: general reduction of growth due to low light and temperature, shortening the production cycle to avoid the last rapid growth stage to prevent the risk of tip blight, promoting xylem mass flow by using downward airflow fans, and spraying Ca spray on lettuce and leaves to increase transpiration rate (28).

Knopp conducted nutrient analyses (Ca, K, and Mg) in experiments to determine the relationship between calcium and tip blight in his study in 2019 to determine the climatic factors that trigger tip blight in lettuce and cultivation methods that reduce tip blight in a controlled environment, and found lower calcium levels in young leaves with tip blight than in young leaves without tip blight, confirming the role of Ca in tip blight formation (29).

4. CONCLUSION

In plant factories and climate chambers where air circulation was insufficient, Cos-type lettuce did not have sufficient air passage due to overlapping of leaves, and calcium applied to the plants gave negative results for tipburn in green lettuce of Thespian variety, while Suntred-type red lettuce showed promising results compared to the control.

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