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Black Cumin Essential Oil Vapor Delays Postharvest Chlorophyll Loss in Garden Cress (*Lepidium sativum* L.)

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ABSTRACT

Purpose: *In this study, the effect of different concentrations of black cumin oil vapor on the quality of garden cress leaves during postharvest storage was investigated.*

Research Method: *Three different doses of black cumin oil vapor including 11.4 ppm (CO1); 22.7 ppm (CO2) and 45.5 ppm (CO3) were used to treat garden cress leaf. The samples only water vapor-treated were taken as control (C). After the treatments, the samples were stored in a cold room set at 2±1°C temperature and 85-90% RH for 15 days. Color values, chlorophyll content, chlorophyll SPAD, electrolyte leakage, and weight loss were determined at three days intervals during storage.*

Findings: *The results of the study have shown that the black cumin oil vapor treatment maintained the green color and increased chlorophyll content of the garden cress leaves. According to these results, it was also found that 22.7 ppm and 45.5 ppm concentrations were more effective than an 11.4 ppm dose.*

Originality/Value: *According to the results, the black cumin oil treatments, especially high doses, have been successful in preventing chlorophyll losses. It is concluded that spraying the black cumin vapor to garden cress before marketing will extend the storage life of garden cress leaves.*

Keywords: *black cumin oil vapor, chlorophyll loss, garden cress leaves, , quality, storage life*

INTRODUCTION

Lepidium sativum L. commonly known as garden cress, the plant belongs to the family Brassicaceae, which contains rich phytochemical component (Zhan et al. 2009), and it is an erect, glabrous annual, 15- 45 cm in height (Mali *et al.,* 2007) cultivated as a salad plant in Turkey. It is known that vegetables belong to the *Brassicaceae* family, are to be highly beneficial to human health. These properties are due to their glucosinolates content as well as rich nutritional quality (Sarıkamış, 2011).

The garden cress plant is rich in glucosinolates. Seeds of the plant contain glucotropaeolin and 2-phenylethyl glucosinolate, which are having anticarcinogenic properties. Also, there are 2-ethyl butyl glucosinolate, methyl glucosinolate, butyl glucosinolate, or glucotropaeolin in leaves (Radwan *et al.,* 2007). Garden cress is also a rich source of polyphenols that are commonly used for medicinal and pharmaceutical purposes (Ullah *et al.,* 2019).

Garden cress is a leafy vegetable that has high water content. Therefore, shriveling, is the most important quality loss during the postharvest period, and is occured due to water-loss. Another important quality loss of garden cress leaves is the yellowing resulting from senescence. The postharvest water loss can be delayed by packaging or coating applications, whereas the yellowing caused by chlorophyll degradation cannot be prevented since it is a natural senescence process (Ferrante *et al.,* 2004; Sağlam, 2015). Chlorophyll does not turn into another color pigment during the yellowing process; on the

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contrary, when chlorophyll breaks down, the yellow pigments already present in the plant become visible (Gross, 1991; Ergün, 2006).

The study of Işık et al. (2009) reported that pre-cooling applications are effective in preventing the loss of green color, air pre-cooling on fresh onions and vacuum cooling on garden cress are effective in delaying yellowing. However, these methods, which are effective for a certain period, need to be developed.

The use of natural compounds of plant origin has increased in recent years. Essential oils, in particular, are highly effective in preventing microorganisms that cause postharvest decay (Sivakumar and Bautista-Baños 2014, Calo *et al.,* 2015; Antunes and Cavaco, 2010; Kasım *et al.,* 2017). Furthermore, the physiological effects and antibacterial properties of essential oils and their potential applications in food industry have been explained in detail in 'review' studies (Burt, 2004; Bakkali *et al.,* 2008; Bajpai *et al.,* 2012).

A limited study has been found about the effect of black cumin oil on quality characteristics of fruits and vegetables during postharvest storage. In one of them, Kahramanoğlu (2018) stated that modified atmosphere packaging (MAP) alone or in combination with black seed oil application have a significant influence on the maintaining of fruit weight. The author also declared that applications of 0.5% black cumin oil especially when combined with MAP, are effective in preventing weight loss, preventing juice content, controlling gray mold development, and decelerate the occurrence of chilling injury. In another study, it was shown that 4% black seed oil decreased the weight loss, fruit decay, fruit firmness, ascorbic acid, and fruit pectin content to 4.357%, 8.333%, 9.667 kg/cm2, 5.923 mg/100mg, and 0.310%, respectively (Majeed *et al.,* 2019). Some postharvest physiological properties have been investigated in the above-mentioned studies.

In this study, the effects of black cumin essential oil vapor applied in different doses on quality and chlorophyll degradation of harvested *Lepidium sativum* L. (garden cress) leaves were investigated during storage.

MATERIALS AND METHODS

Plant Material

The Garden cress plant (*Lepidium sativum* L.) used in the experiment was grown in the research and application greenhouse of Kocaeli University Arslanbey Vocational School. The garden cress plants, which took their full size and reached 8-10 cm leaf size, were removed with their roots and brought to the laboratory. The garden cress leaves were seperated and washed with tap water and the excess water was removed in the centrifugal salad dryer.

Black cumin essential oil vapor treatments

Essential oil vapor treatments were applied to garden cress leaves in a sealed cabinet which has a 40 x 40 x 55 cm internal dimension. The cabinet had two fans with 12 cm diameter and 1500 L/min power. Ten milliliters of diluted black cumin oil was placed in front of the fans, and then the garden cress leaves (max. 5 cm thickness) placed in crates were put into this cabinet. The fans were operated for 15 minutes after the cabinet was closed. Thus, the black cumin essential oil was evaporated and distributed homogenously in the cabinet (Figure 01). Three different doses of black cumin oil vapor were used in the experiment, and distilled water was evaluated as a control. Treatments and black cumin oil vapor concentrations were as below; C: 0 ppm, CO1: 11.4 ppm, CO2: 22.7 ppm, and CO3: 45.5 ppm.

The fatty acids composition of black cumin oil used in the research is given in Table 01.

Packaging and storage

After the black cumin oil vapor treatments, 150 g of garden cress leaves were placed in each polystyrene foam dishes and wrapped by stretch film, and then were stored in a cold room set at 2±1°C temperature and 85-90% relative humidity (RH) for 15 days.

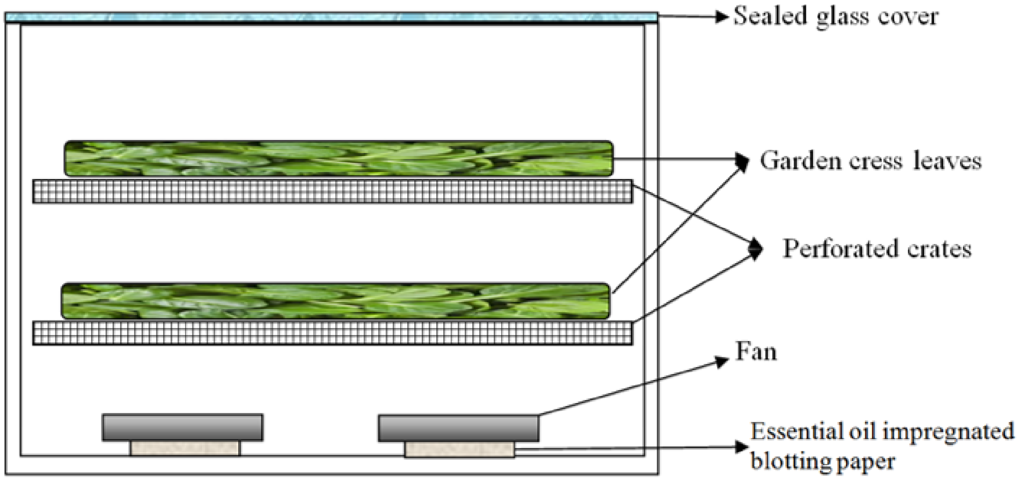

Figure 01: Black cumin oil vapor application assembly

Table 01: The fatty acids composition of black cumin seed oil.

Component	%	Component	%
Erucic acid C22:1	0,09	Palmitic acid C16:0	7,57
Linoleic acid C18:2	32,33	Stearic acid	3,02
Nervonic acid C24:1	0,17	Arachidic acid C20:0	0,32
Oleic acid C18:1	37,28	Eicosanoic (Gadoleik) acid	0,61
Palmitoleic acid C16:1	0,16	Behenic acid C22:0	0,17
Lauric acid C12:0	0,06	Lignoceric acid	0,08
Myristic acid C14:0	0,1	Linolenic acid	17,52

Color determination

Five leaves taken from each replicate were used to measure the leaf color. Color measurement of the leaves was done at three different points using Minolta CR 400 Chroma portable color meter (Minolta Co., Osaka, Japan) that used D65 illumination, and *L**, *a**, *b** color space coordinates (CIELAB) were determined. The color meter was calibrated with a standard white calibration plate at the beginning of the measurements (McGuire, 1992; Lancester *et al.,* 1997). The hue angle and yellowing index were calculated using these data, according to formulas ho= 180+ tan-1 (b^*/a^*) and YI = 142.86 b^*/L^* (Hirschler, 2012), respectively.

Chlorophyll content

The chlorophyll content of the leaves was determined by both chlorophyll-SPAD measurements and spectrophotometric methods.

Chlorophyll-SPAD

Chlorophyll-SPAD measurements were obtained with a chlorophyll meter (SPAD-502 Plus Konica Minolta) from three different points on five garden cress leaves in each replicate.

Spectphotometric measurement

Two grams of the garden cress leaves were homogenized in 30 mL of 80% acetone, then filtered and the final volume was completed to 100 mL. Readings were obtained from the filtrate at wavelengths of 645 and 663 nm and a witness of acetone (Mencarelli and Saltveit, 1988), using a spectrophotometer. From the data obtained, chlorophyll a, chlorophyll b, and total chlorophyll were calculated using the following formulas.

$$Kl_a = \frac{(12.7 \times A_{663}) - (2.6 \times A_{645})}{G} \times F$$

$$Kl_b = \frac{(22.9 \times A_{645}) - (4.68 \times A_{663})}{G} \times F$$

Kl_a: Chlorophyll a (mg/100 g), Kl_b: Chlorophyll b (mg/100 g), A_{645}: Absorbance at 645 nm,

A_{663}: Absorbance at 663 nm, F: Final volume (mL), G: Sample weight (g)

Electrolyte leakage

For this purpose, 1 cm wide strips of garden cress leaves were cut and placed in a 100 mL beaker and washed twice with 50 mL of distilled water. The electrical conductivity was measured after the washed samples were submurged in 50 mL distilled water and left to stand at room temperature for 2 hours. Then the samples were frozen and thawed, and the electrical conductivity value was measured again when the sample temperature reached 18°C. The electrolyte leakage is calculated as a percentage (%) compared to initial values (Kasım and Kasım, 2008).

Texture analysis

The cutting resistance of garden cress leaves was measured using Shimadzu EZ-LX texture analyzer and expressed in N (newton). A 70 mm cutting knife set is used in the device.

Weight Loss

The weight of the samples from triplicates was recorded on the day of harvest and after the designated sampling dates. The weight loss of garden cress leaves was calculated using the following formula:

Weight loss % = (Wi – Ws) Wi x 100.

Where Wi and Ws show the weight of samples at initial and at a sampling period, respectively (Kasım and Kasım, 2018).

Statistical Analysis:

The experiment was established and conducted according to a completely randomized design. There were three replications in every treatment, and each replicate contained 150 g garden cress leaves. The data obtained were subjected to variance of analysis with SPSS 16 package program, and the differences between the averages were compared with the Duncan's Multiple Range test within 5% error limits.

RESULTS AND DISCUSSION

Color a and b* values*

Color *a** and *b** values of samples are given in Figure 02 and 03. Accordingly, the color *a** value, which was -15.9 at the beginning of the experiment increased in all treatment groups during the first three days, and it was also detected that the difference between CO3 and the control group was statistically significant ($p<0.05$), whereas the difference among the different doses of black cumin oil vapor was insignificant. After this period, however, the *a** values of the samples treated with black cumin oil vapor increased and reached -16.0, -15.6 and -15.5 for CO1, CO2, and CO3 respectively, whereas it was decreased (-16.8) in the control group, at the 9th day of storage. The color *a** values of samples treated with black cumin oil decreased between the 9th and 15th days, while it showed an increase in C treatment during this period. The statistical analysis result of these periods also changed similarly on the 3rd day. The differences between C and all cumin oil-treated samples were found to be significant statistically at the level of $p<0.05$.

When the color *b** values obtained in the experiment were examined (Figure 02), it was seen that the *b** values of the samples in the control group were higher than those treated with black cumin oil vapor. The color *b** values also started to increase on the 3rd day in all treatment groups but after this, the *b** values of the control group continued to rise, while it decreased in samples treated with black cumin oil vapor. The *b** values of the control group decreased after the 12th day of storage, however, it started to increase in samples with black cumin vapor oil after the 9th day of storage. The differences between C and black cumin oil vapor treatments was also statistically significant on the 3rd day, but on the 9th and 12th days, it was found that the difference between *b** values of C and CO2 and CO3 was statistically significant whereas the difference between C and CO1 was insignificant ($p<0.05$).

According to CIELAB color coordinate system, -*a** values refer to green color, and if the color -*a** values increase from the center of the coordinate system to out, the green colour slightly turns to light green.

Similarly, the increase of +*b* values corresponds to yellowing (Ly *et al.,* 2020). In this research, it was found that the color *a** values of garden cress leaves decreased after the 3rd day of storage, whereas it increased in the control group. Hence, it can be said that the green color of samples in CO1, CO2, and CO3 is darker compared to the control group. The *a** values of samples treated with black cumin vapor oil were also higher than the initial value of samples from the sixth day and remained at this level during the storage, whereas in the control group, the initial color values could have only been reached at the end of the storage. Similarly, it was found that the *b** values of the control group are higher than both the initial level and in samples treated with black cumin oil, during storage. Thus it can be said that black cumin oil vapor treatment caused darkening in green color, and also the treatments of CO2 (22.7 ppm) and CO3 (45.5 ppm) are better practices in terms of maintaining the green color of samples than CO1 (11. 4 ppm).

No literature found about the effect of black cumin oil on quality parameters on any leafy vegetables, but Viacava et al. (2018) detected that microencapsulated thyme essential oil treatment did not show any significant differences in *(-a*/ b*)* values on lettuces during the entire storage period. Vieira et al. (2018) also found that rosemary, cinnamon, citronella grass, and clove essential oils did not affect on physicochemical attributes of apples. But in the present study, conversely, it was found that black cumin oil vapor caused an increase in the green color of garden cress leaves by the 9th day, and this effect was changed depending on the concentration and time. It is also thought that the reason for the difference between the studies is due to the different essential oil and plant materials used.

Hue (h°) angle color values and yellowing index (YI)

According to the hue angle (*h°*) and YI data (Figure 04 and 05), it was observed that the *h°* values of garden cress leave followed a similar trend of -*a** values, while YI values showed changes similar to *b** values. The differences in hue values of C and the other treatments were also found to be statistically significant at the sixth day and no differences were observed on the other days. Furthermore, the difference of YI values of C and the treated samples was detected statistically significant at the 9th and 12th days of storage. So, both hue angle and YI values showed that the garden cress leaves treated with black cumin oil vapor remain green during the 9 days of storage compared to the control group, and thereafter, the effect declined. Consequently, it can be stated that the black cumin oil vapor affects the color of samples, but this effect declines with extended storage.

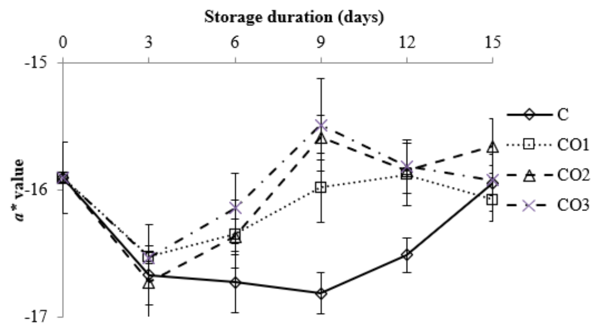

Figure 02: Changes of color *a values of garden cress leave during storage.**

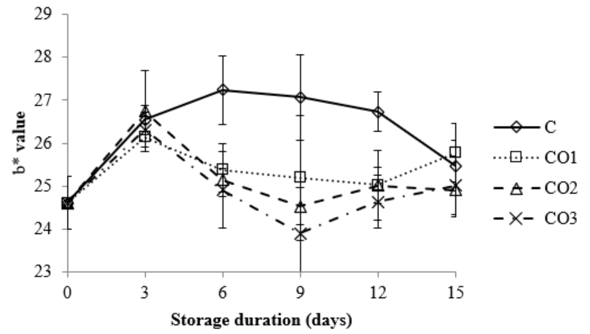

Figure 03: Changes of color *b values of garden cress leave during storage**

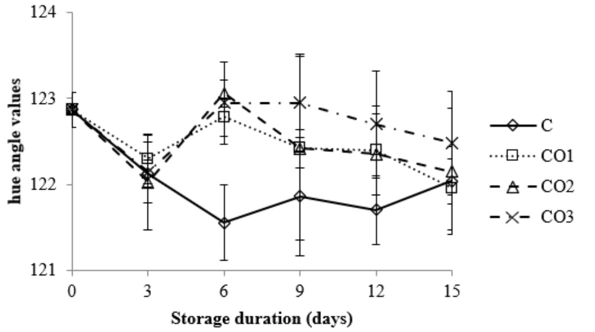

Figure 04: **Changes of color ho values of garden cress leave during storage.**

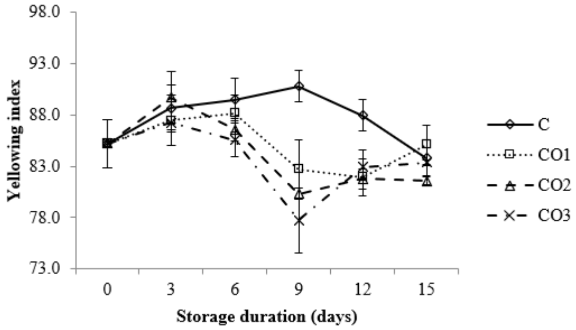

Figure 05: **Changes of color YI values of garden cress leave during storage.**

Chlorophyll SPAD

The amount of chlorophyll-SPAD of the control samples decreased until the ninth day of storage, then increased, even so, remained below the initial value. However, the chlorophyll-SPAD values of samples treated with the black cumin oil vapor were higher than the control group (Figure 06), continuously. In particular, the SPAD values of the samples in the CO3 were higher than the other treatments. However, it was found that the differences between CO3 and C treatments were statistically significant ($p<0.05$) only at the 9th day of storage but differences among black cumin oil vapor treatments were insignificant. Also, no significant differences were found among the treatments on the other days of storage.

In conclusion, chlorophyll-SPAD data showed that the black cumin oil vapor treatments, especially 45.5 ppm dose, increased chlorophyll content of garden cress leaves compared to the control group. But this increase was not found statistically significant throughout the storage period. So it could be said that the black cumin oil vapor can affect the green color of samples according to the result of color measurement, but the SPAD values have not reflected these results. However, it was seen that when the doses of black cumin vapor increased, the SPAD values of samples increased There was no literature found about the effect of black cumin oil on chlorophyll content of the fruits, vegetables, or ornamental plants. However, in one study, it was stated that, the essential oils have not maintained SPAD value, but peppermint, thyme, and black cumin treatment at the dose of 50 mg/L were useful to increase vase life of Alstroemeria (Bazaz and Tehranifar, 2011). Only a few studies were found about the effect of different essential oil on chlorophyll SPAD of different plants. Bazaz et al. (2015) found that the SPAD value of leaves of chrysanthemum cut flower was greater in 100 mg/L thyme essential oil treatments. Foliar spray of 1000 ppm essential oil also increased leaf SPAD value of tomatoes (Souri and Bakhtiarizate, 2019). According to previous studies, it was seen that the higher doses of essential oil have increased SPAD values of different plants. Therefore, it can be suggested that higher doses of black cumin oil vapour could be tested for better performance of harvested garden cress leaves, too.

Chlorophyll a, b and total chlorophyll content

The chlorophyll (chl)-a content of black cumin oil vapour treated samples were significantly higher on the sixth day (Figure 07). Also, CO3 treatment (45.5 ppm) recorded the highest value which was significant to CO1 but not to CO2. The chl-a content of samples continued to increase in C and CO2 treatments, whereas a decrease was seen in the other treatments at the 9th day of storage. Besides, the difference between C and CO2 was found statistically significant, whereas it was insignificant between C and CO1 and CO3 ($p<0.05$). After that time, chl-a content of samples in all treatments firstly decreased then increased. Chlorophyll (chl)-b values decreased at the end of the tested period but it did not have any consistent pattern throughout the storage period. The total chlorophyll content of the samples also changed similar to he chlorophyll-a values (Figure 09). According to chlorophyll-a, b, and total chlorophyll data black cumin oil vapor increased the chlorophyll amount of the garden cress leaves. Chlorophyll pigment, which is considered to be a quality indicator for green vegetables such as garden cress in the postharvest period, gradually breaks down with senescence (Limantara *et al.,* 2015). Therefore, the reduction of chlorophyll disintegration after harvest is an important issue for consumer preference for green vegetables. Gross (1991) and Ergun (2006) stated that the yellow pigment present in leafy vegetables appears only when chlorophyll is broken down. Accordinly, black cumin oil vapor treatments may have preserved the green color, and increased the shelf life of the product by preventing chlorophyll break down in garden cress leaves.

Weight loss

In the present study, it was observed that the weight loss of garden cress leaves in all treatments increased with storage duration where it was significantly higher in black cumin oil vapour treated samples (Figure 10). This increase was found to be the highest in the CO1 group (4.1 %) and followed by 3.72% CO2, 3.66% CO3, and control group (3.14%), at the end of storage. Thus, it indicates that the application of black cumin oil in the form of steam leads to a significant weight loss of samples. It is reported that the most important quality loss in cress leaves after harvest was shrinkage due to water loss, and this loss can be reduced by packaging or coating applications (Ferrante *et al.,* 2004; Sağlam, 2015). However, no literature was found on the effect of black cumin oil vapor on the weight loss of garden cress leaves. Previous studies using basil (500 ppm) and ajowan (250 ppm) essential oil revealed weight loss is increased by increasing storage period in grapevine whereas essential oil treatments declined these traits (Salimi *et al.,* 2013). Similar results were found with cumin essential oil in strawberries by Asghari Marjanlou et al. (2009). But, conversely to the previous studies, in the present study, black cumin oil vapor was found to accelerate the weight loss of packaged garden cress leaves regardless of the applied doses. This high weight loss of garden cress leaves treated with black cumin oil is thought to be due to the higher surface area of the cress compared to grapes and strawberries. On the other hand, the maximum weight loss was 4.1% among all applications, and it has been seen that this weight loss did not affect the visual quality of the product.

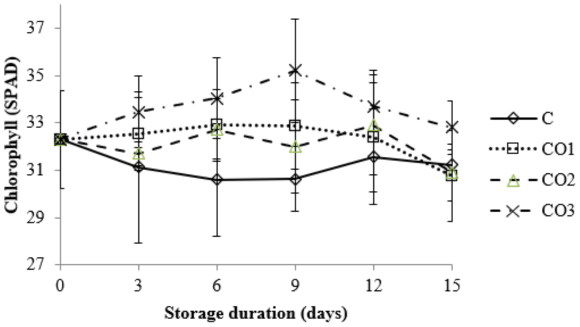

Figure 06: Changes of chlorophyll-SPAD values of garden cress leave during storage

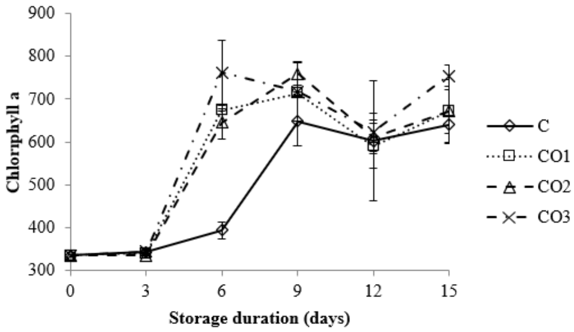

Figure 07: Changes of chlorophyll-a values of garden cress leave during storage

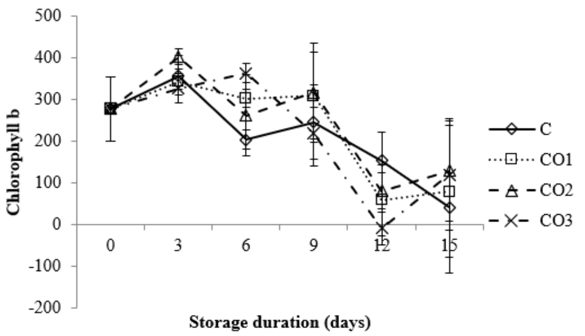

Figure 08: **Changes of chlorophyll-b values of garden cress leave during storage**

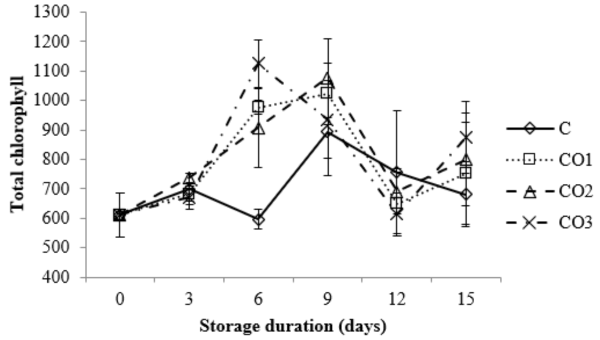

Figure 09: **Changes of total chlorophyll values of garden cress leave during storage**

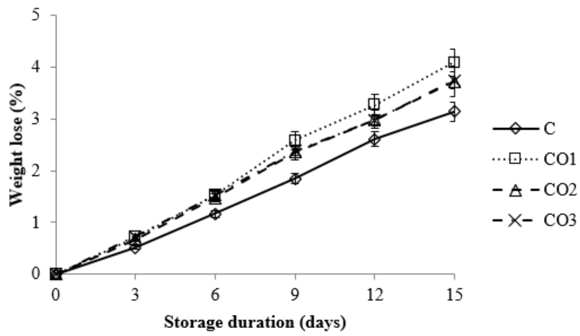

Figure 010: **Changes of weight loss of garden cress leave during storage**

Leaf-cutting resistance (N)

The resistance of the garden cress leaves against cutting in the control group decreased rapidly parallel to the increase of the storage time (Figure 11). Although there was a decline also in the black cumin oil applied groups, it was found to be slower than the control group. At the end of the storage period, the cutting resistance of the CO1 (3.7N) and CO3 (3.3N) treatments was at or near the initial value (3.7N) while the control and CO2 applications were 2.7N and 2.8N, respectively. However, the difference between the treatments found to be statistically insignificant ($p > 0.05$), except for the 12th day, in which the differences between CO3 and C and CO1 were statistically significant. According to these results, it can be said that the black cumin oil vapor had increased the strength of samples. The higher water loss of garden cress leaves may also have increased leathery nature of leaves in these groups, and accordingly, the cutting resistance may also increase.

Electrolyte leakage

Electrolyte leakage means, the content of potassium in the cell membrane to flow into the intercellular space due to cell membrane damage, cell aging, or abiotic stress (Demidchik *et al.,* 2014). Therefore, the measurement of electrolyte leakage provides information about senescence or damage to the product. In the study, at the end of the storage period, the most electrolyte leakage occurred in the control group (6.8), followed by CO2 (5.6), CO1 (4.0), and CO3 (3.5) applications (Figure 12). According to these results, it can be stated that cumin oil vapor treatments delayed senescence of cress leaves, caused less damage on leaves compared to the control group. Especially 45.5 ppm CO3 application was more effective in this sense. As a matter of fact, according to the statistical evaluation results, it was found that the difference between the CO2 and CO3 treatment was statistically significant ($p < 0.05$), whereas the difference between the other black cumin oil vapor and control applications was not significant at the 12th day.

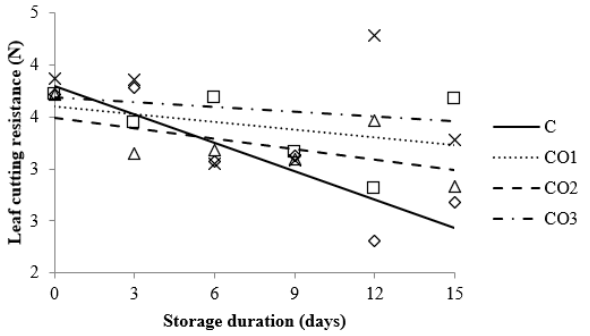

Figure 011: **Changes in cutting resistance of garden cress leave during storage**

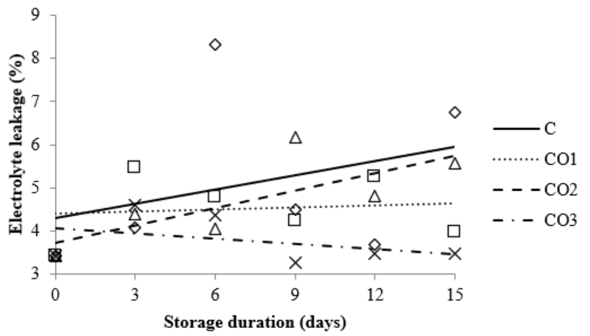

Figure 012: **Changes of electrolyte leakage of garden cress leave during storage**

CONCLUSION

In this research, it was aimed to investigate the effect of the black cumin oil vapor on some visual quality criteria of garden cress leaves during storage. According to the results, different doses of black cumin oil vapor treatments maintain the green color and chlorophyll content of garden cress leaves with the highest positive effect at 45.5 ppm black cumin oil. All treatments are also effective to maintain the cutting resistance of leaves but again 45.5 ppm is more efficient than two other essential oil vapor treatments. Although these treatments caused to increase in the weight loss of samples, they did not affect the salable quality of garden cress. Moreover, they decreased the electrolyte leakage assuring the visual quality of garden cress leaves.

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