The Effects of Some Post-Harvest Organic Acid Treatments on the Storage Quality of Brussels Sprouts

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ABSTRACT

In this study, the effects of ascorbic, folic, and salicylic acid treatments on post-harvest quality losses in ‘Franklin F1’ Brussels sprouts stored at 4°C for 28 days were investigated. Weight loss (%), color values (L*, a*, b*), total soluble solids (TSS, %), pH, titrable acidity (TA, %), total chlorophyll (mg/g), and CO₂ concentration (ppm) were all measured at 7-day intervals. When the findings were compared to the control group, it was observed that all of the treatments were effective in reducing losses in the examined properties. At the end of the storage period, it was found that ascorbic acid was the most effective in terms of weight loss, pH, and TA features, salicylic acid in terms of L* value, and folic acid in terms of preventing pH, total chlorophyll, and CO₂ concentration changes. There has been no research on the effects of ascorbic, folic, and salicylic acid treatments in Brussels sprouts during the post-harvest period, and the goal of this study is to fill in the gaps in the literature and give light on future research. It is thought that determining the appropriate doses of the treatments performed in future studies, as well as examining the efficiency of the treatments in more detail, will be beneficial.

Keywords: Ascorbic Acid, Folic Acid, Salicylic Acid, Post-harvest Storage, Brussels Sprout

1. Introduction

Brussels sprouts are a Brassica species that gets its name from the Belgian city of Brussels, where it was first grown and is now widely consumed. A mutation of *Brassica oleracea* capitata L. Sabuda, a form of winter curly sprout, is assumed to be the source of Brussels sprouts (Anonymous, 2022). Brussels sprouts are high in water and include a lot of vitamins and minerals (85%). As a consequence, post-harvest respiration rates are high (40-60 mg CO₂/kg hour at 5°C), therefore it is critical to preserve the species in line with its post-harvest physiology. As with many other types of vegetables, the most common quality loss in Brussels sprouts is yellowing caused by chlorophyll breakdown. Furthermore, darkening and chilling injuries are common problems when storage conditions are not suitable (Anonymous, 2021). It has been determined that Brussels sprouts are stored at optimum 0°C for 3-5 weeks, but chilling injury occurs at -0.6°C (Cantwell and Suslow, 2002). According to Lyons et al., (1959) post-harvest quality losses in Brussels sprouts storage at 5°C were exhibited by yellowing of the leaves by the 15th day.

Many experiments have been conducted to date to minimize quality losses and extend the storage duration of Brussels sprouts post-harvest. To that end, the effects of hormone mechanisms (Thomas, 1977), different temperatures, and modified atmosphere packaging (Kosiyachinda and Ketsa, 1983), different light intensity during storage (Kasim and Kasim, 2007), edible coatings (Viña et al., 2007), the effects of LED light (Haspaure et al., 2016; Castillejo et al., 2021), and essential oil treatments (Kraśnıewska et al., 2016) were used. When looking through the relevant literature, no literature was found in which the effects of organic acid treatments in the post-harvest period on Brussels sprouts were examined.

Ascorbic acid (AA), a water-soluble vitamin, plays many important roles in plant metabolism. Studies have shown that ascorbic acid plays an important physiological role in reactive oxygen species (ROS) that occur in plants under stress conditions. It has been reported that exogenous ascorbic acid treatments in the post-harvest period are effective in preventing quality losses during storage in many vegetable species such as spinach (Bergquist et al., 2006), beans (Sikora and Świeca, 2018) and broccoli (Bilgin, 2021). Furthermore, the efficacy of ascorbic acid added to edible coatings (Sun et al., 2010; Saleem et al., 2021) or mixed with natural ingredients such as *Aloe vera* has been studied (Sogvar et al., 2016).
Salicylic acid (SA) is a natural compound in the plant metabolism, plant growth regulators group; plant growth (Tufail et al., 2013), stomatal conductivity (Hayat et al., 2010), photosynthesis mechanism (Khodary, 2004), seed germination (Babalar et al., 2007), disease resistance (Delaney et al., 1994). Dempsey et al., 1999 have been reported to be involved in the metabolism of resistance to different stressors (Hayat et al., 2008; Chavan and Sakhal, 2020). In addition, in many different studies, it has been stated that salicylic acid suppresses ethylene production and fungal rot in the post-harvest period in fruits, prevents chlorophyll deterioration, is effective in the preservation of color values in storage products, and prevents enzyme activities that cause quality losses in the post-harvest period in different species (Leslie and Romani, 1988; Romani et al., 1989; Zhang et al., 2003; Babalar et al., 2007; Sayyari et al., 2009; Wei et al., 2011).

Although the effects of folic acid (FA), a water-soluble vitamin, on human health are well known, its role in plants has only lately been discovered. Studies have reported that folic acid regulates gene expression in plants through the riboswitch mechanism, plays a role in chlorophyll biosynthesis, and plays a role in the oxidative stress tolerance mechanism (Al-Said and Kemal, 2008; Rasei et al., 2017; Xu et al., 2021). Studies are reporting that exogenous folic acid treatment in the post-harvest period prevents quality losses in broccoli (Xu et al., 2021; Bilgin, 2021).

In this study, the effects of AA, SA, and FA treatments, each of which is a natural compound and their effects on post-harvest quality losses in different species, were investigated on post-harvest quality losses of Franklin F1’ Brussels sprouts kept at 4°C for 28 days.

2. Materials and Methods

Material

The research material was Brussels sprouts of the Franklin F1’ variety grown in the Sarcakaya district of Eskişehir. Brussels sprouts were harvested in March 2021, during the last harvest period for cultivated ecology. The average head weight of preserved Brussels sprouts at harvest was 10.61 g, the TSS content was 8.15 %, the pH was 7.21, the TEA content was 0.18 %, and the total chlorophyll content was 0.58 mg/g. Homogeneously selected Brussels sprouts were treated with salicylic acid (2 mM), ascorbic acid (2 mM), and folic acid (5 mg L⁻¹). Tween 80 (0.01%) were added to the prepared solution for better adhesive. Brussels sprouts were immersed in the prepared solutions for 10 minutes and left to dry for one hour after immersion in the solutions. The control group was treated with pure water. After all treatments, Brussels sprouts were kept in non-color plastic containers (11x12x7cm) at 4°C for 28 days.

Method

After packaging the Brussels sprouts, the first and last weight values were measured by making weight measurements with a precision scale with a sensitivity of 0.01 on the first day of storage and in each storage period. Color values (L*, a*, b*): It was measured with the aid of a digital colorimeter (Konica Minolta, Japan). Afterward, the preserved products were ground, and the samples were stored at -18°C. The amount of TSS (%) measured with a refractometer and the pH level was measured with a pH meter (HI9321, Hanna, USA) were by using a small amount of water obtained from the ground samples. Amount of TA was determined according to Kowalczyk et al., (2019). The total chlorophyll content was determined by the colorimetric method, taking into account the fresh weights of the frozen samples (Yuan et al., 2010). Measurements were made at wavelengths of 645 and 663nm and the total amount of chlorophyll was calculated according to the formula:

\[
\text{Total chlorophyll (mg/g) = (20.2 \times OD645 nm + 8.02 \times OD663 nm \times V) \times 100 \times W.}
\]

where OD is optical density, V is a volume of the extract (mL) and W is the weight of the sample (g). The carbon dioxide concentration was measured with the help of a carbon dioxide meter (Tartes-AZ 7752) by taking samples from the airtight packages with an injector.

Statistical analysis

The study was carried out with 3 replications and 4 parallels by designing random plots according to a 4x5 factorial experimental design. In the study; the statistical model, the treatment (Control, FA, SA, and AA), and storage time (0, 7, 14, 21, and 28 days) were taken as factors, and whether there was a statistical difference between the treatment and storage time averages and the presence of interaction were investigated. In the statistical analysis of the research, after applying √(X_i+3/8) square root transformation to the weight loss feature, which does not provide homogeneity of group variances, and √(X_i+3/8) to the marketable status feature, two-way analysis of variance (Two-way ANOVA) was applied to all other features (Zar, 2014). IBM SPSS 23 (IBM Corp. Released, 2015) statistical package program was used in statistical analyzes in terms of the features discussed to determine the effect of the treatment topics on the examined features. In the research, Tukey HSD multiple comparison tests at a 5% significance level were determined as statistically significant in terms of the existence of interaction and the differences between which treatment and storage time averages.

3. Results and Discussion

Weight loss occurs as a result of the continued metabolic activity and the water loss in crops during the post-harvest period. As a result, wrinkling, shrinkage, and quality losses occur in the tissues. Table 2 shows that, the effect of the treatments on the weight loss of Brussels sprouts during the 28-day storage period. According to the findings, the difference between the treatment averages was found to be statistically significant (p<0.05). At the end of the storage period, it was determined that the lowest weight loss average occurred in
folic acid treatment (3.3%). Similar to these findings, similar results were found in studies on broccoli, and it was determined that folic acid treatment had a preventive effect on weight loss (Xu et al., 2021; Bilgin, 2021). In addition, when compared with the relevant literature, it is seen that the average head weight is higher than the studies conducted on this country's ecology (Sönmez, 2007; Yılmaz and Sarı, 2019). At the end of the 28-day storage period, it was determined that the average weight loss ranged between 3.3% (FA) and 3.8% (AA). Depending on the treatment effect, Viña et al. (2007) reported 15-25% weight loss on the 30th day of storage, while Kasım and Kasım (2007) reported 3-10% weight loss at the end of the 21-day storage period. It is thought that the difference between the literature and the findings is due to differences in the treatments, variety, and ecological differences.

Changes in the color values of the goods occur in tandem with the loss of quality in the tissues during storage. In the study, day x treatment interaction was determined in all of the color values findings. The most fundamental indicator of decay in Brussels sprouts during the post-harvest period is yellowing, which is caused by the degradation of chlorophyll. Although there is a decrease in L* value during storage, especially in fruit species, when Table 1 is examined, it is seen that there is an increase in L* value in the study, similar to the relevant literature and as in many vegetable species (Bonasia et al., 2013; Jin et al., 2015; Hasperü et al., 2016). On the other hand, there are also kinds of literature stating that the L* value decreases as the storage time increases (Viña et al., 2007; Krasniwksa et al., 2016). When Table 1 is examined, it is seen that the difference between the treatment averages is statistically significant, and all the treatments compared to the control group have a preventive effect on color values. In addition, the existence of day x treatment interaction was determined (p<0.01). At the end of the 28-day storage period, it was determined that the SA treatment was the most effective to prevent the change in L* value. When the changes in a* and b* values in the storage period are examined in Table 1, it is seen that both values increase as a result of chlorophyll degradation as the storage time increases in both values, similar to the relevant literature. At the end of the storage period, the highest a* value was 0.35 and the b* value was 39.25 as a result of local decays in the control group. When the findings were examined, it was determined that the least changes in the a* and b* values at AA treatment, and the a* and b* values were -3.68 and 34.80, respectively. These findings are similar to the findings that ascorbic acid preserves color values in the post-harvest period in different species (Gil et al., 1998; Terdbaramee et al., 2006; Lin et al., 2007; Liu et al., 2014; Sikora and Świeca, 2018).

The changes in visual sensory scores are shown in Table 1. When the findings were examined, it was determined that although folic acid treatment had a preventive effect on changes in features such as weight loss and total chlorophyll amount, the highest average in visual quality scores was in SA treatment. This is because the folic acid solution has a yellow/mustard color due to the color of the folic acid, and this color affects the storage Brussels sprouts. Considering this situation, it is thought that it would be beneficial to determine the effective post-harvest folic acid treatment doses for different species. The findings are in line with studies in different species where SA has been reported to affect the preservation of visual quality in the post-harvest period (Shafiee et al., 2010; Wei et al., 2011; Kant et al., 2013; Chavan and Sakhal, 2020).

Table 1

<table>
<thead>
<tr>
<th>Physical features of ‘Franklin F’ Brussels sprouts during post-harvest storage at 4 °C.</th>
<th>Treatments</th>
<th>Storage Period (Day)</th>
<th>Mean*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Features</td>
<td>0</td>
<td>7</td>
<td>14</td>
</tr>
<tr>
<td>Weight Loss (%)</td>
<td>Control</td>
<td>0.00±0.00</td>
<td>2.29±0.52</td>
</tr>
<tr>
<td></td>
<td>FA</td>
<td>2.02±0.36</td>
<td>2.57±0.56</td>
</tr>
<tr>
<td></td>
<td>AA</td>
<td>1.49±0.34</td>
<td>2.07±0.84</td>
</tr>
<tr>
<td></td>
<td>Mean</td>
<td>0.00±0.00</td>
<td>1.78±0.39</td>
</tr>
<tr>
<td>L*</td>
<td>Control</td>
<td>44.46±0.18Ea</td>
<td>47.24±0.03Ca</td>
</tr>
<tr>
<td></td>
<td>FA</td>
<td>42.29±0.50Ec</td>
<td>46.26±0.07Db</td>
</tr>
<tr>
<td></td>
<td>AA</td>
<td>43.51±0.14Ebb</td>
<td>45.67±0.06Dc</td>
</tr>
<tr>
<td></td>
<td>Mean</td>
<td>44.55±0.30Ea</td>
<td>46.68±0.04Db</td>
</tr>
<tr>
<td>a</td>
<td>Control</td>
<td>-7.45±0.06Ea</td>
<td>-6.32±0.09Da</td>
</tr>
<tr>
<td></td>
<td>FA</td>
<td>-7.62±0.29Eab</td>
<td>-7.13±0.02Dc</td>
</tr>
<tr>
<td></td>
<td>AA</td>
<td>-7.95±0.07Ebb</td>
<td>-6.88±0.03Dc</td>
</tr>
<tr>
<td></td>
<td>Mean</td>
<td>-7.71±0.08</td>
<td>-6.74±0.08</td>
</tr>
<tr>
<td>b</td>
<td>Control</td>
<td>27.01±0.27Ec</td>
<td>30.81±0.99Da</td>
</tr>
<tr>
<td></td>
<td>FA</td>
<td>27.25±0.33Eeb</td>
<td>30.16±0.16Db</td>
</tr>
<tr>
<td></td>
<td>AA</td>
<td>28.05±0.30Eeb</td>
<td>29.89±0.06Dc</td>
</tr>
<tr>
<td></td>
<td>Mean</td>
<td>27.47±0.19Ebb</td>
<td>29.56±0.05Dd</td>
</tr>
<tr>
<td>Sensory Score</td>
<td>Control</td>
<td>5.00±0.00Aa</td>
<td>4.50±0.08Bb</td>
</tr>
<tr>
<td></td>
<td>FA</td>
<td>5.00±0.00Aa</td>
<td>4.47±0.03Bcb</td>
</tr>
<tr>
<td></td>
<td>AA</td>
<td>5.00±0.00Aa</td>
<td>4.67±0.03Bbb</td>
</tr>
<tr>
<td></td>
<td>Mean</td>
<td>5.00±0.00Aa</td>
<td>4.57±0.09Bbab</td>
</tr>
</tbody>
</table>

* The comparison of days within each treatment is shown as A, AB, B, C, D, E. The difference between treatments within each day is shown as a, ab, b, c, d, e. and the comparison of storage period means are shown as x, y, u, and w.
During the post-harvest period, the stored products lose water, become concentrated, and increase in the amount of TSS. In the findings obtained from the study, it is seen in Table 2 that there is an increase in the amount of TSS during the storage period. It was determined that all of the treatments, the effects of which were examined in the study, were effective in preventing the change in the TSS compared to the control group. One of the biochemical changes that occur in stored products is the changes in the pH level of the juice. This feature is also important in varieties where it is common to be served frozen or minimally processed, such as Brussels sprouts. According to findings, the difference between the treatment averages is statistically significant (p<0.05). When the treatment averages were examined, it is seen that there is an increase in the pH level during the storage period. According to the findings, the most important effect in preventing changes in pH level is in the AA treatment.

The changes in TA are shown in Table 2, and it is seen that the amount of TA increases during storage, similar to the relevant literature (Kowalczyk et al., 2019). When the findings are examined, it is seen that the difference between the treatment averages is statistically significant (p<0.05). When the treatment averages were examined, it was determined that the lowest TA average was 0.17% in the SA group. In addition, although the x treatment interaction is not statistically significant, it is seen that the lowest TA increase was observed in the SA-treated group at all storage periods. These findings are compatible with studies that reported the preventive effect of SA treatment on the change in the amount of TA in the post-harvest period in different species (Davarynejad et al., 2015; Bannaiem et al., 2016).

The most important symptom of quality loss in the post-harvest period in many vegetable species with green color is the loss of species-specific color as a result of chlorophyll degradation. It has been reported that chlorophyll content, which is essentially a color pigment, increases the anticarcinogenic impact of Brussels sprouts as well as its effects on aroma (Olivera et al., 2011). Therefore, the amount of chlorophyll is an important feature in terms of both visual quality and its effect on biochemical content, and it is of great importance to prevent its losses in the post-harvest period. In the study, it was observed that the total chlorophyll content decreased during storage.

According to findings, the difference between the treatment averages is statistically significant, and all the treatments compared to the control group have a preventive effect on total chlorophyll amount. In addition, the existence of day x treatment interaction was determined (p<0.01). When the findings are examined, the observation of the lowest total chlorophyll content in the control group at the end of the storage period shows the effectiveness of the treatments. At the end of the storage period, the highest total chlorophyll amount was observed in FA and SA treatments. The findings are compatible with studies reporting the preventive effect of FA (Xu et al., 2021; Bilgin, 2021) and SA treatments prevent chlorophyll degradation (Wei et al., 2011).

The respiration proceeds in the stored crops, as a result of which increases the CO₂ concentration in the storage atmosphere. For this reason, monitoring the CO₂ concentration of the storage atmosphere is of great importance to prevent quality losses. In the species such as Brussels sprouts, which are usually offered to the consumer in closed packages, the increased CO₂ concentration in the limited air mass in the package affects the storage quality negatively.

Table 2

<table>
<thead>
<tr>
<th>Features</th>
<th>Treatments</th>
<th>Storage Period (Day)</th>
<th>Mean¹</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>0</td>
<td>7</td>
</tr>
<tr>
<td>TSS (%)</td>
<td>Control</td>
<td>7.12±0.12</td>
<td>7.50±0.00</td>
</tr>
<tr>
<td></td>
<td>FA</td>
<td>7.00±0.00</td>
<td>7.25±0.14</td>
</tr>
<tr>
<td></td>
<td>SA</td>
<td>7.25±0.14</td>
<td>7.12±0.12</td>
</tr>
<tr>
<td></td>
<td>AA</td>
<td>7.00±0.20</td>
<td>7.25±0.14</td>
</tr>
<tr>
<td></td>
<td>Mean²</td>
<td>7.09±0.07Tz</td>
<td>7.28±0.06z</td>
</tr>
<tr>
<td>pH</td>
<td>Control</td>
<td>5.82±0.11Cb</td>
<td>7.00±0.01Ba</td>
</tr>
<tr>
<td></td>
<td>FA</td>
<td>5.80±0.11Db</td>
<td>6.80±0.04Cab</td>
</tr>
<tr>
<td></td>
<td>SA</td>
<td>5.90±0.12Cab</td>
<td>6.95±0.01Ba</td>
</tr>
<tr>
<td></td>
<td>AA</td>
<td>6.05±0.09Da</td>
<td>6.69±0.03Cb</td>
</tr>
<tr>
<td></td>
<td>Mean³</td>
<td>5.92±0.06</td>
<td>6.86±0.03</td>
</tr>
<tr>
<td>TA (%)</td>
<td>Control</td>
<td>0.16±0.00</td>
<td>0.17±0.00</td>
</tr>
<tr>
<td></td>
<td>FA</td>
<td>0.17±0.00</td>
<td>0.17±0.01</td>
</tr>
<tr>
<td></td>
<td>SA</td>
<td>0.16±0.00</td>
<td>0.16±0.00</td>
</tr>
<tr>
<td></td>
<td>AA</td>
<td>0.17±0.00</td>
<td>0.17±0.00</td>
</tr>
<tr>
<td></td>
<td>Mean⁴</td>
<td>0.16±0.00</td>
<td>0.17±0.00</td>
</tr>
<tr>
<td>Total Chlorophyll (mg/g)</td>
<td>Control</td>
<td>0.58±0.00Ab</td>
<td>0.57±0.00Bb</td>
</tr>
<tr>
<td></td>
<td>FA</td>
<td>0.58±0.00Ab</td>
<td>0.570±0.00Bb</td>
</tr>
<tr>
<td></td>
<td>SA</td>
<td>0.57±0.00Ab</td>
<td>0.570±0.00Bb</td>
</tr>
<tr>
<td></td>
<td>AA</td>
<td>0.58±0.01Aa</td>
<td>0.58±0.01Aa</td>
</tr>
<tr>
<td></td>
<td>Mean⁵</td>
<td>0.58±0.00</td>
<td>0.57±0.00</td>
</tr>
</tbody>
</table>

¹ The comparison of days within each treatment is given as A, AB, B, C, D, E, while the difference between treatments within each day is shown as a, ab, b, c, d. Also, the comparison of the treatment means regardless of the storage time is expressed as Y and Z, while the comparison of the storage periods regardless of the treatment is shown as x, y and z.

² The comparison of day x treatment interaction was determined (p<0.01). When the findings are examined, the observation of the lowest total chlorophyll content in the control group at the end of the storage period shows the effectiveness of the treatments. At the end of the storage period, the highest total chlorophyll amount was observed in FA and SA treatments. The findings are compatible with studies reporting the preventive effect of FA (Xu et al., 2021; Bilgin, 2021) and SA treatments prevent chlorophyll degradation (Wei et al., 2011).
The temporal variation of the CO₂ concentration detected in the storage atmosphere samples taken from the inside of the packages of storage Brussels sprouts is shown in Table 2. According to the findings, close CO₂ concentration values were obtained on the 7th, 14th, and 21st days of storage (between 684-722 ppm), but these values increased to 846 ppm on the 28th day of storage. The findings show that CO₂ concentration increases at the last period of storage (28th day). This situation can be interpreted as the respiratory rate of the storage crop increased during this period and therefore the quality losses accelerated. The significant increase in the changes in weight loss and total chlorophyll (Table 2) content on the 28th day of storage compared to the other days features the relationship between the properties.

4. Conclusion

In this study, the effects of AA, FA, and SA treatments, each of which are natural compounds, on the prevention of quality losses in the post-harvest period in Brussels sprouts were investigated. According to the findings, it was determined that quality losses occurred at a lower rate compared to the control group in all of the treatments, although it varies according to the day and features. With the study, information on the effectiveness of AA, FA, and SA treatments in Brussels sprouts, where studies on preservation are limited compared to many vegetable species, have been brought to the literature. In future studies, it is thought that it will be useful to determine the effective doses of the treatments in terms of species and to examine the effectiveness of the treatments more comprehensively.

5. Acknowledgements

Some of the findings from this study were presented orally at the 4th International Agriculture, Environment and Health Congress.

6. References


