

Effects of Ascorbic and Citric Acids on CIE Color Values of Fresh-cut Apple Cubes

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Abstract: Effects of Ascorbic Acid (AA) and Citric Acid (CA) on the CIE color values of minimally processed apple cubes were investigated. Fresh apples washed and cut into 1.5 cm cubes and treated with AA solution (0.1 and 0.5%, w/v), CA solution (0.1 and 0.5%, w/v), or their combinations (0.1% AA + 0.5% CA, 0.5% AA + 0.1% CA, 0.5% AA + 0.5% CA, w/v). Samples without any treatment were considered as control. All samples were stored in dark at 4°C and RH 90% and color were studied for 9 days. An important reduction in lightness ($p < 0.05$) was observed throughout storage in the fresh-cut apples for all dipping conditions. On the other hand, a^* -, b^* - and chroma generally increased during storage regardless of treatments. Treatment of 0.5% AA and the combination of 0.5% AA + 0.5% CA and 0.5% AA + 0.1% CA resulted in less change in those parameters, turned out to be the most effective to prevent the browning reaction.

Key words: Color, fresh-cut, apple cubes, Ascorbic Acid (AA), Citric Acid (CA), enzymatic browning

INTRODUCTION

With an increasing demand for higher quality of fresh-cut fruits and vegetables, the food industry has focused on the development of new processing technology and techniques for minimally processed fruit and vegetable products (Dawely and Flurkey, 1993; Oluo *et al.*, 2006; Brecht, 1995). But during the processing, peeling, cutting and shredding induce enzymatic browning, softening and microbial contamination. Among those, enzymatic browning has been the major cause of quality deterioration. The color of fruit products has a significant impact on consumers' choice since the color is one of the most vital quality attributes in food products.

Enzymatic browning in fruits is a complex process, catalyzed by Polyphenol Oxidase (PPO), resulting in the formation of *o*-diphenols (slightly colored) and oxidation of *o*-diphenols to *o*-diquinones followed by non-enzymatic formation of melanins (Brecht, 1995). The control of enzymatic browning is frequently achieved through the use of different type of antibrowning agents. The use of chemicals that lower the product pH finds widespread application in the control of enzymatic browning. The most commonly used acidulant is citric acid (Pizzocaro *et al.*, 1993; Santerre *et al.*, 1988; Jiang *et al.*, 1999, 2004). Acidulants are frequently used in combination with other types of antibrowning agents, because it is difficult to achieve efficient browning inhibition solely through pH control. And reducing

agents can also be applied in the prevention of discoloration. Among those, ascorbic acid is probably the most widely used antibrowning agent and in addition to its reducing properties, it also slightly lowers pH (Pizzocaro *et al.*, 1993; Santerre *et al.*, 1988). Reactants are irreversibly oxidized during the reaction, which means that the protection is temporary since they are consumed in the reaction. Also, due to the effect of pH, temperature, enzyme activity, oxygen and substrate concentrations and so on, the effect of ascorbic acid on browning inhibition is temporary (Santerre *et al.*, 1988).

Although there are many reports about ascorbic and citric acids on the quality of fresh-cut apple cubes, there is limited information available on the effect of antibrowning agents such as AA and CA on the CIE (Commission International de l'Eclairage) color parameters of fresh-cut apple cubes during storage. The main objective of the present study was to investigate the CIE color values of fresh-cut apple cubes during storage after treated with ascorbic acid, citric acid and their combinations. Inhibitory effects of various antibrowning agents were examined.

MATERIALS AND METHODS

Fuji (*Malus Domestica* B.) apples were purchased from a local supermarket in 15 kg lots and stored for less than 2 weeks at 4°C prior to experiments. L(+)-ascorbic acid and citric acid used this study were analytical grade.

Treatment and storage conditions: Apples were randomly selected for each experiment. They were initially washed with chlorinated water (125 ppm of active chlorine) for 5 min to prevent surface contamination. After peeling and coring, each apple was cut into uniform cubes of 1.5 cm and then dipped into different solutions (0.1% AA, 0.5% AA, 0.1% CA, 0.5% CA, 0.1% AA + 0.5% CA, 0.5% AA + 0.1% CA and 0.5% AA + 0.5% CA, w/v) for 2 min. Excess liquid was removed manually using a paper towel (Soliva-Forluny and Martin-Belloso, 2003). Samples without any treatment were treated as control. All samples were stored in LDPE (low density polyethylene) bags without sealing in the dark at 4°C, RH 90%, up to 9 days. CIE color characteristics of samples were measured at different times of storage (0, 3, 6 and 9 days).

Color assessment: Color characteristics (CIE $L^*a^*b^*$, C^* and h) were measured using a Minolta Chroma Meter (Model CR-200, Minolta Co., Osaka, Japan) calibrated with a calibration plate using $Y = 94.2$, $x = 0.3131$ and $y = 0.3201$. Color was measured at the same location (six sides of each cube) using 10 apple cubes for each treatment. The results reported in this paper are the mean values of samples accompanied by their standard deviations.

Statistical analysis: The statistical analysis was done using the SAS Statistical Analysis System for Windows v8.1 (SAS Institute, 1996). The means were compared with Duncan's Multiple Range test at 5% level of significance.

RESULTS AND DISCUSSION

The L^* -value is a useful indicator of darkening during storage, either resulting from oxidative browning reactions or from increasing pigment concentrations (Mastrocola and Lericci, 1991; Monsalve-Gonzalez *et al.*, 1993; Sapers and Douglas, 1987; Lozano *et al.*, 1993). The a^* -value is a measure of greenness is highly correlated with color change of apple flesh (Goupy *et al.*, 1995). A decrease in L^* -value and an increase in a^* -value are indicative of browning (Mastrocola and Lericci, 1991; Monsalve-Gonzalez *et al.*, 1993). Sapers and Douglas (1987) also suggested that enzymatic browning at the cut surfaces of apples could be monitored by measuring changes in reflectance L^* - and a^* -values, that b^* -values seemed to be unrelated to the extending of browning. Especially in apple fruit, tissue browning was mostly due to changes in L^* - and a^* -value of the pulp, which caused by the enzymatic action of polyphenol oxidases (Soliva *et al.*, 2002).

The L^*C^*h color space uses the same diagram as the $L^*a^*b^*$ color space, but uses cylindrical coordinates instead of rectangular coordinates. In this color space, L^* indicates lightness and is the same as the L^* of the $L^*a^*b^*$ color space, C^* is chroma and h is the hue angle. The value of chroma C^* (the saturation index) is 0 at the center and increases according to the distance from the center. It is proportional to its intensity, so it is the intensity or purity of the hue. The chroma can be obtained from numerical values of a^* and b^* (SAS Institute, 1996):

$$C^* = \sqrt{(a^{*2} + b^{*2})}$$

Hue angle, h , is another parameter frequently used to characterize color in food products, which is defined as starting at the $+a^*$ axis and expressed in degrees; an angle of 0° or 360° would be $+a^*$ (red) and represent red hue, while angles of 90, 180 and 270° represent yellow, green and blue hue, respectively (Maskan, 2001). The hue angle can be calculated as:

$$H = \tan^{-1}(b^*/a^*)$$

Total color difference can be expressed as a single numerical value, ΔE , which indicates the magnitude of the color difference, but not in what way the colors are different. ΔE is defined by the following equation:

$$\Delta E = \sqrt{\Delta L^{*2} + \Delta a^{*2} + \Delta b^{*2}}$$

Changes in L^* -values: Tristimulus colorimetry of cut surface was used to access the extent of browning with different antibrowning agents. Table 1-3 shows changes in color (L^*) of apple cubes treated with different antibrowning agents, after being stored for 3, 6 and 9 days at 4°C. Dipping solutions and storage time had a significant effect ($p < 0.05$) on the lightness of apples. A significant reduction ($p < 0.05$) in lightness throughout storage was observed for all dipping conditions. A similar reduction of lightness throughout storage time was also reported by Soliva-Fourtuny *et al.* (2002) in Golden Delicious fresh-cut apple and by Raybaudi-Massilia *et al.* (2007) in Fuji apples. These large reduction in color were explained by a greater decompartmentalization of oxidase and by a increased synthesis of phenolic compounds. Likewise, L^* -values were also affected by dipping conditions, where lower values were observed for fresh-cut apples dipped in 0.1% AA, 0.5% AA, 0.5% CA and all combinations in comparison to samples dipped in 0.1% CA and control.

Table 1: Color loss (%) after 3 days of storage

Treatment	CIE values					
	<i>L*</i> (%)	<i>a*</i> (%)	<i>b*</i> (%)	<i>C*</i> (%)	Hue (%)	ΔE^*
Control	4.76±1.28 ^a	14.64±2.09 ^b	12.99±5.15 ^a	11.68±4.66 ^a	2.21±1.04 ^{ab}	5.04±1.30 ^a
0.1% AA	2.20±1.19 ^{bc}	6.25±4.53 ^{bc}	12.36±5.78 ^a	11.71±5.69 ^a	2.43±1.08 ^b	3.56±1.32 ^b
0.5% AA	1.95±1.37 ^{bc}	3.39±2.41 ^e	6.79±3.43 ^b	6.62±2.85 ^{bc}	1.24±1.57 ^{bcd}	2.21±0.67 ^{cd}
0.1% CA	2.65±1.63 ^b	5.63±6.20 ^b	11.26±3.26 ^a	10.02±4.17 ^{ab}	1.10±0.63 ^{bc}	3.64±1.35 ^b
0.5% CA	2.43±1.66 ^b	6.67±5.30 ^{cd}	8.96±4.51 ^{ab}	8.60±5.06 ^{abc}	0.68±0.48 ^d	3.08±1.04 ^{bc}
0.1% AA + 0.5% CA	1.86±1.27 ^{bc}	25.62±8.04 ^a	5.64±3.10 ^b	5.99±3.29 ^c	0.97±0.60 ^d	2.51±0.68 ^{cd}
0.5% AA + 0.1% CA	0.96±0.62 ^c	11.17±4.03 ^{bc}	7.24±3.20 ^b	6.66±3.24 ^{bc}	1.60±1.15 ^{abcd}	2.04±0.69 ^{cd}
0.5% AA + 0.5% CA	1.34±0.67 ^{bc}	9.27±4.85 ^{cd}	5.62±3.43 ^b	5.43±3.43 ^c	0.87±0.58 ^d	1.92±0.76 ^d

Table 2: Color loss (%) after 6 days of storage

Treatment	CIE values					
	<i>L*</i> (%)	<i>a*</i> (%)	<i>b*</i> (%)	<i>C*</i> (%)	Hue (%)	ΔE^*
Control	6.12±1.40 ^a	44.74±7.21 ^a	27.54±7.97 ^a	25.46±7.72 ^a	7.12±0.96 ^c	8.31±1.39 ^a
0.1% AA	3.71±1.34 ^b	22.38±10.01 ^c	18.36±3.79 ^b	17.48±3.63 ^b	3.35±1.36 ^{bc}	5.43±1.37 ^b
0.5% AA	3.80±0.66 ^b	6.63±5.08 ^e	8.81±5.58 ^d	8.92±5.29 ^{cd}	1.76±1.01 ^{cd}	3.90±1.44 ^{cd}
0.1% CA	5.30±1.92 ^a	22.12±4.10 ^c	14.80±5.16 ^{bc}	13.69±5.01 ^{bc}	2.17±0.86 ^{cd}	5.66±1.73 ^b
0.5% CA	3.33±1.08 ^b	15.05±5.49 ^d	14.22±5.46 ^{bcd}	13.75±5.31 ^{bc}	3.93±0.71 ^b	4.49±2.02 ^{bcd}
0.1% AA + 0.5% CA	3.43±1.57 ^b	32.40±8.56 ^b	11.56±4.35 ^{cd}	11.59±4.28 ^c	1.36±0.97 ^d	4.29±0.96 ^{bcd}
0.5% AA + 0.1% CA	3.16±1.39 ^b	9.24±4.52 ^e	11.97±3.83 ^{cd}	9.86±4.03 ^{cd}	2.01±1.14 ^{cd}	3.89±0.89 ^{cd}
0.5% AA + 0.5% CA	2.70±1.83 ^b	7.80±7.5 ^e	6.46±5.43 ^e	5.60±5.10 ^d	2.55±1.11 ^{cd}	3.43±1.12 ^d

Table 3: Color loss (%) after 9 days of storage

Treatment	CIE values					
	<i>L*</i> (%)	<i>a*</i> (%)	<i>b*</i> (%)	<i>C*</i> (%)	Hue (%)	ΔE^*
Control	8.79±1.44 ^a	55.26±6.41 ^a	21.10±7.66 ^a	17.77±7.55 ^a	11.50±2.44 ^a	9.18±1.84 ^a
0.1% AA	4.54±1.13 ^{bc}	34.61±19.35 ^c	18.53±4.75 ^{ab}	17.16±5.00 ^b	3.42±0.68 ^{cd}	5.91±1.76 ^{bc}
0.5% AA	4.14±1.93 ^{bc}	12.84±6.93 ^d	10.47±4.41 ^{cd}	10.41±4.04 ^{bc}	4.49±1.03 ^{bc}	4.28±2.47 ^{cd}
0.1% CA	5.72±1.46 ^b	41.61±5.65 ^{bc}	14.48±7.32 ^{bc}	13.55±7.44 ^{abc}	1.55±0.86 ^d	6.13±1.67 ^b
0.5% CA	4.67±2.35 ^{bc}	38.47±9.73 ^c	13.84±4.73 ^{bcd}	12.62±4.68 ^{abc}	4.85±0.89 ^b	5.73±2.08 ^{bc}
0.1% AA + 0.5% CA	5.15±1.46 ^{bc}	43.92±9.18 ^{bc}	12.52±5.46 ^{bc}	12.61±5.41 ^{abc}	1.35±1.08 ^d	5.59±1.11 ^{bc}
0.5% AA + 0.1% CA	4.26±1.30 ^{bc}	14.71±9.43 ^d	9.17±4.27 ^d	7.64±3.11 ^c	4.33±1.05 ^{bc}	4.25±1.05 ^{cd}
0.5% AA + 0.5% CA	3.40±1.52 ^c	12.37±7.30 ^d	8.22±5.82 ^d	7.09±5.70 ^c	2.68±0.69 ^{cd}	3.75±0.87 ^d

*L**-value of apple of control decreased sharply during the first 3 days and between the 3 and 6 days, browning approached a plateau, which shows the similar trend as mentioned by Rocha and Morais (2003) and Kim *et al.* (1993). *L**-values of apple cubes treated with AA, CA and their combinations were reduced slightly after 6 days of storage and were significantly different from the control except treated with 0.1% CA only. Among all treatments, the combination treatment of 0.5% AA + 0.5% CA was the most effective on inhibiting browning of apple cubes, which reducing *L**-value from 8.79% observed for untreated cut apple to 3.40% of apple cubes at the final day of storage (Table 3). In general, AA, CA and their combination treatments showed antibrowning effect except sample treated with 0.1% CA only. The effectiveness of the antibrowning agents was more noticeable after 9 days of storage.

Changes in *a-values:** A decrease in *L**-value and an increase in *a**-value are indicatives of browning (Mastrocol and Lericci, 1991; Monsalve *et al.*, 1993; Rocha and Morais, 2003), which can be observed in Table 1-3. In

general, more concentration of CA or AA, less of color changed. However, the effectiveness of CA is quiet low when compared with that of AA. There was little changes in *a**-values of the apple cubes, only 12.37% increase, treated with combination of 0.5% AA + 0.5% CA, while there was 55.26% increase in control. 0.5% AA treatment and its combinations with CA appeared to be more effective to prevent the increase in *a**-values. González-Aguilar *et al.* (2004) reported that individual applications of the antibrowning compounds, Ascorbic Acid (AA), Acetyl Cysteine (AC) and Isoascorbic Acid (IAA) reduced color changes and browning of pineapple slices during storage at 10°C. No additional benefits were obtained when these antibrowning compounds were combined. In contrast, better results were obtained in radish slices when Hexyl Resorcinol (HR) and IAA were combined (Gonzlaze *et al.*, 2001). The use of AC combined with citric acid was found to be effective in reducing browning of banana slices stored at 5 and 15°C (Moline *et al.*, 1999). It has been reported that an additive effect occurred when HR, AC and IAA were combined, being more effective in reducing browning and decay than

when applied individually (Monsalve *et al.*, 1995; Sapers and Miller, 1998; Buta *et al.*, 1999; Gunzalez, 2000).

Changes in b^* -values: The change of b^* -value during storage was much greater for control than for those treated with the anti-browning agents (Table 1-3). The b^* -value of control increased considerably at first 6 days up to 27.54% and then decreased afterwards. With increased concentration of AA, changes in b^* -values became less. AA seems to be more effective than CA. In the combination treatment, samples treated with 0.5% (w/v) AA + 0.5% (w/v) CA had least changes in b^* -value.

Changes in C^* -values: Changes in C^* -values were similar to those in b^* -values (Table 1-3). C^* -value for all samples under investigation except 0.5% AA + 0.5% CA increased quickly at first 6 days and then decreased. There was 25.46% increase of C^* -value of control at the 6 days. Except the samples treated with 0.5% AA and combination of 0.5% AA + 0.1% (or 0.5%) CA, C^* -value of other samples seemed to increase at first and began to decrease after 6 days of storage. It appeared that the changes in C^* -values were less remarkable in the samples treated with higher concentration of AA or CA or combination of them. The sample treated with combination of 0.5% AA + 0.5% CA shows the smallest change (7.09%) in C^* -values.

Changes in h -values: Dipping conditions and storage time also significantly ($p < 0.05$) affected the hue angle (h^* values). Apples treated with antibrowning agents regardless of type and combinations maintained lower reduction in hue angle than those of control throughout the storage time. The h -values of control decreased remarkably than those of other treatments (Table 1-3), where there was 11.50% reduction in control after 9 days of storage. On the contrary, samples treated with higher (0.5%, w/v) concentration of AA or CA and combination of them decreased slowly. Interestingly, the effectiveness of AA in the inhibitory effect based on h - measurement was not clear. The smallest changes in h -values are in the sample of 0.1% AA + 0.5% CA and 0.1% CA with 1.35 and 1.55%, respectively.

Changes in total color difference: Changes in total color difference of apple cubes as influenced by each treatment during storage were shown in Fig. 1. It was found that the higher concentration of ascorbic acid or citric acid, the lower total color difference and ascorbic acid was seem to be more effective than citric acid to prevent color changes. The results showed that total color difference of apple cube treated with combination of AA (0.5%, w/v)

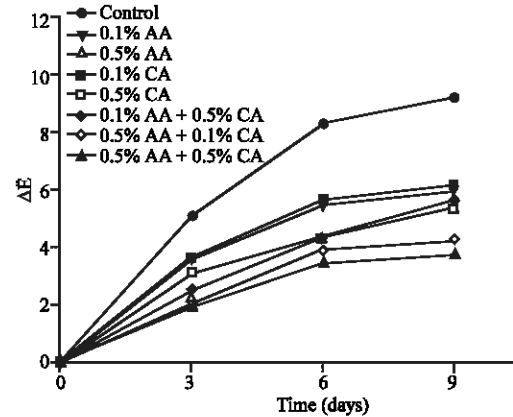


Fig. 1: Total color difference of apple cube with different treatment during storage at 4°C in dark

and CA (0.5%, w/v) was most effective to inhibit browning of fresh-cut apple cubes during storage up to 9 days.

CONCLUSION

Among all treatments, the combination treatment of 0.5% (w/v) AA and 0.5% (w/v) CA was the most effective treatment by objective measurement (highest L^* and lowest a^*). And another combination with 0.1% CA was also effective on inhibition browning of fresh cut apple. The decrease in L^* -value and hue angle and an increase in a^* -value and b^* and chroma may be due to mechanical injury during postharvest handling. This reaction results from the Polyphenol Oxidase (PPO)-catalyzed oxidation of phenolic compounds to *o*-quinones which subsequently polymerize to form dark-colored pigments (Santerre *et al.*, 1988). It was easy to compare browning of apple through compare L^* - and a^* -values in CIE $L^*a^*b^*$ color space. Measurements of L^* - (brightness) and b^* -values (browning or loss of yellow color) clearly showed varying degrees of suppression of browning on cut surfaces that resulted from the antibrowning on cut surfaces that resulted from the antibrowning compounds used. These results demonstrated that 0.5% AA solution is also adequate to prevent browning of cut apples and maintain the fresh-cut apple color for up to 9 days as much as combinations of CA solution are. Thus, the use of 0.5% AA might be a good low cost for maintaining the color quality of apples during storage.

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