



Inhibition of Browning and Decay of Fresh-cut Radishes by Natural Compounds and their Derivatives

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Treatments with inhibitors of enzymatic activity or microbial growth to retard browning and decay of fresh-cut radish slices were evaluated. Slices were dipped in solutions of natural products and their derivatives, 4-hexylresorcinol, isoascorbic acid, N-acetylcysteine and potassium sorbate alone or in combination prior to storage in air for up to 14 days at 10 °C. Inhibition of browning and deterioration without affecting texture of slices was achieved during 14 days in cold storage. From the different solutions evaluated, the combination of 4-hexylresorcinol (0.001 g/L), potassium sorbate (0.05 g/L) and N-acetylcysteine (0.025 g/L) was most effective in preventing browning and deterioration of radish slices for up to 18 days at 10 °C.

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Introduction

Radishes are an important component of many mixed salads. The marketing of fresh-cut salads is limited by a short shelf-life and rapid deterioration of their components due to tissue damage by slicing and similar methods of preparation (Watada, 1997). Slicing accelerates physiological changes such as enzymatic browning, and promotes growth of spoilage microorganisms (Avhenainen, 1996). The increase in microbial population and browning of the cut surfaces causes a deterioration of quality during storage. Observations were made earlier that radish slices started to deteriorate and anthocyanin leakage could occur after 5–7 days at 10 °C, a temperature commonly used in the produce display section of retail markets.

Different methods have been used to prevent deterioration and browning of fresh-cut produce. Traditionally, sulphites have been used to prevent browning, however, their use in processed fruits and vegetables was banned by the Food and Drug Administration in 1986 (FDA, 1986) as a result of adverse reactions developed in certain consumers.

An alternative is to inhibit browning and deterioration by using natural products derived from plants. Various endogenous organic acids and reductants such as ascorbic acid have been proposed or used as potential antibrowning and antimicrobial agents (Wiley, 1994; Avhenainen, 1996). Recently, the use of natural compounds and their derivatives such as 4-hexylresorcinol, N-acetylcysteine, ascorbic acid, iso-ascorbic acid, citric acid, potassium sorbate, calcium chloride and propionate alone or in combination at different concentrations, have been found effective in retarding browning and decay of different fruits and vegetables (Monsalve-Gonzalez *et al.*, 1995; Gunes & Lee, 1997; Kim & Klieber, 1997; Saper & Miller, 1998; Buta *et al.*, 1999; Buta & Abbott, 2000).

The rapid deterioration and browning that occurred on surfaces of radish after slicing, could possibly be diminished by the use of these natural compounds.

In this study, the use of browning and microbial inhibitors alone or in combination to inhibit enzymatic browning and deterioration of fresh-cut radishes was investigated in an attempt to find the most effective treatment.

Material and Methods

Plant material

Radishes (*Raphanus sativus* L.) obtained from a wholesale market near Jessup, MD, were used for this study.

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Table 1 Treatment with different solutions used on radish slices, prior to storage at 10 °C

Solutions	Concentration (g/L)
1. Control	Water
2. HR	0.001
3. ER	0.5
4. HR + ER	0.001 + 0.5
5. HR + ER + KS	0.001 + 0.5 + 0.05
6. ER + KS	0.5 + 0.05
7. HR + KS	0.001 + 0.05
8. HR + ER + KS + AC	0.001 + 0.5 + 0.05 + 0.025
9. ER + KS + AC	0.5 + 0.05 + 0.025
10. HR + KS + AC	0.001 + 0.05 + 0.025

Note: All solutions were adjusted to pH 5.6.

HR = 4-hexylresorcinol; ER = D-isoascorbic acid;

KS = potassium sorbate; AC = N-acetylcysteine.

Radishes were sorted for uniform appearance and absence of physical defects, cleaned and washed in a 200 ppm chlorine solution and dried. Fruits were manually sliced (4.5 mm thick) with a commercial slicing machine Model 827, Berkel Inc., ID. After slicing, anti-browning agents were applied.

Preliminary studies were performed to evaluate the efficacy of different compounds at various concentrations alone or combined. From these we selected 10 treatments which included the reducing agent, D-isoascorbic acid (ER); SH compound, N-acetylcysteine (AC); the competitive inhibitor of polyphenoloxidase (PPO), 4-hexylresorcinol (HR) and potassium sorbate (KS), as an antimicrobial compound (**Table 1**). All of the test solutions were adjusted to pH 5.6 with 11 N KOH.

Radish slices were dipped for 2 min in test solutions, drained and placed in a 15 × 100 mm plastic Petri dish. Control samples were dipped in distilled water. Petri plates were sealed with Parafilm to retard desiccation and slices were stored at 10 °C. For each treatment 20 Petri dishes (4 slices/dish) were used. After 7 and 14 days of storage, 10 Petri dishes from each treatment were sampled and colour, browning, decay symptoms, overall quality and texture were evaluated both visually and instrumentally. The experiment was repeated twice.

Colour. Tristimulus reflectance colorimetry was used to assess the extent of browning in radish slices (Saper & Douglas, 1987). Colour of slices (L^* , a^* and b^* values) was obtained from the centre of each slice using a Minolta CR-300 chroma meter. Ten replications (4 slices per replicate) were prepared for each treatment. A decrease of L^* value indicated a loss of whiteness (brightness), and a more positive a^* value indicated browning had occurred, whereas a more positive b^* indicated yellowing. Color of slices was measured initially and after 7 and 14 days at 10 °C.

Decay and browning index. After colour evaluation, slices from different treatments were evaluated subjectively

for symptoms of decay and browning, using a hedonic scale where; 0 = none, 1 = trace, 2 = slight, 3 = moderate, 4 = severe and 5 = extremely severe according to Moline *et al.* (1999).

Texture. After colour readings and subjective evaluation, texture of same slices was determined as maximum force to puncture the slice to a depth of 2.5 mm with a 4 mm diameter cylindrical probe loaded at 1 mm/s using a universal testing (force/deformation) instrument (Stable Microsystem Texture Analyzer, Surrey England, U.K.). Force deformation curves and maximum rupture strength were recorded and differences among slices from different treatments were analysed.

Statistical analysis. The analysis of variance (ANOVA) and Tukey's multiple range test for comparison of means and least significant differences ($p \leq 0.05$) were performed with the data using the SAS 6.03 system. Data from the Minolta chroma meter were transferred to a laptop computer and stored as a DOS file. The data were then imported to a Lotus 123 spreadsheet, which was then imported into a sigmaStat 5 program. One-way nonparametric Kruskal-Wallis ANOVA ($p \leq 0.05$) was run, and mean comparisons were performed.

Results and Discussion

Only a few of the combinations of antibrowning compounds tested on various fresh-cut fruits and vegetables were found to be effective in retarding browning and deterioration of fresh-cut radishes. Surprisingly, several combinations enhanced the development of browning and yellow coloration as well as increasing decay symptoms. After a series of studies we found that combinations of antibrowning agents could be used as a possible method to reduce decay and browning of radish slices stored at low temperature.

Figure 1 shows changes in colour (L^* , a^* and b^*) of radish slices treated with different antibrowning agents, after being stored for 7 and 14 days at 10 °C. The effectiveness of antibrowning agents alone or in conjunction was evaluated by tristimulus colorimetry of cut surfaces and by visual observations of radish slices. Measurements of L^* (brightness) and a^* (browning) values clearly showed varying degrees of suppression of browning on cut surfaces that resulted from the various treatments. Significantly different color changes resulted from the use of the various combinations of antibrowning agents (**Fig. 1**). In general, during the first 7 days at 10 °C, slight yellowing and darkening of surfaces were observed. However, these symptoms became more noticeable afterwards. Individual application of HR and ER reduced the loss of brightness of radish slices compared with controls (**Fig. 1a**). No additional benefits were obtained when these compounds were combined. Results obtained with the combination of HR and KS were similar to that of HR or ER when applied individually. When KS was

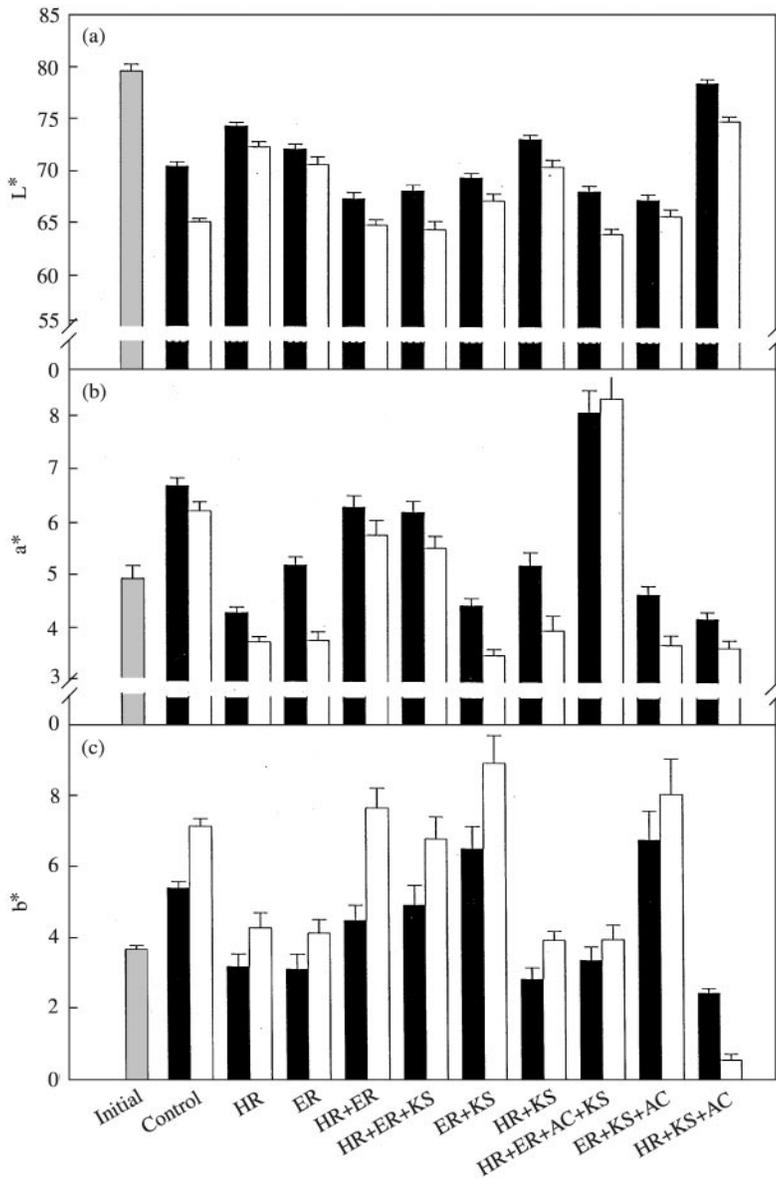


Fig. 1 Colour (L^* , a^* and b^*) changes of radish slices treated with different antibrowning solutions after storage for 7 and 14 days at 10 °C. Each value is the mean of 40 slices. Vertical bars represent standard error \pm SE. See coding explanation in **Table 1**. ■, 7 days at 10 °C; □, 14 days at 10 °C

combined with ER, a reduction in a^* values occurred (**Fig. 1b**), but this treatments also resulted in a development of yellow color in the slices (**Fig. 1c**). In the same manner, dipping radish slices in HR + ER, HR + ER + KS, and ER + KS + AC solutions caused more yellowing during storage at 10 °C. Colour changes came mostly from surface browning when examined visually. The effectiveness of HR + KS in maintaining brightness (L^*) and retarding browning (a^*), was increased with the addition of AC (**Fig. 1a** and **b**). Combinations of HR or ER with AC were not successful (data not shown).

In this study, decay and browning symptoms became more apparent after 7 days of storage at 10 °C (**Fig. 2**). By day 14, severe browning and microbial spoilage were evident on control slices and those treated with HR + ER; HR + ER + KS and HR + ER + AC + KS. Dipping slices in HR, ER, HR + KS and HR + KS + AC solutions were more effective in decreasing

decay than untreated controls and other combinations, throughout the storage at 10 °C. We found a noticeable anthocyanin leakage from the skin of the slices causing red and purple coloration of the interior of the radish slices. This phenomena enhanced the browning index as well as decay of radish slices. Treatments that enhanced anthocyanin leakage also had high a^* values (**Fig. 1b**).

The effect of HR and ER as well as ER + KS in reducing decay symptoms was similar (**Fig. 2a**). Slices treated with these solutions had only slight to moderate browning, even after 14 days at 10 °C. However, the most effective combination was HR + KS + AC, which reduced decay symptoms and browning of radish slices to a much greater extent (**Fig. 2a** and **b**). This treatment maintained a fresh appearance, so at the end of storage, these slices did not show any physiological deterioration, microbial spoilage or browning. It appeared that inhibition of microbial growth was necessary, in addition to

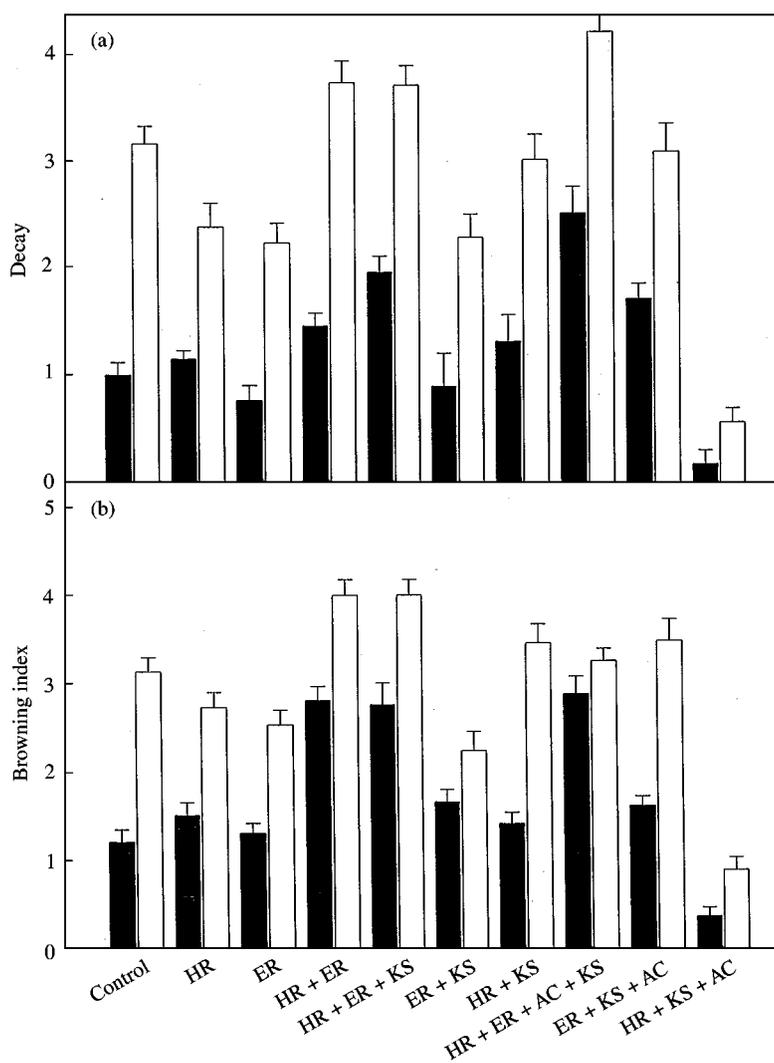


Fig. 2 Decay and browning index of radish slices treated with different antibrowning solutions after storage for 7 and 14 days at 10 °C. Each value is the mean of 40 slices. Vertical bars represent standard error \pm SE. See coding explanation in **Table 1**. ■, 7 days at 10 °C; □, 14 days at 10 °C

antibrowning treatments, to extend storage life. There was an overall decline in firmness after 14 days of storage and no differences associated with the various treatments was measured, other than maintenance of firmness with HR + ER and pronounced softening with HR treatment (**Fig. 3**). Similar results were found on pear slices using different combinations of these antibrowning agents (Buta & Abbott, 2000).

Recent studies found that the applications of HR, a competitive inhibitor of PPOs, and ER, a reducing agent that affects oxidation of phenolics, were effective in reducing browning and decay symptoms of apple and pear slices (Monsalve-Gonzalez *et al.*, 1995; Buta *et al.*, 1999). The effectiveness of these compounds increased when combined. It was surprising that combination of these chemicals did not inhibit browning in radish slices. We expected an additional benefit after combining HR and ER compounds because of the good results observed when applied individually. However, slices treated with the ER + HR treatment presented more symptoms of decay and browning than those treated individually with

ER or HR, or combined with KS. The addition of KS and AC to HR but not to ER suppressed the decay symptoms and browning of radish slices. These results were in contrast to earlier finding of the effectiveness of ER-containing combinations in preventing browning of several fruit and vegetables slices. Buta *et al.* (1999) found that the use of AC in conjunction with ER and calcium propionate was effective in reducing browning and decay of 'Red delicious' apple slices for several weeks. In another study, Saper and Miller (1998) found that HR combined with sodium isoascorbate and calcium chloride inhibited browning of pear slices. 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).

There is no literature available on radish slices treated with antibrowning compounds to prevent browning and decay during storage at low temperature. The results obtained in this study provide a new treatment for radish slices intended to be used in salad preparations. We concluded that shelf life of radish slices could be extended

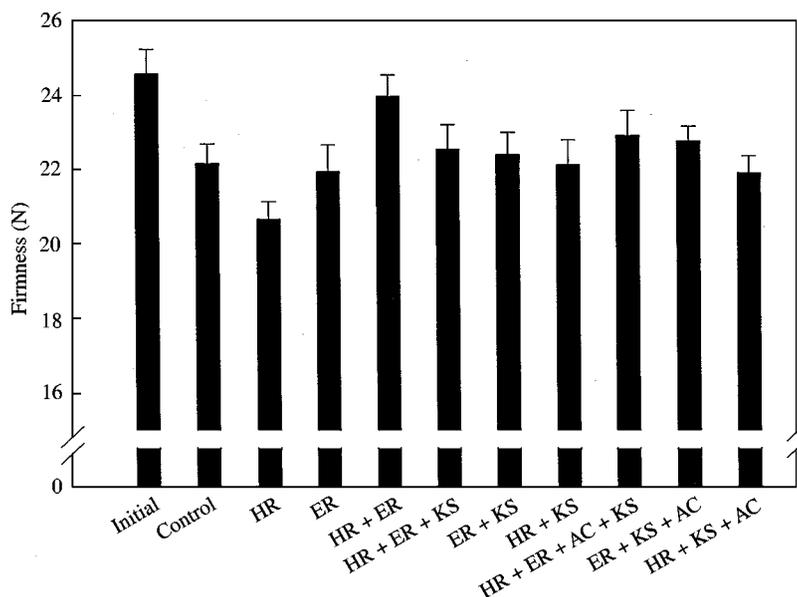


Fig. 3 Texture of radish slices treated with different antibrowning solutions after storage for 14 days at 10 °C. Each value is the mean of 40 slices. Vertical bars represent standard error \pm SE. See coding explanation in **Table 1**

from only several days to nearly 18 days under refrigerated storage by using appropriate combinations of antibrowning agents. The mixture of 4-hexylresorcinol (0.001 g/L), potassium sorbate (0.05 g/L) and N-acetylcysteine (0.025 g/L) was very effective in preventing browning and decay of radish slices for up to 14 days at 10 °C.

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