



Anti-browning methods on fresh-cut fruits and fruit juice: A Review

Afoakwah A. N.^{1*}

¹Department of Food Science and Technology, University for Development Studies, Tamale, Ghana. E-mail: nafoakwah@uds.edu.gh

Article Info

Volume 2, Issue 4, October 2020

Received : 22 October 2019

Accepted : 15 June 2020

Published : 05 October 2020

doi: [10.33472/AFJBS.2.4.2020.27-32](https://doi.org/10.33472/AFJBS.2.4.2020.27-32)

Abstract

Enzymatic browning is an economically important disorder that degrades the sensory characteristics, and prevent consumer from purchasing fresh-cut fruits and vegetables. Prevention and control of enzymatic browning in fresh-cut fruit products is imperative. This has led to extensive studies being conducted and many methodologies being explored with positive outcomes. However, concerns over off-odors, safety of food, has resulted in few browning inhibitors showing the potential for usage in the fresh-cut fruit-industry. A significant goal has been the discovery of natural compounds with health benefits, which has the potential to provide safe and effective control of enzymatic browning in fresh-cut fruits. Studies on effective combinations of diverse treatments reviewed shows that it has the potential to effectively elongate the shelf-life of fresh-cut fruits while inhibiting the activities of polyphenol oxidase (PPO), thereby maintain product quality and safety.

Keywords: *Fresh-cut fruits, Fruit-industry, Enzymatic browning, Polyphenol oxidase activity, Off-odors, Sensory characteristics*

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1. Introduction

Fruits are complex systems, which, after size reduction (pulping, milling, or cutting) are transformed into a mixture of chemical and biochemical active components reacting in aqueous media. Browning of fruits is a major problem in the fruit industry and is believed to be one of the main causes of quality loss during postharvest handling and processing. The mechanism of browning in fruits and fruit products is well-characterized and can be enzymatic or nonenzymatic in origin. Enzymatic browning is the result of fast reactions. Even an optimized processing technology cannot completely avoid the enzymatic browning during pulping and pressing of fruit juice, unless extraordinary care is taken to inhibit oxygen. During enzymatic browning reactions, polyphenol oxidase catalyzes the oxidation of phenols to o-quinones, which are extremely reactive compounds. O-Quinones produced undergo unprompted polymerization to form brown pigments (melanins). These pigments may in turn react with amino acids and proteins leading to improvement of the brown color produced.

Several studies have concentrated on either inhibiting or preventing polyphenol oxidase (PPO) activity in foods. Vámos-Vigyázó (1995) categorized the principles of enzymatic browning inhibition into: Prevention or inactivation of the enzyme, and elimination or transformation of the substrate.

Enzymatic browning is an oxidative reaction; it can be arrested by the removal of oxygen from the cut surface of fruit. However, browning can resumes quickly when oxygen is re-established. Oxygen omission is

* Corresponding author: Afoakwah A. N., Department of Food Science and Technology, University for Development Studies, Tamale, Ghana; E-mail: nafoakwah@uds.edu.gh

by immersion of fresh-cut fruit in syrup, deoxygenated water, or coating fruit with films not permeable to oxygen (McEvily *et al.*, 1992). As copper prosthetic-group of polyphenol oxidase must be present for the enzymatic browning reaction to occur, these chelating-agents capable of removing Cu may be effective to control enzymatic browning deterioration. Inactivation of the polyphenol oxidase by heat-treatments, such as steam blanching, is efficiently useful for the control of browning in fruits that are to be canned or frozen.

Chemical alteration of phenolic substrates, including chlorogenic acid, caffeic acid, and tyrosine, can stop oxidation. Certain chemical compounds react with the products of polyphenol oxidase activity and hinder the development of colored compounds. Other methods, such as the usage of naturally occurring enzyme-inhibitors and ionizing -radiation, have been utilized as substitutes to heat treatment. It must be appreciated that inactivation of enzymes accountable for browning in fruits can be permanent (e.g., heat treatment) or reversible (e.g., use of ascorbic acid).

2. Heat treatments

Though steam blanching is among most active approaches for controlling enzymatic browning in canned or frozen fruits (Vámos-Vigyázó, 1995), it is not a practical substitute for treatment of fresh foods. In such cases, the elimination of oxygen and/or the use of inhibitors ought to be applied. Moreover, blanching must not be utilized as it affects the texture and flavor of fruit-products.

A review on enzyme inactivation using thermal processing methods revealed that enzymes have complex-covalent and non-covalent structures, which are vulnerable to heat-induced degradation and disruption (Adams, 1991). In general, enzyme-inactivation as a function of temperature can be defined by Arrhenius or the activated-complex model. It is known that refrigeration (0-48 °C) impedes browning; but during fruit juice processing the cellular-tissue is destroyed, and lower temperatures are not sufficient to control oxidation. It is also true that 10 s at 90 °C incapacitates polyphenol oxidase (Dimick *et al.*, 1951), which are environments provided during heating of pulps. Yet, in practice longer delay occurs between crushing (or pulping) and heat processing.

Heat-inactivation kinetics of crude polyphenol oxidase from six apple cultivars (Golden Delicious, Starking Delicious, Granny Smith, Gloster, Starckinson, and Amasya) at three temperatures (68 °C, 73 °C, and 78 °C) was examined. The polyphenol oxidase activity firstly improved and then reduced with heat, but followed the first-order kinetic model (Yemeniciogýlu *et al.*, 1997). The researchers ascribed the increase in activity to the existence of latent polyphenol oxidase. Calculation of activation energies (54.7-77.2 kcal/mol) indicated that polyphenol oxidase in apples was generally more heat stable than polyphenol oxidase in other fruits, like banana (Galeazzi *et al.*, 1981), grape (Lee *et al.*, 1983), and pear (Halim and Montgomery, 1978).

3. Treatment using cold temperatures

For every 10 °C reduction in temperature a decrease in the rate of enzyme catalyzed-reactions occurs, this is called temperature coefficient (Q₁₀). This effect was linked to a decrease in mobility and "effective collisions" needed for the establishment of enzyme substrate- complexes and their products. Freezing temperatures of -18 °C or lower are frequently used for the longer preservation of food. Some fruits may be pre-cooled or stored at chilling- temperatures. However, others such as bananas, mangoes, and avocados are vulnerable to chill-injury and should not be kept below their respective critical temperatures (Fennema, 1975). Cold preservation and during delivery and retailing are required for the prevention of browning in fruit, because refrigerated-temperatures are efficient in lowering polyphenol oxidase activity.

4. Chemical inhibitions

Chemical anti-browning agents have been used to stop browning of fruits and fruit products. Anti-browning agents are compounds that either act primarily on the enzyme or react with the substrates and/or products of enzymatic catalysis in a manner that inhibits colored product formation. The enzyme polyphenol oxidase can be inactivated by acids, halides, phenolic acids, chelating agents, sulfites, and reducing agents such as ascorbic acid, quinone couplers such as cysteine, and some other substrate-binding compounds.

The method utilized in the fruit industry to inhibit enzymatic browning is the usage of sulfiting agents. As a reducing-agent, sulfites reduce the o-quinone produced by polyphenol oxidase catalysis to the fewer reactive diphenol, preventing the development of later condensation of complex brown melanins. Inactivation of polyphenol oxidase by application of sulfur dioxide (SO₂) has been successful in preventing enzymatic browning, but its use was controlled by regulations. Sulfites have been linked to allergic reactions, hence the

Food and Drug Administration (FDA) has banned the use of sulfite preservatives in fresh vegetables and fruits (Langdon, 1987). The effect of reducing agents is temporal since these compounds oxidize irreversibly by reacting with pigments, enzymes, and metals. They have the ability to reduce o-quinones. Sulfhydryl compounds transform o-quinones in stable, colorless products.

4.1. Ascorbic Acid (AA)

Ascorbic acid (AA) does not inactivate polyphenol oxidase directly, but acts as a reducing agent and decreases the orthoquinones to dehydroxyphenols. This action continues as long as, the concentration of AA is adequate to maintain lower concentration of quinones. As the concentration of AA decreased, the quinone concentration increases, and causes the formation of the brown pigments. Sapers and Douglas (1987) examined the efficiency of AA in cut-surfaces of apple and pear juices, finding that 40 ppm AA inhibited about 60% in raw Granny Smith juice, but only 20% in Red Delicious juice, after 90 min at 20 °C.

Sapers and Douglas (1987) assessed the efficiency of sodium bisulfite (NaHSO_3) and AA in cut surfaces and apple and pear juices. Results showed that enzymatic browning in apple juice was totally inactivated by the addition of 10 ppm SO_2 . The success of AA and erythorbic acid (EA) in inactivating enzymatic browning at cut surfaces of apple and in raw apple-juice was evaluated by tristimulus colorimetry (Sapers and Ziolkowski 1987). Besides, Lozano *et al.* (1995) determined the color changes of apple pulp treated with the various AA concentrations at 18 °C. It was evident in that study that the effect of a definite amount of AA in apple fruit pulp showed a very well-defined breaking point after which browning proceeds at the usual rate.

5. Non-chemical anti-enzymatic browning agents

5.1. Honey

The usage of honey as a natural browning inhibitor was noted in apple slices, grape juice, and model systems (Oszmianski and Lee, 1990). The browning of apple slices was subdued to a greater extent by using 10% honey, comparable sucrose solution having an equivalent sugar concentration. Analysis of honey indicated that a small peptide was responsible for the inhibition of the polyphenol oxidase. The effectiveness of honey in inhibiting polyphenol oxidase activity varied in accordance with the variety of honey (Chen *et al.*, 1998).

5.2 Aromatic carboxylic acids

Cinnamic acid such as, p-coumaric, ferulic, and sinapic acids, is potent inhibitors of apple polyphenol oxidase (Pifferi *et al.*, 1974; and Walker and Wilson, 1975). Cinnamic acid at a concentration of 0.01% was effective in inhibition of apple juice polyphenol oxidase (Walker, 1976).

5.3. Proteases

Plant proteases such as ficin, papain, and bromelain are sulfhydryl enzymes (Labuza *et al.*, 1990; and Taoukis *et al.*, 1990), are browning inhibitors. Pineapple juice was found inhibit browning in apple rings (Lozano-de-Gonzalez *et al.*, 1993). Bromelain, organic-acids, sulfhydryl compounds, and metallic components of pineapple juice are accountable for the inhibitory effect. polyphenol oxidase activity in plum juice was decreased when the juice was treated in a column containing immobilized proteases (Arnold *et al.*, 1992).

5.4. Irradiation method

Food irradiation is increasingly recognized as a method for reducing postharvest food losses and ensuring hygiene. Food irradiation is effective in securing the long-term preservation of foods through the inactivation of microorganisms. Fruits can be preserved by irradiation, thereby delaying their maturation or sprouting. Browning reduction in tropical fruits by gamma irradiation was reviewed by Thomas (1984 and 1986). Browning was minimized by controlling the dosage level of the applied radiation. However, ionizing radiation at doses exceeding 1 kGy can introduce various types of physiological disorders in food products. Free radicals produced during the treatment of food with ionizing radiation, are capable of reacting with various food constituents and inducing undesirable side effects, such as tissue darkening, lipid oxidation, and decreased vitamin content. Nonenzymatic browning (NEB) reactions of free amino acids and proteins with reducing sugars, such as glucose, may be responsible for this discoloration. The sensitivity of enzymes to ionizing radiation is defined as the dose required inactivating 63% of the original activity of the enzyme, D_{37} . Combined treatments using both irradiation and heat or other methods have demonstrated a synergistic effect.

5.5. Ultrafiltration (UF)

Ultrafiltration (UF) has shown to be effective in stabilizing the color of fruit juices (Flores *et al.*, 1988; and Sims *et al.*, 1989). Moreover, Goodwin and Morris (1991) studied UF as an alternative to sulfiting for the control of enzymatic browning. UF is believed to remove polyphenol oxidase, but not lower molecular weight polyphenols or Maillard-reaction precursors, which could undergo NEB during storage. Galeazzi *et al.* (1981) found that banana polyphenol oxidase fractions had molecular weights >30 kDa, within the range of molecular weight cut-offs for UF membranes.

5.6. High-pressure treatments

High-pressure treatments decrease microbial counts and enzyme activity, and affect product functionality (Farr, 1990; Hoover *et al.*, 1989; and Cheftel, 1991). This provides the basis for the development of new methods preservation of food (Mertens and Knorr, 1992). Effects of high-pressure-treatments on enzymes may link to reversible or irreversible changes in protein structure (Cheftel, 1992). However, loss of catalytic-activity may differ dependent on kind of enzyme, the nature of substrates, and the time and processing temperature (Cheftel, 1992; and Kunugi, 1992). Ogawa *et al.* (1990) stated the effect of high pressure-treatments on pectinesterase and peroxidase activity. Anese *et al.*, (1995) evaluated the effects of high-pressure treatments on enzymes, including PPO from puree and juice of fruits. However, inactivation of enzymes (PPO) is based on the kind of fruit and vegetable products being investigated (Knorr, 1995). Enzyme inhibition may be seen in cell-free extracts (Anese *et al.*, 1995).

6. Conclusion

The heating of the fruit mash or juice immediately after crushing the fruit appears to be the most effective way to control enzymatic browning in many fruit products. The addition of sulfur dioxide, ascorbic acid, or cysteine has been used to retard browning during the heating period. The effect of a definite amount of AA in apple fruit pulp showed a very well-defined breaking point after which browning proceeds at the usual rate. Non-traditional methods, like UF of liquid fruit products, use of supercritical carbon dioxide, or sonication, in combination with heat treatments have been used by researchers since the last decade. Although foreign additives are considered ever more undesirable in foods in general (for health reasons), the inhibition of chemicals will play primary role in the prevention of enzymatic browning, at least in the very near future. Cysteine appears to be a good alternative to the use of sulfite in foods at present.

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Cite this article as: Afoakwah A. N. (2020). Anti-browning methods on fresh-cut fruits and fruit juice: A Review. *African Journal of Biological Sciences*. 2(4), 27-32. doi: 10.33472/AFJBS.2.4.2020.27-32.