

# **Increase of the Shelf-Life of Vegetables**

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## ABSTRACT

Sugar loaf chicory, treated with a solution of ascorbic acid and sodium hypochlorite did not present browning after seven days storage at 3-4 °C. The same treatment red radish gave the same result. The treatment of endive escarole with an aqueous solution of sodium metabisulfite and sodium hypochlorite gave the same type of result. Curly endive was treated with a solution of sodium metabisulfite and sodium hypochlorite as in the case of endive escarole gave the same positive result after seven days storage.

Keywords: Vegetable, shelf life, ascorbic acid, sodium hypochlorite, sodium metabisulfite.

## **INTRODUCTION**

The most important properties of vegetables to be used of human consumption, from the point of view of a consumer, are the appearance, the taste, the consistency, and the nutritional value.

In the estimation of the appearance, color is one of the first attributes to be considered. It can result from pigments naturally present in the foods, such as chlorophylls, carotenoids, and polyphenols or it can result from other compounds obtained from enzymatic and nonenzymatic browning reactions. While the nonenzymatic browning is connected to the "technologic history" of the product, the enzymatic one is strictly connected to the nature of the vegetable and to the presence of oxidative enzymes. The reaction is catalyzed bv polyphenoloxidase (PPO), known also as phenoloxidase, phenolase, monophenoloxidase, diphenoloxidase and tyrosinase. This enzyme is present in all the vegetables but also in mushrooms, in crustaceans, and in some bacteria of genus Streptomyces (Claus & Decker, 2006). Some Authors reported that the reaction products of PPO, quinones, can react with other compounds, such as amino acids, phenols, giving carbohydrates off-flavors (Morton & Macleod, 1986). The interactions of quinones with the proteins can decrease the digestibility and the nutritional value of foods inducing changes of their organoleptic properties (Mayer & Harel, 1991). It has been estimated that 50% of the losses of vegetables is due to the enzymatic browning (Whitaker &

Lee, 1995). Therefore, the control of PPO activity was the object of a widespread activity. It is difficult to limit its activity during postharvest, but we need to do it in order to maintain the economical and nutritional value of the product (Marshall et al., 2000).

Examining in more detail polyphenoloxidase, we note the presence of two type of enzymes, odiphenoloxidase (cathecoloxidase, tyrosinase, phenolase) and p-diphenoloxidase, or laccase, observed mainly in mushrooms. Both enzymes are oxidoreductase, but they are different for some molecular properties and for the substrates they use (Mayer & Harel, 1991). PPO (Rapeanu et al., 2006) shows in its active site a  $Cu^{2+}$  ion linked to six or seven histidine residues and to a cysteine residue (Mayer, 2006). The active site of PPO can be present in three different forms: met-PPO  $(Cu^{2+})$ , deoxy-PPO  $(Cu^{+})$ , and oxy-PPO ( $Cu^{2+}$ ). The latent form of the enzyme is met-PPO and it is reduced to deoxy-PPO by oxidation of a phenol to *o*-quinone (Solomon et al., 1992). PPO is an intracellular enzyme. It was found in organelles connected to the membrane and in the soluble fraction of the cells (Mayer & Harel, 1978). It gives two main reactions: o-hydroxylation of phenol to odiphenol, and oxidation of o-diphenol to oquinones. The active site of the enzyme has to combine with oxygen to give oxy-PPO (Lerch, 1995). The reaction with phenol gives deoxy-PPO.

The catalytic activity of PPO is influenced by environmental parameters such as temperature

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and pH. Temperature is an important factor influencing PPO catalytic activity (Yoruk and Marshall, 2003). It is well known that a decrease of the temperature induces a decrease of the reaction kinetics, while high temperatures destroy the structure of the enzyme. Furthermore, temperature modifications can change the oxygen solubility. The optimal temperature for PPO changes depending on the different origin of the enzyme. Modifications of pH can influence PPO activity. In particular, both acid and basic pH values induce a modification of ionization status of the functional groups in the active site of the enzyme inducing, then, conformational modifications of the active site and an inactivation of the enzyme. The kinetics of enzymatic activity is influenced by pH (Valero & Garcia-Carmona, 1998). Optimal pH value changes in function of the origin of the enzyme and it is within 4.0-8.0 range.

Enzymatic browning does not occur on intact cells, because polyphenols are non in contact with the enzyme. Cuts of the tissues induce the reaction of the substrate with the enzyme producing the characteristic brown color.

Several methods have been reported able to avoid the browning of fruits and vegetables. We observe the presence of both physical and chemical methods: low dose  $\gamma$  irradiation (Prakash et al., 2000), ozone (Beltrán et al., 2005), thermal treatments between 50 and 70  $^{\circ}$ C (Kratky & Vadehra, 2005), peracetic acid (Alvaro et al. 2009), and sodium hypochlorite (Komatsuhara & Honma, 1999; Okishio, 1994). Ascorbic acid was used to avoid a discoloration process (Busta & Brooks, 1974), to maintain the aroma (Ogawa A., 2005), and to avoid browning of lettuce (Shalata & Abushqara, 2004; Chao, 1999). In chicory the browning process was inhibited by using sodium metabisulfite (Protin et al., 1987). Both ascorbic acid (Shalata & Abushqara, 2004) and a mixture of sodium metabisulfite, citric acid and ascorbic acid (Singh, 2002) increased the shelf life of iceberg salad.

In this communication we want to report our results on the best conditions to be used to increase the shelf life of some vegetables (until seven days). Furthermore, we will show that the expensive procedure, used in most the above reported literature, where multiple treatments with different protective agents were conducted in sequence, can be substituted with a unique treatment containing different protective agents. The most used protective agents used at this purpose are acid L-(+)-ascorbic, citric acid, malic acid, and phosphoric acid. The first compound is able to reduce *o*-quinones (Walker, 1977). However, when it was consumed, the browning process started again (Ros et al. 1993). The other agents reduces pH in order to inactivate the enzyme (Richardson & Hyslop, 1985).

The vegetables used in this work were subjected to this treatment:

- 1. Molding of the product in order to eliminate outer leaves.
- 2. Cutting.
- 3. Washing with suitable detergents and antibacterial agents.
- 4. Rinse with potable water.
- 5. Washing with reducing and/or acidulant agents.
- 6. Optional rinse with potable water.
- 7. Centrifugation.
- 8. Optional reduction of the oxygen.
- 9. Storage at 3-4 °C.

In the case of sugar loaf chicory, the product was washed with sodium hypochlorite (0.4 ml in 100 ml water) for 30 seconds at 19 °C. After this treatment the product was washed with potable water and treated with an aqueous solution of ascorbic acid and sodium The solution hypochlorite for 10 minutes. contained 0.35 g of ascorbic acid and 0.4 ml of sodium hypochlorite in 100 ml water. The treatment was performed at 21 °C. The pH of the solution was 4.93. After washing with potable water and centrifugation, the product was stored at 3-4 °C for seven days. After this period no browning evidence was present on the product.

Another vegetable we treated was the red radish. Also in this case the same procedure described above was able to gave the best results. The treatment of endive escarole with sodium hypochlorite as described above and then with an aqueous solution of sodium metabisulfite and sodium hypochlorite for ten minutes at 18 °C gave the same result after a storage at 3-4 °C under vacuum for seven days. The solution contained 0.5 g of sodium metabisulfite and 0.4 ml of sodium hypochlorite in 100 ml water. The treatment of curly endive with sodium hypochlorite as described above and then with a solution of sodium metabisulfite and sodium hypochlorite as in the case of endive escarole

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gave the same positive result after seven days storage.

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**Citation:** *Maurizio D'Auria, "Increase of the Shelf-Life of Vegetables", International Journal of Research Studies in Science, Engineering and Technology, 8(2), 2021, pp. 9-11.* 

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