

A Mini-Review: Enzymatic and Non-Enzymatic Browning

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Abstract - Food quality can be impacted by the browning, a process that gradually changes the colour of food products from white to brown or dark brown over time. There are different kinds of food can be found all around the world including shell fish which is lost because of post-harvest deteriorative reactions. Poly phenol oxidase is the potent enzyme for the colour changes occurred in shells of the fish. A lot of chemicals present today, to inhibit the colour formation like, use of controlled atmosphere packaging; high hydrostatic pressure is processing etc. This mini review enlightens the two major types of browning and its control measures in brief.

Keywords: Enzymatic browning, non-enzymatic browning, Maillard reaction, Caramelization, Ascorbic acid oxidation.

I. INTRODUCTION

Food processing and storage-related browning reactions are a common occurrence and have a significant impact on how foods look, taste, and are nutritionally. In terms of custom and consumer acceptance, browning is regarded as desirable if it improves the look and flavour of a food product, as in the cases of toasting bread and snack foods. Browning is undesirable in many other situations, such as fruits, vegetables, frozen foods, and dehydrated foods, as it produces off flavors and colours. The reduction in the food substance's nutritive value is another notable adverse effect of browning. Given that it lowers consumer acceptability and has a significant financial impact on both the primary food producers and the food processing sector, this issue is very important to the food industry. There are two main mechanisms of browning reactions:

- 1) Non-enzymatic browning (the Maillard reaction, caramelization, and ascorbic acid oxidation)
- 2) Enzymatic browning

The Maillard reaction takes place between compounds with free amino groups, such as amines, amino acids, and proteins, and carbonyl compounds, particularly reducing

sugars (Maillard et al., 1912). This reaction could result in either positive or negative consequences (Ajandouz et al., 2001).

Enzymatic browning of food due to mechanical injury during post-harvest storage or processing is caused by the catalytic action of the enzyme polyphenol oxidase (PPO) (Lee, 1999; Mayer and Harel, 1991). PPO is a very important enzyme for food chemists and processors because its action leads to major economic losses in fresh food (Whitaker, 1996). Heat treatments or the application of sulfites are commonly used methods of inactivating the enzyme.

II. ENZYMATIC BROWNING AND ITS PREVENTION

Browning of different foods as a result of mechanical or physiological damage during postharvest storage or processing is thought to be one of the main causes of quality loss. It is brought on by the oxygen-induced enzymatic oxidation of phenols to quinones by enzymes, usually PPOs (PPOs are found in fruits and vegetables as well as crustaceans like shrimp and prawns). These quinones are highly reactive substances that normally react further with other quinones, amino acids, or proteins, leading to the formation of dark-colored pigments (Richard-Forget et al., 1992; Weemaes et al., 1998; Ozoglu and Bayindieli, 2002; Selles-Marchart et al., 2006; Nunez-Delgado et al., 2007). These products are high molecular weight polymers, called melanins and melanoproteins, which are produced as a result of enzymatic browning (Wong et al., 1990). It is estimated that up to 50% losses in some tropical fruits occur as a result of enzymatic browning (Whitaker and Lee, 1995) although the overall mechanism of plant PPOs is still incompletely understood. Therefore, inhibition studies have gained more importance for these types of reactions in food and vegetable processing technology (Vamos-Vigyazo and Haard, 1981).

2.1 Control of enzymatic browning

Over the years, numerous methods and mechanisms have been created, and the majority of them aim to remove one or

more of the crucial elements—oxygen, enzyme, copper, or substrate—from the reaction.

- 1) The exclusion of oxygen, removal of copper by chelating agents are the ways to prevent browning because the browning reaction acts rapidly upon exposure to oxygen and PPO requires the copper prosthetic group to act in this browning (McEvily et al., 1992).
- 2) Chutintrasri and Noomhorm (2006) studied the thermal inactivation (between 40-90°C) of PPO for 30 min where the PPO activity reduced by 60%.
- 3) A combined effect of heat treatment (70°C for 2 min) and chemicals (ascorbic acid, citric acid, EDTA, sodium metabisulfite) were studied by Almeida and Nagueira (1995).
- 4) Lee et al. (1988) studied the effect of blanching on catalase, lipoxygenase, and PPO activities in green beans where inactivation was done completely at 82°C for 3.5 min.
- 5) Ultrafiltration, many researchers have looked into the alternative method of ultrafiltration for the management of enzymatic browning (Flores et al., 1988; Sims et al., 1989; Goodwin et al., 1991). Based on variations in the molecular weights of its components, this technique is used to separate the enzyme from liquid systems (Drake and Nelson, 1986; Hsu et al., 1987; Padilla and McClellan, 1989).
- 6) High hydrostatic pressure (HHP) processing is another method for the preservation of foods. It is a non thermal preservation technique and causes little or no change in the organoleptic and nutritional quality of product unlike most conventional heat treatments.
- 7) Depending on the amount of pressure and the length of time the product is exposed to, microorganisms and degrading enzymes can be inhibited or inactivated (at mild pressure, around 300 MPa) (Seyderhelm et al., 1996).
- 8) Ballestraet al. (2002) studied the effects of high-pressure treatments (100–500 MPa at 4°C for 10 min) on freshly sliced mushrooms and on liquid extract. They have reported that pressure above 200 MPa results in significant enzymatic browning.
- 9) Manothermosonication (MTS), a combined treatment of heat and ultrasound under moderate pressure, is another alternative to conventional heat treatment in order to inactivate enzymes and microorganisms (Lopez et al., 1994; Vercet et al., 1997; Vercet et al., 2002).
- 10) Supercritical carbon dioxide (SCCO₂) was reported to inactivate peroxidase (Christianson et al., 1984), pectinesterase (Arreola, 1990), and PPO (Zemel, 1989). Chen et al. (1992) studied the inactivation of PPO from Florida spiny lobster, broken shrimp, and potato.

There are many others prevention methods developed by many researchers which also be found effective in food industries.

III. NON-ENZYMATIC BROWNING AND ITS PREVENTION

Foods undergo non-enzymatic browning (NEB) reactions during the heating and cooling processes. Foods undergo both desirable and undesirable chemical and structural changes as a result of these reactions. The most prominently observed changes include the development of a brown colour, the production of flavours or off-flavors, nutritional loss followed by a reduction in ascorbic acid, amino acids, and invert sugars, and the formation of some toxic and mutagenic compounds like imidazols, HMF, acrylamide, advanced glycation end products (pentosidine and argpyrimidine), melanoidines, and HMF. This browning reaction includes i) Maillard reaction ii) Caramelization iii) Ascorbic acid oxidation.

3.1 Maillard Reaction

The Maillard reaction, referred to as nonenzymatic glycation or browning, was characterized in 1912 (Maillard, 1912) which involves the reaction between the carbonyl groups of reducing-sugars with the amino groups of amino acids, polypeptides, proteins, enzymes, nucleic acids, or phospholipids, forming Schiff bases, and followed by their Amadori rearrangement. The end product of the reaction is a brown pigment known as melanoidin. Some negative consequences (some desirable and undesirable effects) of the Maillard reaction have been discussed in various studies (Ferrer et al., 1999; Nakamura et al., 2006).

Desirable

- 1) Formation of antioxidants
- 2) Formation of desirable food flavours and aromas
- 3) Formation of desirable colours

Undesirable

- 1) Loss in nutritional value of protein
- 2) Formation of undesirable colors and flavors
- 3) Formation of reactive oxygen species
- 4) Formation of carcinogenic products
- 5) Formation of mutagenic products
- 6) Formation of advanced glycation end-products

The mechanism of this reaction is taught to occur at seven steps (A–G) with three main steps as initial, intermediate, and final stage (Martins *et al.*, 2001):

I. First stage (colorless, no absorption in the near-UV)

- A. Sugar-amine condensation
- B. Amadori rearrangement

II. Intermediate stage (colorless, or yellow with strong absorption in the near-UV)

- A. Sugar dehydration
- B. Sugar fragmentation
- C. Amino acid degradation

III. Final stage (highly colored)

- A. Aldol condensation
- B. Aldehyde-amine polymerization; formation of heterocyclic nitrogen compounds (melanoidines).

3.1.1 Control of Maillard Reaction

Many strategies have been put forth for regulating the Maillard reaction. Temperature, time, water activity, pH, reduction of reducing sugar and/or amino nitrogen content, high hydrostatic pressure, gas packaging and the use of chemical inhibitors like sulfites, flavonoids, and cations can all be used to control or inhibit the Maillard reaction.

1. Temperature- The rate of browning increases with increasing temperature or time.

2. Water Activity- Because the Maillard reaction requires moisture for optimal activity, it can be inhibited by lowering the moisture content using dehydrating techniques (Sherwin and Labuza, 2003).

3. pH- the Maillard reaction is generally favored at the more alkaline conditions, lowering of the pH might provide a good method of control.

4. Biochemical agents- The use of commercial glucose oxidase-catalase preparations controls the browning (Eskin, 1990).

5. Modified Atmosphere packaging- Effects of modified atmosphere packaging (MAP) on browning in glucose syrups stored at 25°C and 45°C were studied (Rais and Aroujalian, 2007).

6. High Pressure- In unbuffered and buffered media at 60°C either under atmospheric pressure or at 400 MPa, Moreno et al. (2003) studied glucose-lysine model systems over a range of pH values (5-10) in the two different pressure conditions. They claimed that the pressure slowed the Maillard reaction from its initial stages in the buffered media at pH ranges between 8.0 and less.

7. Chemical Inhibitors- Sulfites are the most widely used inhibitors. However, the restrictions to the use of sulfites in foods promoted the scientists to develop alternatives to sulfites. Use of acrylamide (Gokmen and Senyuva, 2007), aminoguanidine (Edelstein and Brownlee, 1992) could inhibit the Maillard reaction both in in vitro and in vivo. In a study published in 2005, Kwak and Lim examined the effects of phenolic acids (ferulic, hydroxybenzoic, syringic, and vanillic acids) and antibrowning agents (cysteine, glutathione, sodium sulfite, pentasodium tripolyphosphate, citric acid, and oxalic acid) on the prevention of browning in a glucose-lysine model system.

3.2 Caramelization

Caramelization is the process of degrading sugars in the absence of proteins and amino acids by heating them above their melting point, which alters their flavour and colour. This reaction could result in the production of bitter, burnt, and unpleasant products if it is not carefully controlled. Caramel has pleasant properties when the reactions are conducted under controlled circumstances. Compared to other browning reactions, caramelization begins at relatively high temperatures and is dependent on the type of sugar used.

Sucrose must be caramelised at a temperature of roughly 200°C. Mutarotation, enolization, isomerization, dehydration, fragmentation, anhydride formation, and polymerization are among the reactions that contribute to the caramelization of sucrose. The pH, temperature, and duration of the heating process all affect how much the reaction happens (Fagerston, 1969).

Bozkurt (1996) investigated how the caramelization reaction affects overall browning in glucose and fructose model systems in the appearance and absence of amino acids

Ajandouzet *al.* (2001) investigated fructose caramelization in both acidic and alkaline conditions. They discovered that at initial pH values ranging from 4.0 to 7.0, a progressive accumulation of intermediate degradation products occurred as a function of time with no lag time, whereas between pH 8.0 and pH 9.0, a high level of fructose degradation occurred during the initial stages of the heating period.

3.3 Ascorbic acid oxidation

Although ascorbic acid is an antioxidant, it contributes to food browning because it is easily oxidised and decomposed under typical storage and processing conditions. As a result, ascorbic acid is responsible for the browning of the majority of fruit juices and concentrates. It is particularly important in the citrus industry.

Oxygen, pH, ascorbic acid concentration, temperature, light, metals, citric acid, and other factors all influence ascorbic acid degradation. The reduced form of L-ascorbic acid is critical in nutrition and food processing. However, once degraded, it produces reactive carbonyl compounds that act as intermediates in nonenzymatic browning of foods. These reactive carbonyl compounds undergo additional reactions, which result in the formation of brown pigments. According to research, L-ascorbic acid decomposition results in the formation of furfural and HMF (Huelin, 1953).

The nonenzymatic browning reaction of L-ascorbic acid is thought to have two possible pathways, oxidative and nonoxidative.

A diverse range of degradation products have been reported in the literature during the nonenzymatic browning reaction of l-ascorbic acid. (Schulz et al., 2007; Huelin, 1953; Yuan and Chen, 1998).

To distinguish the oxidative pathway from the non-oxidative pathway, Schulz et al. (2007) used dehydro-Shinoda-ascorbic acid as the reaction's starting material.

IV. CONCLUSION

The development of anti-browning agents in the food industry is critical to maintaining food product quality. Traditionally, when developing anti-browning agents, effectiveness and cost-efficiency are important factors to consider. However, current anti-browning agent trends must meet consumer demands for natural sources, health benefits, and sustainability. Food ingredients such as onion, pineapple, lemon, grape, and wine, as well as various other dietary components, have been studied for their anti-browning properties. Some of them are biologically active and strongly inhibit PPOs. Furthermore, research is being conducted to determine the anti-browning activities of food byproducts and waste.

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