Enzymatic Browning of Fruit and Vegetables: A Review

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Abstract

Enzymatic browning in fruits and vegetables occurs by exposure to the air after cutting and slicing and in pulped states, mechanical damage during transportation, and thawing of frozen or cold stored foods. Polyphenol oxidase (PPO) and peroxidase (POD) are the main enzymes responsible for browning. PPO is classified as an oxidoreductase enzyme with four atoms of copper as a prosthetic group. It catalyzes the oxidation of functional OH group attached to the carbon atom of the benzene ring of monohydroxy phenols (phenol, tyrosine, *p*-cresol) to *o*-dihydroxy phenols (catechol, dopamine, adrenalin) and dehydrogenation of *o*-dihydroxy phenols to *o*-quinones. The oxidation of phenolic compounds to quinones and production of melanin give rise to a dark color in the foods. The POD is thermostable enzyme that belongs to a group of oxidases that use H_2O_2 as a catalyst for oxidation of phenolic compounds. The POD is related to undesirable changes in flavor, texture, color, and the nutritional quality of foods. The level of PPO and POD varies in fruits and vegetables and their content changes with maturity and senescence depending upon the ratio of bounded and soluble enzymes. Change in color of fruits and vegetables by enzymatic reactions is a major problem during harvesting, transportation, storage, and processing. Color deterioration, off-flavor, and loss of nutritive value in foods are unacceptable to the consumers. The purpose of this chapter is to provide information available in

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the literature on PPO and POD in different fruits and vegetables, their role in browning/color changes, and available prevention methods.

Keywords
Browning · Polyphenol oxidase · Peroxidase · Processing · Storage · Prevention

4.1 Introduction

Browning is the most significant phenomenon that takes place in food during processing and storage. Generally, browning reactions lead to unfavorable changes in the sensory properties, along with decreased market value of various foods; however, browning reactions may also be beneficial as these provide necessary color and flavor to some products such as baked and fried foods, coffee, tea, cocoa, etc. (Whitaker and Lee 1995). The enzymatic browning outcome is loss of functional, nutritional, and organoleptic abilities like softening, darkening, and off-flavor changes (Zawistowsky et al. 1991). The enzymatic browning has been considered as a significant problem leading to economic losses of fruits like apples, pears, bananas, grapes, etc. and vegetables like lettuce, potatoes, mushrooms, etc. (Whitaker and Lee 1995). The enzymatic browning is due to oxidation reaction which is another key reason for food spoilage after microbiological infection (Ioannou and Ghoul 2013). It is a prevalent color reaction occurred among food products (vegetables and fruits), because of the interaction of phenolic compounds, oxygen, and enzymes (PPOs). It has been observed that a large number of fruits and vegetables show browning after a physiological or mechanical injury during harvesting or storage. The development of brown discoloration in a wide variety of fruit and vegetables decreases the consumer tolerability and thus is of considerable economic importance to the primary producer and the food processing trade (Lattanzio et al. 1989; Mathew and Parpia 1971). Handling of fruit and vegetables with injuries in their tissues resulted in the loss of cell compartmentalization (Soliva-Fortuny et al. 2001); consequently, phenolic components get exposed; and their interaction with oxidizing enzymes (Polyphenol oxidase and peroxidase) leads to browning reactions (Degl'Innocenti et al. 2005). These browning reactions in fruits not only reduce the visual quality but also result in loss of nutrients and impairment of flavor leading to decreased consumer acceptability and significant economic losses (Luo and Barbosa-Canovas 1997; Núñez-Delicado et al. 2005). Hence, protection against oxidation in fruits and vegetables during storage or processing has become primacy matter of concern in the food business. Enzymatic browning in vegetables and fruits can be controlled by various methods such as reduction of temperature, heating, packaging, and coating, using an antioxidant, chelating and reducing agents that prevent enzymatic action, limiting the substrates, and/or bleaching the pigments. However, the effectiveness of anti-browning method relies on several factors including concentration, cultivar, and their interaction with more components like pH and application system, etc. (Ghidelli et al. 2013). The present chapter provides brief information available in the literature on causes of browning/color

changes in fruits and vegetables, the role of enzymes in browning, mechanism of browning, and available prevention methods.

4.2 Causes of Enzymatic Browning

The concentration of polyphenol oxidases (PPO) and phenolic compounds present in different fruits and vegetables along with factors like temperature, pH, and availability of oxygen to the tissue governs the rate of enzymatic browning (He and Luo 2007). However, the extent of browning reaction is influenced by the concentration of phenolic compounds and activity of enzymes (Zawistowski et al. 1991). Browning reaction is carried out by specific enzymes known as polyphenol oxidase which bring about oxidation of phenolic compounds. Nevertheless, the contribution of additional enzymes (e.g., peroxidase) to overall browning reaction might be applicable in certain fruits and vegetables (Vámos-Vigyázó and Haard 1981). The main causes of browning are as follows.

4.2.1 Phenolic Compounds

Phenolic compounds or polyphenols are chemical substances found in fruits and vegetables (Singh et al. 2016, 2017a, b). They have a major part in enzymatic browning, as they act as substrates for the enzymes responsible for browning. These compounds are secondary metabolites of plants produced through shikimate pathway by utilizing the intermediates of carbohydrate mechanism (Dixon and Paiva 1995; Singh et al. 2017b). There are a wide variety of phenolic compounds present in fruits that varies with species, the maturity of the cultivar, and environmental conditions along with other circumstances of plants (Singh et al. 2016). They are also associated with fruit quality (Es-safi et al. 2003). Their structure contains an aromatic ring having more than one hydroxyl moieties composed of numerous other substituted components (Singh et al. 2017a, b; Marshall et al. 2000). Few naturally occurring substrates for PPO are found in vegetables and fruits; e.g., in case of apples which are highly susceptible toward enzymatic browning, catechin, chlorogenic acid, epicatechin, etc. are present (Podsędek et al. 2000). Polyphenols play a crucial role in proving color to fruits (apples) and providing taste and flavor to beverages (apple juice, tea), along with being a rich source of antioxidants in plants. They are generally compound organic constituents, having additional phenol group (carbolic acid). In postharvest handling and processing of freshly cut damaged fruits and vegetables, polyphenols are the main cause of enzymatic browning. When the cellular structure of fruits or vegetables is disrupted by cutting or other methods, then these polyphenols are responsible for browning reactions (enzymatic and nonenzymatic). In the instance of enzymatic browning oxidation of polyphenolics is catalyzed by PPO enzymes that results into formation of quinones which then participates in a secondary reaction leading to the development of extremely colored secondary compounds (Loomis and Battaile 1966; Mathew and Parpia 1971; Kahn

1985). However, several biochemical reactions occur as phenolic compounds do not remain stable during processing and storage of foods.

4.2.2 Polyphenol Oxidase and Related Enzymes

Two enzymes are found to have a major part in the oxidative deprivation of polyphenols as these bring about the synthesis of brown-colored polymers known as melanins (Whitaker and Lee 1995; Espín et al. 1998). They are polyphenol oxidases (PPO) and peroxidases (POD). PPO (1,2-benzenediol: oxygen oxidoreductase; EC 1.10.3.1) is an enzyme which contains copper in its structure. Other names of this enzyme are catechol oxidase, diphenol oxidase, catecholase, phenolase, *o*-diphenolase, cresolase, tyrosinase, etc. At its active site, copper is present, which is crucial for the activity of the enzyme. PPO is found in certain microbes (bacteria and fungi) in a large number of plants, few arthropods, as well as mammals. This enzyme is linked with dark pigmentation in the organism to provide a protective role (Mayer and Harel 1979). PPO catalyzes two simple reactions: (1) phenolic substrate is hydroxylated at *o*-position, next to a prevailing hydroxyl group (monophenol oxidase activity), and (2) *o*-benzoquinones are formed by oxidation of diphenols (diphenol oxidase activity). In each of the abovementioned reactions, molecular oxygen is utilized as a co-substrate. PPOs discovered till date have the capability to transform *o*-dihydroxy phenols to *o*-benzoquinones, but every PPO does not have the capability to hydroxylate monophenols. The substrate specificity of PPO varied depending on the enzyme source from which it is extracted. The molecular weight of this enzyme ranges from 57 to 62 kDa (Hunt et al. 1993). Other enzymes with polyphenol oxidase activity are peroxidases (POD; EC 1.11.1.7) which accomplish mono-electron oxidation on a wide variety of composites in the presence of hydrogen peroxide (Dunford and Stillman 1976). Even though peroxidases are broadly dispersed in the plant kingdom, their part in the enzymatic browning of vegetables and fruits is not clear, as the interior level of hydrogen peroxide in plants restricts the activity of peroxidases. It is supposed that even though peroxidase may also lead toward enzymatic browning, their role is still unanswered (Nicolas et al. 1993) and depends on the availability of hydrogen (Amiot et al. 1992). It has been suggested that PPO could act as an agent to increase the POD activity, because of the synthesis of hydrogen peroxide at the time of oxidation of phenolic components in PPO-catalyzed reactions (Richard-Forget and Gauillard 1997; Subramanian et al. 1999).

4.2.3 Temperature

Temperature is a significant factor which influences the rate of enzymatic browning. Low temperature maintained during storage prevents browning of fruits and

vegetables. After thawing, the enzyme activity resumes. Enzymatic activity of (PPO) polyphenol oxidase stops at temperature below 7 °C although it is not deactivated. Hence refrigeration and chilling are practiced to inhibit spoilage in fruits and vegetables during circulation and retailing. Chilling is another treatment that is often used for fruits like berries, bananas, mangoes, and avocados and vegetables like broccoli, peas, spinach tomatoes, etc. The sensitivity of PPOs to thermal treatment varies depending on the source, yet the temperature of 70–95 °C has been reported to destroy the activity of PPOs (Chutintrasri and Noomhorm 2006; Ndiaye et al. 2009; Özel et al. 2010). A short exposure of 70–90 °C temperature is sufficient to obtain partial or total inactivation of the enzymes.

4.2.4 pH

Like other factors, pH also plays a significant part in the browning of fruits and vegetables. Optimal pH for the action of browning enzymes is 5.0–7.0, and it decreases below pH 3.0. The high acidic content of fruits such as oranges, lemons, and lime prevents enzymatic browning. Polyphenol oxidase enzyme activity is inhibited in the presence of acids (http://www.fao.org).

4.3 Mechanism of Enzymatic Browning

Enzymatic browning involves the action of PPO enzyme in the existence of oxygen. The substrates (mainly polyphenols) that take part in enzymatic browning are present in plastids, whereas the enzymes are located in the cytoplasm. During processing of fruits and vegetables, when the tissue gets injured, plastids are ruptured, and PPOs come in interaction with the substrates (Mayer and Harel 1979). After that, PPO catalyzes the hydroxylation of the monophenols following another step of oxidation of *o*-diphenols into *o*-quinones in the presence of molecular oxygen. The abovementioned reaction is catalyzed in the presence of PPOs which contain two copper moieties at its active site. After this, another step of nonenzymatic polymerization of the quinones takes place; as a result of which high-molecular-weight pigments, i.e., insoluble and complexed dark-colored compounds known as melanins, are formed (Fig. 4.1) (Peñalver et al. 2005; Queiroz et al. 2008). The difference in color of pigments extensively in their hue and intensity relies on polyphenols from where they initiate and environmental causes of the oxidation process during their formation (Nicolas et al. 1994). PPOs from plant sources have wide substrate specificities, and they are capable of oxidizing a range of mono-, di-, or polyphenolic components. Certain PPOs hydroxylate the monophenols to yield *o*-dihydroxy phenols, and they are further enzymatically oxidized to yield *o*-benzoquinones; they are highly unstable and reactive.

Afterward, nonenzymatic reaction with molecular oxygen gives rise to auxiliary reactions of formation of complex products such as indole 5,6-quinone from

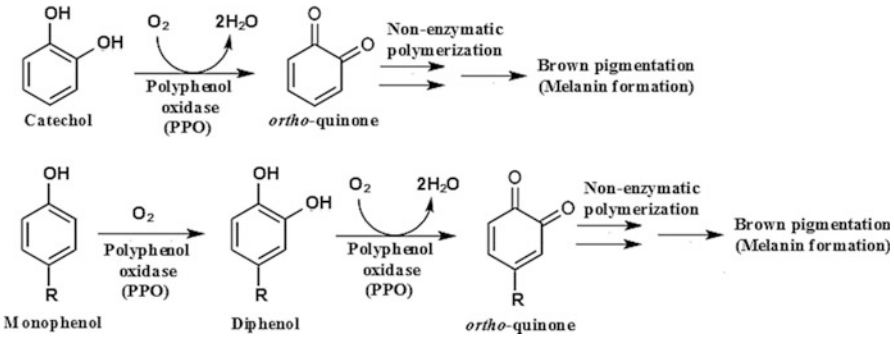

Fig. 4.1 Brown pigment (melanin) formation from phenolic compounds

tyrosine. Then *o*-benzoquinones covalently react with another polyphenol to provide intensive colored compounds ranging from red, yellow, green, blue, to black. *O*-benzoquinones upon their reaction with thiol compounds and aromatic amines together with those in proteins provide an abundant range of products, consisting high-molecular-weight protein polymers (Matheis and Whitaker 1984).

Mechanism-based enzymatic states have been proposed to explain the activity of PPO (Solomon et al. 1992; Espín et al. 1998). The structure of copper active site of PPO enzyme is present in three isoforms, namely, *oxy*-PPO[Cu(II) Cu(II) O_2], met-PPO [Cu(II) Cu(II)], and deoxy-PPO [Cu(I) Cu(I)] (Nirmal and Benjakul 2012). The suggested mechanism for hydroxylation and dehydrogenation reactions with phenols occurs by discrete pathways although they are associated with a common deoxy-PPO intermediate (Whitaker and Lee 1995; Whitaker 1995). Molecular oxygen binds to two Cu(I) groups of deoxy-PPO to give oxy-PPO. The distance between the bond of molecular oxygen joining the two Cu(II) groups is typical of a peroxide.

The two Cu(II) groups of oxy-PPO then bind with oxygen atoms of the two hydroxyl groups of catechol to form the O_2-catechol-PPO complex (Solomon et al. 1992). The catechol is oxidized to *o*-benzoquinone, and the enzyme is reduced to met-PPO. Another catechol molecule binds to met-PPO and gets oxidized to *o*-benzoquinone, and the enzyme gets reduced to deoxy-PPO. The *o*-hydroxylation of a monophenol occurs by reaction with only oxy-PPO. The in vitro reaction starts with met-PPO by reducing it to deoxy-PPO to avoid lag phase. Deoxy-PPO binds with molecular oxygen to yield oxy-PPO. The monophenol binds with one of the Cu (II) groups via the oxygen atom of the hydroxyl group to give the O_2-monophenol-PPO complex. Consequently, the hydroxylation of *o*-position of the monophenol by an oxygen atom of the molecular oxygen of the O_2-monophenol-PPO complex gives catechol, which then disassociates to give deoxy-PPO to complete the cycle. Only the first cycle of hydroxylation of a monophenol needs initiation at the met-PPO, and after that sequential mechanism begins with deoxy-PPO (Whitaker and Lee 1995; Whitaker 1995).

4.4 Control of Enzymatic Browning

Browning of plant foods by enzymes results in 50% loss of tropical fruits. Control of enzymatic browning has many economic and quality benefits in fruits and vegetables. Enzyme browning can be prevented by following methods:

4.4.1 Heating and Cooling

Temperature has a significant impact on the rate of biochemical reactions as well as the activity of enzymes. The sensitivity of PPOs to thermal treatment varies depending on the source, yet the temperature of 70–95 °C has been reported to destroy the activity of PPOs (Chutintrasri and Noomhorm 2006; Ndiaye et al. 2009; Özel et al. 2010). Blanching is the commonly employed method for prevention of enzymatic browning wherein steam blanching offered inferior results in the context of POD inactivation and heterogeneity in contrast to water blanching (Shivhare et al. 2009). However, destruction of several heat-sensitive vitamins and loss of delicate texture and aroma make blanching disadvantageous. The application of microwave energy for blanching is beneficial as microwave blanching better conserves the nutritional value of products. Alternatively, a treatment with superheated steam with sprays of microdrops of hot water can be used for blanching as it gives better results for potatoes in association with the traditional methods of blanching (Sotome et al. 2009). In addition, freezing may also be used for prevention/control of enzymatic browning as freezing decreases the availability of water for taking part in enzymatic reactions leading to reduced activity of PPOs (Lavelli and Caronni 2010). Zhou et al. (2008) showed that browning by oxidation and polymerization of phenols could be avoided by storing the fruit at low temperatures. Despite, freezing can be a useful technique for prevention of browning if the product needs not to be thawed since enzymatic browning happens very quickly in produce after thawing which may alter food quality (Ioannou and Ghoul 2013). Thus, freezing has been employed in combination with other methods such as blanching or dipping to increase product shelf life (Gossinger et al. 2009).

4.4.2 Heat Shock Method

The high-temperature short-time (HTST) method commonly known as heat shock method generally consisted of washing at a temperature of 45–70 °C for not more than 5 min. By this method, the activity of PPO is inhibited; thereby it is proven beneficial in the prevention of browning and preservation of foods. Loaiza-Velarde et al. (1997) described that heat treatment (50–60 °C) of freshly chopped lettuce inhibits enzymatic browning. Murata et al. (2004) also stated prevention of browning in lettuce by heat treatment, limiting the increase of polyphenols along with enhanced organoleptic properties in vegetables. Vegetables like lettuce and celery originally have less concentration of preformed phenolic compounds.

Accumulation of these compounds after cutting occurs due to induced synthesis, and it results in browning (Hisaminato et al. 2001). Heat shock treatment (45 °C for 90 s) inhibits browning in fresh-cut lettuce by redirecting synthesis of proteins away from the production site of wound-encouraged enzymes of phenolic metabolism and near to the site of synthesis of safe heat-shock proteins (Saltveit 2000; Martin-Diana et al. 2005).

4.4.3 High-Pressure Processing

Enzymes can be deactivated by exposing food products to high pressure in the range 3000–8000 bars without affecting nutrients and flavor related with conventional thermal processing treatments (Rico et al. 2007; Palou et al. 2000). However, the integrity of porous products is affected by their use on vegetables. Air limited in the food matrix is exposed to expansion and compression during decompression and pressurization, hence disturbing food tissues and creating this operation inappropriate for fresh fruits and vegetables.

4.4.4 Chemical Anti-browning Agents

The chemical anti-browning agents are classified based on their role in various groups like reducing agents, antioxidant agents, acidifying agents, and chelating agents. Antioxidants inhibit the origination of browning upon reaction with oxygen. They also play part in decreasing the chances of degenerative syndromes and the oxidative damage associated with it (Singh et al. 2015, 2017a, b). The antioxidants check chain reaction and prevent the formation of melanins by reacting with the intermediate products (Lindley 1998), whereas reducing compounds inhibit browning by decreasing the *o*-quinones back to their parent phenolic compound or by causing irreversible inactivation of PPOs. Hexylresorcinol, *N*-acetyl cysteine, erythorbic acid, ascorbic acid, cysteine hydrochloride, and glutathione are main antioxidants which have been studied for preventing browning of fruits (Oms-Oliu et al. 2006; Arias et al. 2007; Ioannou and Ghoul 2013). Hexylresorcinol prevents polyphenol oxidase activity by a competitive type (Jiménez and García-Carmona 1999) or a slow-binding inhibition mechanism (Jiménez and Garcáa-Carmona 1997). It showed an inhibition effect at a concentration of 0.04 mg/g in mango purees (Guerrero-Beltrán et al. 2005). Ascorbic acid (vitamin C), in addition to acidification, controls PPO activity by reducing *o*-quinones to their polyphenolic substrates (McEvily et al. 1992; Guerrero-Beltrán et al. 2005). However, the effect of this vitamin C is temporary as it gets oxidized permanently by intermediates such as endogenous enzymes, pigments, and copper (Queiroz et al. 2008). Kojic acid has anti-browning effect owing to its ability to inhibit PPO activity by interfering with the uptake of O_2 required for reaction and bleach melanin because it chemically reduced colored pigments to colorless one (Chen et al. 1991; Queiroz et al. 2008). It showed an anti-browning effect in apple juice at applications ranging from 1 to

4 mM (Iyidogan and Bayiindirli 2004). Son et al. (2001) reported that kojic acid had the higher inhibitory activity against browning in apple slice than other phenolic acids (caffeic, ferulic, chlorogenic, coumaric, cinnamic, and gallic acid), yet its use in the food industry is not widespread as its large-scale production is difficult and costly. Cysteine, a sulfur-containing amino acid, is also an effective compound for the prevention of browning reactions as it reacts with *o*-quinones intermediate to produce colorless and stable products (Iyidogan and Bayiindirli 2004; Dudley and Hotchkiss 1989). Cysteine was found to be active against inhibition of the activity of PPO in mango puree at 0.2 mg/g concentration (Guerrero-Beltrán et al. 2005) and in avoiding the browning of apple juice at concentrations between 1 and 4 mM (Özoglu and Bayiindirli 2002; Iyidogan and Bayiindirli 2004).

Chelating agents form complexes with PPOs or react with its substrates leading to decrease in enzymatic browning. These compounds reduce enzymatic browning because of their capability to form a complex with Cu present in enzyme structure. The most commonly used chelating agent in fruit processing is a citric acid which not only reduces the pH but also chelates copper present in the active site of PPOs, thereby inactivating the enzyme (Son et al. 2001). Similarly, benzoic, cinnamic, and oxalic acids also prevent PPO activity by forming a complex with copper at the enzyme's active site (Tong et al. 1995; Marshall et al. 2000) because of their affinity to form metal complexes with a copper ion.

Acidifying agents, e.g., citric acid, ascorbic acid, and glutathione, control browning by lowering the pH of the system since optimum pH for PPO action ranges from 5 to 7.5; the lesser values prevent enzymatic activity. Acidifying agents decrease pH value below 3.0 at which PPO becomes inactive (Richardson and Hyslop 1985). Acetic acid, citric acid, malic acid, malonic acids, and tartaric acid are the main compounds; however, these (except citric acid, which also acts as a chelating agent) are rarely used for the inhibition of enzymatic browning in fruits and vegetables.

On the other hand, calcium salts, e.g., calcium propionate, calcium lactate, calcium ascorbate, calcium chloride, etc., prevent the destruction of cell compartments by exerting strengthening of cell walls hence preventing the contact of PPO with polyphenols in the vacuole (Quiles, et al. 2007; Guan and Fan 2010; Khunpon et al. 2011). The grouping of calcium with ascorbic acid was described to inhibit membrane and cell breakdown and control activity of PPO in impaired cells, whereas damage of compartmentalization has previously happened (Toivonen and Brummell 2008).

Since different chemical anti-browning agents work on different mechanisms, these may be combined to achieve a better control of enzymatic browning (Zocca 2010). Suttirak et al. (2010) used oxalic acid, ascorbic, and citric acid as anti-browning agents and reported that browning of fresh-cut apples and mango could be effectively eliminated using oxalic acid together with citric or ascorbic acid. Özoglu and Bayiindirli (2002) showed that the use of cysteine, cinnamic, and ascorbic acid together had a synergistic effect in hindering browning of apple juice. Similarly, Guerrero-Beltrán et al. (2005) showed a synergistic effect of ascorbic acid (1 mg/g) or cysteine (0.3 mg/g) with 4-HR (0.04–0.08 mg/g) in

decreasing PPO activity and better stability of color in mango puree at the time of storage. A treatment of fresh-cut pears with hexylresorcinol (0.01%), ascorbic acid (0.5%), and calcium lactate (1%) was also reported to stabilize their color for 30 days (Dong et al. 2000).

4.4.5 Other Methods

The use of osmotic dehydration for concentration has been reported to decrease enzymatic browning (Convey et al. 1983). Coating of foods with edible films can enhance their shelf life by exhibiting a barrier toward gases, water vapor, and light along with the incorporation of active components such as antimicrobials, antioxidants, and flavors (Tien et al. 2001; Shevkani and Singh 2015).Coating of freshly cut apples by whey protein isolate-beeswax improved the shelf life of foods by inhibiting PPOs (Perez-Gago 2003). The combination of physical methods (blanching) with chemical anti-browning treatments has tried to inhibit enzymatic browning in vegetables and fruits (Premakumar and Khurduya 2002, Severini et al. 2003; Yadav 2008, Guan and Fan 2010). Saengnil et al. (2006) applied warm water dipping and oxalic acid for controlling enzymatic browning in litchi. They showed that hot-water dips followed by oxalic acid treatment inhibited browning during storage by reducing PPO and POD activities. Furthermore, the enzymatic browning can also be prevented/controlled by developing transgenic lines that have a reduced amount and activity of PPOs (Coetzer 2001; Rodov 2007). An innovative technique to control the PPO activity is the use of antisense techniques (Bird and Ray 1991). Antisense RNAs have been found to selectively block the expression of the gene of plant enzymes, such as peroxidase and polygalacturonase in tomatoes (Sherf et al. 1993). The expression of PPO in potatoes has been reduced by using vectors carrying antisense PPO cDNAs (Bachem et al. 1996).

4.5 Conclusion

Enzymatic browning is the second main reason for the quality deterioration in fruits and vegetables. PPO exists in a wide range of fruits and vegetables. It is liable for enzymatic browning in fresh horticultural crops during processing conditions like cutting, slicing, etc. that damages the cells. Physical methods for preventing enzymatic browning consist of refrigeration, freezing, blanching, and modification of product atmospheres like MAP, whereas the chemical methods for preventing enzymatic browning comprised the practice of using certain agents like sodium bisulfite, ascorbic acid, and chelating agents or use of antioxidants and agents for firmness, etc. However these treatments could also be applied in different combinations for more efficacy, after optimizing them for different species and cultivar. However to solve some problems, modest techniques, such as the use of

edible coating films which actively inhibit both browning and textural declining, can also be used. New methods for preventing enzymatic browning are based on the usage of antisense RNA technology.

4.6 Future Perspectives

The business of fresh-cut fruits and vegetables is frequently increasing because of shoppers and buyer's response. People determine the quality of freshly cut fruits on the basis of freshness and appearance during buying. Processing methods like peeling, cutting, grating, etc. change the structural integrity of vegetables and fruits leading to some detrimental effects on quality like the development of off-flavor, browning, and breakdown of texture. Browning of fruits and vegetables due to enzymatic oxidation of phenolic compounds leads to a reduction in quality parameters in terms of appearance as well as nutritionally with the development of poisonous compounds. The unfavorable enzymatic browning in foods is a matter of great concern and is required to be examined by using effective enzyme inhibitors. Components which are accomplished to inhibit enzymatic browning in food harvests through the means of interfering tyrosinase-mediated reactions or through the decrease in the level of *o*-quinones to *o*-diphenols have been recognized. Safety is of primary concern for an inhibitor to be used in the food production. There is a need for continuous exploration related to improved inhibitors from natural sources so that they are free of any detrimental side effects. Edible coatings can be used to protect active constituents like flavors, colorants, anti-browning agents, spice nutrients, and antimicrobial components. Coatings increase the shelf life and decrease the risk of flourishment of pathogens on food surfaces. Though particular research on freshly cut fruits are inadequate and their industrial utilization is still under process, a new advancement in edible coatings is beneath, with the primary purpose of addition and/or organized release of active components with nanotechnological answers like multilayered systems and nano-encapsulation. These days, nanotechnology is applied to boost the nutritive features of food with nanoscale nutrients, additives, and nano-sized distribution systems for bioactive components (Bouwmeester et al. 2009). Use of nano- and microencapsulation of active composites with edible coverings helps to manage the release of active components under definite conditions (Lopez-Rubio et al. 2006), thus keeping them away from heat, moisture, and other extreme circumstances and improving their stability and feasibility. However, the usage of silver nanoparticles (Ag NPs) may offer an upcoming substitute toward current harmful and expensive enzymatic browning lowering, antibacterial agents, and antioxidants. Ag NPs exhibited a substantial decline in enzymatic browning with an enzymatic browning reduction index of 8.4 (Khan et al. 2016). In recent studies, chitosan nano-encapsulation was found to be effective as it increases the PPO inhibitory activity of ascorbyl palmitate (Kim et al. 2013).

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