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Critical Analysis of Polyphenol Oxidase: A Major Contributing Factor in Enzymatic Browning Reaction

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Enzymatic browning takes place due to chemical reactions in foods. The reaction occurs in enzymatic browning is mainly caused by enzymes like polyphenol oxidase (PPO), peroxidase (POD) and tyrosinase (Hamdan et al. 2022). The oxidation of phenols to quinines in the presence of oxygen is due to these enzymes. However, this reaction occurs only if the enzymes, phenolic compounds, and oxygen come together. Tyrosine, catecholase, phenolase, and polyphenol oxidase are the enzymes involved in enzymatic browning (McEvily et al. 1992). The role of browning in foods is very crucial. It is generally used to determine raw product shelf life. Hence, it is important to inhibit the enzymatic reactions in raw foods after harvesting. Enzymatic browning mainly occurs in foods during harvesting, transportation, storage, and processing. Also, it affects the nutritional values and texture of the foods.

Why it is undesirable?

The color is a very important attribute of fruits and vegetables which attracts the customers to buy the products. Hence, fruits and vegetables which turn brownish to blackish in color decrease the palatability and attractiveness, declining market acceptability as a result. Apart from that, it also causes undesirable post-harvest changes such as nutritional, biochemical, functional as well as sensory in fruits and vegetables (Navina et al. 2023). That is why, it is important to prevent this reaction in fruits and vegetables to extend their shelf life and to increase their beneficial factors as well.

How Enzymatic Browning can be controlled?

Due to enzymatic browning, 50% of total tropical fruits are lost. Hence, the controlling enzymatic browning has become a crucial factor to preserve the nutritional as well as the shelf life of the products. There are various ways (Singh et al. 2018) to prevent enzymatic browning and it is listed below.

Heating and cooling

Temperature, the rate of biochemical reactions and the enzymatic activity is inversely proportional. It has been reported that at a temperature of 70-95˚C, PPOs are sensitive to it and gets destroyed easily. Blanching, which is a common method for inhibiting enzymatic activity can be used to prevent browning. This method destroys the enzymes, such as PPO, to prevent browning. However, it is disadvantageous because heat-liable vitamins are destroyed and there is even loss of texture and aroma of the product. The application of microwave energy to blanch the products is very advantageous as it prevents loss of nutrients. Apart from that, freezing technique is also used in preventing browning. In this method, the water particles are frozen and turns into solid, i.e., ice crystals. This thus, results in the reduced activity of PPOs.

Heat Shock Method

Heat shock method is usually done at temperature 45-70˚C but not more than 5 mins. Here, the PPO is inhibited and helps in controlling the browning of foods. Vegetables like lettuce and celery have low levels of phenolic compounds. This is why, accretion of compound occurs after cutting due to synthesis of protein which leads to browning. At 45˚C for 90 sec, this method hinders fresh cut lettuce from browning by altering the protein synthesis.

High Pressure Processing

High Pressure Processing (PPO) is a preservation which uses the application of non – thermal technique to deactivate the enzymes and inactivate harmful pathogens by the use of pressure (Balasubramaniam et al. 2008). At a pressure range of 3000-8000 bars, enzymes get deactivated. This method works on mainly two principles, i.e, Le Chatelier's principle which states that any phenomenon accompanied by a decrease in volume is enhanced by

pressure. And Isostatic Principle which states that pressure is uniformly distributed throughout the entire sample, whether in direct contact with pressuring medium or insulated from it in a flexible container. This method is popular since this does not affect the nutrients present in the food as well as the flavour of products like the thermal treatments.

Chemical Agents

The anti-browning agents which are chemical in nature are used in preventing browning (McEvily et al. 1992). They are classified into groups and they are as follows.

Reducing agent

The ability of reducing agents or antioxidants to chemically reduce endogenous or enzyme-produced O-quinones to diphenols (colorless) or to irreversibly react with O-quinones to form stable colorless products is what prevents browning. However, since PPO enzyme does not directly react with ascorbic acid, it depletes enzymatic browning by reducing oxidized substrates. Ascorbic acid is a widely used anti-browning agent. Anti-browning property of ascorbic acid is due to reducing activity mechanism. The examples of reducing agent include sulphiting agents, ascorbic acid and analogs, cysteine, glutathione.

Chelating agents

Chelating, or sequestering, compounds bind to many of the minerals found in food and are required as cofactors for the activity of various enzymes. This prevents food products from deteriorating due to a variety of enzymatic processes that occur during processing and storage. In the context of PPO-catalyzed browning, chelating compounds are believed to either bind to the copper in the active site of PPO or reduce the amount of copper available for integration into the holoenzyme. The examples of chelating agent include ethylenediaminetetraacetic acid [EDTA], citric acid, tartaric acid, glycine, pyrophosphoric acid, tripolyphosphoric acid

Acidulants

The aim of acidulants is to maintain the media's pH below the necessary level for optimal catalytic activity. Depending on the enzyme's source and the particular substrate, polyphenol oxidase activity's optimal pH varies, however in most situations it has a pH range between 6 and 7. PPO formulations are reportedly inactive below pH 4 according to a number of sources. The examples of Acidulants include acetic, adipic, citric, fumaric, lactic, malic, phosphoric and tartaric acids, and glucono-delta-lactone.

Conclusion

Enzymatic Browning in fruits and vegetables is a natural phenomenon ad occurs only after post-harvest due to chemical reactions. There are several enzymes but Polyphenol Oxidase plays a major role in it. The oxidation of Polyphenol oxidase in the presence of oxygen converts the enzyme into a compound called Quinones which are mostly harmful in nature and an undesirable characteristic of fruits and vegetables products. In order to inhibit or prevent the formation of this compound, several chemical and non-chemical methods can be used. This helps in keeping the nutrients intact as well as increasing the shelf-life of the fresh fruits and vegetables. Thus, subsequently increases the attractiveness as well as palatability of the food products as well.

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