

# APPLE BROWNING

Eileen Hemmati, Farzanegan 2 high school, Tehran, Iran

## ABSTRACT

This problem is about apple turning brown after being cut or bruised. We need to find ways of preventing browning. To find chemical reactions and parameters which are important in this phenomenon, Several experiments are designed. Some of the substances such as citric acid, and salt are studied and the duration which it takes the color of the fruit (apple) is changed. The objective of the present work was to quantify enzymatic browning and PPO activity and identify and quantify target polyphenols in apple. Fruit which contains acid, it is helpful in stopping the enzymatic browning and somehow paralyzes the polyphenol cells.

**Keywords :** Enzymes, Polyphenols, Browning, Fruit

## ARTICLE INFO

Participated in PYNT 2017

Advisor: Dr. Mohammad Qorbani

Accepted by Ariaian Young Innovative

Minds Institute, AYIMI

<http://www.ayimi.org>, [info@ayimi.org](mailto:info@ayimi.org)

## 1. Introduction

Polyphenols are one of the most important and certainly the most numerous among the groups of phytochemicals present in the plant kingdom.

Currently, over 8000 phenolic structures have been identified, of which more than 4000 belonging to the class of flavonoids, and several hundred occur in edible plants.

However, it is thought that the total content of polyphenols in plants is underestimated as many of the phenolic compounds present in fruits, vegetables and derivatives have not yet been identified, escaping the methods and techniques of analysis used, and the composition in polyphenols (Fig. 1) for most fruits and some varieties of cereals is not yet known.

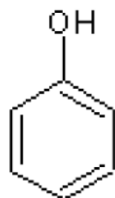


Fig. 1: Polyphenol molecule

The term polyphenols refers to a wide variety of molecules that can be divided into many subclasses, subdivisions that can be made on the basis of their origin, biological function, or chemical structure. Chemically, they are compounds with structural phenolic features, which can be associated with different organic acids and carbohydrates. In plants, the most part of them are linked to sugars, and therefore they are in the form of glycosides. Carbohydrates and organic acids can be bound in different positions on polyphenol skeletons.

Among polyphenols, there are simple molecules, such as phenolic acids, or complex structures such as condensed tannins, that are highly polymerized molecules.

## 2. Material and Methods

### 2.1. Polyphenol Oxidase

Polyphenol oxidase (tyrosinase) (TY) is a bifunctional, copper-containing oxidase having both catecholase and cresolase activity (Malmström and Rydén 1968) (Fig. 2).

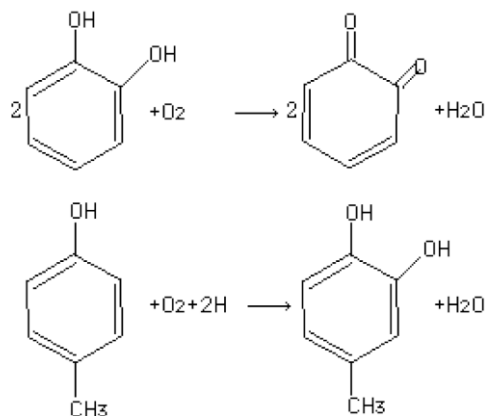


Fig. 2: Polyphenol oxidase

Jolly et al. (1974) refer to it as oxygen and 4 electron-transferring phenol oxidase. It is responsible for browning reactions throughout the phylogenetic scale.

Although a tyrosinase from *Neurospora crassa* has been purified (Fling et al. 1963), most work has been done with the mushroom enzyme, even though yields and consistency are poor; its multiplicity was shown by Smith and Krueger (1962). Bouchilloux et al. (1963) obtained four enzymes. See review by Nelson and Mason (1970).

**Inhibitors:** Compounds that complex with copper. The enzyme is also inhibited competitively by benzoic acid with respect to catechol and by cyanide with respect to oxygen (Duckworth and Coleman 1970).

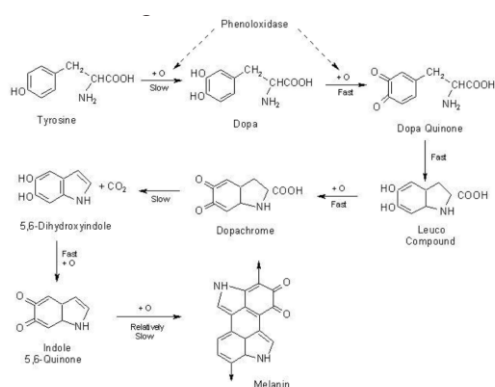
**Activity:** Polyphenol oxidase is an oxygen transferring enzyme. Besides using O<sub>2</sub> to catalyze the dehydrogenation of catechols to orthoquinones and the orthohydroxylation of phenols to catechols, a peroxidase activity has been reported on by Strothkamp and Mason (1974). Kinetic studies have been reported by Kertész et al. (1971). See also the review by Malmström and Rydén.

**Specificity:** A large number of parasubstituted catechols are oxidized (Duckworth and Coleman 1970).

**Stability:** The lyophilized preparation is stable for 6-12 months when stored at -20°C.

### 2.2. Enzymatic Browning

Enzymatic browning is one of the most important reactions that occur in fruits and vegetables, usually resulting in negative effects on color, taste, flavor, and nutritional value. The reaction is a consequence of phenolic compounds' oxidation by polyphenol oxidase (PPO), which triggers the generation of dark pigments. This is particularly relevant for apples, which are rich in polyphenols and highly susceptible to enzymatic browning. The objective of the present work was to quantify enzymatic browning and PPO activity and identify and quantify target polyphenols in apple [*Malus ×sylvestris* (L.) Mill. var. domestica (Borkh.) Mansf.] pulp in the cultivars (cvs.) Aori27, Elstar, Fuji, and Mellow at three fruit developmental stages (FDS). Enzymatic browning showed significant among cvs. and FDSs and interaction between both factors. PPO activity showed significant difference among cultivars and FDSs. A significant difference was evidenced for polyphenol content among cultivars and FDSs with interaction between both factors (Fig.3).



## 3. Experiment

### 3.1. Lemon Juice

Lemon juice is another traditional and popular way of preventing this procedure. Lemon juice with the pH of 2 to 2.60 is considered as a fruit which contains acid, which is really helpful in stopping the enzymatic browning and somehow paralyzes the polyphenol cells. But there's a problem! We need a general way to prevent, but lemon juice changes the taste and color of the apple slices (Fig. 4).

**WHAT MAKES LEMONS SOUR?**  
The sour taste of lemons is caused by the presence of organic acids. The major acid in lemons is citric acid, which makes up around 5 to 6% of the lemon's juice.  
Other acids are also present, although in much lower concentrations than citric acid. Malic acid is one of these, present at around 2% of the concentration of citric acid.

**VITAMIN C, LEMONS & SCURVY**  
Lemons contain high levels of vitamin C, also known as ascorbic acid. The levels in lemons are around 50mg per 100g, and a pair with oranges and kiwis can provide the amount of Vitamin C.  
Vitamin C deficiency can lead to scurvy, a disease that causes loss of teeth, jaundice, and eventually death. In the 1700s, all British ships were required to provide a lemon juice ration to seamen to guard against this disease.

Time (Min)	Vitamin C (mg)
15	9.0
30	8.8
45	8.0
60	7.0
90	7.0
105	5.0
120	5.0
135	4.0
150	3.0
165	3.0
180	2.0
195	2.0

Fig. 4: Stopping the enzymatic browning vs time by Lemon juice

### 3.2. Tap Water

You probably know water's chemical description is H<sub>2</sub>O. A water molecule consists of one atom of oxygen bound to two atoms of hydrogen. The hydrogen atoms are "attached" to one side of the oxygen atom, resulting in a water molecule having a positive charge on the side where the hydrogen atoms are and a negative charge on the other side, where the oxygen atom is. Since opposite electrical charges attract, water molecules tend to attract each other, making water kind of "sticky." The side with the hydrogen atoms (positive charge) attracts the oxygen side (negative charge) of a different water molecule. All these water molecules attracting each other mean they tend to clump together. This is why water drops are, in fact, drops! If wasn't for some of Earth's forces, such as gravity, a drop of water would be ball shaped -- a perfect sphere. Even if it doesn't form a perfect sphere on Earth, we should be happy water is sticky. Water is called the "universal solvent" because it dissolves more substances than any other liquid. This means that wherever water goes, either through the ground or through our bodies, it takes along valuable chemicals, minerals, and nutrients (Fig. 5).

Pure water has a neutral PH about 7, which is neither acidic nor basic.

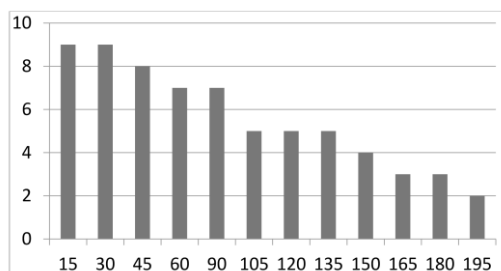


Fig. 5: Stopping the enzymatic browning vs time by Tap Water

### 3.3. Citric Acid

As we know citric acid is a nearly strong acid which could destroy our apple So I used one with higher pH which is 4.8, but I used it in two forms, crystals and liquid. To estimate what form works the best, I tested both of them. The pH of this acid is 4.8 and the chemical Formula C<sub>6</sub>H<sub>8</sub>O<sub>7</sub>.

We used 50 g of Crystal form of Citric acid and sprinkled it all over the bare part of apples. It ruins the surface and gives it a chemical taste which makes it uneatable and it is more harmful than the liquid form.

In other experiment we poured 50 g acid on the surface. It makes red color but after 1 hour it stayed still and it was better than crystals (Figs.6 and 7).

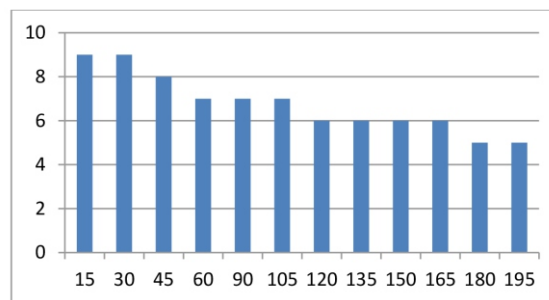


Fig. 6: Stopping the enzymatic browning vs time by liquid citric acid

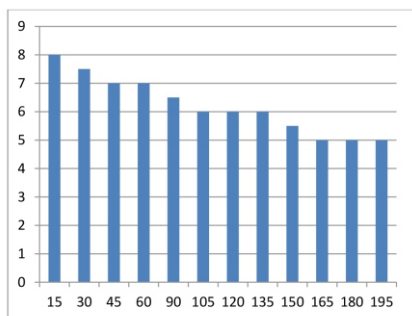
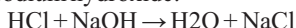


Fig. 7: Stopping the enzymatic browning vs time by crystal citric acid

### 3.4. Salt

Sodium chloride is the most common salt, the common table salt used for the seasoning of food. It forms in cubic crystals apparent even in table salt. It occurs commonly in the mineral form, halite, also called rock salt. It can be formed by the acid-base reaction of hydrochloric acid and sodium hydroxide:



As a solid, sodium chloride is crystalline and forms a cubic lattice. The bonding of the sodium and chlorine atoms is one of the classic examples of ionic bonding. In aqueous solution it ionizes to  $\text{Na}^+$  and  $\text{Cl}^-$  ions and forms an electrically conducting solution.

The ionic bonding of  $\text{NaCl}$  can be visualized in terms of Lewis diagrams.

When the diameters of sodium and chlorine and their ions are measured, they offer some confirmation of the picture of ionic bonding.

When sodium loses an electron to form  $\text{Na}^+$ , its effective size decreases to about half. When chlorine gains an electron to produce the octet structure  $\text{Cl}^-$ , its size increases to almost double (Shipman, et al).

Sodium chloride exists on the earth in great abundance in sea water and is an important part of the fluid electrolytes of humans and other living organisms (Fig. 8).

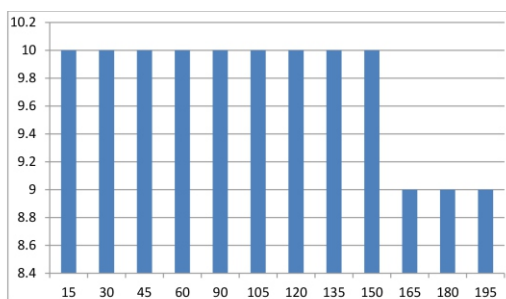


Fig. 8: Stopping the enzymatic browning vs time by salt

### 4. Results

Due to diagrams salt was the substance we were looking for, by a small calculation we realize that it's the best possible way. Because when we sprinkle a small amount of this chemical on the surface, it keeps it white for a long time and has 2cm penetration after 3:45 hours. But the disadvantage in that it soaks in the water and makes it like jelly.

So the solution changes, we need a physical way of preventing which is blocking the oxygen which is physical. Because it's general and can be used by companies and for

shipping this fruit to long distances.



Fig. 9: Different slices with salts to prevent browning

### 5. Conclusion

After all the progress in our experiments, still we haven't found an adequate and general chemical substance that can be used in some jobs such as shipping fruits like apple or pear over long distances.

But the answer is a physical change and not a chemical one. Because any chemical change, changes the substance and that physical change that I considered, is blocking the oxygen because oxygen helps the process of oxidizing the polyphenols by polyphenol oxidase so it needs to be taken away.