Determination of Ascorbic Acid Level in Bell Peppers

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Abstract

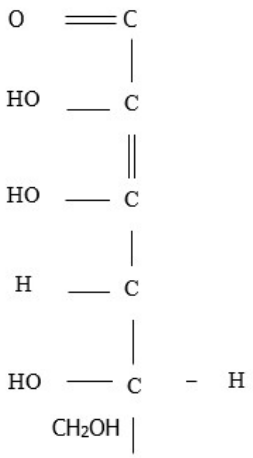
The research work determined the vitamin C levels in green and red bell pepper fruits sold in Kontagora market. The analysis of vitamin C level was done by iodimetry titration method. Vitamin C is required by human body to improve the immune system and can be obtained from the bell peppers. Bell pepper has different colours according to the level of maturity, ranging from green, yellow, orange, and red. Differences in colour also make possible differences in vitamin C content. The results show that vitamin C level in Red bell pepper juice contain more ascorbic acid (1.19mg/ml) than the Green bell pepper (0.33mg/ml), indicating that different stages of bell pepper maturity have different vitamin C levels. Therefore, this research recommends that based on the result and comparison with other literatures, further study should be carried out on the effect of heat on the ascorbic level in bell peppers, the amount of calories in bell peppers and also the amount of ascorbic acid level in yellow bell peppers.

Keywords: Green and Red bell peppers, Iodimetry, Titration and Vitamin C

Introduction

Ascorbic acid (vitamin C) is a naturally occurring organic compound with antioxidant properties. It is a white solid, but an impure sample can appear yellowish. It dissolves well in water to give mildly acidic solutions with molecular mass of 176.12gmol^{-1}, molecular formula $C_6H_8O_6$ and melting point of 190 to 192 degree Celsius (Lachapelle, 2010). According to Wardlaw (1996) by definition vitamin C dissolves in water. In addition, the vitamin B and vitamin k function as parts of co-enzymes i.e., molecules that help enzymes to function. Although vitamins themselves yield no energy to the body, they often facilitate energy yielding chemical reaction, vitamin A D E and K dissolved in organic solvent whereas vitamin B and C dissolved in water. It was originally called L-hexuronic acid, but, when it was found to have vitamin C activity in animals ("vitamin C'' being defined as a vitamin activity, not then a specific substance), the suggestion was made to rename L-hexuronic acid. The new name for L-hexuronic acid is derived from a (meaning "no'') and scorbutus (scurvy) i.e. the disease caused by a deficiency of vitamin C. Because it is derived from glucose, many animals are able to produce it but humans require it as part of their nutrition. Other vertebrates which lack the ability to produce ascorbic acid include primates, guinea pigs, teleost fishes, bats and some birds, all off which require it as a dietary micro nutrient that is, in vitamin form. (Lachapelle and Drouin, 2010).

The structure of ascorbic acid was elucidated by (Finar, 1981; Andrea, 2013) as shown below.



Also, one of the most important vitamins in our diets is vitamin C (which is also known as ascorbic acid) found in many fruits and vegetables, vitamin C is also easily damaged or destroyed. Also vitamin C is a water soluble vitamin that has many functions in the body. Ascorbic acid (vitamin C) is powerful reducing agent (antioxidant) and it likely that all of its, biochemical and molecular functions relate to this property. In humans, vitamin C acts as an electron donor for eight enzymes, of which three are involved in collagen hydroxylation (including aspects

of nor epinephrine, peptide hormone and tryrosine metabolism) are involved in carnitine biosynthesis (Procop and Kiviikko 1995). Humans and primates lack a key enzyme, L-3 gulonolacton oxidase, necessary for the biosynthesis of vitamin C. Vitamin C main function in chemistry appears to be balancing or setting the redox potential, the oxidized (dehydroascorbic acid) is readily reduced by the tripeptide gluthathione, and its main biological rate may be maintenance of gluthathione in tissues in its oxidized form, (Johrison *et al.*, 1993).

A bell pepper fruit are grown around the world and is used for colon, flavour and aroma. Some countries have used sweet pepper for thousands of years. Now, it is mostly commonly grown in Hungary (Europe). Bell peppers has been used for various conditions, including nausea, vomiting, and the desire to drink alcohol. In this research, it will be scientifically test the amount of vitamin C in vitamin C tablet solution as the control sample and in bell pepper (that is, green and red) to determine the amount of vitamin C. Bell pepper contains nutrients such as carbohydrates, proteins, fats, minerals (potassium, calcium, phosphorus, and iron), and vitamins (vitamin A, vitamin B, and vitamin C) (Nadeem *et al.,* 2011). Vitamin C is also known as ascorbic acid that can be found in nature almost in all plants, especially fresh vegetables, and fresh fruits, so called fresh food vitamins (Singh and Kumari, 2015).

A mixture containing sweet pepper reduce oxidative stress and also help in the reduction of food and energy intake, as well as body weight by decreasing hunger and appetite food. (Podman, 1998). There are several methods in determining vitamin C levels, namely: iodimetry titration method (Bekele and Geleta, 2015; Tareen *et al.,* 2015), 2,6-dichloroindophenol titration method (Shrestha *et al.,* 2016), voltammetry method (Pisoschi *et al.,* 2008; Pisoschi *et al.,* 2011), spectrophotometer method and chromatography method (Al Majidi and Al Quburi, 2016). Spectrophotometric methods and chromatographic methods require an instrument in the measurement of vitamin C. The simplest and fastest method for vitamin C analysis is the titration method which can also give similar analytical results to spectrophotometric method and chromatography method (Ullah *et al.,* 2012). Foodstuffs were complex matrix that contains many reducing agents other than vitamin C. The iodimetry titration method uses an iodine solution as the standard solution and not effective for measuring vitamin C levels in foodstuffs, because iodine not selective to vitamin C and oxidize all reducing agents besides of vitamin C. The 2,6-dichloroindophenol titration method use 2,6-dichloroindophenol solution as the standard solution and most widely used for determining vitamin C levels in foodstuffs, because 2,6-dichloroindophenol selective to vitamin C (only oxidize vitamin C) and not oxidize others reducing agents (Tantray *et al.,* 2017).

Hence, the purpose of this research is to determine the level of ascorbic acid in bell peppers *"Capsicum Annnum"* by iodometric titrimetric method. Finally, the results of this research will help doctors to encourage patients that are suffering from scurvy to take plenty of bell pepper and also encourage the agriculturists to boost the production of the bell pepper.

Material and Methods

Materials and Reagents

- Bell pepper fruit (100cm^3)
- Distilled water
- Vitamin c tablet as standard solution (1g)
- Motar, pestle and sieve
- Iodine solution
- Starch solution (1g)
- Hydrochloric acid (HCl) 1M

Apparatus

- 50cm^3 burette and resort standard
- Two small bottles with droppers
- Two 10ml graduated cylinder
- 25cm^3 graduated cylinder
- Two 250cm^3 conical flask
- 100ml volumetric flask
- 250cm^3 of Erlenmeyer flask/ volumetric flask

Preparations of Reagents:

Preparation of vitamin C standard solution

Vitamin C standard solution was prepared by dissolving one tablet of vitamin C (10mg) in 75 cm^3 of distilled water, and allowed to dissolved completely which was further diluted with distil water to 100 cm^3 level marks. This solution is now the standard solution with vitamin C concentration of 1.0 mg/ml.

Preparation of Starch

1g of starch powder was weighed using weighing balance. 100 ml of water was measured and kept on heating. A paste of starch was mixed in a watch glass, the paste was added to the 100ml of boiling water which was kept on heating and stirred. When all the paste was added, it was allowed to boil for a few seconds. And was also removed from heat immediately and allow to cooled.

Preparation of Iodine Solution

0.6g of potassium iodide was weighed using weighing balance and dissolved in 500cm^3 of distilled water. Another 0.6g of iodine crystal (I_2) was weighed and dissolved in 50ml of ethyl alcohol (ethanol) contained in a beaker. The two solutions were mixed and diluted to 1 liter with distilled water in a volumetric flask.

Preparation of 1M Hydrochloric Acid

8.3cm^3 of concentrated HCl was measured and carefully added to 50cm^3 of distilled water. Then it was diluted with distilled water to 100cm^3 level mark volumetric flask.

Sample Extraction (Bell peppers)

140g of each of the bell pepper fruits (red and green) were weighed using weighing balance from the bulky selected samples. Thereafter, they were pounded in a mortar and squeezed into a beaker. The pepper juice was transferred into a sieve and filtered successfully. Highly coloured filtrate was obtained, a clean solution of 100cm^3 of bell pepper juice each was used for the analysis.

Experimental Procedures

Titration of Iodine Solution against Vitamin C Standard Solution

10 ml of vitamin C standard solution was measured using graduated cylinder and placed in a conical flask, 20 ml of distilled water was added to the vitamin C solution contained in the flask. 2 drops of 1M HCl and 15 drops of the starch solution were added. The burette was filled with the prepared iodine solution, and held on the resort stand, and titrated against the vitamin C standard solution contained in the conical flask, drop wise and shake at an interval until a permanent colour of blue-black was obtained. The above procedure was repeated three (3) times to obtained an average titre value.

Titration of iodine solution against the red bell pepper extraction

10ml of red bell-pepper juice was measured using graduated cylinder and placed in a flask, 20 ml of distilled water was added to the pepper juice and also 2 drops of 1M HCl and 15 drops of starch solution were added. The burette was filled with the prepared iodine solution and held on the retort stand, and titrate against solution contained in the conical flask, drop wise and shaken at an interval until a permanent colour of blue-black was obtained. This was repeated three (3) times to obtain an average titre value.

Titration of iodine solution against the green bell-pepper extraction

10ml of green bell-pepper juice was measured using graduated cylinder and placed in a flask, 20 ml of distilled water was added to the pepper juice and also 2 drops of 1M HCl and 15 drops of starch solution were added.

The burette was filled with the prepared iodine solution and held on the retort stand, and titrate against solution contained in the conical flask, drop wise and shaken at an interval until a permanent colour of blue-black was obtained. This was repeated three (3) times to obtain an average titre value.

Results

The result of the analysis ware shown in Table 1, 2 and 3

Table 1: Iodine solution against vitamin C standard solution

Reading	1st	2nd	3rd
Final reading (cm^3)	37.40	41.50	40.30
Initial reading (cm^3)	0.00	0.00	0.00
Volume of Iodine used (cm^3)	37.40	41.50	40.30

Average volume of Iodine used

$$\frac{37.40 + 41.50 + 40.30}{3} = \frac{119.2}{3} = (39.70 cm^3)$$

Table 2: Iodine solution against Red Bell pepper

Reading	1st	2nd	3rd
Final reading (cm^3)	50.00	47.60	44.00
Initial reading (cm^3)	0.00	0.00	0.00
Volume of Iodine used (cm^3)	50.00	47.60	44.00

Average volume of Iodine used

$$\frac{50.00 + 47.60 + 44.00}{3} = \frac{141.6}{3} = 47.20 cm^3$$

Table 3: Iodine solution against Green Bell pepper

Reading	1st	2nd	3rd
Final reading (cm^3)	14.00	11.50	13.50
Initial reading (cm^3)	0.00	0.00	0.00
Volume of Iodine used (cm^3)	14.00	11.50	13.50

Average volume of Iodine used

$$\frac{14.00 + 11.50 + 13.50}{3} = \frac{39.00}{3}$$

$$= 13.00\text{cm}^3$$

From the value obtained in Table 1 above, the titration of iodine solution against vitamin C standard solution, the proportion can now be used to determine the amount of vitamin C in Red and Green bell pepper juice sample.

Recall that the standard vitamin C solution was 1.0mg/ml (10mg).

Using the relation above;

39.7cm^3 ——— 10.0mg for standard solution

47.2cm^3 ——— Xmg for the Red bell pepper

39.7cm^3 ——— 10.0mg

47.2cm^3 ——— Xmg

$39.7 \times X = 47.2 \times 10.0$

$39.7X = 472$

$X \approx 11.9$ mg

Therefore 11.9 mg of vitamin C per 10 ml of Red bell pepper juice.

To get the milligram of vitamin C in one millilitre of Red bell pepper fruit juice we simply divide the value obtained above by10 i.e.

$11.9/10 = 1.19$ mg/ml

For Green Bell pepper

Using the relation above;

39.7cm^3 ——— 10.0mg

13.00cm^3 ——— Xmg

$X \approx 3.3$mg

Therefore 3.3mg of vitamin C per 10ml of Green bell pepper juice.

To get the milligram of vitamin C in one millilitre of Green bell pepper fruit juice we simply divide the value obtained above by10 i.e.,

$$\frac{3.3\text{mg}}{10\text{ml}}$$
$$= 0.33\text{mg/ml}$$

Discussion

According to George *et al* (1990), in human being's ascorbic acid helps in the reduction ferric (Fe^{3+}) to ferrous iron (Fe^{2+}), a necessary stage before Iron is absorbed. Prospective cohort studies indicate that higher intake of vitamin C from either diet or supplements are associated with a reduced risk of cardiovascular diseases (CVDs), including coronary heart disease and stroke. There is evidence that regular use of vitamin C supplements shortens the duration of common cold, but the effect in cold treatment may be limited. From the findings of this research; it was found that there is significant difference between the values of ascorbic acid (vitamin C) obtained from red and green bell peppers.

From the calculations in Table 2 and 3 above, it shows that Red bell pepper juice contain more ascorbic acid (1.19 mg/ml) than the Green bell pepper (0.33 mg/ml). The differences in their values may be due to the gain of ascorbic acid in their ripening process during oxidation (Katlner *et al.,* 1979). Also, after the harvest of fruits and vegetables, then a number of vitamins can be lost during the storage stage, depending on temperature, exposure to air and sun, and storage time. The higher the temperature, the longer exposed to air and sun, the longer it is stored, the more vitamin C is lost (Ibrahim, 2016). Changes in physical properties and chemical properties in fresh fruits and vegetables common during maturation are changes in colours and acid levels (Tosun *et al.,* 2008). The longer the fruits and vegetables are stored in the open, the vitamin C levels will decrease as a result of the oxidation process. So that the titration process during testing should be done quickly to prevent the oxidation of vitamin C in order to provide more accurate results of vitamin C (Bieniasz *et al.,* 2017).

However, it shows that more enzymes are required in the Green pepper formation than in Red pepper, which affects the level of ascorbic acid in both juices. Above all, the significance difference in their ascorbic acid level will not affect human consumption because it is favourable to human health if compared with concentration of the standard solution. Furthermore, these levels of vitamin C in both peppers if consumed per day can contribute to the needed concentration required by human body as recommended by WHO, standard of vitamin C consumption per day which is 60mg/l (USDA, 2009)

Conclusion

Based on the results obtained in this research work; red bell pepper fruits contain higher level of vitamin C than green bell pepper fruits. Bell pepper fruit (Capsicum annum) production could be encouraged even in commercial quantity, bell pepper fruit is highly rich in vitamin C. There is no harm in consuming the green bell pepper fruits; this is because the level of vitamin C (Ascorbic acid) in such fruits is within the recommended intake of 60 mg/day by the standard organization of Nigeria (SON).

Recommendation

Based on the results and comparison with other literatures, further study should be carried out on the effect of heat on the ascorbic level in bell peppers, the amount of calories in bell peppers and also the amount of ascorbic acid level in yellow bell peppers.

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