Study of the Applications and Uses of Ascorbic acid (Vit. C) Estimated in Some Different Foods in the Laboratory and Determined by HPLC Techniques

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Abstract

The study aimed to highlight the importance of using Vitamin C in some food applications, which was estimated in some fresh fruits and vegetables. I collected 8 samples from local markets: Ananas comosus, Malus domestica, Psidium guajava, Citrus limon, Capsicum annuum, Brassica oleracea var. italica, Spinacia oleracea and Solanum lycopersicum. Using the standard iodine solution titration method, the amount of the vitamin was estimated. The amount of vitamin in guava, according to the results of the study was 224.7 mg/100 g, the highest amount in fruit samples, while red pepper in vegetables recorded 119.8 mg/100 g. When adding certain concentrations of Vitamin C to pickles and jams in order to enhance the flavor, taste and health aspect, the best concentration for the period 1-3 days when added to bananas through the Maillard reaction, color, taste and flavour is 5mg/100 gm, in contrast to the concentrations 10 and 15 mg/100 gm which recorded poor results in the potato and carrot test while the concentration 5 mg/100 gm Vitamin C recorded the best results for pickled cucumbers for a period of 1-7 days and was negative for the Maillard reaction It was good for color, taste and flavor. This was shown through the results of the sensory evaluation of the included samples, as they were accepted and welcomed by the residents. The tests were conducted in the laboratories College of Science /Al-Mustansiriya University for the period from 5/10/2024 - 20/11/2024.

Keywords: Vitamin C, food applications, fruits, pickles, jums

.1Introduction

Vitamin C is one of a large group of vitamins found in nature, whether naturally or synthetically, and is also called Ascorbic acid. Given the extreme importance of this vitamin for living organisms, specifically humans, the vitamin plays a prominent role in metabolic processes within the body and provides it with vital functions, including eye protection, bone building, immunity, and antioxidants. As for antioxidants, they also help in the formation of important collagen in healing wounds and cracks[1]. The body is provided with Vitamin C by eating vegetables and fruits, as the body cannot produce it naturally. A healthy and healthy body consumes 200-250 milligrams of Vitamin C daily and the excess is excreted from the body with urine, as it is a water-soluble vitamin. It should be consumed in ideal quantities that do not negatively affect human health. Ascorbic acid does not contain a carboxyl group (COOH) in its composition, and the reason for its acidity is the presence of the enediol or dienoil group[6].

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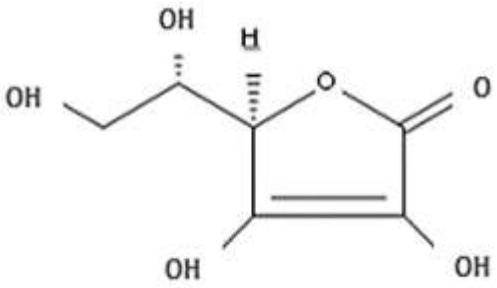

Figure 1. Structural structure of ascorbic acid

Ascorbic acid exists in two structural forms that are extremely important. The first is reduced (L-Ascorbic acid) and is more active, while the second form is oxidized (L-Dehydroascorbic acid) as in figure (2).

Vitamin C in fruits and vegetables was estimated using various methods including spectrophotometric (UV/VIS) measuring devices[10].

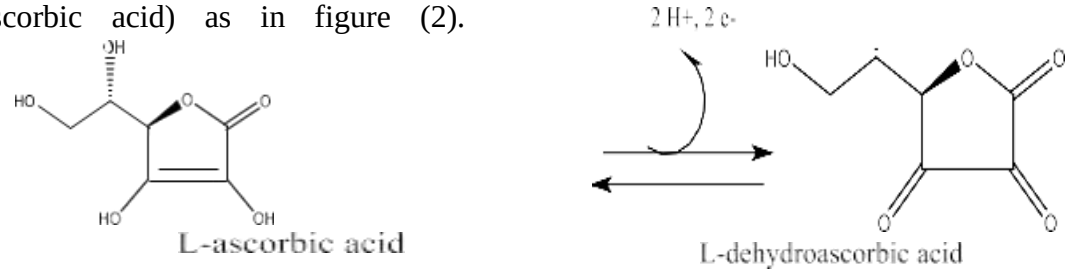

Figure 2. Reduced and Oxidized for Ascorbic acid

The demand has increased recently for foods and other products that contain good functional properties to reduce some diseases or get rid of some health problems. Among these compounds with functional properties is ascorbic acid, which has an important role in various food products due to its prominent role in improving texture, color, taste, flavor and as a preservative in some products[5.[

Ascorbic acid is considered one of the organic acids that is safe for health if taken in ideal quantities via tablets or through food. Anything in excess has harmful effects and serious consequences[8.[

.2Materials and Methods

.2.1Sample collection:

Some samples were collected from different farms in the governorates of Iraq and others were from local markets Ananas comosus, Malus domestica, Psidium guajava, Citrus limon, Capsicum annuum, Brassica oleracea var. italica, Spinacia oleracea and Solanum lycopersicum. for the period from 5/10/2024 - 20/11/2024. The tests were conducted in the laboratories College of Science /Al-Mustansiriya University.

.2.2Sample preparation :

The samples to be estimated were washed with lukewarm water several times to remove dirt and other particles. After that, the damaged and inedible parts were removed, cut into small cubes and then ground with an electric blender. The resulting solution was filtered to obtain a clear liquid on which tests were conducted in order to estimate the quantity Vitamin C.

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Table 1. Sources and types of samples used in the study

No. Sample	Name	Kind	Source
1	Psidium guajava	Fruit	local markets
2	Citrus limon	Fruit	Farm
3	Ananas comosus	Fruit	local markets
4	Malus domestica	Fruit	Farm
5	Spinacia oleracea	Vegetable	Farm
6	Solanum lycopersicum	Vegetable	Farm
7	Brassica oleracea var. italica	Vegetable	local markets
8	Capsicum annuum	Vegetable	local markets
Total	8		

.2.3Preparation of stock and standard solution of Ascorbic acid:

As shown in the table (2) Prepare the ascorbic acid standard solution by dissolving 0.01 g of ascorbic acid standard (BASF) in a 0.5% oxalic acid conical flask of 200 ml and then supplement the volume to 100 ml of the same solution. to get scared (5, 10, 15, 20, and 25) ml/ 100g.

Table 2. Standard Ascorbic Acid

Concentration (ml/100g)	Absorbance
5	0.03
10	0.05
15	0.07
20	0.09
25	0.11

2.4

.Standard curve for Ascorbic Acid (Vitamin C:(

Standard curve was determined by concentrations vs absorption of Ascorbic acid by 2 ml of each standard solution and placing it in a test tube, then 0.3 ml of KMnO4, the absorbance reading was 530 nm against blank by spectrophotometer. As shown in the figure (3).

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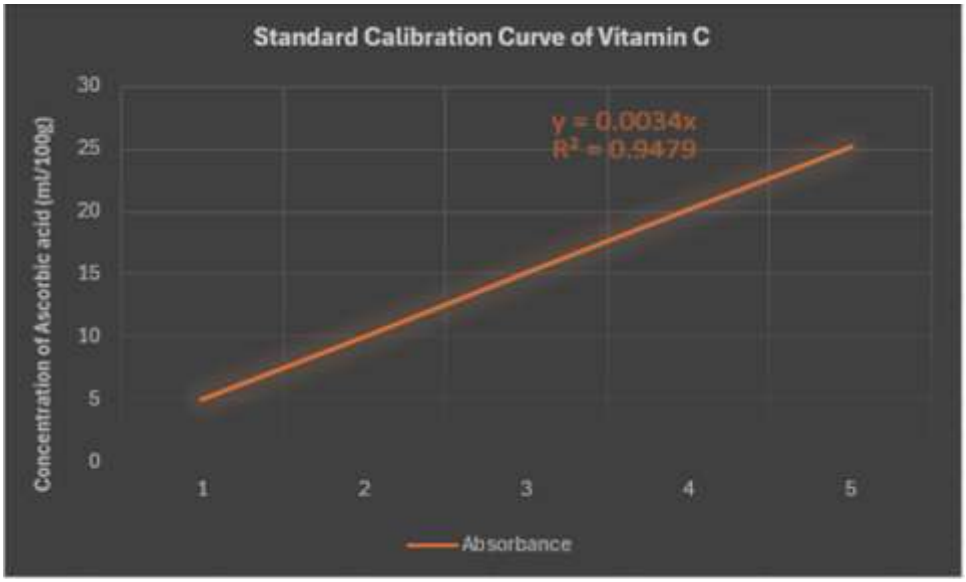

Figure 3. Standard calibration curve of vitamin C

2.5. Determination the amount of Ascorbic acid by titration:

As shown in figure (3) the method for determining acid using titration depends on oxidation and reduction in the presence of iodine solution. Ascorbic acid is oxidized to dehydrogenated ascorbic acid, as shown in the equation, and iodine is reduced to iodide. Appearance of a blue color as a result of the reaction of starch with iodine is evidence of acid oxidation[3].

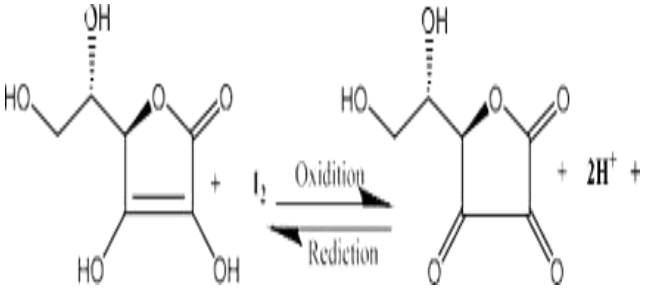

Figure 3. The calibration process through the equation and the appearance of the blue color

10 ml of the clear liquid was taken and placed in 250 ml volumetric flask. Add 5 ml of 1% starch solution, then add 100 ml distilled water to mixture, the mouth of the flask was covered with cellphone or tinfoil. Brought a glass burette and filled it with 0.1 N iodine solution. The burette was covered with cellophane or tinfoil to prevent light from entering to preserve the solution. Inserted the burette into the opening of the flask after piercing the lid and the process of titrating the mixture began by using iodine inside the burette until the blue color appeared indicating the completion of the titration. The test is repeated three times and the rate is taken, after which the amount of ascorbic acid is calculated through the following relationship[11]:

1 ml of 0.1 N iodine solution is = 0.008807 ascorbic acid. And through the equation:

1ml of I_2 = 176.14 / 1000∗10∗2 = gm of ascorbic acid

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Where the number (176.1400) represents the molecular weight of ascorbic acid.

Including the weight of the acid in grams in the titrated solution = the average volume of iodine consumed x 0.008807[4].

2.6. Diagnosis of Ascorbic acid using HPLC technology

HPLC technology was used in the laboratories of the College of Science /Al-Mustansiriya University in order to diagnose the estimated acid in the samples subject to the study.

2.7. Ascorbic acid Applications:

One of the distinctive properties of ascorbic acid is that it prevents browning, color, taste and flavor. Samples of bananas, potatoes, carrots, pickles such as (cucumber, turnips and cauliflower), jams like a (Raspberry, strawberry and fig).

3. Result and Discussion

In order to obtain ideal and accurate results, 10 ml of the banned vitamin was calibrated by dissolving two commercial vitamin tablets in 100 ml of distilled water, which contains 250 grams in each tablet. The recovery rate reached 80%, as shown in table (2) and figure (4).

Table 3. Shows the amount of Ascorbic acid (mg/100g & 10g) and average of iodine

No. Sample	Standard Deviation (SD)	Amount of Ascorbic acid (mg/100 g)	Amount of Ascorbic acid)g/10g)	Average size of iodine
1	3.1×10^{-3}	228.31	0.0228	**2.45**
2	1.8×10^{-3}	39.82	0.0040	**0.48**
3	0.27	79.27	0.0079	**0.93**
4	6.9×10^{-4}	8.45	0.0084	**0.09**
5	1.7×10^{-3}	130.50	0.0131	**1.92**
6	7.3×10^{-3}	20.12	0.0020	**0.17**
7	2.4×10^{-4}	89.23	0.0089	**1.02**
8	**2.8**	**127.76**	**0.0128**	**1.84**

Results showed ascorbic acid in guava fruit reached 228.3 mg/100g and apples recorded 8.4 mg/100 g. While in vegetables, it was 130 mg/100g in spinach, while in tomatoes it was 20 mg/100 g and the results were remarkably similar in a study[14].

The reason for the convergence of these results, which were within the ideal and permissible limits according to the joint expert committee between the World Health Organization (WHO) the Food and Agriculture Organization (FAO) and the Scientific Committee for Food and Drugs of the European Union (SCF) which set the permissible concentration for daily consumption of ascorbic acid at 100 mg/kg per day was the good storage period, the complete ripening of the fruit and its lack of exposure to oxidative conditions[2].

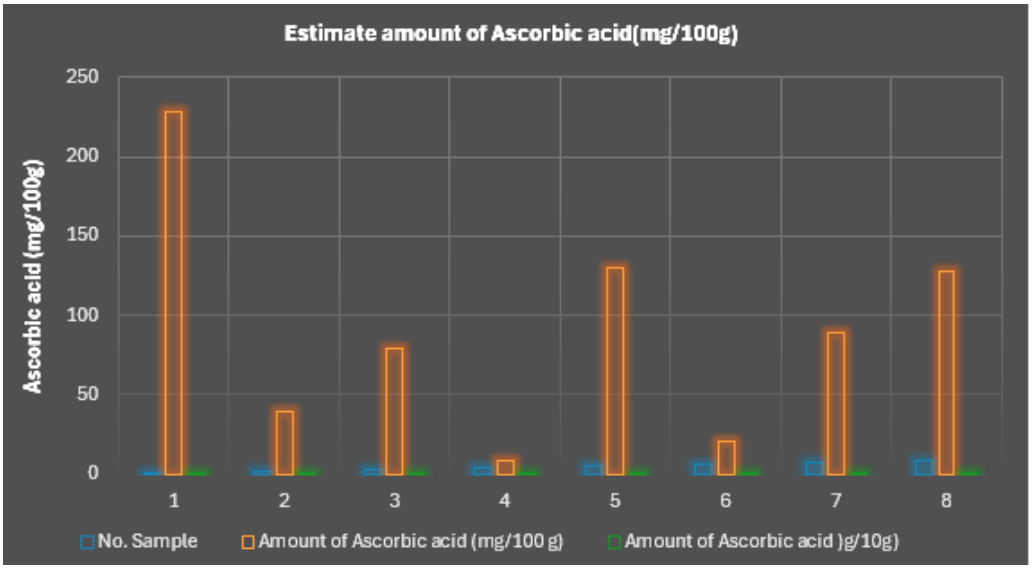

Figure 4. Estimate amount of Ascorbic acid in fruit and vegetable(mg/100gm)

The results in the table (4) show the concentration of ascorbic acid by spectroscopic and titration methods. Guava is a fruit and spinach is a vegetable. The highest content of Vitamin C was recorded for both methods, while apples and tomatoes recorded the lowest amount of Vitamin C in both methods. As shown in table (4) and figure (5).

Table 4. Concentration of Vitamin C by spectrophotometer and titration methods (mg/100g)

Sample codes	Sample	Spectrophotometer Method (530 nm)	Titration Method
1	Psidium guajava	52.33	**2.45**
2	Citrus limon	13.67	**0.48**
3	Ananas comosus	19.31	**0.93**
4	Malus domestica	8.25	**0.09**
5	Spinacia oleracea	40.88	**1.92**
6	Solanum lycopersicum	10.05	**0.17**
7	Brassica oleracea var. italica	22.85	**1.02**
8	**Capsicum annuum**	**34.72**	**1.84**

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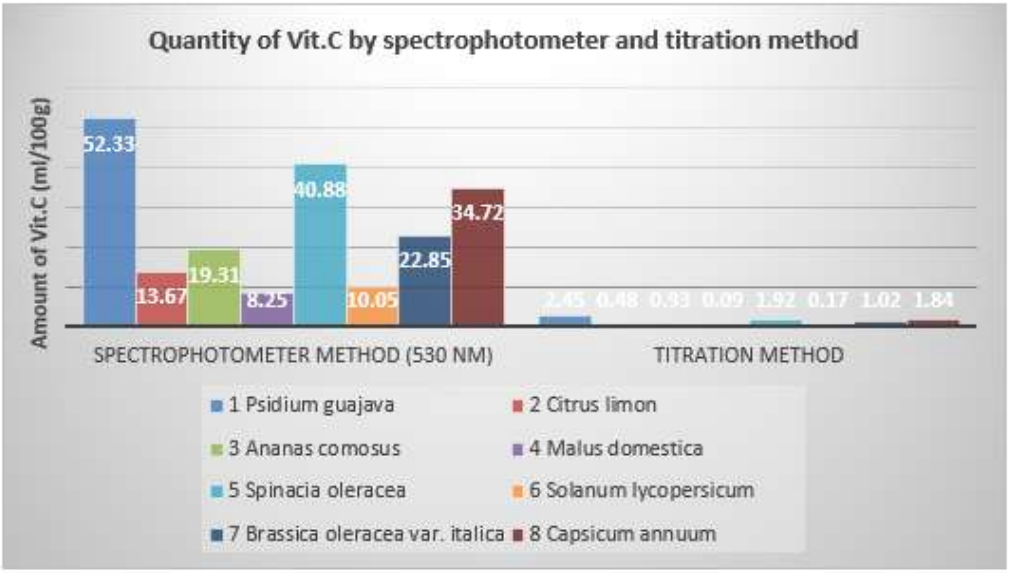

Figure 5. Amount of Vitamin C by spectrophotometer and titration methods(mg/100gm)

The important reason for the difference in the amounts of vitamin C in different foods is the type and quality of the food. If it is fresh, the amount of the vitamin is high compared to cooked, raw, and poor-quality foods[13]. There are also some factors that have a negative effect on the vitamin such as temperature, humidity, and direct light which may lead to the disintegration of the bonds in the functional structure of the acid contents which causes it to lose its ideal properties. Also, the shelf life of the product, such as long periods of time, leads to poor quality and the loss of many of the product's properties, including Vitamin C, nutritional value, color, taste and texture[12].

As shown in figure 6 Ascorbic acid (Vitamin C) was diagnosed by HPLC technology through calculating the retention time for eight samples at a wavelength of 280 nm from lowest to highest retention time over two tests. The first sample A showed a time of 5.159 and 5.233 minutes as the lowest detention time respectively, while the last sample H recorded a time of 17.004 and 17.140 minutes respectively, as the highest detention time. The reason for the difference is due to the quality of the food substance, the elements included in the composition and the purity of acid or Vitamin within the food[1].

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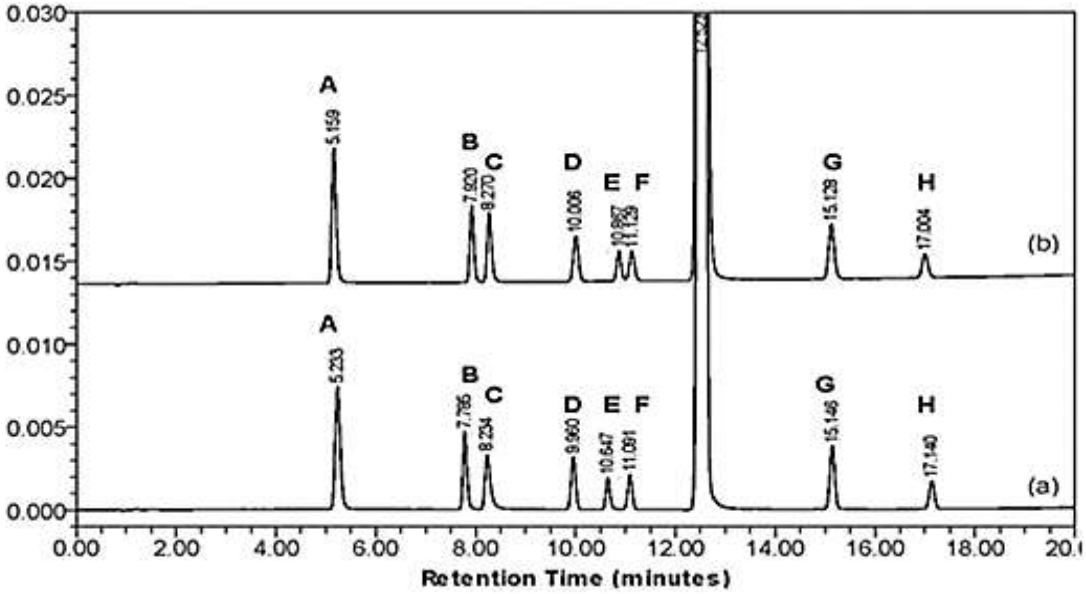

Figure 6. Diagnosis of Ascorbic acid (Vitamin C) using HPLC technology within two tests(a. standard / b. samples) from the lowest to the highest time for samples

As shown in the table (5) the concentration of Vitamin C 5mg/100 gm was the best concentration for the period 1-3 days when added to bananas through the Maillard reaction, color, taste and flavour, in contrast to the concentrations 10 and 15 mg/100 gm which recorded poor results in the potato and carrot test while the concentration 5 mg/100 gm Vitamin C recorded the best results for pickled cucumbers for a period of 1-7 days and was negative for the Maillard reaction It was good for color, taste and flavor, but the two concentrations 10 and 15 mg/100g had poor results as a result of the increased concentration. The concentration of 5 and 10 mg/100g of Vitamin C in the raspberry and strawberry jams test recorded the best concentration compared to the concentration 15 mg/100g in the fig jam[9].

Increasing the concentration leads to an increase in acidity, as well as a clear change in the bitter taste, which negatively affects the quality of the food and thus its acceptance and palatability by the consumer, due to the occurrence of chemical reactions within the food material, resulting in the emergence of organic compounds and ketones with undesirable odors and tastes[15].

Table 5. Concentration of Vitamin C by spectrophotometer and titration methods (mg/100g)

Sample	Concentrate of Vitamin C (mg/100g)	Time (Day)	Reaction	Color	Taste	Flavor
Bananas	5	1-3	Maillard	yellow	Good	**good**
Potatoes	10	1-3	Maillard	brown	Bad	**bad**
Carrot	15	1-3	Maillard	brown	Worst	**worst**
Pickled Cucumber	5	5-10	Maillard	green	Good	**good**
Pickled Turnips	10	5-10	Maillard	brown	Bad	**bad**
Pickled Cauliflower	15	5-10	Maillard	brown	Worst	**worst**
Raspberry jam	5	10-15	Maillard	Dark red	Good	**good**
Strawberry jam	10	10-15	Maillard	Red	Good	**good**
Fig jam	**15**	**10-15**	**Maillard**	**Natural**	**Bad**	**bad**

4. Conclusions :

The amount of vitamin C (Ascorbic acid) was measured in different samples of fresh and local fruits and vegetables such as guava, lemon, apple, pineapple, spinach, red pepper, broccoli and tomatoes. The quantities varied according to the type and quality of the product. Use different concentrations of the acid, 5, 10 and 15 mg/100g for varying periods between 1-15 days adding it to some various foods and observing the changes that occur. The concentration of 5 mg/100 g was the best in terms of stability of color, taste, flavor, and Maillard reaction compared to the rest of the concentrations.

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