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Determination of the total amount of vitamin C in some packaged juices from local companies in Libya

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Abstract

Fruit juices are known to contain many organic acids, the most important of which is ascorbic acid (vitamin C), because of its nutritional values and benefits for human health and disease prevention. These acids are found in fresh fruits from which the juice is made or are added as a nutritional supplement during manufacturing. This study aims to evaluate the quality of some local packaged juices available in the Libyan market for 5 local companies: (Al-Rayhan, Al-Mazraa, Judy, Al-Morouj, and Zain), with the results of previous studies and Libyan standards. in terms of the total amount of vitamin C, due to the large use of these packaged juices in the diet of adults and children in particular, as samples of packaged juices of different types (pineapple, orange, pear, guava, peaches, grapes, and others) available in various commercial markets and in a random manner after a storage period ranging between 6-10 months from the date of production, at a rate of 3 samples for each. It was analyzed spectrophotometrically using a molecular absorption spectrometer for visible and ultraviolet radiation (JENWAY 6405 UV/Vis. Spectrophotometer). The resultes shown that the amount of vitamin C in all samples was with in the limit and conformed to Libyan standards, and the best in vitamin C content was at Al-Rayhan Company and the least was at Zain Company.

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Keywords: Ascorbic acid, Vitamin C, packaged juices, spectrophotometric method, standard curve, (UV/Vis. Spectrophotometer)

تقدير الكمية الكلية لفيتامين ج في بعض العصائر المعلبة لشركات محلية في ليبيا

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الملخص

عصائر الفواكه معروفة باحتوائها على العديد من الاحماض العضوية وأهمها حمض الاسكوربيك (فيتامين ج)، تحتوي العصائر على قيم غذائية وفوائد عامة لصحة الانسان كالوقاية من الأمراض. توجد هذه الاحماض في الفاكهة الطازجة التي يصنع منها العصير أو تضاف كمكمل غذائي أثناء التصنيع، تهدف هذه الدراسة الى تقييم جودة بعض العصائر المعلبة المحلية والمتوفرة في السوق الليبي لعدد 5 شركات محلية هي (الريحان، المزرعة، جودي، المروج، وزين)، ومقارنتها مع المواصفات القياسية الليبية، من ناحية الكمية الكلية لفيتامين ج، وذلك نظرا للاستخدام الكبير لهذه العصائر المعلبة في النظام الغذائي للكبار والاطفال خصوصا. تم تجميع عينات العصائر المعلبة لأنواع مختلفة تشمل (أناناس، برتقال، كمثرى، جوافة، خوخ، عنب، وغيرها) المتوفرة في الأسواق التجارية الليبية و بطريقة عشوائية بعد فترة تخزين تراوحت ما بين 6–10 أشهر من تاريخ الإنتاج و بمعدل 3 عينات لكل منها، و تم تحليل العينات بالطريقة الطيفية بواسطة جهاز مطياف الامتصاصي الجزيئي للأشعة المرئية و الفوق بنفسجية (JENWAY 6405 UV/Vis. Spectrophotometer)، و قد تبين من خلال النتائج الكيميائية المتحصل عليها أن كمية فيتامين ج في جميع العينات كانت ضمن الحد المسموح به ومطابقة للمواصفات القياسية الليبية، و كان أفضلها في شركة الريحان و أقلها في شركة زين.

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الكلمات المفتاحية: حمض الاسكوربيك، فيتامين ج، العصائر المعلبة، الطريقة الطيفية، المنحنى القياسي، (*UV/Vis. Spectrophotometer*).

Introduction

Juice is a natural, non-fermentable but fermentable liquid of one or more types of healthy, ripe fruits or vegetables containing all or part of the pulp, free of seeds, peels and coarse fibers, and treated with one of the appropriate preservation methods if it is not intended for consumption immediately after its preparation, provided that it maintains its natural characteristics. Chemical and nutritional. In this case, the phrase “fruit juice prepared from concentrated juice” must be mentioned, and the word “fruits” must be replaced with the names of the fruit or concentrated fruits from which the juice is made [1].

packaged juices can be classified into nectar and drinks. Nectar means fruit pulp juice, to which an appropriate amount of sugar solution and a suitable organic acid such as citric acid or malic acid are added. Ascorbic acid may be added to it as an antioxidant, and it is preserved so that the resulting product, i.e. nectar, becomes ready for consumption without the need for any desalination, acidification, or dilution with water [1], [2]. While drink **is a** fruit juice filtered from peels, seeds, and coarse fibers, not fermented, to which an appropriate amount of organic acid and an amount of sugar are added until the sugar concentration in it reaches between 40-60%, and it is preserved by one of the appropriate preservation methods such as pasteurization, freezing, or adding chemical preservatives. Therefore when consumed, it must be diluted with an appropriate amount of water so that the sugar concentration in it reaches the required level of acceptable sweetness desired by the consumer, which is between 10-13%. The drink is made from many fruit juices, for example orange, lemon, raspberry, strawberry, and pomegranate [1], [2].

Human health is very important for our survival and vitamins can help us maintain a healthy diet. They serve as essential components of specific coenzymes involved in metabolism and other specialized activities. Vitamin C is an important vitamin for human nutrition

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that is provided by fruits and vegetables. Commonly known as ascorbic acid, ascorbic acid (AA) is the main biologically active form of vitamin C and is a powerful reducing agent that accelerates hydroxylation reactions in several ways. It is also a cofactor in the biosynthesis of collagen, and is essential for the optimal functioning of many enzymes. Including the enzymes prolinehydroxylase and lysinehydroxylase [3], [4], [5]. Ascorbic acid, as shown in Figure (1), is a white powder, its chemical formula is $C_6H_8O_6$, molar mass is 176.12 g/mole, density is 1.65 g/cm^3, melting point is 190-192°C, and it is a weak acid that exists in the form of two compounds, one of which can be transformed into the other form. They are L-ascorbic acid, which is considered a strong reducing agent, and L-dehydroascorbic acid, which is considered an oxidized ascorbic acid derivative, as in Figure (2). The ascorbate ion is the active moiety in vitamin C, and commercial vitamin C is considered a mixture of ascorbic acid. and sodium ascorbate [6]. Many animals and plants can biosynthesise ascorbic acid in their bodies from glucose, but most mammals, including humans and a few other animals (such as monkeys, chimpanzees, and guinea pigs), cannot synthesize it, as they suffer from a deficiency of the L-enzyme. Therefore, gulonolactone oxidase is not created in their tissues, and therefore they depend on external sources to obtain ascorbic acid (vitamin C), which can be plant sources, including fruits and vegetables, or animal sources, which include some meat and fish, as well as various types of animal milk. and pharmaceuticals [7],[8].

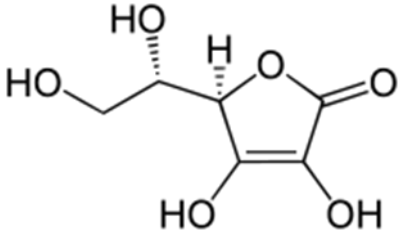

Figure 1 Structural formula of ascorbic acid

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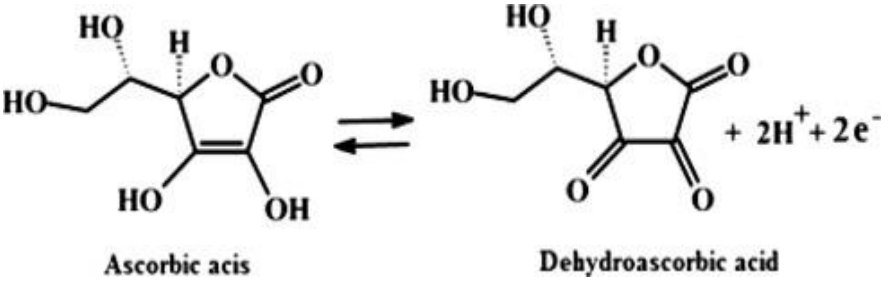

Figure 2 Forms of ascorbic acid

Vitamin C is sensitive to heat, light and storage, and it can be partially or completely destroyed in food due to storage or overcooking, and refrigeration helps reduce its loss from food [9], [10], [11], [12], [13],[14]. The importance of vitamin C lies in that it helps in the manufacture of collagen, which is important for the building blocks of the skin, cartilage, tendons, ligaments, and blood vessels. It has an important role in healing wounds and maintaining bones and teeth. It works as an antioxidant, as it oxidizes itself to protect other nutrients from oxidation[3], [15], [16]. Many researches and studies indicate that the content of vitamin C is linked to lower levels of cardiovascular disease and blood pressure. Vitamin C deficiency also affects many physiological systems and causes bleeding gums, loss of hair and teeth, and pain and swelling in the joints. The vitamin C deficiency of rare in developed countries [17], [18], [19],[20]. The recommended amount of vitamin C is 60 mg/day or more for men and women, and it is higher for smokers than for non-smokers as shown in **Table 1**, given that smokers are exposed to increased oxidative stress resulting from toxins contained in cigarettes, and they generally have low concentrations of vitamin C in the blood [20],[21]. Vitamin C is considered a low-toxic compound, but in high doses, more than 2000 mg per day, it can cause diarrhea, gas, and stomach disorders [20], [21].

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Table 1 A summary of the recommended amounts of vitamin C according to the World Health Organization and other international organizations, and Libyan standards [20], [21], [22], [23]

Age stage	*Age*	*Males (mg/day)*	*Females (mg/day)*
Babies	0-6 months	20	20
Children	1-3 years	15	15
Children	4-8 years	25	25
Children	9-13 years	45	45
Teenagers	14-18 years	75	65
Adults	19 years and older	90	75
Smokers	19 years and over	125	110

Many studies were conducted previously on estimating the amount of vitamin C in juices and soft drinks. A study was conducted in Iraq [24] which the volume of carbon dioxide gas, total solids, ash, moisture, organic matter, acidity, citric acid, vitamin C, caffeine, Sodium chloride and some mineral elements in 49 samples of juices. The results of the study were within the permissible limits in some variables, some of which were higher than the permissible limit. Vitamin C was also estimated in Romania [25], and the effort method was used for estimation, it was found that the vitamin content of orange juice was 29.39 mg/100mL, while lemon juice contained 33.97 mg/100mL, and Fanta lemonade contained 10.21 mg/100mL of vitamin C. In Nigeria[26] it was also found that the vitamin C content of lemon, orange, and grape juice reached 305.75 mg/L, 612.15 mg/L and 454.57 mg/L, respectively. Another study was conducted in Romania [27] to estimate vitamin C in juices. Pulse voltage measurement was used for estimation in this study. The study found that the amount of the vitamin ranged from 6.83 mg/100mL in Fanta to 54.74 mg/100mL in lemon juice, another study was also conducted in Misurata, Libya [28], to estimate the contents of some samples of local juices and drinks from the markets of Misurata city. The percentage of vitamin C ranged between 0.023 and 0.038%, a study was also conducted using a simple UV spectroscopic method to estimate total vitamin C (ascorbic acid + dehydroascorbic acid) in different fruits and vegetables in the Koya

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region of the Kurdistan Region [29], the spectrophotometric method involved the oxidation of ascorbic acid to dehydroascorbic acid by bromine water in the presence of acetic acid. After coupling with 2,4-dinitrophenylhydrazine at 37°C for approximately three hours, the solution was treated with 85% H_2SO_4 to produce the red color compound. The absorbance was then measured spectrophotometrically at 280 nm. The vitamin C content was 1.868 to 51.74 mg/10 g in fruits and 0.841 to 17.416 mg/10 g in vegetables. The current study aims to estimate the amount of vitamin C in different samples of various types of packaged juices from five local companies available in commercial stores in Libya, and to evaluate their quality by comparing them with the Libyan standards, and to know their value in the diet of children and adults. The content of vitamin C and its health benefits.

Methodology

Sample collection

Samples of packaged juices of different types (orange, peach, pear, guava, grape, and others) were collected from five different local companies available in the Libyan market (Al-Rayhan, Al Mazraa, Judy, Al Morouj, and Zain) from different stores in sabratha city in a random manner after a storage period. They ranged between 6-10 months from the date of production and an average of 3 samples for each.

Analysis method

It is possible to estimate the vitamin C content in packaged fruit juices by several methods, including direct titration with iodine solution [30], [31],[32], titration with dichloro-lindophenol solution, and the use of capillary electrode with UV-VIS and chromatographic analysis [HPLC] [33], [34], [35], [36] and others. However, in this study the spectroscopic method; wasemployed using the JENWAY 6405UV/Vis molecular absorption spectrometer for visible and ultraviolet radiation Spectrophotometer due to its speed, simplicity, and availability in most laboratories, in addition to its accurate results[37] [38], [39], [40],[41], [42], [43]. UV-Vis Spectroscopy (or Spectrophotometry) is a quantitative

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technique used to measure how much a chemical substance absorbs light, This is done by measuring the intensity of light that passes through a sample with respect to the intensity of light through a reference sample or blank. This technique can be used for multiple sample types including liquids, solids, thin-films and glass.

Materials

- Bromine water 5%.
- Thiourea solution 10%.
- A solution (metaphosphoric acid 5% - acetic acid 10%), which is prepared by dissolving 15 g of solid metaphosphoric acid in a mixture of 40 ml of glacial acetic acid and 450 ml of distilled water, then the solution is filtered and stored in 500 ml Standard volumetric flask.
- 2,4-dinitrophenylhydrazine solution.
- Sulfuric acid 85%.
- Glacial acetic acid 10%.
- Standard solution of vitamin C (ascorbic acid).
- Samples of the various packaged juices mentioned above.
- Distilled water.

Work steps

Preparation of the stock solution of vitamin C

A basic vitamin C solution (stuck solution) with a concentration of 500ppm was prepared by dissolving 0.05 g of standard ascorbic acid crystals in a little distilled water, then transferring the resulting solution to a standard 100 mL standard volumetric flask and fill it with distilled water to the mark.

Preparation a series of standard solutions

A six standard solutions (standard solution series) were prepared at concentrations of (5, 10, 15, 20, 25, 30) ppm from the stock solution, through the dilution rule, in 100 mL standard volumetric flask, and 6 drops of bromine water were added to each standard volumetric flask. Stir well to ensure that the ascorbic acid (vitamin C) is oxidized to dehydroascorbic acid, and then add 6 drops of thiourea solution to get rid of excess bromine water and obtain a clear solution. Then add 1 mL of glacial acetic acid and 1 mL of 2 ,

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4 -dinitrophenylhydrazine solution to all the standard volumetric flasks, then all the previously prepared standard flasks were placed in a water bath at 37°C for 3 hours to complete the reaction (thermostatic reaction), then all the standard flasks were cooled in an ice bath for half an hour, then finally 10 mL of 85% concentrated sulfuric acid was added to all the standard flasks, then all were fill them with distilled water to the mark.

Determination of the maximum absorption wavelength λ_{max}

The maximum absorption wavelength λ_{max} of the colored complex was determined using a JENWAY 6405UV/Vis device. Spectrophotometer for the standard solution of 15ppm, in order to avoid deviation from the straight line relationship of Beer-Lambert law , so it was (λ_{max} = 490 nm).

Preparation of the standard curve for a vitamin C standard solutions series

The standard curve for a series of standard solutions of vitamin C at the maximum wavelength λ_{max} was prepared by plotting the values of concentrations versus absorption as in **Table (2)**.

Preparation of different packaged juice samples solutions and measure their absorption at the maximum absorption wavelength (λ_{max} = 490 nm).

Sample solutions were prepared by transferring 4 mL of each juice sample into a 20 mL standard volumetric flask, then fill them to the mark with a solution (5% metaphosphoric acid - 10% acetic acid), then transferring the resulting solutions to a 100 mL standard volumetric flask and adding 6 drops of bromine water while stirring, then 6 drops of thiourea solution and the same additions were added with the same steps were followed with standard solutions, then the absorbance of the packaged juices samples solutions was measured at the same wavelength (λ_{max} = 490 nm) against the Plank.

Calculations

- The straight line equation obtained from the standard curve in **Figure (3)** is as follows: **Y=0.0164X-0.0054**

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- The amount of vitamin C in the sample solutions was calculated in PPm (mg/1000ml) according to the following law: **Ascorbic acid in sample solution (PPm) = (Abs. – 0.0054) / 0.0164**
- The amount of vitamin C in the sample solutions was calculated in units (mg/100ml) using the dilution law. The results were as in **Tables (3), (4), (5), (6), (7)** and **(8)**.

•

Results

Calibration curve results

Table 2 Standard curve results

Concentration(PPm)	Absorption
Plank	0
5	0.100
10	0.180
15	0.230
20	0.321
25	0.423
30	0.501

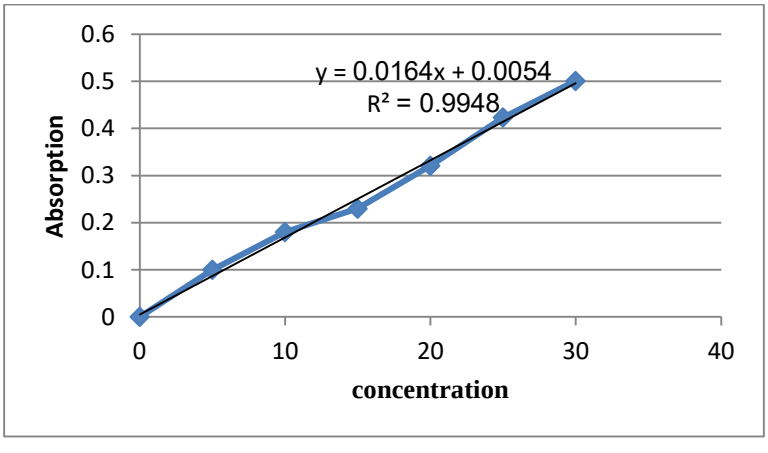

Figure 3 Standard curve of vitamin C at λ_{max} 490 nm

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Sample analysis results

Table 3 Quantities of vitamin C calculated by the chosen method for the studied packaged juice samples of Al-Rayhan Company

Type of packaged juice	*The amount of vitamin C listed on the Package (mg/100mL)	Calculated amount of Vitamin C (mg/100mL)
Grapes	10	7.56 ± 0.09
Orange	35	28.55 ± 0.02
Pineapple	30	22.08 ±0.18
Pears	10	8.27 ±0.00
Guava	10	8.51 ±0.05
Lemon with mint	25	21.0 ±0.13
Peach	10	9.09 ±0.10

*This quantity was found only for Al-Rayhan Company.

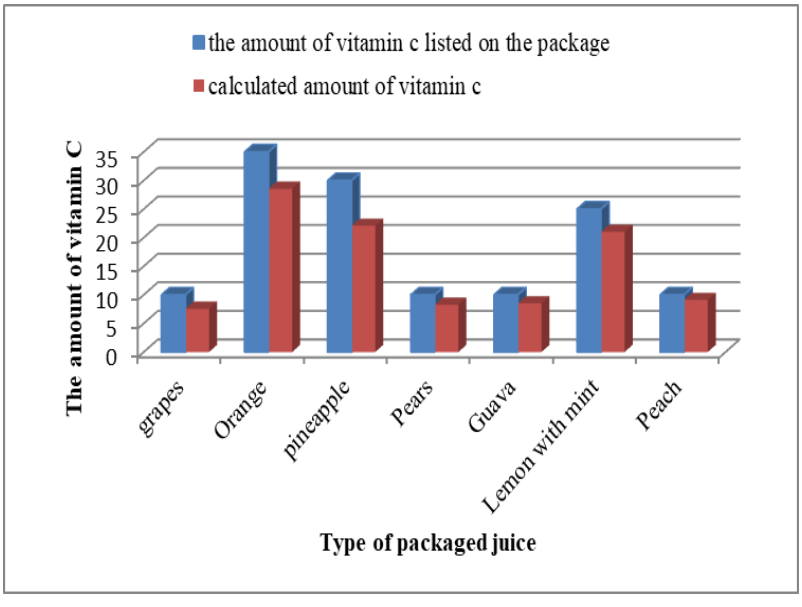

Figure 4 The amount of vitamin C calculated by the chosen method and that listed on the package as mg/100ml for Al-Rayhan Company

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Table 4 Quantities of vitamin C calculated by the chosen method for the studied packaged juice samples of Al-Morouj Company

Type of packaged juice	Calculated amount of Vitamin C (mg/100mL)
Peach	8.56 ± 0.06
Orange	30.55 ± 0.12
Guava	20.08 ± 0.09
Pineapple	8.27±0.17
Grapes	8.51± 0.03
Pears	8.46± 0.00

Table 5 Quantities of vitamin C calculated by the chosen method for the studied packaged juice samples from judy Company

Type of packaged juice	Calculated amount of Vitamin C (mg/100ml)
Peach	7.41±0.13
Pineapple	13.45±0.06
Pears	14.51±0.00
Grapes	8.30±0.12
Guava	8.05±0.09

Table 6 Quantities of Vitamin C calculated by the chosen method for the studied packaged juice samples of Al Mazraa Company

Type of packaged juice	Calculated amount of Vitamin C (mg/100ml)
Peach	9.23±0.13
Orange	24.56±0.10
Grapes	8.26±0.07
Pineapple	15.43±0.11
Pears	8.22±0.00
Guava	10.56±0.07

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Table 7 Quantities of Vitamin C calculated by the chosen method for the studied packaged juice samples of Zain Company

Type of packaged juice	Calculated amount of Vitamin C (mg/100ml)
Grapes	6.25±0.15
Pears	7.34±0.00
Cocktail	15.10±0.11
Mango	9.03±0.10
Peach	7.22±0.10
Guava	9.54±0.13
Pineapple	8.15±0.09

Table 8 Comparison of results of estimating vitamin C content for the same type of juice from different companies

Type of packaged juice	Vitamin C content (mg/100ml) for different local companies				
	Al-Rayhan**(1)**	Al-Morouj**(2)**	Judy**(3)**	Al-Mazraa**(4)**	Zain**(5)**
Pineapple	22.08± 0.18	8.27± 0.17	13.45±0.06	15.43±0.11	8.15±0.09
Grapes	7.56± 0.09	8.51± 0.03	8.30±0.12	8.26±0.07	6.25±0.15
Peach	9.09± 0.10	8.56± 0.06	7.41±0.13	9.23±0.13	7.22±0.10
Pears	8.27± 0.00	8.46± 0.00	14.51±0.00	8.22±0.00	7.34±0.00
Guava	8.51± 0.05	20.08± 0.09	8.05±0.09	10.56±0.07	9.54±0.13

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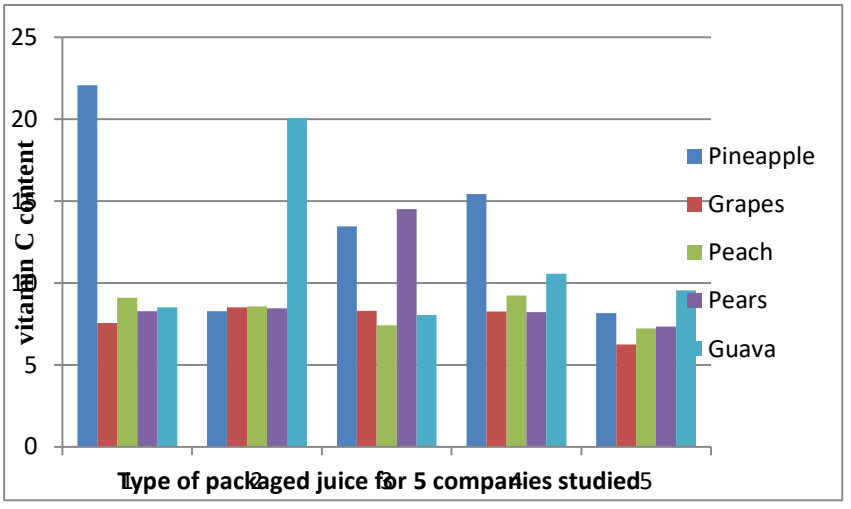

Figure 5 Comparison of the results of estimating the vitamin C content mg/100ml for different types of packaged juices for 5 studied companies

Discussion of results

The total amount of vitamin C in the packaged juices samples studied were all good and conformed to Libyan standards and international organizations, including the World Health Organization. Al-Rayhan Company's samples were the highest in vitamin C concentrations (28.55mg/100ml in Orange juice, 22.8mg/100ml in Pineapple juice and 21.0mg/100ml in Lemon with mint) as shown in **Table (3)** and **Figure (4)**, and the juices samples that least in vitamin C concentrations were for Zain Company's samples (6.25mg/100ml in Grapes juice, 7.34mg/100ml in Pears juice and 7.22mg/100ml in Peach juice as shown in **Table (7)**.

We noticed from **Table (3)** and **Figure (4)** that there is a slight difference between the amount of vitamin C recorded on the packages and that calculated with chosen method for Al-Rayhan Company juices. In all cases, the calculated amount was small compared to the recorded amount, meaning that the vitamin is unstable, and this is because it is sensitive and Its quantity may be affected by several factors, as mentioned previously in introduction. From **Table (8)** and **Figure (5)**, we noticed that the largest amount

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of vitamin C was in the pineapple juice of Al-Rayhan Company 22.08mg/100mL, then the guava juice of the Al-Morouj Company 20.08mg/100ml, and the least amount was in the juice. Grapes for Zain 6.25 mg/100L.

Conclusion

Some fruit packaged juices available locally in commercial markets in Libya are good sources of the vitamin C under study as its quantity in some samples reached 22.08 mg/100ml and 14.51 mg/100ml and they were generally consistent with the quantities recommended by international health organizations, as previously mentioned. That is why all the studied juices samples were good quality and had a good amount of vitamin C. Therefore، they can be included in the diet of children and adults.

Acknowledgments

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