

Determination of Vitamin C (Ascorbic Acid) Concentration by Redox Titration in Some Fruits Cultivated and Sold in Enugu Metropolis of Nigeria

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Abstract

This work was done to determine the concentration of Vitamin C in some mature ripped fruits and canned fruit juice sold in some selected areas within Enugu metropolis in order to sensitize the public on the importance and the needed correct quantity of Vitamin C which the body needs for proper well-being. Some ripe raw fruits and packaged fruit juice were bought from Topland and Garriki markets in Enugu State of Nigeria and the determination of Vitamin C concentration by Redox titration was carried out on them. Aliquot of each sample (20ml) solution were prepared and transferred into a conical flask (250ml). Distilled water (150ml) was added and starch indicator solution (1ml) was also added. Each sample solution was titrated with iodine solution (0.005M). The endpoint of the titration was identified as the first distinct trace of a dark blue-black colour due to the starch-iodine complex. Titration was repeated with further aliquots of sample solution until concordant results (titres agreeing within 0.1 ml) were obtained and the results of the analysis given as: Pineapple Fruit 122.0mg/100ml; Orange Fruit 102.5mg/100ml; Vitamin C tablet 100.73mg/100ml; Ribena Juice 52.98mg/100ml; Cucumber Fruit 37.21mg/100ml; Happy Hour Juice 29.0mg/100ml; Orange Juice 20.1mg/100ml; Lucozade Boost Juice 18.60mg/100ml; Pineapple Juice 12.67mg/100ml. The result of the analysis can help anyone who wishes to take Ascorbic acid (Vitamin C), in making a proper decision in order to meet up with the World Health

Organization recommendation of atleast 45 milligrams per day; 300 milligrams per week for healthy living.

Keywords: Ascorbic acid, Redox titration, concentration, World Health Organization

Introduction

Vitamins are a group of small molecular compounds that are essential nutrients in many multi-cellular organisms, and humans in particular. L-Ascorbic acid, first isolated as a pure substance by Albert Azent-Gyorgi and Charles Kingin (Romero, et al., 1992), is an anti-oxidant and free radical scavenger found ubiquitously in fruit and vegetables, such as citrus fruits (oranges, lemons, limes, tangerines, etc.), melons, tomatoes, peppers, broccoli, and green leafy vegetables such as spinach, potatoes and turnips. Its quantitative determination is especially important in the production of wine, beer, milk, soft drinks and fruit juices, where it can be a quality indicator (Gerrior & Zizza, 1994). It plays essential roles in the human diet and its importance to growth and repair of tissues in all parts of human body cannot be over-emphasised. It is necessary in collagen formation, an important skin proteins, scar tissue, tendons, ligaments, and blood vessels (Wilson, Baguley, Wall, Jameson, & Findlay, 2014). Vitamin C is essential for the healing of wounds, and for the repair and maintenance of cartilage, bones and teeth¹³. Since Vitamin C is easily oxidized, storage and cooking in air leads to the eventual oxidation of Vitamin C by oxygen in the atmosphere (Sidibé, et al, 1996).

Vitamin C is the most familiar of all the nutrients because of its widespread use as a dietary supplement. Vitamin C is very critical to human beings, and all mammals can use their own cells to make Vitamin C. Vitamin C is needed for the growth and repair of tissues in our body and because the body does not store Vitamin C in its tissue, we need to consume it on regular basis (Combs, 2001). Vitamin C improves the absorption of iron from plant based foods and helps to strengthen our immune system to work properly protecting us from diseases (WHO, 2004).

Signs of Vitamin C Deficiency

The signs of Vitamin C deficiency include: Dry and splitting hair; Gingivitis or inflammation of gums, bleeding of gums, rough, dry and scaly skin; reduction of wound healing rate, easy bruising, nose bleed; decreased ability to ward off infections (WHO Model List of Essential Medicines (19th List), 2015). The most severe form of Vitamin C deficiency is scurvy. Scurvy leads to the formation of liver spots on the skin, spongy gums and bleeding from mucous membranes. A person with this ailment looks pale, feels depressed and is partially immobilised. In advanced scurvy there are open wounds which subsequently lead to loss of teeth and eventually death (WHO Model List of Essential Medicines (19th List); 2015). Also, low levels of Vitamin C may lead to high blood pressure, gallbladder disease, stroke, cancer and atherosclerosis, the build-up of plaque that can lead to heart attack and stroke.

Health Benefits of Vitamin C

Immunity

A healthy dose of Vitamin C protects our body from infections and maintains healthy bones and teeth. It also improves our body's wound healing ability and boosts our immune system against bacteria, viruses and other infections. Not only that it is a well-known component of our immune system, it is also necessary for collagen formation, the main structural protein found in connective tissue. Vitamin C taken orally helps wound to heal faster and better. Applied topically, it protects the skin from free radical damage by exposure to UV rays(Boudrant,1990).

Hypertension

Vitamin C has been shown to lower blood pressure and decreases the probability of hypertension, as well as serious health problems that may accompany high blood pressure(WHO; 2004).

Lead Toxicity

Vitamin C helps to lower the level of lead in our blood drastically. Lead toxicity can lead to various behavioural and developmental problems, especially in children living in urban areas. This leads to learning disabilities, lowered Intelligence Quotient (IQ) and stunted growth in children. Adults may suffer from kidney damage and high blood pressure owing to lead toxicity (Gerrior,& Zizza, 1994).

Weight Loss

Individuals with adequate Vitamin C status oxidize 30% more fat during a moderate exercise than individuals with low Vitamin C status; thus Vitamin C depleted individuals are more resistant to fat mass loss. That means that one gets the most out of one's fat-burning workout if one's Vitamin C levels are high. Vitamin C fruits and juices are proven to help people lose fat and maintain healthy weight. Popular dieticians and nutritionists always include Vitamin C fruits and vegetables in diet charts because there is scientific evidence that shows that eating Vitamin C rich foods on a regular basis helps to lose weight significantly(Stationery Office London Scientific Advisory Committee on Nutrition, 2011). Doctors have found that eating Vitamin C fruits helps the hormone called insulin to keep the body's blood sugar level from getting too high (hyperglycemia) in order to reduce fat(WHO, 2004).

Stress

Vitamin C helps the body to deal with stress by reducing the elevated levels of stress, hormone and cortisol(Levine, Dhariwal, Welch, Wang,& Park, 1995).

Cataracts

The eye pupils require Vitamin C to function properly. Vitamin C deficiency can lead to cataracts, where the lens becomes increasingly opaque, causing blurry vision and leads to blindness in adults (WHO Model Formulary, 2008). High intake of Vitamin C fights against cataracts and increases the amount of blood flow to the eye. 1000 mg Vitamin C per day stops cataracts in their tracks and improves vision.

Dietary Recommendations of Vitamin C by National Agencies

In 2000, the North American Dietary Reference Intake chapter on Vitamin C updated the Recommended Dietary Allowance (RDA) to 90 milligrams per day for adult men and 75 mg/day for adult women (Stationery Office London Scientific Advisory Committee on Nutrition, 2011).

The U.S. National Centre for Health Statistics conducts biannual National Health and Nutrition Examination Survey (NHANES) to assess the health and nutritional status of adults and children in the United States.

The 2013-2014 survey (Wilson, et al, 2014) reported that for adults aged 20 years and older, men consumed an average 83.3 mg/day and women 75.1 mg/day. This means, from the findings, that half the women and more than half the men are not consuming the RDA for Vitamin C. The same survey stated that about 30% of adults reported they consumed a Vitamin C dietary supplement or a multi-vitamin/mineral supplement that included Vitamin C, and that for these people total consumption was between 300 and 400 mg/day (Wilson, et al., 2014).

In 2000, the Institute of Medicine of the U.S. National Academy of Sciences set a tolerable upper intake level (UL) for adults, as 2,000 mg/day. The amount was chosen because, as they said, human trials had reported diarrhoea and other gastrointestinal disturbances at intakes of greater than 3,000 mg/day (WHO Model Formulary, 2008). This was the Lowest-Observed-Adverse-Effect Level (LOAEL), meaning that other adverse effects were observed at higher intakes. The European Food Safety Authority (EFSA) reviewed the safety question in 2006 and reached the conclusion that there was not sufficient evidence to set a UL for Vitamin C.

For U.S. food and dietary supplement labelling purposes, the amount in a serving is expressed as a percent of Daily Value (%DV). For Vitamin C labelling purposes, 100% of the Daily Value was 60 mg, but as at May 27, 2016, it was revised to 90 mg to bring it into agreement with the RDA. A table of the old and new adult Daily Values is provided as Reference Daily Intake. Food and supplement companies are expected to have complied with the change because they were ordered to comply before January 1, 2020.

Materials and Method

Preparation of Iodine Solution (0.005M)

Potassium Iodide, KI (2.00g) and Iodine Crystal (1.30g) were accurately and carefully weighed using analytical weight balance respectively and distilled water (20ml) was added to it, shaken and allowed to dissolve in a 250ml Beaker. The Iodine solution formed was transferred to an empty 1 litre volumetric flask, making sure that all traces of the solution was rinsed into the volumetric flask; the solution was made up with distilled water to the 1000ml mark on the flask.

Preparation of Starch Indicator solution (0.5%)

Soluble Starch (0.25g) was accurately weighed with analytical weight balance and transferred to a 250ml conical flask and 50ml of distilled water was added. The Solution was heated, with periodic stirring, at 79⁰C for 5 minutes. Care was taken not to exceed the stated temperature. The solution was allowed to cool to room temperature before use.

Preparation of Food/Drink Sample

Industrial packaged fruit juices (Orange, Pineapple, Ribena, Lucozade Boost, and Happy Hour) were strained through cheese cloth to prevent entry of unseen pulp or seeds.

For analysis of Vitamin C tablets (EMZOR and M&B tablets), a single tablet of Vitamin C was dissolved in 200ml of distilled water (in a volumetric flask).

Juice squeezed from ripe fresh fruit (Orange, Pineapple, and Cucumber) was strained through cheese cloth in order to remove seeds and pulp which might block pipettes.

Sample of fresh ripe fruit (100g) was blended in a food processor together with 50ml of distilled water. After blending, the pulp was strained through cheese cloth, and was washed into a volumetric flask (Romero, et al, 1992).

Titration Procedure

Aliquot of the sample solution (20ml) prepared above was transferred into an empty conical flask (250ml), distilled water (150ml) was added and Starch indicator solution (1ml) was added.

Sample was titrated with Iodine solution (0.005M). The endpoint of the titration was identified as the first distinct trace of a dark blue-black colour due to the starch-iodine complex.

Titration were repeated with further aliquots of sample solution until concordant results (titres agreeing within 0.1 ml) were obtained.

The titrations were done in triplicates and the mean values (ml) with standard deviations recorded as Mean±SD: Orange Fruit Sample = 11.57±0.3; Orange Juice Sample = 2.27±0.1; Pineapple Fruit Sample = 13.77±0.3; Pineapple Juice Sample = 1.43±0.2; Cucumber Fruit Sample = 4.20±0.1; Ribena Juice Sample = 5.98±0.2; Lucozade Boost Juice Sample = 2.10±0.1; Happy Hour Juice Sample = 3.27±0.5; Vitamin C Tablet Sample = 11.37±0.5.

Calculation of the Vitamin C Concentration in the Samples

Note: 1ml of Iodine (I₂) liberates 0.00886g of Vitamin C

For each sample

1ml of I₂ = 0.00886g of Vitamin C

R (ml) of I₂ =?

= R x 0.00886 = Y g of Vitamin C

But 0.001g = 1mg

Therefore, Y / 0.001 = Z mg/100ml

Note: **R** is the Titre value.

Y is the mass of Vitamin C.

Z is the Concentration of Ascorbic acid in milligram per 100ml sample.

Results and Discussion

From the results of the determinations, it is observed that the raw ripe fruits and packaged fruit juice each has the following concentration:

- Pineapple Fruit: 122.0mg/100ml;
- Orange Fruit: 102.5mg/100ml;
- Vitamin C Tablet: 100.73mg/100ml;
- Ribena Juice: 52.98mg/100ml;
- Cucumber Fruit: 37.21mg/100ml;
- Happy Hour Juice: 29.0mg/100ml;
- Orange Juice: 20.1mg/100ml;
- Lucozade Boost Juice: 18.60mg/100ml; and
- Pineapple Juice: 12.67mg/100ml.

The analysis shows that the fruit with the highest concentration of Ascorbic Acid is Pineapple Fruit followed by Orange Fruit while the least in concentration is Pineapple Juice. The results got from the determination of Vitamin C in fresh ripe fruits and canned fruit juices using the Microfluorometric Method, as reported in (Vanderslice, Higgs, Hayes, & Block, 1990) and Liquid Chromatography (Romero, et al., 1992), give a slight higher difference in values, because their methods of determining the total Vitamin C values are different, when compared to the Titrimetric procedure (Levine, et al., 1999) used in this study, which gives only the ascorbic acid values. The titrimetric method was used because it is more cost effective than the other methods. Since most of

the fruits used are seasonal fruits, one may start asking how one would get the recommended Vitamin C for the body; say when those fruits are no more found in the farm. In such cases, it would be advisable to resort to Vitamin C tablet because it contains about 100.73mg of Vitamin C or think of the combination of any of the listed fruit juices that will sum up to a value greater than or up to 45mg/100ml as recommended by the World Health Organization.

Conclusion

The recommendations for Vitamin C intake by adults as set by various National Agencies (Levine, et al, 1999) go thus:

- 40 milligrams per day: the United Kingdom Food Standards Agency.
- 40 milligrams per day: India National Institute of Nutrition, Hyderabad.
- 45 milligrams per day; 300 milligrams per week: The World Health Organization.
- 80 milligrams per day: The European Commission Council on nutrition labelling.
- 90 mg/day (males) and 75 mg/day (females): Health Canada 2007.
- 90 mg/day (males) and 75 mg/day (females): United States National Academy of Sciences.
- 100 milligrams per day: Japan National Institute of Health and Nutrition.

Looking at these recommendations, one can see that if one needs to maintain the recommended amount of Vitamin C, as stipulated by the World Health Organization (WHO), at least, the 45mg/day should be consumed.

Looking at the results got from the analysis, one can decide to be taking one tablet of Vitamin C daily or a can of Ribena Juice or a Pineapple Fruit or an Orange Fruit. If Happy Hour Juice, Lucozade Boost Juice, Cucumber fruit, Pineapple Juice, or Orange Juice has to be taken, then about two of any of them should be appropriate per day.

Recommendations

It is recommended that:

- ✓ Everybody is made to know the importance of taking fruits rich in Vitamin C because of the benefits derived from them.
- ✓ The government at all levels should make sure that fruits rich in Vitamin C be made available in most of the streets of all the states of the federation and should be cheap so that even the poor people can afford to buy it.
- ✓ Every family should form the habit of incorporating fruits in their meal so that it will give them all the necessary nutrients especially the C vitamin that boosts the immune system, so as to maintain healthy living.
- ✓ The Standard Organization of Nigeria (SON) and National Agency for Food, Drug Administration and Control (NAFDAC) should always make sure that each of the packaged or canned fruit juice produced and sold in Nigeria has up to the required concentration of Ascorbic Acid that the human body needs.

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