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# Effect of Temperature and Storage on Vitamin C Content in Fruits Juice

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## Abstract

The amount of Vitamin C content of some fresh fruit degradaded with time was investigated; Pineapple, Orange, Watermelon and Tomato stored under difference conditions. The juices from the samples were extracted and vitamin C content in the juice was analysed at room temperature, $40^0$C and at seven (7) days storage by indophenols titration method. The result revealed that vitamin C content is more in Pineapple juice compared to Orange, Watermelon and Tomato juice as shown in Table 1 in this order; 49.38 ±1.875, 39.75 ±1.000, 27.50±1.250 and 19.13 ±1.630 mg/100ml respectively. The fresh juice were boiled at $40^0$C and vitamin C content was determined. Result shows that; there is decreased in Vitamin C content after boiling as shown in Table 1; 26.25 ±1.750, 23.00 ±1.520, 16.70±1.420 and 10.83 ±2.600 mg/100ml respectively. The fresh juices were also stored at room temperature for 7 days and vitamin C content was determined. Result revealed that; there is loss of vitamin C content in fresh juice but not as much as the boiled juice Table1; 42.13 ±0.875, 31.50 ± 1.000, 21.63 ±1.625 and 15.63±0.625 mg/100ml respectively. The rate at which vitamin C is loss during storage depend on the type of storage methods employed.

## Keywords

Scurvy, Antioxidant, Indophenol Titration, Vitamin C in Fresh Fruits, Vegetables, Stability and Damaturu

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## 1. Introduction

Vitamin C a component of food, need by all animals especially Humans to prevent scurvy, a disease of the gums, bones and blood vessels and to increase the body's resistance to infection. Ascorbic acid acts as an antioxidant, a nutrient that chemically binds and neutralizes the tissue-damaging effects of substances in the environment known as free radicals. As a result, ascorbic acid is vital for the growth and maintenance of healthy bones, teeth, gums, ligaments and blood vessels. Because of its role in the formation of collagen, the body's major building protein, a central component of all body organs. Therefore, the determination of Vitamin C becomes increasingly important in areas such as Biochemistry, Pharmacology, and Nutrition. Vitamin C occurs naturally in many fruits and vegetables, particularly in tomatoes, citrus fruit, cantaloupe, broccoli, spinach, green peppers, cabbage, pineapple, melons, Mangoes and potatoes. The vitamin is easily destroyed by cooking or canning foods and by exposure to air and light. A healthy diet generally contains sufficient quantities of ascorbic acid, but the body requires more of the vitamin after serious injury, major surgery, burns and when exposed to extremes temperature. At risk for deficiency are smokers, women taking contraceptives containing the female sex hormone estrogens and people who live in cities with high levels of carbon monoxide from traffic? There is conflicting evidence that taking large doses of ascorbic acid will either prevent the common cold or reduce the severity of its symptoms. Ascorbic acid is an organic compound of carbon, hydrogen and oxygen. Pure ascorbic acid is a white solid and is made synthetically from the sugar dextrose. It is used both in vitamin supplements and as a food preservative.

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Vitamin C (Also referred to as L- ascorbic acid) is the Lactose -2,3,-dienol-L-gluconic acid and is an odourless, white solid having the chemical formula $C_6 H_8O_6$. Vitamin C is the L-enantiomic form of ascorbic acid which also encompasses the oxidation product of dehydroascorbic. It participates in numerous biochemical reactions, suggesting that vitamin C is important for body process from repair (Rickman et al, 2007). Vitamin C or L- ascorbic acid or L-ascorbate is an essential nutrient for humans and certain other animal species, in which it function as a vitamin. In living organisms, ascorbate is an antioxidant since it protects the body against oxidative stress (Schorah et al, 1996). Ascorbate (an ion of Ascorbic acid) is required for a range of essential metabolic reactions in all animals and plants. It is made internally by almost all organism; notably mammalian group exceptions are most or all of the order Chiroptera (Bats) and one of the two major primate suborder Anthropoidea.

Ascorbic acid, in turn, is essential for the activity of Prolylhydroxylase which convert proline residues in collagen to hydroxyproline.(Bethlehem, Campbell and Farrel, 2013). Analyses of the effect of Nigerian market storage conditions on ascorbic acid, titratable acidity and pH values of selected tetrapak packaged citrus fruit juice in Nigerian markets samples were carried out from 3 months to 10 months from the day of production. The results showed that there was gradual decrease in ascorbic acid contents and pH values with increase in storage period irrespective of the brands (Johnson, et al., 2013). The aim of the research work is determined the amount of vitamin C content in different fresh fruit and find out the rate of degradation with time and temperature.

## 2. History of Vitamin C

Vitamin C also known as ascorbic acid is a small carbohydrate molecule first identified in the 1920s by Albert Van SzentGyorgyi, who discovered that it was able to prevent and cure scurvy. Scurvy is a pathological life threatening condition suffered by people who do not have access to fruits or vegetables for long periods of time. A decade earlier, Kazimierz Funk had prepared a list of nutritional factors called vitamins, whose deficiencies cause severe diseases in humans. In his list, Funk used the Letter “C” to designate a factor still unidentified, but known to prevent scurvy. Later on, SzentGyorgyi and Haworth chemically identified C as Ascorbic acid, and named it so because one of the most popular drugs in human history.

Why is this molecule so well known? Apart from its deficiency causing scurvy in humans, vitamin C is also vitally important to other species. Neither animals nor plants can live without vitamin c and it is therefore suprising that some animals (some fishes and birds, and a few mammals, including guinea pigs and human) have lost the capability to produce it over the course of evolution.(Grano and Tullio, 2007).

## 3. Properties of Vitamin C

Ascorbic acid is a colourless and odourless crystalline substance, slightly sour in taste and optically active. Only the L-Isomer has antiscorbutic properties. It is soluble in water and alcohol but practically insoluble in chloroform, ether and light petroleum. It has a melting point about $190^0$C ($374^o$F) (with decomposition), boiling point about $553^0$C ($1027^0$F), density 1.694 g/cm$^3$, molar mass 176.12g/mole, $P^H$ 3 (5% sol), vapour density autoignition $660^0$C, reactivity O, stable under ordinary conditions. It is readily oxidized, particularly in the presence of Copper and Iron but not of Aluminum. It is for this reason that the foods cooked in copper utensils lose ascorbic acid quickly. This vitamin is also rapidly destroyed by alkalis but is fairly stable in weak acid solutions. Therefore, baking soda has a deleterious effect but steam cooking destroys very little amount of ascorbic acid. However, freezing has no detrimental effect on this vitamin. Citrus fruits and tomato juices may be canned with but little loss of ascorbic acid. On account of its easy oxidization nature, the ascorbic acid is a powerful reducing agent. The oxidative nature is applied locally by women across Nigeria for bleaching, washing and cleansing their jewelries (El-Ishaq and Abdullahi, 2010).

## 4. Biochemical Role of Vitamin C

It was noted in the early experiment of Holst and Frohlich and confirmed by many workers, that defective formation of connective tissue was a primary pathological feature of experimental scurvy, and at one time it was believed that this lesion could account for most of the known pathological sequelae of the disease. The potential hemorrhages, the breakdown of gum tissue and the impairment of wound repair tissue. Attempts to characterize the biochemical modus operandi of vitamin C in preventing scurvy, therefore, centered initially on the metabolism of collagen, the essential glycoprotein component responsible for imparting strength to connective tissue.

By the 1970s, there was suggestive evidence that the biochemical lesion was located in the hydroxylation of the proline and lysine components of the collagen polypeptide and that vitamin C had an essential role in the process. The hdroxylases involved in collagen biosynthesis (Prolyl-4-hydroxylase, prolyl-3- hydroxylase, and lysyl hydroxylase) require ferrous iron as a cofactor, and it appears that vitamin C, a powerful biological reductant, has an almost obligatory role in maintaining the ferrous iron in the reduced form. Thus emerged a simple and reduction role of vitamin C in preventing the emergence of the main clinical features of scurvy (Kiple, 2000).

Vitamin C also has specific roles in the copper-containing hydroxylases and the α-ketoglutarate-linked iron-containing hydroxylases. It also increases the activity of a number of other enzymes in vitro, although this is a non-enzymic effect as a result of its action as a reducing agent and oxygen radical quencher (Murray et al, 2012).

Vitamin C is for humans and other primates, the guinea pig, bats, Passeriformes birds, and most fishes and invertebrate; other animals synthesize it as an intermediate in the uronic acid pathway of glucose metabolism. In those species for which it is a vitamin, there is a block in the pathway as a result of absence of guluconolactone oxidase. Both ascorbic acid and dehydroascorbic acid have vitamin activity (Berg, Tymoczko, stryer and Clarke, 2002).

# 5. Excretion

The average half-life of ascorbic acid is believed to be between 16 and 20 days. Its half-life is inversely related to intake. The water-soluble properties of vitamin C lead to urinary excretion of the vitamin C Metabolites including dehydroascorbate (DHAA). Oxalic acid, 2-O-methyl ascorbate and 2- ketoascorbitol are also excreted from the body via the urinary system, the kidneys play a major role in vitamin C excretion and retention. Dehydroascorbate and ascorbic acid can be reabsorbed by the kidney tubules as long as body pool levels are equal to or less than 1500mg. levels within the body that are 1500mg or less will result in no urinary excretion of vitamin C. as levels increase above 1500mg the reabsorption efficiency of the kidneys decreases. Thus, body pools levels from 1500 to 3000mg relate to tissue saturation of the vitamin. Plasma ascorbate levels between 0.8 and 1.4 mg/dl are considered the renal threshold. Above these levels, vitamin C will be excreted rather than reabsorbed by the kidney (Dekker, Van Doornen and Kemper, 1998).

# 6. Methodology

## 6.1. Sample Collection and Preparation

Fresh fruits *Ananas comosus* (Pineapple), *Citrus sinesis* (Orange), *Citrllus lanatus* (Watermelon) and *Lycopersicum esculentum,* (Tomato) were purchased from Damaturu Central Market, Nigeria. The fruits were washed thoroughly with distilled water, and the juices were extracted manually using juice squeezer. Samples were filtered to remove pulp and seed and stored in already labeled plastic containers, till further use.

## 6.2. Determination of Vitamin C (Indophenol Titration Method)

CHEMICALS: Ascorbic acid, 2,6-Dichloroindophenol (DCIP), Metaphosphoric acid and Acetic acid are of analytical grade supplied by Sigma (Madrid, Spain).

50cm$^3$ un-concentrated juice was pipette into 100cm$^3$ volumetric flask. 25cm$^3$ of 20% acetic acid was added as stabilizing agent and was diluted to 100cm$^3$. 10cm$^3$ was pipette into a conical flask, and 2.5cm$^3$ acetone was added. It was titrated with 2, 6-Dichloroindophenol (DCIP) Standard solution. A faint pink colour that was persisted for about 15 seconds was observed. The amount of dye used in the titration was determined volumetrically and used in the calculation of the vitamin C content mg/ 100ml in the fruit samples. (AOAC, 2006)

## 6.3. Statistical Analysis

The result were expressed as the mean of triplicate ± standard error of the mean (SEM) at α = 0.05 (α=5%).

# 7. Results

Concentration of Vitamin C (Mg/100ml) of Fresh Fruits Juice before Storage, at temperature of 40$^0$C, and at 7 Days Storage/shelve live are shown in the Table 1.bellow:

**Table 1.** Concentration of vitamin c (mg/100ml) of fruits juice.

| Sample | Room Temperature | Temperature of 40$^0$C | 7 Days Storage |
|---|---|---|---|
| Pineapple | 49.38 ±1.87 | 26.25 ±1.70 | 42.13 ± 0.87 |
| Orange | 39.75 ±1.00 | 23.00 ±1.52 | 31.50 ±1.00 |
| Water melon | 27.50 ±1.25 | 16.70 ±1.42 | 21.63 ±1.62 |
| Tomato | 19.13±1.63 | 10.83± 2.60 | 15.63 ±0.62 |

The Results are Mean of triplicate ± SEM.

# 8. Discussion

The result obtained from the research work shows differences in values of vitamin C at different temperature and shelve life: 49.38 ±1.875, 26.25 ±1.750 and 42.13 ±0.875mg/100ml for fresh, boiled and a week shelve life values respectively. The difference in vitamin C content of fresh juice and boiled juice is 23.13 mg/100ml (46.84%). However, the stored juice is having vitamin C content of 42.13 mg/100ml (7.2 mg) difference from fresh juice. The pineapple had the highest vitamin C content followed by Orange, Watermelon, and Tomato. This consistent with reports that, climate, especially temperature affect vitamin C level. Light exposure was found to promote browning in pineapple juice, therefore, the longer the exposure, the greater the loss of vitamin C (Maria, Encarna and Kader, 2006). Areas with cool nights produce fruits with higher vitamin C levels. Hot tropical areas produce fruit with lower levels of vitamin C (Padayatty, et al., 2003).

Squeeze washing reduced the Vitamin C content of some Nigeria Green leafy Vegetables; Ugu from 62.50mg/100g to 6.47mg/100g (89.65%) and Bitter leaf from 42.49mg/100g to 4.28mg/100g (89.90%). Squeeze-washing followed by boiling of bitter leaf reduced the vitamin C content from 42.40mg/100g to 2.18mg/100g recording the highest loss of 94.90% when compared with other processing methods (Babalola, Tusgbobo and Daramola, 2010)

The vitamin C content in fresh orange juice shows difference values to difference temperature and shelve life, 39.75 ± 1.000, 23.00 ± 1.520 and 31.50 ±1.000 mg/100ml for fresh, boiled and a week shelve life respectively. The difference in vitamin C content of fresh orange juice and boiled juice is 16.75 mg/100ml (42.14 %). However, the stored juice is having vitamin C content of 31.50 mg/100ml (8.25 mg) difference of 20.75% from fresh juice. Vitamin C decreases during the ripening process. The immature fruit has the highest levels. The position on the tree also affect vitamin C levels, fruit positioned on the outside of the tree and the south side have a higher levels, shaded inside fruit has lowest (Davey, Montagu and Inze, 2000).

The fresh watermelon juice shows a difference in vitamin C content at difference temperature and shelve life which are 27.5 ±1.250, 16.70 ±1.420 and 21.63 ±1.625 mg/100ml for fresh, boiled and a week shelve life respectively. The difference in vitamin C content of fresh watermelon juice and boiled juice is 10.80 mg/100ml (39.27%). However, the stored juice is having vitamin C content of 21.63 mg /100ml (5.87mg) difference 21.35% from fresh juice. Heat and water reduce vitamin C content, cooking reduce the vitamin C content in fruit juice because vitamin C content is sensitive to heat, water, and air. The vitamin C is first leached out of the fruit into the water, and then degraded by the heat (Murcia, et al., 2000).

The rate at which vitamin C content decreased in fresh tomato juice shows a difference values at difference temperature and shelve life which are 19.13 ±1.630, 10.83 ±2.600 and 15.63 ±0.625 mg/100ml for fresh, boiled and a week shelve life values respectively. The difference in vitamin C content of fresh juice and boiled juice is 8.30 mg/100ml (43.39%). However, the stored juice is having vitamin C content of 15.63 mg/100ml (3.50mg) difference from fresh juice. Early maturity fruit has higher levels of vitamin C than late maturing types, for instance, green tomato has the highest levels of vitamin C than the red tomato this is because the longer the exposure, the greater the loss of vitamin C (Rickman, Barret and Bruhn, 2007).

## 9. Conclusion

High temperature has effects on vitamin C content of fruits, blanching in hot water can cause an appreciable loss in vitamin C that is thermally labile. Ascorbic acid oxidase needs to be inactivated; this prevents enzyme-catalyzed reaction during processing. It can be inffered from results that the lower the temperature, the higher the availability of vitamin C in fruit juice. It is better to maintain or store vitamin C in a place below the room temperature

Many chemical reactions contribute to the loss of storage life of vitamin C and hence the chemical deterioration of fruits. The majority of these reactions are enzymatically driven while others are chemical reactions that occur because of the senescence. This involves colour, flavor, and odour changes that result from a chemical reaction between the constituents of the fruits. The biochemical reactions occurring over the storage period together with microbial action in all the fruit juices resulted in pH changes. Ascorbic acid is readily oxidized hence, the use of Metaphosphoric acid and Acetic acid to suppress metabolic activity upon disruption of the cell and to precipitate proteins. (Frank, et al., 2004).

This study supports the common perception that fresh is often best for optimal vitamin C content, as long as the fresh undergoes minimal storage at room temperature or heated. Loss of vitamin C with time differs from one fruit to other under similar storage environments. While boiled samples cause significant loss of vitamin C in the fruits (Maria, Encarna and Kader, 2006). For example, handling, storage, boiling, and oxygen is the most destructive ingredient in juice should be discouraged from being display in the hot weather above room temperature in order to maintain production concentration.

## References

[1] Abubakar, El-Ishaq and Bukar, A. Abubakar. (2012). Biochemistry for Tertiary Institutions. Management Network Resources.Damaturu.p123.

[2] Babalola, O.O, Tugbobo, O.S, and Daramola, A.S. (2010). Effect of Processing on the Vitamin C Content of Seven Nigerian Green Leafy Vegetables. Advance Journal of Food Science and Technology. 2(6). pp303-305.

[3] AOAC. (2006).Official Method 967.21e Ascorbic Acid in Vitamin Preparations and Juices: 2,6-Dichloroindophenol Titrimetric Method.Official Methods of Analysis of the Association of Official Analytical Chemists.

[4] Berg, JM, Tymoczko, JL, Stryer L. Biochemistry. 5th edition. New York: W H Freeman; (2002). Available from http://www.ncbi.nlm.nih.gov/books/NBK21154/

[5] Bethlehem Brown Campbell and Farrel Torrell. (2013).Introduction to Organic and Biochemistry.8th ed. Brooks/Cole. CENGAGE Learning. Belmont. USA.

[6] Davey, MW, Van Monatgu M, Sanmatin M, Kanellis A, Smirnoff N, Benzie IJJ, Strain JJ, Favell D, Fletcher J. (2000). Plant L-ascorbic acid: Chemistry, Function, Metabolism, Bioavailability and Effects of Processing. J Sci Food Agric.; 80: 825–860.

[7] Duyff, F. Ruurd, Joan Van Bosch, D Martin Laman, Bert-Jan Potter Van Loon, Wim H.J.P. Linssen. (2000) Neuromuscular Findings in Thyroid Dysfunction: A Prospective Clinical and Electrodiagnostic Study. J Neurol Neurosurgery Psychiatry; 68750-755 doi:10.1136/jnnp.68.6.750

[8] Frank, A.A., Cluster, T.J., Arakaki, C., and Murphy, S.P. (2004). Vitamin C and Flavonoids Level of Fruits and Vegetables Consumed in Hawaii. Journal of Food Composition and Analysis, 17, 1-35.

[9] Grano, A. De Tullio M.C. (2007). Ascorbic Acid as a Sensor of Oxidative Stress and a Regulator of Gene Expression: The Yin and Yang of vitamin C. Med Hypoth 69: 953-954.

[10] Johnson, Ojo Rotimi; Yetu, Abaya Jemimah; Oloruntoba, Akintayo Christopher; Samuel, Seriki. (2013). Effects of Nigerian Market Storage Conditions on Ascorbic Acid Contents of Selected Tetrapak Packaged Citrus Fruit Juice. Journal of Agricultural & Biological Science ; Feb 2013, Vol. 8 Issue 2, p179.

[11] Kiple, F. Kenneth. (2000). Cambridge World History of Food. 2 Volume 2. Cambridge University Press. page-1120.

[12] Padayatty, S.J., Katz, A, Wang Y et al., (2003). Vitamin C. Journal of American Coll.Nutr.2003.22; pp18-35.

[13] Maria, I.Gil, Encarna Aguayo, and Adel A. Kader. (2006). Quality Changes and Nutrient Retention in Fresh-Cut versus Whole Fruits during Storage. J. Agric. Food Chem. Vol 54 (12).pp4284-4296.

[14] Murcia, M. A., Lopez-Ayerra, B., Martinez-Tome, M., Vera, A. M., & Garcia-Carmona, F. (2000). Evolution of Ascorbic Acid and Peroxidase During Industrial Processing of Broccoli. Journal of the Science of Food and Agriculture, 80, 1882–1886.

[15] Murray, R. K., Bender D. A., Botham K. M., Kennelly P. J., Rodwell V. W., Weil P. A. (2012). A Lange Medical Book: Harper's Illustrated Biochemistry. New York, NY: Mc Graw Hill Medical

[16] Rickman, J.C, Bruhn, C.M, and Barrett DM. (2007). Nutritional Comparison of Fresh, Frozen, and Canned fruits and Vegetables II. Vitamin A and Carotenoids, Vitamin E, Minerals and Fiber. Journal of the Science of Food and Agriculture; 87: 1185-1196.

[17] Schorah, CJ,C. Downing, A. Piritsi et al., (1996). Total Vitamin C, Ascorbic Acid, and Hydro Ascorbic Acid Concentration in Plasma of Critical Ill Patients, Am J Nutr 63:760-765.