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COMPARATIVE ANALYSIS OF VITAMIN C IN FRESH FRUITS JUICE OF *Malus domestica*, *Citrus sinens*i, *Ananas comosus* AND *Citrullus lanatus* BY IODOMETRIC TITRATION

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Abstract: Vitamin C, also known as ascorbic acid, is a water soluble vitamin that is regarded as one of the safest and most effective nutrients. Vitamin C can be found in most fruits and vegetable. The objective of the study is to determine vitamin C content in four commercial fruits (Apple, Orange, Pineapple and Watermelon). Vitamin C content of the fresh fruits was determined titrimetrically. The method of determination was cheap, accurate and can be used for routine analysis. Apple vitamin C content was used as control (7.94 ± 0.13mg/100ml). The highest amount of vitamin C was in orange (10.13± 0.10mg/100ml) higher than that of apple followed by pineapple (6.40±0.18mg/100ml). However, watermelon had the lowest amount of vitamin C (4.08±0.12mg/100ml). There is a significant difference in vitamin C content among the fruits ($p < 0.05$). The method of determination was cheap, accurate and can be used for routine analysis.
Keywords: Vitamin C, iodometric titration, apple, orange, pineapple, watermelon.

Introduction

Vitamin C or L-ascorbic acid, or simply ascorbate (the anion of ascorbic acid), is an essential nutrient for humans and certain other animal species. Vitamin C refers to a number of vitamers that have vitamin C activity in animals, including ascorbic acid and its salts, and some oxidized forms of the molecule like dehydroascorbic acid. Ascorbate and ascorbic acid are both naturally present in the body when either of these is introduced into cells, since the forms interconvert according to pH. Vitamin C is a cofactor in at least eight enzymatic reactions, including several collagen synthesis reactions that, when dysfunctional, cause the most severe symptoms of scurvy (Food standards agency, 2007). In animals, these reactions are especially important in wound-healing and in preventing bleeding from capillaries. Ascorbate may also act as an antioxidant against oxidative stress (Padayatty *et al.,* 2003). However, the fact that the enantiomer D-ascorbate (not found in nature) has identical antioxidant activity to L-ascorbate, yet far less vitamin activity (Aboul-Enein *et al.,* 1999) underscores the fact that most of the function of L-ascorbate as a vitamin relies not on its

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antioxidant properties, but upon enzymic reactions that are stereospecific. "Ascorbate" without the letter for the enantiomeric form is always presumed to be the chemical L-ascorbate. Ascorbate (the anion of ascorbic acid) is required for a range of essential metabolic reactions in all animals and plants. It is made internally by almost all organisms; the main exceptions are most bats, all guinea pigs, capybaras, and the Anthropoidea (i.e., Haplorrhini, one of the two major primate suborders, consisting of tarsiers, monkeys, and humans and other apes). Ascorbate is also not synthesized by some species of birds and fish. All species that do not synthesize ascorbate require it in the diet. Deficiency in this vitamin causes the disease scurvy in humans (Food standards agency 2007; University of Maryland Medical Center 2007; Higdon *et al* 2006). Ascorbic acid is also widely used as a food additive, to prevent oxidation.

Fruit is a part of a flowering plant that derives from specific tissues of the flower, one or more ovaries, and in some cases accessory tissues. Fruits are the means by which these plants disseminate seeds. Many of them that bear edible fruits, in particular, have propagated with the movements of humans and animals in a symbiotic relationship as a means for seed dispersal and nutrition, respectively; in fact, humans and many animals have become dependent on fruits as a source of food (Lewis and Robert, 2002). Fruits account for a substantial fraction of the world's agricultural output, and some (such as the apple and the pomegranate) have acquired extensive cultural and symbolic meanings.

In common language usage, "fruit" normally means the fleshy seed-associated structures of a plant that are sweet or sour and edible in the raw state, such as apples, oranges, grapes, strawberries, bananas, and lemons. On the other hand, the botanical sense of "fruit" includes many structures that are not commonly called "fruits", such as bean pods, corn kernels, wheat grains, and tomatoes (Schlegel and Rolf, 2003; Mauseth and James, 2003).

Apple (*Malus domestica*) It is one of the most widely cultivated tree fruits, and the most widely known of the many members of genus *Malus* that are used by humans. Apples grow on deciduous trees which are large if grown from seed, but small if grafted onto roots (rootstock).

Orange (*Citrus sinesi*) The orange (specifically, the sweet orange) is the fruit of the citrus species *Citrus sinensis* in the family Rutaceae(Citrus sinensis (L.) Osbeck (pro sp.) (maxima reticulata) sweet orange". *Plants.USDA.gov*).The fruit of the *Citrus sinensis* is considered a sweet orange, whereas the fruit of the *Citrus aurantium* is considered a bitter orange. The

orange is a hybrid, possibly between pomelo (*Citrus maxima*) and mandarin (*Citrus reticulata*), which has been cultivated since ancient times (Nicolosi *et al.,* 2000).

Pineapple (*Ananas comosus*) is a tropical plant with edible multiple fruit consisting of coalesced berries (Dictionary.reference.com. Retrieved 6 December 2009). It is the most economically significant plant in the Bromeliaceae family (Geo *et al., 2003*). Pineapples may be cultivated from a crown cutting of the fruit (Pineapple Working Group-International Horticultural Society, Retrieved 15 August 2010).

Pineapples are consumed fresh, cooked, juiced, and preserved, and are found in a wide array of cuisines. In addition to consumption, in the Philippines the pineapple's leaves are used to produce the textile fiber piña- employed as a component of wall paper and furnishings, amongst other uses.

Watermelon (*Citrullus lanatus*) is a vine-like flowering plant originally from southern Africa. It is a large, sprawling annual plant with coarse, hairy pinnately, lobed leaves and white to yellow flowers. It is grown for it edible fruit, which is a special kind of berry referred to by botanists as pepo. The fruit has a smooth hard rind, usually green with dark green stripes or yellow spots, and a juicy sweet interior flesh, usually deep red or pink, but sometimes orange, yellow, or white with many seed.

Materials and Methods

Sample collection and preparation

Four ripe fruits were purchased from keffi local market Nasarawa state, Nigeria. The fruits include Apple (*Malus domestica*), Orange (*Citrus sinensi*), Pineapple (*Ananas comosus*) and Watermelon (*Citrullus lanatus*). The fruits were blended with a blender each and was filtered using a muslin cloth and made up to 100ml with distilled water.

Preparation of Reagents and estimation of ascorbic acid

1% starch indicator solution was prepared by adding 0.50g soluble starch to 50ml of distilled water. Mix well and allow cooling before use.

Iodine Solution was prepared by mixing 5.00 g potassium iodide (KI) and 0.268 g potassium iodate (KIO3) were dissolved into 500 ml beaker with 200 ml of distilled water. 30 ml of 3molar sulfuric acid was added into the beaker and then diluted with distilled water until 500 ml solution.

Vitamin C Standard Solution was prepared by dissolving 0.250 g ascorbic acid in the beaker with 100 ml distilled water. The solution was transferred into 250 ml volumetric flask and diluted to 250ml with distilled water.

Standardization of iodine solution with the vitamin C standard solution was by pipetting 25ml of vitamin C solution into a 125 ml Erlenmeyer flask. 10 drops of 1% starch solution were added and then titrated against iodine solution until blue-black colour was observed. Titrations were repeated in triplicates. The volume of each fruit sample used was measured and the concentration of ascorbic acid per 100ml fruits was calculated using:

Concentration of ascorbic acid used in mg/100ml

$$= \frac{\text{Concentration } (\text{mg/ml}) \text{ standardard}}{\text{Weight of samplesin gram}} \times 1000$$

Results and Discussion

A total of four fruits were studied, Vitamin C content of the four types of Fruits are tabulated in Table 1. It is clear that from the result obtained in the titrimetric method of vitamin C determination, Apple was used as reference standard and was lower than orange but was significantly higher than both pineapple and watermelon. This implies that both pineapple and watermelon has the lowest vitamin C content and orange has the highest vitamin C content from this analysis. Since the iodometric titration method is based on the oxidation-reduction reaction, a number of other reducing substances in foods (besides ascorbic acid) could interfere with the determination. Many molecules (e.g., phenols, sulphydryls, and triose reductones) and ions (e.g., ferrous, cuprous, or sulphite) are able to reduce the DCIP dye (Pachla *et al.,* 1985), and therefore giving rise to falsely high titration results. Generally, interferences can be overcome by adjusting the pH and other reaction conditions so that most other materials react only very much more slowly than does ascorbate (Bender, 1982). Another major practical problem associated with the titrimetic method of DCIP is the difficulty in ascertaining end-point when the food extracts are coloured, especially reddish-purplish colors.

Table 1: Result of vitamin C (mg/100ml) in the four fresh fruits.

Fruits	Scientific name	mean value of vitamin C (mg/100ml)
Apple	*Malus domestica*	7.94±0.13
Orange	*Citrus sinensis*	10.13±0.10
Pineapple	*Anana comosus*	6.40±0.18
Watermelon	*Citrullus lanatus*	4.08±0.12

The study has quantified the differences in vitamin C values obtainable by the iodometric titration for a number of Nigerian indigenous, and readily available fruits. There is considerable variation in these differences, depending on the amount of DHAA present in the

food. It is clear that the use of the iodometric titration method would result in underestimation of total vitamin C activity in a food. However, if only ascorbic acid values are required, the titrimetric procedure as described would give good results for most samples. The exception would be those that yield highly intense coloured extracts. Careful titration using ether extraction should be carried out for these foods. Attention should also be paid to ensure that no interfering substances are contributing to the titration results. In spite of these potential difficulties, the titration method has become widely used due to its convenience. Rapid determinations may be carried out with simple laboratory equipment. In the comparative studies reported by Wills *et al* (1983) and Bradbury and Singh (1986), there was fairly good agreement in ascorbic acid levels estimated by the simple iodometric titration method and the more sophisticated liquid chromatography procedure or a number of fruits.

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