

Determination of Ascorbic Acid Content in Different Fruit Juices Under Various Storage Conditions Using Iodometric Titration

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Abstract - This study investigated the impact of storage conditions room temperature, heat and cold on the levels of ascorbic acid (vitamin C) of chosen fruit juices like lemon, orange, apple, tomato and mango. Vitamin C was quantified by iodometric titration and the concentration of each fruit was recorded for the three conditions. the results revealed significant discrepancies both among the different fruits and the storage methods. Lemon juice always maintained the maximum ascorbic acid content of 2.1 at room temperature, 2.0 heated and 2.05 refrigerated, followed by orange at 1.8, 1.72 and 1.76 respectively. Mango has 1.1, 1.0 and 1.07, and apple at 0.92, 0.83 and 0.88 were moderately present, while tomato contained the lowest levels 0.72, 0.64 and 0.71. a common trend suggested that warming reduced ascorbic acid content in all fruit juices, validating vitamin C is heat labile nature. alternatively refrigeration preserved ascorbic acid content significantly better than room temperature and warming with values closer to initial concentrations.

Keywords - Introduction, Structure and Properties of Vitamin C, Applications in Human Diseases, methodology, Result, Discussion, Conclusion.

INTRODUCTION

Fruits have been an integral part of human diets as well as dietary supplements for centuries. They supply adequate amounts of water, vitamins, minerals, and organic compounds, all of which are essential for our bodies to function properly [1]. fruits are also a good source of antioxidants, which mop up free radicals. fruits are very perishable and spoil easily, so preservation becomes essential to maintain them fresh for a long period [3]. Orange, apple, grape and similar fruit juices are some of the beverages derived from fruits. Less fatty, nutrient-rich beverages, juices are packed with vitamins, minerals, and naturally occurring phytonutrients that promote healthy living, and the human body utilizes fruit juices to facilitate detoxification [4]. Fruits are industrially processed to manufacture fruit juices, jams, jellies, etc. Juices occur in their original concentrations or processed states. fruit juice is primarily produced by mechanically pressing fresh fruits or water extraction. The majority of commercially available juices are fat-free, concentrated nutrient drinks with high content of vitamins, minerals and naturally occurring phytonutrients [5]. Vitamins are essential and extremely crucial for the existence and sleek operations of human bodies. Vitamin C cannot be produced or stored in human body thus the intake of ascorbic acid on daily basis is required to maintain a healthy body. vitamins do not give direct energy but they work in

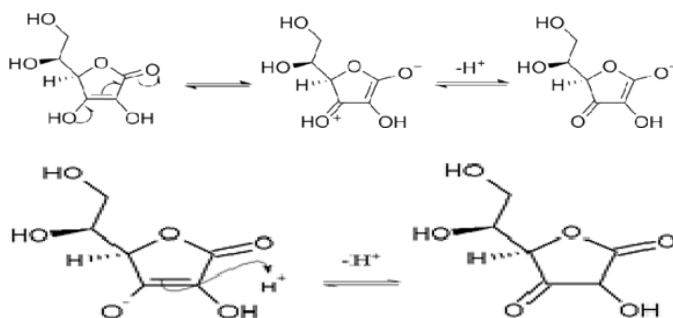
combination with other compounds in the body to provide energy.

Vitamin C is predominantly contained in vegetables and fruits such as strawberries are regarded to be one of the richest in ascorbic acid [6]. vitamin C of up to 80 mg per 100 g exists in fresh strawberries. Strawberry juices, which are ready to use sources of ascorbic acid for human consumption, might be prepared from strawberries with ease. but during storage, fruit juices usually lose their ascorbic acid due to easy oxidation. Many variables such as exposure to sunlight, pH 5, dissolved oxygen level, metal ion presence, sugar presence and storage temperature, influence this process of oxidation. Ascorbic acid is very sensitive to various processing conditions and is thermolabile [7].

The main reason why strawberry juices lose their nutritional value and prolong their shelf life is the loss of ascorbic acid during storage. Manufacturers need to exercise due care to process and store their items in such a manner that maximizes the vitamin C content for consumers, studying the kinetics of ascorbic acid loss in strawberry juices during storage is critical to understanding the mechanism of ascorbic acid degradation. fruit juices shelf life is established by the rate constant and kinetic order. kinetic models are also not restricted to objective, rapid and inexpensive assessments of food quality [8]. but can also be utilized to forecast the effect of various experimental

parameters on important nutritional values. Various kinetic investigations in literatures attempted to approximate the rate constants for the vitamin C degradation in fruit juices. Various kinetic models like zero-order, first order, pseudo-first order, and second order kinetic reactions have been utilized effectively to explain vitamin C degradation. Nevertheless, few kinetic studies of strawberry juice ascorbic loss during storage have been reported. The objective of the present study is to assess the degradation kinetics of vitamin C in fresh home-made strawberry juices after 8 hour storage at room and refrigerated temperature and in addition to examine the influence of storage temperatures and added sugar on strawberry juice vitamin C loss. Direct iodimetric titration was employed to analyze the vitamin C level in strawberry juices. The technique is cheap, effective and easy with regards to the equipment and reagents [9].

Fruit juice is a popular beverage that is frequently promoted for its health advantages especially due to its high vitamin C content. but it is generally accepted that ascorbic acid concentrations are higher in freshly squeezed juices than in processed and packaged juices which may be pasteurized and stored under different circumstances [10]. however because commercial fruit juices are exposed to air and undergo temperature changes during storage and transit some do not maintain their original vitamin C levels by the time ingestion. therefore, measuring the vitamin C concentration of various fruit juices both commercial and fresh is crucial to assessing their nutritional claims. this type of investigation also aids in determining how various packing materials, storage circumstances and preservation techniques impact the stability of vitamin C [11] A six-carbon molecule chemically bonded to glucose, ascorbic acid ($C_6H_8O_6$) contains a lactone ring and two hydroxyl groups in enediol form that imbues it with its reducing properties. It is therefore susceptible to oxidation, especially in alkaline and aerobic conditions. Ascorbic acid, the reduced form of the molecule, and dehydroascorbic acid, its oxidized form, both of which possess vitamin C activity in man [12].



Mechanism of Ascorbic Acid

It is deterioration can be enzymatically catalyzed by ascorbate oxidase and by metal ions of copper and iron, further adding to its instability upon processing and storage [13]. The determination of its actual concentration in juices upon consumption, therefore, allows for very meaningful judgment on the capability of processing methods in maintaining nutrient quality. There are a number of intrinsic and extrinsic factors that can affect the rate of degradation of vitamin C in fruit juices. These include the Increased temperatures promote the oxidation of ascorbic acid. Pasteurization, although essential for microbial safety, can lower vitamin C content and oxygen exposure are starts the oxidative decomposition of ascorbic acid. Inadequate packaging and frequent opening of juice packs will enhance oxygen exposure. but the pH levels of acidic media (low pH) are more conducive to ascorbic acid stability, since most fruit juices naturally contain acids and it assist to decrease degradation. Though light exposure is ultraviolet radiation can stimulate photo oxidation particularly in transparent packaging [14].

In understanding these variables is crucial for improving conservation methods and proper choice of the packaging materials to reduce nutrient loss. while newer techniques such as HPLC are highly accurate, iodometric titration is very much in vogue due to its ease of operation and low cost, especially for educational and field laboratories. It is not instrumental intensive and can produce good results if proper procedure is taken care of. In addition, it is versatile to various types of samples, such as fresh and processed fruit juices and can be employed to estimate vitamin C contents within a reasonable error margin [15]. Regardless of its shortcomings like interference from other reducing substances in complicated matrices proper sample handling and control experiments may inhibit such occurrences. Thus, iodometric titration proves to be a good technique for comparative investigations of vitamin C content in different samples of juice [16].

Vitamin C also known as ascorbic acid is a vital water soluble antioxidant that plays an essential role in numerous physiological processes in the human body. first isolated in 1928 by Albert Szent-Györgyi, who later received a nobel prize for this work vitamin C gained significant attention for its role in preventing scurvy a disease historically prevalent among sailors deprived of fresh fruits and vegetables [17]. today vitamin C is widely recognized not only for its antioxidant properties but also for its crucial role in collagen synthesis, immune function, wound healing, iron absorption and neurotransmitter production [18]. As an essential nutrient vitamin C cannot be synthesized by the human body and must be obtained exogenously through the diet. fruits and vegetables are the primary natural sources with certain fruits providing

exceptionally high concentrations of this vitamin. the Recommended Dietary Allowance (RDA) for vitamin C varies by age and sex with adult males requiring approximately 90 mg/day and adult females 75 mg/day smokers require an additional 35 mg/day due to increased oxidative stress [19].

Given its biochemical importance and widespread occurrence in plant based foods the determination of vitamin C content in fruits has been a subject of scientific interest for decades. numerous analytical methods including iodometric titration, UV visible spectrophotometry and High Performance Liquid Chromatography (HPLC) have been employed to quantify ascorbic acid levels accurately [20]. the content of vitamin C in fruits can vary greatly due to species differences, ripeness, storage conditions, geographical origin and processing methods for example tropical fruits such as guava, acerola and amla (Indian gooseberry) contain significantly higher concentrations of vitamin C than commonly consumed fruits like apples, bananas or grapes. but understanding the variation in vitamin C content among different fruits is essential for multiple reasons. From a nutritional perspective, it helps consumers make informed dietary choices especially in populations at risk of deficiency. from an agricultural and industrial standpoint, it aids in the selection of fruit varieties for food processing, fortified products and nutraceuticals. moreover the stability of vitamin C during storage and processing is a key concern for maintaining nutritional value in commercial fruit products [21].

This review seeks to analyze and compare the vitamin C content in various fruits, examining not only their nutritional significance but also the factors affecting their ascorbic acid concentration by evaluating both common and exotic fruits, this study aims to provide comprehensive insights into their roles as dietary sources of vitamin C and to support food scientists, nutritionists and health conscious individuals in optimizing vitamin C intake through natural sources.

The Structure and Properties of Vitamin C

Structure of Ascorbic Acid

Vitamin C is an acidic poly hydroxyl compound comprising carbon, hydrogen and oxygen with a molecular formula of $C_6H_8O_6$ and an IUPAC name of 2,3,4,5,6-pentahydroxy-2-hexenoide4-lactone with a molecular weight of 176.1 g/mol. the structural formula is shown in fig. 1. the structure of vitamin C contains enediol groups of which C3–OH exhibits strong acidity. this is influenced by the conjugation effect ($pK_a \frac{1}{4}$ 4.17). In contrast C2–OH is weakly acidic due to intramolecular hydrogen bonding ($pK_a \frac{1}{4}$ 11.75). accordingly vitamin C exists in the form of anion under physiological conditions. the structure of VC contains conjugated structures, which result in ultraviolet absorption. the maximum absorption

wavelength is 245 nm which provides a method for the detection of vitamin C content.

The double bond between C2 and C3 yields two electrons, which are subsequently lost by vitamin C to generate AA free radicals (semi hydroascorbic acid). the majority of these free radicals have a lifetime of less than a millisecond and exhibit reduced activity [22]. when vitamin C loses its second electron and forms a substance that is more stable than the ascorbate radical DHA. Both AA free radicals and DHA can be reduced to AA. Once the five-membered loop of DHA is hydrolyzed and broken to produce 2,3-diketo-lgulonic acid this process is irreversible the reduction of DHA to AA is no longer possible but the detailed oxidation process of VC is shown in fig. 2. some biological processes within the human body may result in the production of highly reactive and potentially harmful free radicals, and these free radicals can be removed by vitamin C. In this process vitamin C itself is converted to less active ascorbate radicals. these results indicate that AA is an effective antioxidant and free radical scavenger [23].

Oxidation of ascorbic acid.

Oxidation of ascorbic acid are approved by the National Medical Products Administration (NMPA) annually. the vitamin C related preparations approved by NMPA from 2021 to 2023 are shown in to reduce vitamin C degradation, achieve targeted drug delivery and improve therapeutic efficacy, researchers have developed different drug delivery systems to deliver vitamin C such as polymeric nanoparticles, liposomes, microemulsions and micelles fig. 3. nanoparticles made of natural polymers have low toxicity high biocompatibility and sufficient degradability [24]. Chitosan is a kind of hydrophilic polysaccharide. Chitosan based drug delivery systems have the following advantages that increased solubility controlled drug release, enhanced drug targeting and improved absorption [25]. adopted the ionotropic gelation method and used tripolyphosphate as a cross linking agent to prepare vitamin C chitosan nanoparticles which realized the encapsulation of vitamin C within the nanoparticle. the obtained nanoparticles improved the stability of vitamin C in the gastrointestinal tract and showed consistent vitamin C release for up to 48 hours [26].

Liposomes are phospholipid bilayer vesicles composed of amphiphilic molecules which can load hydrophobic and hydrophilic molecules and have the advantages of low toxicity and high biocompatibility [27, 28]. Orally administered vitamin C is easily degraded in the gastrointestinal tract and liposomes can provide a hydrophilic hydrophobic interface to avoid vitamin C degradation [29,30]. Liposomes can also reduce gastrointestinal interference and prolong the release of vitamin

c [31,32]. used chitosan coated liposomes to deliver ascorbic acid and folic acid which improved the antioxidant efficacy of the preparation. therefore the use of liposome delivery technology can improve the bioavailability of vitamin C to a certain extent and avoid the risks associated with intravenous administration [33,34]. microemulsion is a transparent thermodynamically stable colloidal system formed spontaneously from oil, water, and emulsifier, with an average particle size of 10 to 100 nm.

The microemulsion has low surface tension, high interfacial tension and solubilization properties [35]. In addition to the above nano preparations, currently, researchers use vitamin C to develop new drug delivery systems to achieve targeted drug delivery [36]. used polyethylene glycol phosphatidylethanolamine to prepare palmitoyl ascorbate micelles which improved the solubility of palmitoyl ascorbate and increased the accumulation of palmitoyl ascorbate at the tumor site through the high permeability and enhanced permeability and retention effect (EPR effect) of solid tumors showing good antitumor activity.

It used AA derivatives as liposome ligands and used vitamin C to bind to the receptor ligand of GLUT1 and SVCT2 to deliver drugs targeted to the brain, indicating that AA has the potential to enhance brain targeting of drugs in the central nervous system. Luo et al used ascorbate coupled polylactic acid hydroxyethyl copolymer to promote oral drug absorption through SVCT1 [37]. but used AA derivative ascorbic acid 2-phospho-6-palmitate trisodium to form micelles and used as drug carriers to improve drug skin permeability. in general these drug delivery systems address the limitations of vitamin C namely its poor stability and strong hydrophilicity through enhanced encapsulation and targeted drug delivery thereby improving the absorption of vitamin C in vivo. however, the current industrial technologies impose constraints on the large scale clinical application of these delivery systems [38].

Vitamin C Promotes Collagen Formation

VC plays a pivotal role in the posttranslational modification of procollagen a crucial coenzyme factor in collagen biosynthesis [39]. Vitamin C facilitates the synthesis of collagen a fundamental protein in the extracellular matrix by regulating the structure and secretion of procollagen [40]. Procollagen is synthesized in the endoplasmic reticulum and consists of integrity. specifically proline and lysine residues undergo hydroxylation and conversion to hydroxyproline and hydroxylysine, respectively. this hydroxylation process requires ascorbic acid as a cofactor [41]. Collagen has a triple helix structure and the presence of hydroxyproline is essential to stabilize this structure [42]. without hydroxyproline,

fibroblasts cannot secrete collagen properly. hydroxylysine is necessary for collagen cross linking and its absence can also lead to structural instability. a computer simulation experiment was conducted to study the effect of vitamin C on amino acid sequence and interaction forces during collagen formation [43]. In the presence of vitamin C collagen was synthesized, while in the absence of vitamin C hydroxyproline dissociates from prolyl 5-hydroxylase and the reaction stops therefore, VC can be considered a cofactor for prolyl-5-hydroxylase which is responsible for the conversion of proline to hydroxyproline. VC is conducive to the hydroxylation process which facilitates the formation of a stable triple helix structure in collagen and promotes collagen formation [44].

Vitamin C Promotes Hydroxylation

Hypoxia-inducible factor-1 is a nuclear protein with transcriptional activity that plays an important role in physiological processes such as erythropoiesis, cell survival and angiogenesis [45]. hypoxia-inducible factor-1 can induce the formation of new blood vessels around hypoxic cells and tissues thus promoting cell survival. consequently HIF-1 exerts a significant influence on fetal development yet it also serves as a key promoter of a range of pathological conditions, including inflammatory diseases, lung disease, heart disease, diabetes and cancer [46]. HIF-1 consists of an active α subunit and a structurally expressed β subunit. Proline residues on the HIF α subunit are hydroxylated by the prolyl hydroxylase domain (PHD) and undergo rapid degradation by ubiquitin protease under normal oxygen conditions. under hypoxia conditions, prolyl hydroxylase domain is inhibited [47]. vitamin C plays an important role in the hydroxylation of proline residues by prolyl hydroxylase domain. prolyl hydroxylase domain is a nonheme iron-dependent dioxygenase, its catalytic activity can be enhanced by vitamin C [48,49]. In vitro experiments have shown that vitamin C has a significant inhibition effect on HIF-1 and can block hypoxia-inducible factor-1 induced gene transcription thereby delaying the development of the disease. In summary by influencing the PHD to regulate hypoxia-inducible factor-1 levels of ascorbic acid can diminish symptoms and delay the progression of the disease [50].

Ascorbic Acid Enhances Immune Function

Vitamin C regulates immune function by enhancing various cellular functions of both the innate and acquired immune systems, regulating redox sensitive cell signaling pathways or directly protecting important cellular structural components [51]. the immune system is composed of immune organs like (bone marrow, spleen and lymph). Then the immune cells for examples (lymphocytes, mononuclear phagocytes and neutrophils) and immunologically active substances is

antibodies and complements that protect the host from a range of pathogens [52]. the immune system can be divided into two categories: nonspecific immunity and specific immunity. These categories can be further delineated into three main aspects: physical barriers such as skin and mucosa, immune cells and antibodies [53].

Vitamin C has been shown to protect the skin from environmental oxidative stress by promoting clearance of oxidants [54]. thereby strengthening the physical skin barrier. the infiltration of neutrophils into infected tissues is the early step of innate immunity. VC can accumulate in phagocytes such as neutrophils, enhance the chemotaxis and phagocytosis of neutrophils, produce reactive oxygen species (ROS) and eventually kill microorganisms [55]. Oxidants can activate nuclear factor κ B (NF- κ B) which triggers a signaling cascade that leads to the continued synthesis of oxides and other proinflammatory mediators [56]. VC has been shown to reduce the production of oxidants and the activation of NF- κ B in dendritic cells in vitro [57]. In addition research has demonstrated that VC enhances the immune system response to infection by stimulating T lymphocyte proliferation increasing cytokine production and promoting immunoglobulin synthesis [58]. The Changing team at West Lake University found that vitamin C can facilitate plasma cell differentiation and humoral immune response by enhancing TET2/3 mediated DNA demethylation [59]. a chronic deficiency of vitamin C can lead to impaired immune function, rendering individuals more susceptible to infection. the inflammatory and metabolic demands associated with infection can significantly reduce the body's ability to absorb vitamin C therefore, timely and appropriate supplementation of ascorbic acid can help enhance the bodies immune response [60].

Vitamin C Improves Fat Metabolism

The fatty acids produced by the hydrolysis of fat in the human digestive tract are activated as fatty acyl CoA in the endoplasmic reticulum and the outer membrane of mitochondria. the enzymes that catalyze fatty acid oxidation are located within the mitochondrial matrix therefore, the activated fatty acyl CoA must enter the mitochondria to be reduce trivalent iron to ferrous thereby promoting the absorption and utilization of iron [61]. The extent of this reduction is influenced by the pH value. The studies have shown that in the range of pH 2.6 to 6.0, the reduction rate of vitamin C decreases as the pH increases. at a pH range of 6.8 to 7.4, AA is unable to effectively reduce the trivalent iron [62]. subsequent studies demonstrated that duodenal cytochrome B is an iron regulatory protein with iron reductase activity which plays a critical role in dietary iron absorption [63]. Several studies identified duodenal cytochrome B promoting iron reduction in a

dependent manner indicating that vitamin C is primary role is in promoting iron dissolution. furthermore, studies demonstrated that the administration of vitamin C a few hours before the consumption of an iron containing meal did not increase iron absorption, suggesting that ascorbic must be taken with iron to promote iron absorption [64,65].

II. APPLICATIONS IN HUMAN DISEASES

Vitamin C has many preventive and therapeutic effects in the field of medicine. this section will mainly discuss its application in cataracts, cancer, CVD and skin disease Fig.1.

Cataracts

Cataracts are a leading cause of vision impairment and blindness worldwide [66]. Population projections indicate that by 2025, cataracts could affect 40 million individuals [67]. The underlying mechanisms of cataract formation remain unclear but oxidative stress is a prominent hypothesis. the free radicals generated by oxidative stress such as glutathione (GSH), superoxide dismutase and ROS will damage the lens composition and resulting in lens opacity and thus lens damage [68]. although cataract surgery is considered to be one of the safest surgeries, the prevention of cataracts has become a research focus due to the complications that can arise from the procedure including recurrent or persistent inflammation, glaucoma and posterior capsule opacification [69].

The balance between antioxidants and free radicals determines the state of appropriate physiological function if the level of free radicals rises uncontrollably, oxidative stress will appear [70]. The literature indicates that VC as a nonenzymatic antioxidant, it may have certain preventive effects on cataracts although this is limited to nuclear cataracts [71]. nuclear cataracts are located in the center of the lens and are usually caused by advancing age [72]. Vitamin C is present in high amounts in the aqueous humor and is therefore thought to play a role in protecting the lens from oxidative stress in the aqueous humor [73]. A statistical survey has shown that dietary intake of ascorbic acid can reduce the risk of age related cataracts [74]. moreover several studies and case control studies have shown that individuals with a high dietary intake of VC exhibit a reduced risk of developing cataracts [75,76]. however it is worth noting that the vitamin C has the potential to generate free radicals intermediates and strong oxidizers which can inflict damage to the biological tissue. it has been demonstrated that VC can facilitate the formation of advanced glycation end products which are responsible for the chemical aging of lens proteins [77,78]. Besides, high doses of VC have been shown to increase the prevalence of age related cataracts in women

consequently, the effectiveness of vitamin C for cataracts may be limited to a role in the prevention of nuclear cataracts [79].

Tumor Treatment

Epidemiological evidence suggests that VC or foods rich in VC may play a role in cancer prevention as they have been shown to reduce the incidence of various tumors [80]. For example, demonstrated that administering a high dose of VC daily was able to maintain optimal levels of AA within 48 hours while simultaneously downregulating the activity of the HIF-1 pathway within tumor tissue. Nevertheless, there is a lack of consensus regarding the efficacy of ascorbic acid in treating tumors. Two previous studies yielded disparate results [81,82]. Cameron and Pauling showed that high-dose vitamin C improved the average survival rate of patients with advanced cancer. Conversely, two clinical trials by Creagan and Moertel indicated that vitamin C did not confer a benefit in cancer treatment. A deeper examination of vitamin C pharmacokinetics may elucidate the discrepancy in outcomes between the two administration methods.

In the initial experiment intravenous administration was used, while in the latter oral administration alone was used when doses of oral vitamin C exceed 200 mg the absorption of VC decreases, urinary excretion increases and the bioavailability of ascorbic acids decreases [83]. In contrast intravenous administration bypasses intestinal absorption, resulting in a plasma concentration of vitamin C that can reach pharmacological levels and thus exert efficacy [84]. Given that AA is susceptible to a pH dependent autooxidation reaction to produce hydrogen peroxide (H₂O₂) (H₂O₂ is toxic to a variety of tumor cells) [84]. High dose intravenous AA can be used as a prodrug to deliver H₂O₂ to tumors thereby treating cancer [85]. Potential mechanisms of action for vitamin C in cancer therapy include:

AA is an important free-radical scavenger. The oxidation of a large dose of AA to DHA has been demonstrated to increase ROS, trigger oxidative stress and lead to the apoptosis of tumor cells [86].

III. METHODOLOGY

Materials and Reagents

- Fresh fruit juice samples
- muslin cloth
- lemon pineapple, apple)
- 0.005 M Iodine solution (I₂)

- 0.1 M Sodium thiosulfate solution (Na₂S₂O₃)
- 1% Starch solution (indicator)
- 1 M Sulfuric acid (H₂SO₄)
- Distilled water
- Analytical balance
- Burette, pipette, conical flask, beakers
- Filter paper and funnel

Sample Preparation

Fresh samples of fruits were obtained from a regular market in Gangrar and Chittorgarh, India. The samples were washed and classified into three groups for storage with each group containing ten samples of each fruit. The first group of fruits was refrigerated for 24 hours and allowed to settle. The second group of fruits was heated for 10 minutes at a temperature of 80 °C and allowed to cool while the third group was stored at an average room temperature of about 25 °C for 12 hours. The fruit samples were washed again with distilled water, peeled and sliced into transversely and squeezed. The juice obtained was filtered with a muslin cloth. The fruit samples were sliced into smaller portions and squeezed. The juice obtained was also filtered.

Preparation of Reagents

- Iodine solution: Prepared by dissolving 30g iodine crystals and 12 g potassium iodide in distilled water.
- Starch indicator 1 g of soluble starch was dissolved in 100 mL of hot distilled water and allowed to cool before use.
- Sodium thiosulfate solution: Prepared by adding 2.48g and dissolving it in 100ML of distilled water and standardized before titration.
- Sulfuric acid: Diluted appropriately with distilled water to get 1 M concentration.

Iodometric Titration Procedure

- 10 mL of fruit juice was pipetted into a conical flask.
- 10 mL of 1 M sulfuric acid was added to acidify the sample.
- 10 mL of iodine solution was added; the iodine reacts with ascorbic acid in the juice.
- The flask was kept in the dark for 5 minutes to ensure complete reaction.
- The unreacted iodine was titrated with 0.1 M sodium thiosulfate until the brown color faded to pale yellow.
- A few drops of starch solution were added and also the solution turned blue black.
- Titration continued until the color disappeared, indicating the end point.
- The volume of sodium thiosulfate used was recorded for analysis.

- Calculation of Ascorbic Acid Content.

Result

Table of at room temperature

S/N	Vol of Sample	Burette Initial Read	Burette Final Read
0	10 MI	0 MI	50 MI
1	Lemon	0	23.2
2	Orange	0	20.1
3	Apple	0	10.3
4	Tomato	0	8
5	Mango	0	12

Table of hot sample

S/N	Vol of Sample	Burette Initial Read	Burette Final Read
0	10 MI	0 MI	50 MI
1	Lemon	0	22.6
2	Orange	0	19.2
3	Apple	0	9.3
4	Tomato	0	7.1
5	Mango	0	11.3

Table of refrigerator

S/N	Vol of Sample	Burette Initial Read	Burette Final Read
0	10 MI	0 MI	50 MI

1	Lemon	0	22.9
2	Orange	0	19.7
3	Apple	0	9.8
4	Tomato	0	7.9
5	Mango	0	12

Calculation for find the level of ascorbic acid in each sample.

Formula

$$\frac{M_1 V_1}{1} (I_2) = \frac{M_2 V_2}{2} (\text{thio})$$

Table of at room temperature level of ascorbic acid

S/N	Vol of Fruits Sample	Level Of Ascorbic Acid
0	10 MI	0 MI
1	Lemon	2.1
2	Orange	1.8
3	Apple	0.92
4	Tomato	0.72
5	Mango	1.1

Table at hot level of ascorbic acid

S/N	Vol of Fruits Sample	Level Of Ascorbic Acid
0	10 MI	0 MI

1	Lemon	2.0
2	Orange	1.72
3	Apple	0.83
4	Tomato	0.64
5	Mango	1.0

Table of refrigerator level of ascorbic acid

S/N	Vol of Fruits Sample	Level Of Ascorbic Acid
0	10 Ml	0 Ml
1	Lemon	2.05
2	Orange	1.76
3	Apple	0.88
4	Tomato	0.71
5	Mango	1.07



Discussion

Analysis of the ascorbic acid in various fruit juices under different storage conditions room temperature, heat and refrigerator showed significant variations in the concentration of vitamin C in the various fruits as well as the retention of vitamin C under different temperature regimes of each fruit demonstrated a unique ascorbic acid pattern is validating that the content of the vitamin C not only varies with the fruit but is also very sensitive to the storage conditions.

Lemon (Citrus limon)

Lemon juice had the highest ascorbic acid concentration compared to other samples with values of 2.1 L at room temperature, 2.0 L under heat and 2.05 L under chilled conditions. lemon's rich content of vitamin C is in line with reports that citrus fruits contain a high concentration of ascorbic acid a vital antioxidant substance. the mild decrease in concentration under heating conditions reveals that vitamin C is thermolabile experiencing oxidative degradation upon exposure to heat. in contrast, refrigeration preserved the concentration near that noted at room temperature and suggests that cold storage retards enzymatic and non enzymatic oxidation reactions.

Orange (Citrus sinensis)

Orange juice was the second richest in vitamin C in this research the record levels of 1.8 at room temperature, 1.72 when it was heated and 1.76 under refrigeration. like lemon, orange recorded a drop in ascorbic acid content when heated again indicating the instability of the compound to heat. refrigeration reduced the loss and maintained the vitamin C content to near its initial value. these observations accord with earlier findings with orange emerging as a major food source of vitamin C although still somewhat lower in concentration than lemon.

Apple (Malus domestica)

Apple juice also had quite low concentrations of vitamin C with readings of 0.92 at room temperature, 0.83 on heating and 0.88 on refrigeration. in contrast to citrus fruits, apple juice is also reported to possess moderate levels of ascorbic acid which explains the low readings. loss under heat further highlights the sensitivity of vitamin C to heat degradation. refrigeration, though not entirely precluding loss, maintained the concentration better than exposure to heat. these findings indicate that though apple juice is rich in vitamin C it is not an excellent source compared to lemon or orange.

Tomato (Solanum Lycopersicum)

Tomato juice had the lowest ascorbic acid content among the fruit juices with values of 0.72 at room temperature, 0.64 under heat and 0.71 under cold storage. The low values are in line with the established composition of tomato juice which has lower amounts of vitamin C than that found in citrus fruits. However, tomato still makes a contribution to the vitamin C requirement especially when eaten fresh. The loss of vitamin C on heating is significant since tomato-based foods are usually cooked during processing. This implies that the high heat treatment processes like boiling or pasteurization could lower the nutritional value of tomatoes in terms of vitamin C retention.

Mango (*Mangifera indica*)

Mango juice showed intermediate vitamin C levels in comparison to the other samples with 1.1 at room temperature, 1.0 after heating and 1.07 after refrigeration. Although higher than in apple and tomato, mango had much lower levels of vitamin C than citrus. Again, heating lowered the concentration, while refrigeration maintains the levels better. These findings emphasize that mango, though prized for flavor and other vitamins like carotenoids, is not quite as good a source of ascorbic acid as citrus fruits while both clear trends were seen among all fruit samples that heating always decreased vitamin C content, while refrigeration preserved it best. This trend is in accordance with the known chemical property of ascorbic acid being a heat-labile compound subject to oxidative deterioration. The slightly higher refrigeration values compared to room temperature values such as lemon 2.05 to 2.1, orange 1.76 to 1.8, indicate the stabilizing influence of low temperatures on the vitamin C molecules, retarding the rate of degradation reactions.

Recommendation

This study was intended to highlight the importance of proper storage conditions to preserve the nutrient content of fruit juices for domestic and industrial consumption. So, refrigeration is strongly advised to minimize vitamin C loss. Citrus fruits such as lemon and orange, should be included in the diet as the most reliable sources of vitamin C. These results provide consumers, food industry and health authorities with practical knowledge on the best storage and consumption of fruit juices to ensure maximum preservation of this essential vitamin.

IV. CONCLUSION

This study examined the impact of various storage techniques: room temperature, heat, and refrigeration on the levels of ascorbic acid in orange, lemon, apple, tomato, and mango juices. The results indicated that both the fruit type and storage conditions have a significant impact on vitamin C content.

These results suggest that the best way to keep vitamin C in fruit juices is through refrigeration. For the consumer, this implies that chilled or freshly prepared juices are the best options for nutritional purposes. For the food and beverages sector, this highlights the need for cold storage and nonthermal preservation methods in ensuring products retain high nutritional levels among fruit products. This research offers experimental proof that all fruits do not contain the same amount of vitamin C and that storage conditions significantly impact the preservation of this important nutrient. In order to preserve the health benefits of fruit juices, refrigeration must be given top priority, exposure to heat minimized, and citrus fruits promoted in the diet.

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