Comparison of Vitamin C Contents in Commercial Tea Brands and Fresh Tea Leaves

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Summary: Vitamin C contents were investigated in 14 tea brands collected from local market in Peshawar and one brand from National Tea Research Center at Mansehra, N.W.F.P. Pakistan, using UV-Visible spectrophotomtry. In all the tea samples, high amount of Vitamin C was found in green tea than black tea leaves. The fresh tea leaves collected directly from plant was found to have the same amount of Vitamin C as the green tea leaves. The small amount of vitamin C in black tea may be due to its processing as vitamin C is highly sensitive to heat, light and decomposes frequently.

Introduction

Vitamins are necessary for the metabolism, which cannot be prepared in sufficient amount by the human organisms [1] and therefore, must be obtained from other sources such as food and medicine. Vitamin C is an important vitamin taking part in various biological activities such as electron transport reactions, hydroxylation, oxidative catabolism of aromatic amino acids, etc. Due to its bioactivity and as antioxidant, it has wide variety of applications such as in pharmaceutical, chemical, cosmetic and food industries.

Vitamin C is a water-soluble vitamin, which plays an important role in preventing illnesses such as scurvy and the common cold. This vitamin is found in high concentration in citrius fruits, black barrie, melons, tomatoes, green peppers, cabbages and green vegetables [2].

In case of biological activity vitamin C participates in metabolic process as enzyme cofactor of propyl and lysil hydroxylyases, thereby producing hydroxyproline and hydroxylysine which participate in the collagen formation [3]. It also acts in amino acid metabolism and biosynthesis of suprarenal hormone, thereby minimizing the stress effect. Ascorbic acid also increases the organism resistance against microorganism. Deficiency of vitamin C provokes fatigue and debility of blood vessel, teeth, bone, some escorbate indication, difficult the hurt cicatrization, the growth, the reproduction and the lactation [1, 3, 4-6]. Vitamin C is widely used as medicine and also added in manufactured foods to act as antioxidant to conserve the product for a long time [7]. However, the excess of ascorbic acid can cause gastric irritation and diarrhea, giving as metabolic product the oxalic acid, which in turn can cause renal problems [1, 3, 4-6].

Various methods have been employed for its measurement, such as spectrometry [8, 9], thermometric titrimetry [10], HPLC [11], a kinetic method [12], various modified electrodes [13-15], and sol-gel [16].

The spectrophotometric method is the most common method used in laboratory. Reagents as molybdate and other compounds react with ascorbic acid giving color compounds; however, some dyes present in food and medicine sample can interfere in its determination. Thus the sample needs pre-treatment to eliminate this effect. The direct spectrophotometry in UV region is also widely used, since the ascorbic acid absorbs in this region presenting a maximum absorbance at 243 nm in strongly acidic media and in 265 nm in neutral media. However, the majority of these methods [17-21] monitor ascorbic acid at 243 nm, since the ascorbic acid is more stable in acidic media due to less hydrolization of its lactone ring. Therefore, many compounds present in medicinal plant samples absorb in UV region and its effects need to be eliminated.

The aim of the present work was to determine the concentration of vitamin C in commercial tea brands (black and green) as well as the fresh tea

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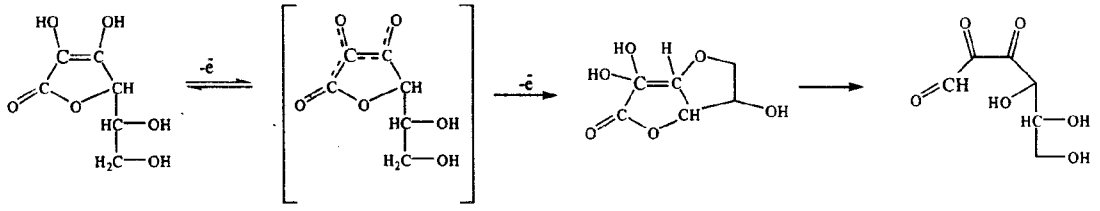
leaves obtained from National Tea Research Center at Mansehra, N.W.F.P. Pakistan using UV-visible spectrophotometry.

Results and Discussion

The solubility of vitamin C in aqueous solution was monitored in different parameters like pH, oxygen, time and temperature. All solutions for the study were prepared in 0.05 M oxalic acid - 0.02 M EDTA solution. These solutions were stored in brown volumetric flasks.

The acidity of vitamin C is based on its enol group ionization from C^3 and C^2 atoms, with their pka value is 4.17 and 11.57, respectively [22]. The undissociated vitamin C present in solution with pH lower than 2, have maximum absorbance at 243 nm. At pH 4 above 50 % of the molecules are dissociated and has maximum absorbance obtained at 250 nm. While from pH 5 to 10 almost all vitamin C is completely dissociated [23].

Vitamin C (Ascorbic acid) is stable in solid form, but oxidized in solutions by dissolved oxygen, according to the following equation.



L-Ascorbic Acid Ascorbic free Radical Dehydroascorbate 2,3-Diketogulonic acid

The factors which effect these reactions are temperature, solvent, light, pH and metal ions (Cu^{2+}, Ag^{+} and Fe^{3+}) as reported in the literature [24, 25].

Vitamin C is water soluble vitamin, plays an important role in preventing illness such as scurvy and common cold. Tea is the frequently used drink on daily base in most of the population in the world particularly in Pakistan and can make up deficiency of the vitamin C in their body and thereby minimizing the risk of these diseases.

In the present study concentration of vitamin C was determined in various tea samples. For this purpose we categorized the tea samples into five different groups i.e., branded and unbranded black tea, unbranded green tea, lemon grass and fresh tea leaves. The mean value of vitamin C contents for the four-branded black tea samples is 164.76 mg/100 g. The results showed that the concentration of vitamin C in the sample 1 of the branded black tea is 181.74 mg/100 g and that of sample 2 is 146.08 mg/100 g. The other two values are 159.20 mg/ 100 g and 172.02 mg/ 100 g in sample 3 and sample 4, respectively Table-1. The mean value for the unbranded black tea samples is 114.92 mg/100 g. The results showed that the concentrations of vitamin C in sample 5, 6, 7, 8 and 9 are 95.56, 103.64, 120.62, 124.94, and 129.84 mg/100 g, respectively [Table-1]. The amounts of vitamin C present in tea is different due to the variety of leaf, growing environment, processing, manufacturing, particle size of ground tea leaves and infusion preparation [26]. As vitamin C is unstable to heat and light, it may decrease vitamin C contents during processing. Due to the fermentation or oxidation process of tea leaves to black tea, convert the simple flavonoids to more complex verities called theaflavins and thearubigins [27].

A special attention was focused on the role of vitamin C present in the tea samples, which has

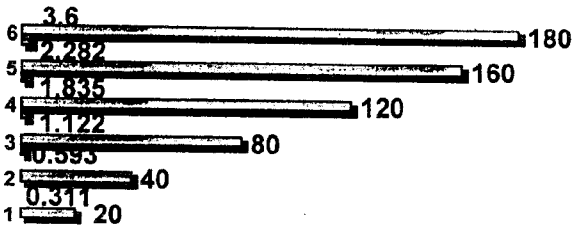


Chart No. 1: Various standards and their concentration.

excellent antioxidant properties [28, 29] and which can also influence the long-term stability of green tea catechins [30].

We also examined the vitamin C contents of four unbranded green tea samples obtained from the

Table-1: Determination of vitamin C in various tea brands

S. No.	Sample name	Concentration (mg/ 100 g)
1	Suprem^{bb}	181.74^{mbb}
2	Lipton^{bb}	146.08^{mbb}
3	Tapal^{bb}	159.20^{mbb}
4	Tetley^{bb}	172.02^{mbb}
5	Kenya plane^{ub}	95.56^{mub}
6	Kenya danidar^{ub}	103.64^{mub}
7	Bangladesh^{ub}	120.62^{mub}
8	Indonesia^{ub}	124.94^{mub}
9	India^{ub}	129.84^{mub}
10	Brazil^{ug}	193.62^{mug}
11	India^{ug}	177.78^{mug}
12	Indonesia^{ug}	182.14^{mug}
13	Vietnam^{ug}	191.68^{mug}
14	Lemon grass^{lg}	38.40^{mlg}
15	Fresh tea leaves^{ft}	187.00^{mft}

bb: Branded black.
ub: Unbranded black.
ug: Unbranded green.
lg Lemon grass.
ft: Fresh tea leaves.
mbb: Mean branded black tea = 164.76 mg/ 100 g.
mub: Mean unbranded black tea = 114.92 mg/ 100 g.
mug: Mean unbranded green tea = 186.305 mg/ 100 g.
mlg Mean lemon grass = 38.40 mg/ 100g.
mft Mean fresh tea leaves = 187.00 mg/ 100 g.

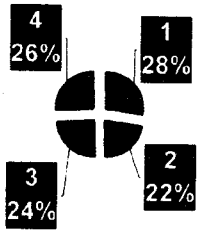


Chart No. 2: Concentration of vitamin C in Branded black tea leaves

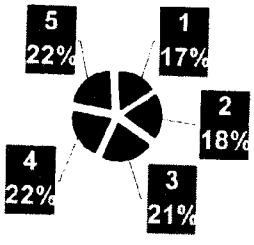


Chart No. 3: Concentration of vitamin C in unbranded black tea leaves.

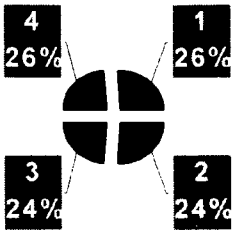


Chart No. 4: Concentration of vitamin C in unbranded green tea leaves.

open market. The mean value for green tea samples is 186.305 mg/100 g. Higher value 193.62 mg/100 g was observed in sample 10 and lower value 177.78 mg/100 g in sample 11. The concentration of vitamin C in sample 12 and 13 is 182.14 and 191.68 mg/100 g, respectively Table-1. The small amount of vitamin C in black tea may be due to its processing. The concentration of vitamin C in lemon grass is very low, 38.40 mg/100 g, showing lower concentration of vitamin C than the rest of all tea samples. The fresh tea leaves contain high amount of vitamin C, 187.00 mg/100 g, Table-1.

Green tea is manufactured by drying fresh tea leaves; therefore, its composition resembles that of fresh tea leaves contains high amount of vitamin C [31]. The processing of green tea is simple that's why it contains more vitamin C.

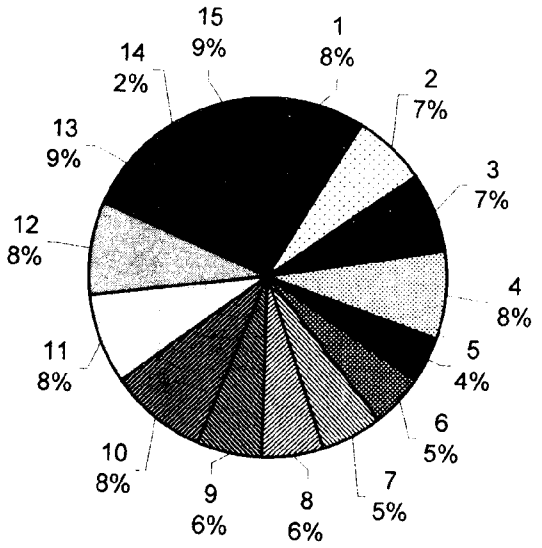


Chart No. 5: Concentration of vitamin C in various tea samples.

Experimental

Reagents

All reagents used in the present study were of analytical grade. Ethylene diamine tetra acetic acid (EDTA) and sulphuric acid were purchased from Merk BDH, ammonium molybdate AR (Winlab), oxalic acid (E. Merk, Germany) and were used without any further A USP grade vitamin C reference standard (RS) from May and Baker's Ltd., (England), and De-ionized distilled water were used throughout the investigations.

Instrument

Hitachi UV-vis Spectrophotometer model U-2000 (Japan), with a 1.0-cm optical path quartz cell were used for spectrophotometric measurements of vitamin C at a wavelength of 760 nm.

Sampling

Fifteen different tea samples were taken for vitamin C analysis. Among them 14 samples were obtained directly from the main local market in Peshawar, N.W.F.P. Pakistan, including branded as well as unbranded black/green teas, and lemon grass. The remaining one was collected directly collected from National Tea Research Center at Mansehra, N.W.F.P. Pakistan. The fresh tea leaves were dried at room temperature, powdered and analyzed for vitamin C content determination.

Preparation of stock solutions

Ammonium molybdate (5% w/v) solution

5 g of ammonium molybdate was dissolved in 25 cm^3 de-ionized water and then makeup the volume with de-ionized water in a 100 cm^3 volumetric flask.

Oxalic acid (0.05 M) - EDTA (0.02 M) solution

0.63 g oxalic acid dissolved in 20 cm^3 de-ionized water, add 0.7448 g EDTA with shaking on water bath and then make up the volume with de-ionized water in a 100 cm^3 volumetric flask.

Sulphuric acid (5 % v/v) solution

5 cm^3 of concentrated sulphuric acid (95%) was added to de-ionized water and make up the volume in a 100 cm^3 volumetric flask.

Meta phosphoric acid - acetic acid solution

Dissolve with shaking 3 g of meta phosphoric acid pellets in 20 cm^3 acetic acid on water bath and make up the volume with de-ionized water in a 100 cm^3 volumetric flask.

Standard vitamin C (0.1% w/v) solution

0.1 g of vitamin C was dissolved in 100 cm^3 of 0.05 M oxalic acid and 0.02 M EDTA solution freshly prepared.

Preparation of working Standard Solutions of vitamin C

Take 0.5, 1, 2, 3, 4 and 4.5 cm^3 of standard vitamin C (0.1% w/v) solution in a separate 25 cm^3 brown volumetric flasks and added 4.5, 4, 3, 2 and 0.5 cm^3 of oxalic acid (0.05 M) and EDTA (0.02 M) solution in each brown volumetric flask in the same manner respectively. Then added separately meta phosphoric acid - acetic acid 0.5 cm^3, sulphuric acid (5% v/v) solution 1 cm^3 and ammonium molybdate (5% w/v) solution 2 cm^3 in each brown volumetric flask and then make up the volume 25 cm^3 with de-ionized water.

Preparation of Blank solution

Without adding vitamin C (0.1 % w/v), add all other reagents in the same way as that in working standard and make up the final volume 25 cm^3.

Preparation of sample solutions

1 g of each tea sample was taken in a 25 cm^3 in Erlenmeyer flask and 10 cm^3 of 0.05 M oxalic acid and 0.02 M EDTA solution was then added. These samples were then kept for overnight.

After 24 h, the samples were filtered through 0.45 μm filter paper. From each sample 2.5 cm^3 were transferred to 25 cm^3 brown volumetric flask and to each sample 2.5 cm^3 of oxalic acid (0.05M) - EDTA (0.02 M) solution was added and the remaining reagents such as 2 cm^3 of ammonium molybdate (5% w/v), 1 cm^3 sulphuric acid (5% v/v) and 0.5 cm^3 meta phosphoric acid - acetic acid, were added in each sample in the same way. Finally make up the volume with de-ionized water in 25 cm^3 volumetric flask.

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