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Vitamin C and Phenolic Compounds in Fruit Peel and Pulp Waste

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Abstract Fruit wastes have attained considerable interest in the modern trading industry. Globally, almost 50% of the fruits agriculturally produced are processed in the beverage industry for juices, and almost 25 million tons of fruit wastes are being generated only from citrus fruits. In Pakistan, in the beverage industry, are being processed 34% of Citrus fruits, so a large volume of waste is being produced annually. The basic aim of the research work was to evaluate the potential bioactive components especially vitamin C and phenolic contents. The fruit wastes of five species of the citrus family selected for the research. Vitamin C and the Phenolic contents were estimated by High-performance liquid chromatography and Folin Ciocalteau respectively. The Grapefruit showed the highest contents of vitamin C i.e. 114.92 ± 0.36mg/100g and 110.56 ± 0.415mg/100g in pulp and peel respectively. Similarly the peel of oranges showed highest contents of phenolic compounds as compare to the peels of the other members of citrus family i.e. 155.1165 ± 0.1355mg/100g while the pulp of fruiter showed the highest concentration of phenolic compounds i.e. 119.9342 ± 0.194mg/100g as compare to the pulp of other citrus members. Due to the presence of a reasonable amount of Vitamin C and phenolic contents in fruit peel and pulp samples of citrus family, it can be concluded that the result of the present research would be fruitful for the treatment of deficiencies of these bioactive compounds.

Keywords: *RDA, GDP, GAE, UV-Visible, Nutraceutical*

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1. Introduction

Pakistan is one of those countries who produce a variety of fruits due to its rich and fertile soil and availability of all the weathers. Pakistan's agriculture produces 20.9% of its gross domestic product (GDP) and supports 67.5% of the total rural populations are being associated with agriculture as a profession [1]. Pakistan is among the top ten leading producers of citrus fruits where as Brazil is at the top position. In spite of the low yield of oranges per hectare i.e. 9.5 tons, Pakistan is the potential cultivator of 95% of the world's orange production [2]. The fruits are being used as a source of various nutrients viz. minerals, carbohydrates, vitamins, etc. The fruits are the source of vitamins that help in vision improvement [3]. Citrus peels are being a rich source of antioxidants such as phenolic contents have health promotion effects [4,5,6,7]. Vitamin C (Ascorbic acid) plays an influential role as an enzyme complement, cofactor, co-substrate, reducing agent and antioxidant in various biochemical reactions [8]. The recommended daily allowance (RDA) of ascorbic acid is 15-45mg/day for 1-13years of age, and 65-75mg/day for 14-18years of age [9]. The intake of ascorbic acid should be enhanced during infectious problems, pregnancy and lactation period [9]. The problem of scurvy is caused by the deficiency of ascorbic acid for a long time. The spectrum of scurvy is quite varied which include dental, dermatological, bone and systematic manifestation [10]. Ascorbic acid is involved in the hydroxylation of lysine and proline in protocollagen, which is most vital component for the synthesis of collagen; hence the deficiency of ascorbic acid leads to unavailability of collagens and abnormal connective tissues, resulting in the problem of scurvy [11]. The prevention of scurvy by using the citrus fruits has also been described, later on ascorbic acid was first isolated in 1928 [12]. Clinical findings develop after a period of three months of intake of diet deficient of ascorbic acid. Musculoskeletal findings show knee, ankle and wrist pain along with muscular pain and hemarthrosis. Other symptoms of oral infection include hypertrophy, bleeding of gums, and inflammation of supporting tissues of teeth, as it is inferred that teeth contribute the gateway to the microbes into the gingiva [13]. As researches show that there is a close relation between vitamin C of pulp of citrus fruits and hydrophilic antioxidant activity so pulp of

citrus fruits might be valuable [14,15]. Similarly phenolic contents are also present in various tissues of fruits in varied amounts but highest in the peels of the citrus fruits [16-19]. But the variation in the phenolic contents depends upon the location of cultivation, soil and citrus species [20]. The leaves of Citrus limon are also a good source of phenolic compounds along with flavonoid contents [21, 22] and peels and fleshes of certain vegetables such as cucumber, potato, carrot and pumpkin have free radical scavenging capacity due to the presence of phenolic contents [23] but it was also estimated that the contents of phenols were higher in the peels of ripened C. limon as compare to the peel of unripen [24]. The main aim of the present research is to evaluate the fruit wastes and to estimate the concentration of ascorbic acid as well as phenolic contents in the fruit peels and pulps which are considered as agro-waste, and to use these fruit wastes as a cheapest source of ascorbic acid and phenolic contents.

2. Materials and Methods

2.1. Materials

Raw material was obtained from various beverage industries in various areas of Punjab, Pakistan. The fruits waste was washed by distilled water carefully. Then the washed fruit wastes were dried under shade in highly hygienic conditions and then in hot air oven. The dried fruit waste was in the form of peel and pulp along with seeds. That dried sample of fruit peels and pulps were grinded to powder form. Then the powder was preserved in air tight jars and stored at 4℃ until analyzed.

2.2. Methods

2.2.1. Extraction and Analysis of Ascorbic Acid

Ascorbic acid was extracted by metaphosphoric acid and acetic acid solution method and estimated by high performance liquid chromatography. The detail of solution preparation and extraction of ascorbic acid for analysis was given below. [25] 100 ml of de-ionized distilled water and 20 ml acetic acid were taken in a 250ml volumetric flask. Then added 7.5g metaphosphoric acid to it and stir to dissolve. After dissolving process, the volume was made up to the mark with distilled water to dilute the mixture to 250 ml. Then filtered the mixture through fluted filter paper into a bottle and closed the bottle with a stopper or lid and refrigerated until use.

Standard solution of ascorbic acid was prepared by dissolving 100 mg of L-ascorbic acid in metaphosphoric acid (0.3 M) and acetic acid (1.4 M) solution at the final concentration of 1mg/ ml.

The ascorbic acid was extracted by homogenizing 10 g of the sample in solution containing 20 ml meta-phosphoric acid (0.3 M), and acetic acid (1.4 M). The mixture was taken in a conical flask and wrapped with aluminum foil and agitated at 100rpm with an orbital shaker for 15 minutes at room temperature. Then filtered the mixture through Whatman No. 4 filter paper, and 30 µl of this extract was used immediately in triplicates for HPLC analysis. To identify the peak of vitamin C on the chromatogram, the techniques; comparing the retention time and spiking test with that of L-ascorbic acid were used.

2.3. Total Phenolic Contents

The concentration of Total phenolic contents was determined by the Folin-Ciocalteau method. [25]

The principle of this method is that phenolic compounds are oxidized to phenolates by reagent at alkaline pH in a standard solution of Na_2CO_3 resulting in blue colored complex.

The solutions required for the determination of total phenolic contents are:

1. 10% w/v solution of Folin Ciocalteau
2. 7.5% w/v solution of sodium carbonate

About 1.5mℓ of above Folin-Ciocalteau was added in 300µℓ of sample, followed by the addition of 1.2mℓ of above Na_2CO_3. Then, allowed the mixture to stand in the dark for 90min. Then the readings of absorbance of blue color at 760nm on UV-Vis spectrophotometer against blank (distilled water) were determined. The total phenolic concentration (mg/mℓ) were estimated in triplicate and extrapolated from a standard curve, constructed by using Gallic acid as a standard. Then, expressed the results as Gallic Acid Equivalent (GAE).

3. Result and Discussion

3.1. Vitamin C

Table 1 and Graph 1 show the values and standard deviations for Vitamin C in pulp and peel from five different fruits.

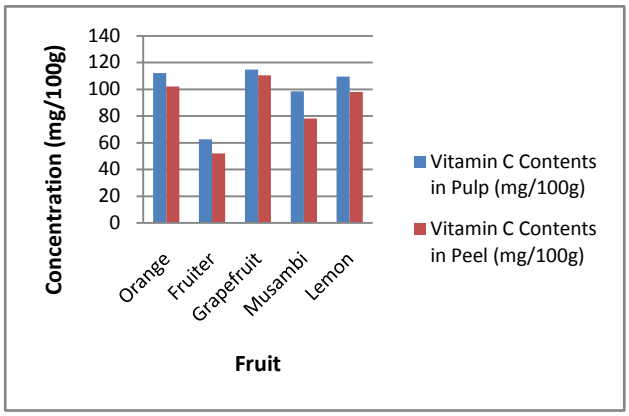
Table 1. Contents of Vitamin C in pulp and peel of Citrus Fruits

Fruit	Vitamin C Contents in Pulp (mg/100g)	Vitamin C Contents in Peel (mg/100g)
Orange	112.33 ± 0.45092	102.23 ± 0.689
Fruiter	62.58 ± 0.505	52.123 ± 0.255
Grapefruit	114.92 ± 0.36	110.56 ± 0.415
Musambi	98.63 ± 0.640	78. 164 ± 0.176
Lemon	109.61 ± 0.5	98.133 ± 0.875

The contents of vitamin C in the citrus family viz. Orange, Lemon, Grapefruit, Musambi and Fruiter were quantified as shown in Table 1 and graph 1.

Graph 1 and Table 1 reveal that pulp and peel of grapefruit have highest contents of Vitamin C as compare to the pulp and peel of other member of citrus family [26]. The result is also comparable with a research work which shows that Grapefruit and oranges have higher contents of vitamin C as compare to lemon [27]. As the Table 1 reveals that the pulp and peel of Grapefruit has highest contents of vitamin C i.e. 114.92 ± 0.36mg/100g and 110.56 ± 0.415mg/100g respectively while orange pulp and peel showed vitamin C concentration at second position with 112.3 ± 0.45092mg/100g and 102.23 ± 0.689mg/100g respectively. On the other hand, fruiter pulp and peel showed lowest content of vitamin C i.e.

62.58 ± 0.505mg/100g and 52.123 ± 0.255mg/100g respectively.



Graph 1. Contents of Vitamin C in pulp and peel of Citrus Fruits

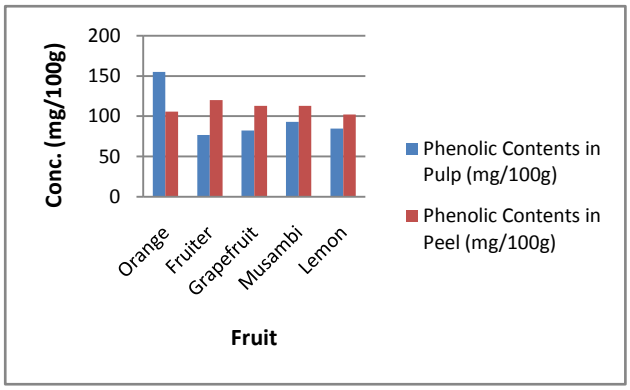
3.2. Total Phenolic Contents

Table 2 and Graph 2 show the values and standard deviations for Total phenolic in pulp and peel from five different fruits.

Table 2. Total phenolic contents in fruit wastes (pulp and peel)

Fruits	TPC Concentration (mg/100g)	
	Peel	Pulp
Orange	155.1165 ± 0.1355	105.862 ± 0.129
Fruiter	76.54385 ± 0.11835	119.9342 ± 0.194
Musambi	82.40749 ± 0.2013	112.8993 ± 0.09315
G. Fruit	92.962 ± 0.1811	112.8993 ± 0.0706
Lemon	84.75294 ± 0.22015	102.3438 ± 0.33465

The contents of phenolic compounds in the fruits of citrus family, viz. Orange, Lemon, Grapefruit, Musambi and Fruiter quantified by UV-Visible spectrophotometry.



Graph 2. Total phenolic contents in fruit wastes (pulp and peel)

Table 2 and graph 2 revealed that the peel of oranges showed higher contents of phenolic compounds as compare to the peels of the other members of citrus family i.e. 155.1165 ± 0.1355mg/100g. Similarly the pulp of fruiter showed the highest concentration of phenolic compounds i.e. 119.9342 ± 0.194mg/100g. On the other hand peel of the fruiter showed lowest total phenolic contents i.e. 76. 54385 ± 0.11835mg/100g and pulp of lemon showed total phenolic contents i.e. 102.3438 ± 0.33465mg/100g.

The results obtained in the present work revealed that the agro wastes especially peels and pulps of the citrus fruits particularly oranges, fruiters, musambi, grapefruits, and lemon contain an appreciate amount of vitamin C as well as phenolic contents. Vitamin C present in pulp is correlated with antioxidant activities [28]. These wastes are the suitable natural resources for the formulation of nutraceuticals which are recommendable as food additive over artificial food additives to overcome the deficiency of these food components. According to a research work [29] phenolic contents are a vital part of food for health and nutritional benefits. From Table 1, it was clear that inner part of the fruits i.e. pulp was rich in vitamin C contents as compare to the outer part i.e. peel of the respective fruits. But the concentration of vitamin C in the fruits which were selected by a group of researchers from Nepal was far lesser than the concentration of vitamin C estimated in the present work [30]. Similarly if we compare the result of a research work with the present work then it would be clear that the contents of vitamin C in the fruit wastes is far greater than its contents in the fresh juices [31,32] but the evaluation of vitamin C in citrus fruit by a research work showed the higher concentration as compare to previous compared work [33].

Table 2 represents that the phenolic contents were higher in concentration in the pulp parts of the fruits as compare to the peels of the respective fruits. According to a research work, the peels of Citrus grandis show high concentration of total phenolic contents [34]. This valuable concentration of bioactive compounds in the fruit wastes indicated that these fruit waste materials could be an inexpensive and ready source of vitamin c and phenolic contents which might be useful for greater oxidation power in food and pharmaceutical industries. The concentrations of the bioactive compounds in the agro wastes of fruits represented that the contribution of vitamin C in the citrus fruit parts is greater than that of the phenolic contents. In a general view, this finding concluded that the wasted parts of citrus fruits particularly peel and pulp could be considered as a best source of compounds such as vitamin C and phenolic compounds which are involved in radical scavenging properties in the living organisms.

4. Conclusion

The research work concludes that the fruit wastes, which are being considered as wastes and are much difficult to get rid of, are the potential source of many bioactive compounds such as ascorbic acid and phenolic contents. These fruit wastes exhibit more than the sufficient amounts of these compounds so it can be concluded that these fruit wastes can be used for many beneficial purposes such as for the formulation of a nutraceutical to overcome the problems caused due to the deficiency of these compounds in humans.

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