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Physicochemical, antioxidant and enzymes activities of grape fruit peel and pomace enriched functional drinks

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Abstract: Experiment was conducted to determine the proximate, minerals, antioxidant capacities and enzymes activities of grape fruit peel and grape fruit pomace along with sensorial evaluation of functional drinks. In this milieu, values of grapefruit peel and pomace powder for moisture, fat, crude protein, carbohydrate, crude fiber, ash, and NFE were recorded as 10.85±1.34,8.9±0.08 , 9.27±0.03, 7.69±0.02, 60.22±2.32, 50.33±2.1, 6.13±0.02, 6.13±0.01, 2.97±0.01 ,2.16±0.01 ,10.56±1.97, 24.97±2.4, respectively whilst in time intervals highest TPC for peel (118.66±8.9) mg/g was observed in 60 min followed by (102.33±7.6) mg/g at 90 min and (82.02±5.5) mg/g at 30 min respectively Whereas, the recorded TPC for pomace at 30, 60 and 90 minute were (112.73±9.1) mg/g has observed in 60 min followed by (97.21±7.9) mg/g at 90 min and (84.55±5.8) mg/g at 30 min respectively. Among the time intervals highest flavonoids contents of peel were at 60 min 52.3±1.9% followed by 52.51±1.7% at 90 min and minimum 50.72±1.4% at 30 min. The highest ABTS value was observed for peel content 248.33±5.6 µg/ml in ethanol extract followed by methanolic extract 212.11±4.4 µg/ml least in water extract 152.5±3.2 µg/ml. The means reviewed FRAP activity highest value for ethanol in peel and pomace were (92.66±5.3 µg/ml Fe2+/g) & (82.47±4.2 µg/ml Fe2+/g) followed by methanol (86.33±4.1 µg/ml Fe2+/g) & (76.83±3.4 µg/ml Fe2+/g) and least in water (66.46±2.2 µg ml Fe2+/g) &(54.24±2.1 µg/ml Fe2+/g) respectively. The color acceptability varied significant effect between 7.49 to 7.55 in T0 to T3. Likewise, storage imparted more significant decline from 7.72 to 7.30 at 0th to 60th days, respectively. The flavor scores were 7.59, 7.41, 7.26 and 7.53 in T0, T1, T2 and T3 respectively. The overall acceptability of drink was significantly increase from initiation (0th) day to termination (60th) day as 7.68 to 6.9.

Key words: Grape fruit; Pomace; Antioxidant indices; Enzymes activities; Functional drinks.

Introduction

Today, there is increasing demand for natural bioactive compounds as people express more concern about their health, especially in connection with health-giving diets. Epidemiological researches hint that increased dietary intake of phytochemicals, specifically polyphenols, is linked with a reduced risk of a multitude of chronic diseases. In this connection, fruits of the Citrus genus are regarded as a healthful source of bioactive compounds such as vitamins, carotenoids, fiber, and phenolic compounds (1). Agricultural citrus fruits formulation including oranges, mandarins, lemons, bergamots, limes, pummelos, and grapefruits, has greatly elevated in the last decades, reaching over 100 million metric tons per year around the globe (2). Almost a third of citrus fruits involve to synthesize fresh juice or citrus-based drinks. The citrus fruits juice yield covers half of the fruit weight, and thus a very large amount of pulp and peel waste is synthesized around the globe every year. It has been found that peels are the centered sources of polyphenols in citrus fruits (3).

Citrus fruits are enriched with essential vitamins, minerals, fibers and bioactive phytochemicals, such as alkaloids, carotenoids, nitrogenous compounds and polyphenolics. Citrus waste contains soluble sugar, starch, fiber including cellulose, hemicellulose, lignin and pectin, ash, fat and protein and many bioactive compounds. Peel residues from sweet and bitter oranges, lemons, and mandarins have proved to be main source of phenolic acids and flavonoids (4). These bioactive compounds are firmly associated with medicinal properties including antiallergenic, anti-atherogenic, anti-inflammatory, antimicrobial, anticarcinogenic, antithrombotic, cardio protective, and vasodilatory effects (5). Among citrus fruits, grapefruits are differentiated with unique sensory quality of sweet and tart taste and playing a role as an antioxidant. Grapefruits (Citrus paradisi) are medium-sized, subtropical fruit trees that belong to the family of Rutaceae. Grapefruit, a hybrid of pomelos (C. maxima) and sweet oranges (Citrus sinensis) was first discovered in the 18th century. Different varieties of grapefruits vary in hue from white to red depending on the presence or absence of lycopene (6).

Grapefruits contain several phytochemicals such as flavonoids, carotenoids, limonoids, organic acids, pectin, and folate. The main flavonoids in grapefruit are narirutin, naringin, hesperidin, neohesperidin, didymin, and poncirin. these phytochemicals have anti inflammatory, antiproliferative, anticarcinogenic, and antimicrobial properties (7). Additionally, flavonoids have characteristic presence of hydroxylgroups,which makes these compounds potent antioxidants. Optimum intake of antioxidants is positively correlated with

health benefits such as prevention of certain cancers and cardiovascular diseases (8). Grapefruit byproducts such as peel and pomace may provide a health benefit beyond the traditional nutrients they contain, as well as prevent diet-related diseases, e.g. metabolic syndrome, type II diabetes, coronary heat disease, obesity, hypertension, certain types of cancer, gastrointestinal diseases and osteoporosis (9).

Materials and Methods

Procurement of raw material

The present study was conduct in the Nutritional Lab of Institute of Home and Food Science, Government College University, Faisalabad. Grapefruit were collected from local market of Faisalabad.

Proximate analysis

Proximate analysis of grape fruit peel and pomace were carried out for moisture content, crude protein, crude fat, crude fiber, ash and NFE according to their respective methods as described in (10).

Analysis of extracts

Total polyphenols contents

The solution of gallic acid was added with various concentrations in the methanol 20, 40, 60, 80 and 100µg/ml and a standard curve is prepared. In a test tube 200 µl extract were taken along with the 150 µl diluted folin ciocalteu and 1.35 ml of distilled water. The mixture was allowed to stay for 5 minutes. 6% sodium carbonate (1.5 ml) was added in the test tube. After that the test tube was kept in the dark place where the temperature was 22°C for 60 to 90 minutes. Spectrophotometer spec Cord 200 plus UV visible was used to measure the total phenolic components of the extract at 765nm (11).

Antioxidant activity

The antioxidant activity of the extract was observed by using assay based on coupled oxidation of β carotene and linoleic acid Taga et al., (12) and Bocco et al., (13). 20 mg of β carotene was dissolved in the 400 mg tween20, 40 mg of linoleic acid and 20 ml of chloroform. In the 0.10 ml of sample, 3 ml of prepared emulsion was added after the removal of chloroform and then it was placed in the water bath for up to 120 minutes. Spectrophotometrically the oxidation of β carotene was determined at 470nm.

Free radical scavenging activity (DPPH assay)

Free radical scavenging activity was determined accordance to the platform set by Heimler et al*.,* (14). The 2 ml of extract was taken in the test tube and only 1 ml of DPPH, which was diluted with ethanol (0.025 g DPPH and 100 ml ethanol), was added and then the test tube was incubated at the room temperature for the maximum time of 30 minutes. By using the spectrophotometer, the absorbance rate was noted at 517 nm at every 3 minutes for at least 60 minutes. Following formula was used to calculate the percent inhibition.

FRSR % = 100 (AB – AA /AB)

AB= absorbance of blank sample (t = 0 min)

AA= absorbance of tested extract solution (t = 60 min)

Ferric reducing antioxidant power (FRAP)

The test of FRAP was conducted according to the method set by Rabeta & Faraniza (15). The extracts of peel and pomace were taken 0.5 ml in the test tube and mixed with the phosphate buffer 1.25ml, 0.2 M, and 6.6 pH and potassium ferricyanide 1.25 ml, 1%. Incubation was done and after that 1.25 ml of TCA which is 10% and ferric chloride 0.1% were added in the mixture and placed for at least 10 minutes at the room temperature. The absorbance of the sample was measured at the 700nm.

Functional drink

During the phase of product development, three treatments of functional drinks were prepared with different ratios of peel and pomace with the small amount of venila essences for the flavor under fully hygienic environment. Treatments were labeled. T_0 was a control for the purpose of comparison. T_1 sample was prepared by 250ppm peel only. T_2 was prepared with 500 pomace only, T_3 was prepared by combining the 250 peel and 750 pomace. The drinks were prepared without adding any artificial color and flavors.

Sensory evaluation

The GFBP was subjected to sensory evaluation by trained taste panel using nine-point hedonic scale system (9 = extremely; 1 = dislike extremely) as described by Meilgaard et al. (16). Sensory evaluation regarded attributes like color, flavor, sweetness, sourness and overall acceptability was performed. Hedonic response was judged in Sensory Evaluation Laboratory of Institute of Home and Food Sciences, Govt College University, Faisalabad.

Results and discussion

Proximate analysis of grapefruit peel and pomace

Proximate analysis of any product is key factor for evaluating the quality of raw material. Grapefruit peel and pomace powder were subjected to different quality traits and revealed moisture, fat, crude protein, carbohydrate, crude fiber , ash, and NFE as For Grapefruit peel and pomace powder the values obtained were10.85±1.34,8.9±0.08 , 9.27±0.03, 7.69±0.02, 60.22±2.32, 50.33±2.1, 6.13±0.02, 6.13±0.01, 2.97±0.01 ,2.16±0.01 ,10.56±1.97, 24.97±2.4 respectively in (Table 1). The results of the current findings regarding proximate analysis are in line with the observed variations by Ebana RUB et al. (17). They observed protein, fiber, ash, fat and moisture of the grapefruit peel from 5.5% to 4.5%, 8.0% to 7.0%, 10% to 8.0%, 2.4% to 2.0% and 6.0% to 5.0% respectively. One of the peers Edet et al.,(17) analyzed the moisture, ash, fat, carbohydrate, protein and fiber contents of Grapefruit peel sample and observed the moisture content in grapefruit peel was 11.86, fat 6.6, ash 3.9, carbohydrate 71.8, protein 10.71 fiber 7.5%. Likewise, Ali et al. (18) carried out the proximate profiling of grapefruit byproducts and revealed moisture, crude fat and ash from 6.80 ± 01, 2.50 ± 0.5 and 6.90 ± 01% respectively.

Antioxidant analysis of extracts

Antioxidant activity of GFBP can be assessed by

Table 1. Proximate results of Grapefruit peel and pomace.

Proximate	Peel Composition (%)	Pomace Composition (%)
Moisture	10.85±1.34	8.9±0.08
Crude protein	9.27±0.03	7.69±0.02
Carbohydrate	60.22±2.32	50.33±2.1
Crude fat	6.13±0.02	6.13±0.01
Ash	2.97±0.01	2.16±0.01
NFE	10.56±1.97	24.79±2.4

Table 2. Total phenolic content of grapefruit peel and pomace (mg/g).

Grape fruit peel					Grape fruit Pomace				
Solvent	**Time 30**	**Time 60**	**Time 90**	**Mean**	**Solvent**	**Time 30**	**Time 60**	**Time 90**	**Mean**
Ethanol	101±7.01	145±8.2	115±7.3	120.33±9.9	**Ethanol**	95.95±9.3	137.75±8.7	109.25±8.4	114.31±9.4
Methanol	97±9.1	115±9.7	105±9.7	105.66±9.1	**Methanol**	92.15±9.2	109.25±9.3	99.75±9.3	100.38±8.6
Water	69±6.9	96±9.6	87±8.7	84±8.4	**Water**	65.55±6.5	91.2±9.1	82.65±8.4	79.8±4.9
Mean	82±5.5	118.66±8.9	102.33±7.6		**Mean**	84.55±5.8	112.73±9.1	97.21±7.9	

Table 3. DPPH scavenging of grapefruit peel and pomace%.

Grapefruit Peel					Grapefruit Pomace				
Solvent	**Time 30**	**Time 60**	**Time 90**	**Mean**	**Solvent**	**Time 30**	**Time 60**	**Time 90**	**Mean**
Ethanol	64.82±6.2	82±8.2	71±7.1	71.66±4.8	**Ethanol**	58.28±5.8	77.08±7.7	66.74±6.6	67.36±6.3
Methanol	59±5.9	76±7.6	67±6.7	67.33±4.4	**Methanol**	55.46±5.5	71.44±7.7	62.98±6.2	63.29±5.9
Water	52±5.2	60±6.0	59±5.9	57.3±3.8	**Water**	48.88±4.8	56.4±5.6	55.46±5.5	53.58±4.2
Mean	57.66±5.3	72.66±7.3	65.66±5.9		**Mean**	54.20±5.2	68.30±6.7	61.72±5.8	

measuring total phenolic content, flavonoids, DPPH, ABTS and FRAP.

Total polyphenolic contents of grapefruit peel and pomace

The observed TPC content in peel were, 120.33±9.9,105.66±9.1,84.01±8.4 mg/g in ethanol, methanol and water, respectively. Likewise, trend was observed for pomace highest in ethanol (114.31±9.4 mg/g) followed by (100.38±8.6 mg/g) methanol and least in water (79.8±9.4 mg/g). However, in time intervals highest TPC for peel (118.66±8.9) mg/g was observed in 60 min followed by (102.33±7.6) mg/g at 90 min and (82.02±5.5) mg/g at 30 min respectively Whereas, the recorded TPC for pomace at 30, 60 and 90 minute were (112.73±9.1) mg/g has observed in 60 min followed by (97.21±7.9) mg/g at 90 min and (84.55±5.8) mg/g at 30 min respectively. (Table 2). TPC showed the total antioxidant capacity of the product that enhance its credential and therapeutic agent. The TPC estimation of current product are in line with the concluded of Chu et al. (19) and Sun et al. (20) observed total phenolic contents of grapefruit peel 13.1±0.21 mg/g. Later, Oboh and Rocha (29) carried out antioxidant properties of grapefruit peel samples through different indices like TPC DPPH assay and observed ethanol perform better as compare to other solvents. They observed the peel exhibited promising antioxidant activity traits of ethanol solvent were 1.4±0.14 and 1.8±0.08 mg/g.

DPPH scavenging activity of grapefruit peel and pomace

The observed DPPH content in peel were 72.66±7.3,65.66±5.9,57.66±5.3% at 60 min,90 min and 30 min respectively. Likewise, trend was observed for pomace highest at 60 min (68.30±6.7%) trailed by (61.77±5.8%) at 90 min and least at 30 mins (54.20±5.2%). However, among solvents highest DPPH for peel (77.66±4.8%) was observed in ethanol followed by (67.33±4.4%) methanol and (57.3±3.8%) in water respectively. Whereas, the recorded DPPH observed for pomace in ethanol were (67.36±6.3%) trailed by (63.29±5.9%) methanol and water (53.58±4.2%) respectively. Table (3)

The result of present investigation is strengthened by the carried out the DPPH activity of grapefruit peel and pomace and observed highest in peel and also notice the better performance of ethanol in their extract by Herald et al. (21) 2.5-1000 µg/mL DPPH radical scavenging activity was determined in grapefruit peel using ethanol as solvent. Later, Kedare and Singh, (22) observed DPPH scavenging of grapefruit peels were 110.98± 13.76%. Many factors can be influenced on the DPPH assay, for example, the interaction between antioxidants, reaction time and interference compounds.

Earlier Kumaran & Joel Karunakaran (23). evaluated the DPPH activity of grapefruit peel and pomace values 56.85% to 83.87% and 20.59% to 33.51% respectively. According to Alanon et al. (24) also observed DPPH scavenging of grapefruit peel 86.76±8.40 that study indicated that the extraction time, temperature and solvent are the factors responsible for variations in peel extract contents.

Flavonoids contents of grapefruit peel and pomace

The observed flavonoid contents of peel were 53.3±1.3,52.51±1.4,50.1±1.1% in ethanol, methanol and water respectively. Similar, trend was observed for pomace highest in ethanol (50.63±1.3%) followed by methanol (49.89±2.9%) and least in water (47.64±2.9%). Among the time intervals highest flavonoids content of peel were at 60 min 52.3±1.9% followed by 52.51±1.7% at 90 min and minimum 50.72±1.4% at 30 min. Likewise, in pomace highest flavonoids observed at 60 min (50.09±2.3%) follow by at 90 min (49.89±1.6%) and lowest at 30 min (48.18±1.4%) respectively in

Table (4).

The results of present research were comparable with the earlier findings of Angelon et al. (24). They observed *that* ethanolic extract showed better perform for total flavanols extraction as compare to methanol and water. One of their peers, Abou-Arab et al. (25) concluded that the flavonoids varied from variety to variety. They reported that the flavonoid contents were 455.83±3.82 mg QE/100 g whilst, the flavonoid contents for methanolic extract of were 486.67±12.83 mg QE/100 g. Moreover, Lagha Benamrouche and Madani, (26) reported that total flavonoid contents were 1.29±0.02 mg QE/g in grapefruit peel.

ABTS value of grapefruit peel and pomace

The highest ABTS value was observed for peel content 248.33±5.6 µg/ml in ethanol extract followed by methanolic extract 212.11±4.4 µg/ml least in water extract 152.5±3.2 µg/ml. Similarly, in pomace same trend was observed highest in ethanolic extract (230.95±5.9 µg/ml) followed by methanolic extract (197.16±4.2 µg/ml) whilst water extract exhibited least ABTS activity (144.46±2.2 µg/ml). Considering the time intervals, highest ABTS activity of peel and pomace were detected at 60 min by 261.33±6.4 and 243.04±7.8 µg/ml, respectively followed by 90 min by 196.66±5.4 and182.9±6.4 µg/ml respectively. However, the initial time intervals 30 min showed the least value in peel 157.66±2.3 and pomace 146.63±3.8 µg/ml mentioned in Table (5).

The results of ABTS are in harmony with the findings of Xu G et al. (27) reported 122.34 ± 6.22 µg/ml of ABTS in grapefruit peel sample. In current study the differences in extraction are due to variations in extraction temperature as they used 45°C instead of 60°C. Earlier Re et al. (28). assessed ABTS inhibition of the ethanolic and methanolic extracts of grapefruit peel were 5.1±0.32 and 3.8±0.21 (mg/ml). Later, Oboh and Rocha (29) also reported ABTS scavenging ability (6.09 mmol./TEAC g). They also reported a significant effect of time on the ABTS activity and deduced that polyphenolic yield was dependent on the solvent and extraction time. Moreover, in other study Re et al. (28) reported ABTS value 2.24 ±0.12(mM TE/100g GS) in grapefruit peel.

FRAP activity of grapefruit peel and pomace

The means reviewed FRAP activity highest value for ethanol in peel and pomace were (92.66±5.3 µg/ml Fe2+/g) & (82.47±4.2 µg/ml Fe2+/g) followed by methanol (86.33±4.1 µg/ml Fe2+/g) & (76.83±3.4 µg/ml Fe2+/g) and least in water (66.46±2.2 µg ml Fe2+/g) & (54.24±2.1 µg/ml Fe2+/g) respectively. Furthermore, for time intervals FRAP activity of peel and pomace were highest at 60 min (88.33±4.2 µg/ml Fe2+/g) & (78.61±4.3 µg/ml Fe2+/g) followed by at 90 min (79.33±4.6 µg/ml Fe2+/g) & (70.60±3.2 µg/ml Fe2+/g) and least were at 30 min (72.33±3.1 µg/ml Fe2+/g) & (64.37±2.5 µg/ml Fe2+/g) respectively mentioned in table (6).

In the current study both peel and pomace showed promising antioxidant activity. However, peel elutriated better performance as compare to the pomace. Among the solvent extraction ethanol perform better followed by methanol and water. In the time intervals 60 min perform better effect as compare to 90 and 30 min. It was concluding the grapefruit by products (GFBP) have capacity to analyzed in therapeutic agent. In the later studies González et al. (30) FRAP of the grapefruit peels were 636.94±45. whereas in some other studies the grapefruit by products exhibited strong FRAP activity by the earlier findings of Azman et al. (31) FRAP of the grapefruit peel are 1.76± 0.07(mM TE/100g GS). One of the more research Benzie et al. (32) tested FRAP value of grapefruit peel and pomace 60.30±30 and 71.57±0.60 µg/ml Fe2+/g. However, the respective fractions in extraction due to polarity of the solvent and

Table 4. Flavonoids of grapefruit peel and pomace%.

Grapefruit Peel					Grapefruit Pomace				
Solvent	**Time 30**	**Time 60**	**Time 90**	**Mean**	**Solvent**	**Time 30**	**Time 60**	**Time 90**	**Mean**
Ethanol	54.33±1.6	55.9±1.6	53.9±1.4	53.3±1.3	**Ethanol**	47.18±1.2	53.10±1.3	51.61±1.5	50.63±1.3
Methanol	54.33±1.7	53.1±1.4	50.12±1.3	52.51±1.4	**Methanol**	51.61±1.5	50.44±1.2	47.61±1.4	49.89±2.9
Water	48.16±1.6	49.2±1.3	51.7±1.8	50.1±51.1	**Water**	45.75±1.6	46.74±1.1	50.44±1.6	47.64±2.9
Mean	50.72±1.4	52.73±1.9	52.51±1.7		**Mean**	48.18±1.4	50.09±2.3	49.89±1.6	

Table 5. ABTS value of grapefruit peel and pomace µg/ml.

Grapefruit Peel					Grapefruit Pomace				
Solvent	**Time 30**	**Time 60**	**Time 90**	**Mean**	**Solvent**	**Time 30**	**Time 60**	**Time 90**	**Mean**
Ethanol	196±9.8	313±7.4	236±4.6	248.33±5.6	**Ethanol**	182.28±6.7	291.09±4.6	219.48±9.7	230.95±5.9
Methanol	154±5.7	285±7.9	197±3.4	212±4.4	**Methanol**	143.22±4.3	265.05±6.7	183.21±6.8	197.16±4.2
Water	123±4.7	186±8.9	157±2.1	152.5±3.2	**Water**	114.39±5.8	172.98±7.6	146.01±4.9	144.46±2.2
Mean	157.66±2.3	261.33±6.4	196.66±5.4		**Mean**	146.63±3.8	243.04±7.8	182.9±6.4	

Table 6. FRAP test of Grapefruit peel and pomace µmol Fe^{2+}/g.

Grapefruit Peel					Grapefruit Pomace				
Solvent	**Time 30**	**Time 60**	**Time 90**	**Mean**	**Solvent**	**Time 30**	**Time 60**	**Time 90**	**Mean**
Ethanol	82±4.2	105±6.7	91±5.0	92.66±5.3	**Ethanol**	72.98±3.4	93.45±5.4	80.99±4.9	82.47±4.2
Methanol	79±3.1	94±5.1	86±4.6	86.33±4.1	**Methanol**	70.31±4.3	83.56±4.5	76.54±3.8	76.83±3.4
Water	56±5.3	66±2.5	61±2.1	66.46±2.2	**Water**	49.84±4.2	58.74±2.2	54.29±2.7	54.29±2.1
Mean	72.33±3.1	88.33±4.2	79.33±4.6		**Mean**	64.37±2.5	78.61±4.3	70.60±3.2	

nature of the grapefruit by products fractions.

Enzyme inhibitory activity (IC_{50}) (mg protein/ml)

The DPP-IV IC_{50} values of the intact camel milk proteins (GMP), and camel protein hydrolysate are provided in Table (7). In the present study alcalase 9h (A9), followed by alcalase 6h (A6) and papain 3h (P3) generated hydrolysates displaying highest DPP-IV inhibitory activity. Similar results were reported by Nongonier and Fitz Geraland (33) where DPP-IV G50 value for commercial inhibitor diprotin was 0.001 mg/ml recorded. For instance, peptic hydrolysates from bovine caprine alpha-lactalbumin had comparatively similar DPP-IV IC50 values to hydrolysates of present study Lacroix & Li chan (34). Moreover, higher DPP-IV IC50 than the present GMPHS were observed for tryptic hydrolysates of caprine and bovine CN Zhang *et al*. (35). Postprandial glucose (PPG) level is an important control point in early treatment of diabetes. As shown in table (7). Even the intact camel milk possesses a strong and inherent PPA inhibitory activity. Similar results were reported by El et al. (36). Bioactive peptides derived from natural sources could possess highly potent enzyme inhibitory activities without any side effects (37). Pancreatic lipase is the most important enzyme responsible for digestion of dietary fat, so its inhibition can have beneficial effects in overweight and obese individuals Birari & Bhutani. (38). PPL IC50 for commercial inhibitor orlistat was found to be 0.03mg/ml by Mudgil et al. (39).

Sensory evaluation of functional drinks

Hedonic response is predictable for a product acceptance and marketability. Good sensory response certifies consumer acceptance and confidence on the developed product. The functional and nutraceutical drinks were evaluated for various sensory attributes including color, flavor, sweetness, sourness and overall acceptability.

The color acceptability varied significant effect between 7.49 to 7.55 in T0 to T3. Likewise, storage imparted more significant decline from 7.72 to 7.30 at 0^{th} to 60^{th} days respectively mentioned in table (8). The statistical analysis for flavor revealed non-momentous differences during storage and treatments. The flavor score were 7.59, 7.41, 7.26 and 7.53 in T0, T1, T2 and T3 respectively. The flavor score also affected by storage decreased from 7.53 to 7.37 at 0^{th} to 60^{th} day depicted in table (9). The highest sweetness was assigned to control (T0) on 7.23 followed by T3 (7.19),T1(7.16) and T2(7.07).The sweetness also revealed during storage from 7.28 to 7.04 at initiation (0^{th}) to at the termination (60^{th}) day revealed in table (10). Means for sourness elucidated non-significant variations from 7.38±0.04 to 7.3±0.03 in T_0 and T_3, respectively. Sourness was also decline during the storage at 0^{th} day recorded were 7.44±0.04 that reduced to at 60^{th} day 7.1±0.01 stated in Table (11). The overall acceptability of drink was significantly increase from initiation (0^{th}) day to termination (60^{th}) day as 7.68 to 6.9. The recorded overall acceptability in T0,T1, T3,T4 were 7.45,7.22,7.17,7.3 respectively mentioned in table (12).

From the results it is evident that the grapefruit by products application did not imparted any decline effect to the product. The acceptable color value of polyphenol-based product is well documented in the study of Igual et al. (40) they described that grapefruit peel polyphenols impart darker color to the drink due to the presence of coloring pigment.In general, the color parameters of frozen-stored functional drink slightly changed during the 2 months, as was also observed by Shim and Kim (41). Mishra et al. (42) documented non-significant differences in the flavor and overall acceptability of ascorbic acid enriched functional drink during storage. However, the color affected significantly with storage and treatment. The physical properties of grapefruit peel functional drink were significantly affected by conventional heating treatment. In any case, the color changes were considered non-perceptible from the sensory point

Table 7. Enzyme inhibitory activity (1C50) (mg protein/ml).

Samples	DPP-IV	PPA	PPL
Peel ethanol	1.05±0.03	0.014±0.02	0.054±0.03
Peel methanol	0.78±0.02	0.010±0.01	0.022±0.02
Peel water	0.42±0.01	0.004±0.06	0.009±0.04
Pomace ethanol	0.23±0.01	0.009±0.07	0.031±0.05
Pomace methanol	0.14±0.01	0.006±0.06	0.012±0.01
Pomace water	0.02±0.01	0.002±0.03	0.07±0.02

Table 8. Color of the functional drinks.

Days	Treatment				
	T0 control)	**T1(peel)**	**T2(pomace)**	**T3 (peel pomace)**	**Mean**
0	7.74±0.05	7.70±0.02	7.72±0.02	7.75±0.01	7.72±0.02
30	7.50±0.04	7.54±0.04	7.52±0.01	7.56±0.03	7.53±0.04
60	7.25±0.05	7.34±0.05	7.28±0.01	7.36±0.0.1	7.30±0.014
Mean	7.49±0.01	7.52±0.06	7.50±0.02	7.55±0.06	

Table 9. Flavor of the functional drinks.

Days	Treatments				
	T0 (control)	**T1(peel)**	**T2(Pomace)**	**T3(peel, pomace)**	**Mean**
0	7.70±0.07	7.46±0.04	7.33±0.03	7.65±0.06	7.53±0.05
30	7.60±0.06	7.42±0.04	7.25±0.03	7.51±0.05	7.44±0.04
60	7.48±0.04	7.35±0.03	7.20±0.02	7.45±0.04	7.37±0.03
Mean	7.59±0.03	7.41±0.01	7.26±0.04	7.53±0.09	

Table 10. Sweetness of the functional drinks.

Days	Treatments				
	T0(control)	**T1(peel)**	**T2(pomace)**	**T3(peel, pomace)**	**Mean**
0	7.35±0.03	7.28±0.02	7.20±0.02	7.31±0.03	7.28±0.08
30	7.26±0.02	7.16±0.01	7.06±0.01	7.22±0.02	7.15±0.07
60	7.10±0.01	7.05±0.01	6.95±0.05	7.06±0.06	7.04±0.03
Mean	7.23±0.05	7.16±0.06	7.07±0.08	7.19±0.07	

Table 11. Sourness of the functional drinks.

Days	Treatments				
	T0(control)	**T1(peel)**	**T2(pomace)**	**T3(peel, pomace)**	**Mean**
0	7.6±0.03	7.4±0.04	7.29±0.02	7.5±0.03	7.44±0.04
30	7.36±0.03	7.24±0.02	7.08±0.04	7.25±0.02	7.23±0.03
60	7.2±0.02	7.13±0.01	6.92±0.01	7.15±0.05	7.1±0.01
Mean	7.38±0.04	7.25±0.05	7.09±0.04	7.3±0.03	

Table 12. Overall acceptability of functional drinks.

Days	Treatments				
	T0	**T1**	**T2**	**T3**	**Mean**
0	7.85±0.08	7.61±0.03	7.55	7.69±0.06	7.68±0.02
30	7.37±0.01	7.15±0.06	7.09	7.22±0.05	7.21±0.05
60	7.14±0.09	6.92±0.01	6.87±0.02	6.99±0.04	6.98±0.04
Mean	7.45±0.03	7.22±0.04	7.17±0.07	7.30±0.06	

of view. Later, Chen et al. (43) also reported non-momentous differences for sensory traits in grapefruit peel drink storage.

The research results have proven that grape fruit peel and grape fruit pomace are not only a rich source of polyphenols but also show enzyme activities. The resultant powders show significantly higher antioxidant properties (total polyphenols concentration and antioxidant capacity). Functional drinks show higher sensory attributes during storage intervals. Functional drinks include better product quality characteristics (physicochemical, and sensory quality), enhanced phytochemical profile, and improved storage stability. The developed drinks could be promoted as a nutraceutical product with multiple benefits to the consumers.

Conflict of interest

There is no conflict of interest among authors.

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