

Evaluating health benefits of wheat flatbreads with different fruit peels; determination of anti-oxidant, nutritional value and sensory characteristics

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ABSTRACT

Background: Fruit peels including pomegranate, orange and lemon are rich sources of antioxidant and other bioactive compounds. These fruit peels can be utilized for many health benefits. **Methods**: This descriptive analytical study was carried out in Department of human nutrition, University of Agriculture, Peshawar from 2018-2019. The aim was to determine the nutritive value, antioxidant activity and sensory characteristics of wheat flatbread incorporated with pomegranate, orange and lemon peels. The incorporated flatbreads (chapatti/roti) were made by replacing wheat flour with 5% of peels powder of each fruit. **Results**: The data revealed that addition of fruit peels powder increased the moisture, ash, fats and fiber contents but reduced the protein and carbohydrate contents of the flatbreads. The minerals analysis showed that lemon peel incorporated flatbread was relatively superior in iron content only, whereas, the pomegranate had relatively lower minerals content except magnesium. The total phenolic content and antioxidant activity of flatbreads was also increased with incorporation of peels powder. All the sensory attributes of all the incorporated flatbreads was also increased to get potential nutritional and health benefits of the peels. This can be utilized for health benefits in variety of diseases.

Keywords: Antioxidants, Antioxidant Activity, Fruits, Health Benefits, Wheat Flour

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Introduction

The antioxidant capacity of food can be used as an indicator of the beneficial effects on human health.¹ Antioxidants, such as flavonoids, phenolic acids, vitamin C, vitamin E and tannins, have different biological properties including anti-carcinogenic, antiatherosclerotic effects and reduction in coronary diseases. Foods with antioxidant properties are considered as good quality, valuable and anti-aging foods.² Fruit peels that become wastes, consequently add to the current severe pollution problem. It has become difficult to properly dispose-off the fruit wastes, which is produced largely by food processing industries and at household level as well.

It has been observed that fruit peels are natural sources of many important nutritional substances such as antioxidants, dietary fiber, carbohydrates and anti-carcinogenic agents.³ It is, therefore, possible to utilize fruit peels as dietary fiber and antioxidant food additive to produce valuable food products for human being.

The peel content of the pomegranate fruit is about 50% of the total fruit weight. The pomegranate peel is gaining popularity due to its potential health benefits.



It is an important source of minerals especially, sodium, calcium, phosphorus, potassium and magnesium. It has complex polysaccharides and high levels of different ranges of bioactive compounds such as phenolic, proanthocyanidin, flavonoids and ellagitannin.3 Due to presence of these bioactive compounds, the pomegranate peel has higher antioxidant activity as compared to the pulp extract.⁴ Orange peel is a rich source of vitamin C, fiber, and many nutrients, including phenolics and flavonoids. It has been used as a traditional medicine in certain parts of the world for relieving stomach discomfort, skin inflammation, ringworm infections, aiding in neuro protection and improving heart health. A number of antioxidants have been found in citrus peels, which are involved in metal chelation and free radical scavenging activities. It is needed to find the active phytochemicals in peel of oranges because reactive oxygen species are very beneficial for many diseases such as, neurodegenerative diseases, cardiovascular dysfunction, cancer and in process of ageing.⁵ Lemon peels demonstrate a wide spectrum of biological anti-diabetic, actions including antibacterial, antifungal, anticancer and antiviral activities. There are two layers in a lemon peel. The outermost layer is called as zest, which contains essential oils including primarily limonene (90%), citral (5%) and small amounts of alphaterpincol, cintronelene, gernanyl acetate and linayl along with B complex vitamins. Its use could dissolve gall stones. Also, it has anticancer properties.6

Nowadays the oxidative stress is one of the serious problems. The air pollution, smoking, UV radiation and poor lifestyle lead to cells' damage and, ultimately, many diseases. The addition of antioxidants to foods is required to prevent oxidative stress. Certain fruit peels such as pomegranate, orange and lemon are attractive sources of natural antioxidants. The quality of locally prepared and consumed flatbread (called as roti or chapatti) can be nutritionally improved by incorporation with different fruit peels. Therefore, the current study was designed to examine the nutritive, anti-oxidative and organoleptic characteristics of wheat flatbreads incorporated with pomegranate, orange and lemon peels' powder.

Methods

This descriptive analytical study was carried out in the Department of human nutrition, University of Agriculture, Peshawar from 2018-2019. The study was approved by ethical review board of The University of Agriculture via letter FNS – ECHSRB dated 5th January 2018. Fresh fruits of pomegranate, orange and lemon were purchased from the fruit shops of nearby market. Wheat flour (extraction rate: 82%) was also procured in bulk from the local market for preparation of locally consumed wheat flatbread (**roti/chapatti**).

The procured fruits were first properly washed with tape water and peeled off. The pomegranate peels were dried in oven at 50 °C for 48 hrs. Similarly, orange and lemon peels were dried out in an oven for about 24 hours at 50°C. The dried peels were grinded in the laboratory grinder to bring in powder form. The peels' powder of pomegranate, orange and lemon were put in separate pre-labeled and air tight plastic containers. The plastic containers were wrapped with aluminum foil for further analysis.

Wheat flour was incorporated with a specific quantity of each of the fruit peels' powder on dry weight basis i.e. 5% of the wheat flour in a sample was replaced with each fruit peel powder separately. The formulations of the control and incorporated wheat flour samples are presented in Table 1. The control and incorporated wheat flour samples were kneaded with equal volume of water (80 ml) and, finally, flatbreads (Roti/Chapatti) were made for further analysis.

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Ingredients	Control flatbread	Pomegranate flatbread	Orange flatbread	Lemon flatbread						
Wheat flour	100 g	95g	95g	95g						
Pomegranate peel powder	0 g	5 g	0 g	0 g						
Orange peel powder	0 g	0 g	5 g	0 g						
Lemon peel powder	0 g	0 g	0 g	5 g						
Water	80 ml	80 ml	80 ml	80 ml						

Table 1. Formulation of control and incorporated flatbreads

Moisture, ash, crude protein, fat and crude fiber of the samples were determined according to the standard procedures of AOAC (2003). Carbohydrates content was estimated as under: Carbohydrates (%) = 100 - sum of the percent content of the moisture, ash, crude protein, fat and crude fiber. One gram of dried sample was placed in eppendorf centrifuge tubes with 1% HCL solution. These tubes



were placed in water bath shaker for 2 hours in dark. Then the samples were centrifuged at 3500 rpm for about 20 minutes. Supernatant was collected further analysis.

Antioxidant activity was determined by DPPH (**2**, **2diphenyl-1-picrylhydrazyl**) method. First, 0.1M DPPH stock solution was made and placed in dark to avoid exposure of light. Then, 2.85 ml DPPH solution was taken in another flask from stock solution and 150 μ l of sample extract was added. The solution was then allowed in the dark for 30 minutes. Then, the absorbance was measured by using

spectrophotometer at 517 nm wavelength. The results were expressed as the percent inhibition (% I) of antioxidant activity. Antioxidant activity was then measured as under:

$I (\%) = [(A_c - A_s)/A_c] \times 100$

Total phenols were determined by using Folin-Ciocalteu reagent method. The Folin-Ciocalteu reagent was first diluted 10 times with Milli-Q water. Then 0.2 ml of sample extract was added to 0.8 ml of Folin-Ciocalteu reagent (diluted). After 2-3 minutes, 2 ml of 15% (w/v) sodium carbonate solution was added. By adding Milli-Q water, the mixture was made upto 5 ml. By using spectrophotometer, the absorbance was measured at 760 nm against blank (Milli-Q water). Gallic acid was used as standard (0 - 0.5 mg/ml) and results were expressed as mg of gallic acid equivalent (GAE) per g of sample.

Mineral (Zn, Fe, Cr, Ca, Mg, K, Na) contents of control and incorporated flatbreads were determined. These minerals were determined through atomic absorption spectrometry. Samples were prepared by using the procedure of AOAC (2000). 1 gram dried samples placed in digestion tubes. Ten ml of nitric acid was added to the tubes and left for overnight. Then 4-5 ml of per chloric was added to it and digestion was carried out.

Sensory evaluation of the control and incorporated flatbread samples was performed by 50 consumerbased untrained subjects of both genders. The 9-points hedonic scale was used for sensory evaluation (Jones et al., 1955). All the four flatbreads including control flatbread, pomegranate flatbread, orange flatbread, and lemon flatbread were evaluated for texture (in mouth), color, appearance, taste and overall acceptability.

The collected data was statistically analyzed using Statistical Package for Social Sciences (SPSS, 2011; version 20). The data of different variables was presented as mean \pm standard deviation (SD) of duplicate observations. One-way analysis of variance (ANOVA) with LSD was used for determining significant differences among means at p <0.05. For sensory evaluation, One-way ANOVA followed by posthoc analysis was used to determine the difference among mean values at p < 0.05.

Results

The proximate composition of samples is shown in Table 2.The data revealed that addition of fruit peels powder increased the moisture, ash, fats and fiber contents but reduced the protein and carbohydrate contents of the flatbreads. The results showed a significant increase (p < 0.05) in moisture content of all the sample and control flatbreads. By comparing the flatbreads, it was observed that only the lemon bread had significantly higher moisture content as compared to control and other flatbread samples (p < 0.05). No significant difference was found in ash content of all the three sample flatbreads with control bread (p >0.05). Protein content was highest in lemon flatbread followed by orange and pomegranate flatbreads, respectively. All the incorporated flatbreads had significantly higher fat content than the control (p <0.05). The pomegranate incorporated flatbread held the first position in fat content. The fiber content was highest in lemon incorporated flatbread as compared to all other flatbreads (p < 0.05). The comparative flatbread samples analysis of showed that pomegranate flatbread had significantly the highest carbohydrate content followed by orange and lemon peel incorporated flatbreads (p < 0.05).

	Proximate composition (%) §					
Samples	Moisture	Ash	Protein	Fat	Fiber	Carbohyd rate †
Wheat (control)	46.1 ± 0.1^{b}	1.8 ± 0.4^{cd}	9.7±0.2 ^a	2.8±0.4 ^{cd}	2.1±0.1e	37.9±0.5 ^h
Pomegranate	45.7 ± 0.1^{b}	1.3±0.4 ^d	5.9 ± 0.1^{i}	4.1±0.1 ^c	1.8 ± 0.4^{ef}	41.4±0.3 ^d
Orange	46.8 ± 0.6^{b}	2.1±0.1 ^{cd}	6.4±0.1 ^d	6.8±0.4 ^b	0.8 ± 0.4^{f}	37.3±0.6 ^h
Lemon	49.2± 0.1 ^a	1.8 ± 0.4 ^{cd}	7.0±0.1 ^h	6.5±0.1 ^b	5.8 ± 0.4^{d}	29.8±0.1e

Table 2: Proximate composition of wheat bread and incorporated flatbread samples*

* Each percent value is the mean ±SD of two independent measurements.



Values in the same column with different letters show statistically significant difference between the means at p < 0.05 tested by one-way ANOVA with LSD. Carbohydrates content was estimated by subtracting the sum of moisture, ash, protein, fat and fiber contents from 100.

The result of total minerals content is presented in Table 3. Minerals analysis reveal that lemon peel

incorporated flatbread showed highest amount of calcium, magnesium, sodium and chromium whereas, iron was mainly found in orange incorporated flatbread. The pomegranate incorporated flatbread had relatively lower minerals content except magnesium.

Samplas		Minerals content (mg/L) §					
Samples	Iron	Zinc	Calcium	Magnesium	Sodium	Potassium	Chromium
Wheat (control)	0.40 ± 0.00^{cd}	0.21±0.01 ^c	4.0±0.0 ^h	2.9±0.02 ^b	1.1±0.0g	43.1±0.7 ^b	0.15 ± 0.01^{b}
Pomegranate	0.35 ± 0.07^{d}	0.21±0.01 ^c	24.5±0.0f	2.9±0.01 ^b	1.5±0.0 ^d	42.6±1.0 ^b	0.11±0.00 ^{cd}
Orange	0.60 ± 0.00^{b}	0.21±0.01 ^c	53.2±0.0 ^d	2.8±0.01°	1.4 ± 0.0^{e}	44.6±2.9 ^b	0.13 ± 0.00^{bc}
Lemon	0.50±0.00 ^c	0.21±0.01c	54.0±0.0c	2.9±0.02b	2.0±0.0c	44.3±2.6 ^b	0.17±0.01ª

Table 3: Minerals content of wheat bread and incorporated flatbread samples *

* Each value is the mean±SD of two independent measurements.

Values in the same column with different letters show statistically significant difference between the means at p < 0.05 tested by one-way ANOVA with LSD.

The data pertaining to antioxidant activity and phenolic content is presented in Table 4. The incorporated flatbreads had significantly higher antioxidant activity than the control bread (p < 0.05). The comparative analysis of incorporated flatbreads showed that both the orange and lemon peels incorporated flatbreads had no significant difference with each other in antioxidant activity (p > 0.05); but, both had significantly higher antioxidant activity than the pomegranate peel incorporated flatbread (p < 0.05).

Table 4 also indicated that all the incorporated flatbreads had significantly higher phenolic contents than the control bread (p < 0.05). The comparative analysis of incorporated flatbreads revealed that both the orange and lemon peels incorporated flatbreads had no significant difference with each other in phenolic contents (p > 0.05); but, both had significantly higher phenolic contents than the pomegranate peel incorporated flatbread (p < 0.05).

Table 4: Antioxidant activity and Total phenolic content of wheat bread and incorporated flatbread samples

Samples	DPPH (µmol T.E./100 gm) *	Total Phenols (mg GAE/100 g) *	
Wheat (control)	214.3±3.0 °	27.1±1.5 g	
Pomegranate	238.5±3.8 d	34.6±1.1 ^f	
Orange	284.6±4.2 °	38.2±2.2 ^e	
Lemon	275.6±6.0 °	38.7±1.1 e	

* Each value is the mean \pm SD of two independent measurements. The different letters with values indicate the significant difference (p < 0.05) in means, which was tested by one-way ANOVA with LSD.

Results of the sensory evaluation in Table 5 showed that lemon incorporated flatbread was mostly liked by the panelists and got maximum score in texture, taste and overall acceptability. The orange incorporated flatbread stands second by sensory attributes. Whereas, pomegranate incorporated flatbread failed to improve sensory attributes of wheat bread

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	Sensory score *§				
Flatbread Samples	Appearance	Texture	Taste	Overall acceptability	
Wheat (control)	8.0±1.0 ^a	6.9±1.0 ^b	7.2±1.0 b	7.5±0.9 ^b	
Pomegranate	6.5±1.2 °	6.1±1.1 °	5.9±0.9 °	6.0±1.0 c	
Orange	7.7±1.0 ab	7.1±0.9 b	7.8±0.9 a	7.8±0.9 ab	
Lemon	7.3±1.2 ^b	7.6±0.9 a	8.1±0.8 a	8.0±0.8 a	

* Each value represents the mean score of 50 panelists.



Values in the same column with different superscripts shows statistically significant difference between the means at p < 0.05 tested by one-way ANOVA with tukey.

Discussion

Oxidative stress has been extensively explored worldwide. Generally, it is believed to be harmful because the free radicals attack bio-molecules such as proteins, lipids and DNA. Therefore, oxidative stress can be more properly defined as "a state of imbalance between oxidative forces (free radicals) and their stabilizing agents (the antioxidant)". More simply, oxidative stress is the condition when levels of free radicals largely exceed the antioxidants in body. 7 Oxidative stress has been linked to a number of lifestyle-related diseases, such as diabetes mellitus, atherosclerosis. ischemic diseases, hypertension, Alzheimer's disease, malignancies, age-related macular degeneration and chronic obstructive pulmonary disease.8 A substantial dietary intake of antioxidants has proven to be effective in preventing oxidative stress and the related health hazards.9

The pomegranate peels contains multiple ingredient that has a variety of health benefits not limited to antibacterial, anti-cancer, cardio protective, and effective in many other metabolic disorders. Punicalagins found in pomegranate peel has shown to have antioxidant and anti-inflammatory properties.³ Ellagic acid and catechins has been effective in treating neurodegenerative disorders cancer, and cardiovascular diseases. Furthermore, Gallic acid, Ellagic acid have strong anti-oxidant properties.¹⁰ Thus the addition of pomegranate peels in food can be effectively use to reduce oxidative stress in human body, and subsequently will minimize the risk of development of metabolic disorders and cancer.

The citrus fruits are known worldwide for its antioxidant properties, nutritional values and its flavors. Many studies have been published that shed light on the anti-oxidant and free radical scavenging properties of citrus peels.^{11, 12} literature shows that citrus peels particularly orange and lemon contains high quantities of phenolic compounds, dietary fibers and phytochemical agents which have proven anti-oxidant and anti-inflammatory properties and is effective in treating obesity, diabetes, dyslipidemia and cancer.¹³ In our study we the anti-oxidant activity, total phenolic and mineral content of lemon, orange and pomegranate peels have been identified. Furthermore, addition of these peels in diet enhances the overall acceptability as compared to normal wheat. Thus fruits peels can be utilized to improve overall health, as an add-on to treat variety of disease and most importantly will reduce pollution.

Conclusion

It is therefore concluded that highly nutritious wheat flatbread can be prepared by incorporation of different fruit peels powder for the production of highly nutritious food products and these products will also have role in prevention of many chronic diseases.

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