BIOCHEMICAL PROPERTIES OF MAIZE BRAN WITH SPECIAL REFERENCE TO DIFFERENT PHENOLIC ACIDS

Muzzamal Hussain\(^a\), Aiza Qamar\(^b\), Farhan Saeed\(^c\), Rizwan Rasheed\(^d\), Bushra Niaz\(^a\), Muhammad Afzaal\(^b\), Zunaira Mushtaq\(^b\), and Faqir Anjum\(^d\)

\(^a\)Department of Food Science, Government College University, Faisalabad, Pakistan; \(^b\)Department of Human Nutrition & Dietetics, Riphah International University Lahore, Faisalabad, Pakistan; \(^c\)Department of Botany, Government College University, Faisalabad, Pakistan; \(^d\)Administration Department, University of the Gambia, Serrekunda, Gambia

ABSTRACT

Maize bran is a milling by-product having many nutritional and functional properties. Functional properties are attributed to the presence of some bioactive compounds. Among those, ferulic acid is a natural phenolic compound and abundantly present in maize bran. It has numerous advantages and generally has nutraceutical properties in many food products. In the current research, the nutritional & bioactive profiles of two different maize brans (PEARL-11 and FSH-810) were explored with special reference to different phenolic acids. Moreover, the alkaline-hydrolysis method was used to extract ferulic acid from maize brans using NaOH and then extracted by precipitating with ethanol. The extraction method was optimized using two different concentrations (0.5 M NaOH and 1 M NaOH) at 40°C & 60°C, respectively, and was analyzed to evaluate the optimization effects on extraction yield. Furthermore, obtained extracts were analyzed by High-Performance Liquid Chromatography (HPLC). Results showed that maize bran (PEARL-11) contains higher dietary fiber, total phenolic and flavonoid contents as compared to FSH-810. Moreover, DPPH and FRAP contents ranged from 78 to 86% and 26.8 to 22.6 mg TE/100 g in FSH-810 and PEARL-11, respectively. HPLC results showed maximum amount of ferulic acid (3.89 ± 0.07 mg/g) in maize bran (PEARL-11) at these conditions (0.5 M NaOH concentration; 60°C hydrolysis temperature) whilst other phenolic acids such as diferulic and \(p\)-coumaric acid were 0.79 ± 0.012 and 0.42 ± 0.003 mg/g at 0.5 M NaOH concentration and 60°C hydrolysis temperature, respectively. Furthermore, the extraction yield of ferulic acid from maize bran (PEARL-11) of NaOH concentration (0.5) with hydrolysis temperature (60°C) was increased. Conclusively, it is observed that maize bran is the cheapest and naturally rich source of different phenolic acids (especially ferulic acid) among other cereal bran.

INTRODUCTION

Maize is a leading cereal with high production and productivity worldwide.\(^[1]\) Maize kernel contains numerous phytochemicals especially phenolic acids that are higher in quantity than other cereals.\(^[2]\) After the milling, maize bran (60–70 g/kg) was obtained as a by-product.\(^[3]\) Maize bran is a low-priced source of dietary fiber and natural antioxidants at the food level. It offers essential functional...
properties for products with low calories.\cite{4} It is physically consisting of pericarp, testa and aleurone layer remaining endosperm tissues.\cite{2,5} It also contains 22% cellulose, 10 to 14% protein, 9 to 24% starch, 5% phenolic acids (mainly ferulic and di-ferulic acid), 2% ash and 2–3% lipid.\cite{6}

The most dominant group of phenolic compounds is mainly present in the maize bran cell wall.\cite{2} These phenolic compounds and other natural antioxidants present in maize bran act as free radical scavengers as well as metal chelating agents. Ferulic acid is a natural phenolic antioxidant compound chemically known as (4-hydroxy-3-methoxycinnamic acid). In maize bran, the bioavailability of ferulic acid is higher among the other cereals.\cite{7} Chemically, ferulic acid dimers and trimers are crosslinked with arabinoolans, mainly present within the cell walls. Maize bran is also comprised of higher ferulic acid contents that are linked to hydroxyl via an ester linkage to arabinosyl units.\cite{2} Maize bran phenolic compounds especially ferulic acid is mostly bound to cell wall components.\cite{7} These compounds have different physiological characteristics, together with antioxidant, anti-inflammatory, antiallergic, antimicrobial, antiaging, anticancer, antiviral and free radical scavenging actions and also inhibits coronary disease, liver cholesterol, lowers serum, and enhances sperm viability.\cite{2} Moreover, these phenolic compounds play a crucial role in the rigidity of the cell wall and the formation of other important secondary metabolites including vanillin, sinapic acid, coniferyl liquor, diferulic acid and curcumin.\cite{8}

Ferulic acid is widely utilized in the food manufacturing, medicinal and cosmetic industries.\cite{9} It is a less toxic phenolic compound and can be easily metabolized in the human body. It can be used to preserve foods because of its antioxidant and antimicrobial activities.\cite{2} In many countries, ferulic acid has been approved as a food additive and used as a natural antioxidant in foods, beverages and cosmetics. Several physiological functions of ferulic acid have been identified such as antioxidant, anti-inflammatory, anti-thrombosis, antimicrobial, and anti-cancer activities. In many pharmaceutical industries, ferulic acid was commercially produced by natural sources.\cite{2,9} Maize bran is a rich source of phenolic compounds, that having effects on several chronic disorders.\cite{10} Ferulic acid is released through the action of Feruloyl esterase from the maize bran. It is an active natural antioxidant with potential applications in the food and pharmaceutical industries.\cite{11} The alkaline extraction method was also used for the extraction of ferulic acid and its dehydrodimers from maize bran. The current research work was performed to explore the biochemical composition & antioxidant activity of maize bran and also optimized the extraction method of ferulic acid through optimization of NaOH concentration and hydrolysis temperature.

**MATERIAL AND METHODS**

*Procurement of raw material*

Two different varieties (PEARL-11 and FSH-810) of maize brans were procured from Maize Research station, Ayub Agricultural Research Institute (AARI) Faisalabad. Mega-zyme Total Dietary Fiber assay kit (K-TDFR-100A) was procured from Bray Business Park Bray, Co. Wicklow A98 YV29, Ireland and chemicals were procured from the scientific stores of Faisalabad-Pakistan.

*Biochemical composition*

Maize brans samples (PEARL-11 and FSH-810) were evaluated for proximate composition such as moisture, crude fat, crude protein, ash, crude fiber, and NFE (nitrogen-free extract) by their respective methods as shown below.

Total dietary fiber contents (soluble and insoluble) were also determined by their respective methods. Antioxidant activity of maize bran samples was also evaluated by phenolic acids contents, flavonoids contents, DPPH (2,2-diphenyl-1-picrylhydrazyl) and FRAP (ferric reducing antioxidant power).
**Moisture content**

Moisture content was estimated by following the procedure described in AACC\textsuperscript{12} method No. 44–15.02, and maize brans (PEARL-11and FSH-810) were evaluated for moisture content using a hot air oven at a temperature of 105 ± 5°C.

**Ash content**

Ash content of maize bran determined by following method no. 08–01.01 in AACC.\textsuperscript{12} Oven-dried samples were taken in pre-weighed crucibles and charred on a burner until incineration in the muffle furnace, temperature of 550°C was held until the sample was transformed into grayish-white residue. The residue was cooled at room temperature then weighed. The percentage of ash content was calculated by the following expression.

\[
\text{Ash} \% = \frac{\text{Ash weight}}{\text{Sample weight}} \times 100
\]

**Crude protein content**

The nitrogen content of maize bran sample was determined by using Kjeldahl method as defined in method No 46–10.01 of AACC.\textsuperscript{12} The percentage of nitrogen was determined with the following expression:

\[
\text{Nitrogen} \% = \frac{\text{Amount of 0.1 N H}_2\text{SO}_4 \times 0.0014 \times 250 \times \text{Wt. of the sample}}{\text{Wt. of bran sample}} \times 100
\]

The percentage of protein was estimated by multiplying % nitrogen with a factor of 5.7

**Crude fat**

According to AACC\textsuperscript{12} method no. 30–10.01, the soxhlet apparatus was used for evaluating crude fat. For this reason, 5 g of bran with petroleum ether was extracted at a concentration rate of 2–3 drops/sec. at 8 hours. The residue of the extraction flask was dried at 100°C for 30 min after ether distillation. Fat content was estimated by the formula given below.

\[
\text{Crude fat} \% = \frac{\text{Wt. of ether extract}}{\text{Wt. of bran sample}} \times 100
\]

**Crude fiber**

The fiber content was evaluated by removing fat and moisture from the sample that was digested with 1.25% H\textsubscript{2}SO\textsubscript{4}, then treated with 1.25% NaOH solution. Fiber content was estimated through the Fibertech machine. After charring, the sample was put into a muffle furnace at 550°C till white ash was left. Crude fiber value was estimated by AOAC,\textsuperscript{12} technique no. 32–10.01. The fiber content was measured according to the formula:

\[
\text{Crude Fiber} \% = \frac{\text{Wt. of reduces left} - \text{Wt. of Ash/Fresh sample wt.}}{\text{x 100}}
\]

**NFE (Nitrogen free extract)**

NFE Content was estimated as per the given expression:

\[
\text{NFE} = 100 - (\text{Moisture} \% + \text{Crude Fat} \% + \text{Crude Protein} \% + \text{Crude Fiber} \% + \text{Ash} \%)
\]

**Dietary fiber analysis**

Maize bran samples (PEARL-11and FSH-810) were analyzed for total, soluble and insoluble dietary fiber according to AACC\textsuperscript{12} method No. 32–07.01 and 32–20.01 by using Mega-zyme Total Dietary Fiber assay kit (K-TDFR-100A).
**Total phenolic content**

The total phenolic content of maize bran samples (PEARL-11 and FSH-810) was evaluated using the method defined by Chaovanalikit and Wrolstad.[13] Total phenolic content was spectrophotometrically analyzed using the Folin–Ciocalteu colorimetric method. A calibration curve was constructed using gallic acid (0–36 mg/100 mL), and the absorbance was measured at 750 nm. The concentration of total phenolic compounds was expressed as mg of gallic acid equivalents (GAE) per 100 g.

**Total flavonoid content**

The total flavonoid content of maize bran samples (PEARL-11 and FSH-810) was determined by using the protocol described by Sultana, Anwar & Przybyski.[14] 1 mg sample containing 0.1 g/mL of dry matter was placed in a 10 ml volumetric flask, then 5 ml of distilled water was added followed by 0.3 mL of 5% NaNO₂. After 5 min, 0.6 ml of 10% AlCl₃ was added. After another 5 min, 2 ml of 1 M NaOH was added and volume was made up with distilled water. The solution was mixed and absorbance was measured at 510 nm.

**Antioxidant potential**

The antioxidant potential of maize bran (PEARL-11 and FSH-810) was evaluated by using DPPH assay with the process illustrated by Yen & Chen[15] and FRAP (ferric reducing antioxidant power) test was assessed by the protocol demonstrated by the Benzie and strain.[16]

**Extraction of Ferulic acid (Alkaline Hydrolysis method)**

The extraction of ferulic acid was elucidated by using the method described by Zhao et al.[17] The method was optimized according to laboratory conditions, two different concentrations of NaOH i.e., 0.5 and 1 M and two hydrolysis temperatures 40°C and 60°C were used for each maize bran sample (Table 1). Two maize bran samples PEARL-11 and FSH-810 (25 g) were suspended in two different concentrations of (0.5 M and 1 M NaOH 500 ml) and two different hydrolysis temperatures (40°C; 60°C), stirred at 300 rpm for 4 h on a hot plate, ensuring hydrolysis completely. The hydrolyzed sample was neutralized with 6 M HCL after cooling down. The polysaccharides were precipitated by adding 95% ethanol. Under the vacuum, the precipitate was separated from the Buchner funnel by filtration. The remaining ethanol was eliminated from the rotary vacuum evaporator filtrate to about one-third of the initial amount, ensuring that the ethanol concentration did not decrease below 25%. This resulted in a brown extract containing ferulic acid is to be produced. The extract was finally passed through a 0.5 µm filter before HPLC analysis (Figure 1).

| Optimized factor | Maize bran samples | Concentration of NaOH (M) | Temperature of hydrolysis (°C) |
|------------------|--------------------|---------------------------|-----------------------------|
| 1                | PEARL-11           | 0.5                       | 40                          |
| 2                | --                 | 1                         | 40                          |
| 3                | --                 | 0.5                       | 60                          |
| 4                | --                 | 1                         | 60                          |
| 5                | FSH-810            | 0.5                       | 40                          |
| 6                | --                 | 1                         | 40                          |
| 7                | --                 | 0.5                       | 60                          |
| 8                | --                 | 1                         | 60                          |
HPLC analysis

The HPLC analysis for the quantification of ferulic acid and other phenolics was performed by Stavova, et al.\(^{[18]}\) method with slight modifications. Briefly, the following standard operating conditions were adjusted a reverse phase HPLC system (PerkinElmer, Series 200. USA) column (150 mm × 4.6 mm, particle size 2.7 nm) at 45°C. The composition of the mobile phase was acetonitrile (solvent A) and Formic acid 0.5% (v/v) (solvent B). The data was collected through the data system UV/vis detector (model 481) at the wavelength of 278 nm. Obtaining peaks of different phenolics were compared with the standard/ reference values for quantification and confirmation.

Statistical analysis

The significance level of treatment over varieties was determined by Complete Randomized Design (CRD) and analysis of variance (ANOVA), followed by the least significant difference (LSD) according to the methods of Steel et al.\(^{[19]}\) For the purpose, triplicate samples were used regarding each parameter.
RESULTS AND DISCUSSION

Biochemical composition of maize bran samples

Maize bran samples PEARL-11 and FSH-810 were analyzed for moisture content, crude protein, crude fat, crude fiber, NFE, and crude ash content. The mean values of each parameter are presented in Table 2. Moisture content is an important aspect to control the physicochemical properties of maize bran. It is also important in determining the shelf life of any cereal product. The physicochemical activity of maize bran depends upon the moisture content present in it. Moisture content is an effective parameter for maintaining the quality of maize bran. Mean values of moisture content were 10.6 ± 0.2% and 9.5 ± 0.1% in PEARL-11 and FSH-810, respectively. Mean values of moisture contents were also mentioned in Table 2. The results of the current study were correlated with the results of Munguti et al.\cite{20} Hussain et al.\cite{21} who explored that the moisture content of maize bran was 10–12%.

The crude ash content of bran samples represented inorganic residues after the combustion of organic material which contains a small amount of minerals. Mean values regarding crude ash contents were shown in Table 2. The mean values of ash content were 1.8 ± 0.01% and 2.2 ± 0.02% in PEARL-11 and FSH-810, respectively. In the previous view, Carvajal-Millan et al.\cite{6} described ash content of maize bran similar to current research work, whereas Munguti et al.\cite{20} determined that the maize bran contained 2.9 ± 1.3% ash content. Rose et al.\cite{3} described that the ash content in the maize bran was 0.6%.

Crude protein is estimated after measuring the nitrogen content of a food. Mean values of protein content in maize bran varieties (PEARL-11 and FSH-810) were 11.8 ± 0.07% and 9.8 ± 0.04%, respectively. Maximum protein content was observed in FSH-810. In the present research work, the results of the protein content are following Hespell,\cite{22} who reported that maize bran contained 10–13% of crude protein content. Rose et al.\cite{3} also presented almost similar results i.e., 11.5%. Carvajal-Millan et al.\cite{6} found that the protein content of maize bran was 11.8%. Protein is important for body development and repair, as well as maintaining excellent health. It is also provided energy. Current results showed that maize bran is a cheap and good source of protein.

The crude fat contents were observed in PEARL-11 (6.4 ± 0.1%) and FSH-810 (7.3 ± 0.4%). In present study, the results of fat content were in accordance with Asuk et al.\cite{23} who depicted that maize bran contained 11.6% fat content. Another study by Mutayoba et al.\cite{24} also showed nearly similar results of 10.07% fat content in maize bran. The crude fiber content may be related to grain shape and size and bran portion thickness in grain. The crude fiber content is positively associated with the level of bran in the maize grain. The crude fiber contents were 8.2 ± 0.4% and 7.8 ± 0.3% in PEARL-11 and FSH-810, respectively. The higher fiber content of 8.2 ± 0.4% was observed in PEARL-11. Results of the current research were very closely related to the result of Munguti et al.\cite{20} who reported that the crude fiber content of maize bran varied from 6.2%. Mutayoba et al.\cite{24} explored the crude fiber content of 6.02% in maize bran. Noblet and Le-Goff\cite{25} reported that crude fiber content was 5.2% in maize bran.

Likewise, NFE contents were 60.9 ± 1.8% and 62.8 ± 2.2% in PEARL-11 and FSH-810 respectively. The highest NFE content was observed in FSH-810 (62.8 ± 2.2%). The study results are inline with Asuk et al.\cite{23} and Munguti et al.\cite{20} who described that NFE content ranged from 58.53% to 69.1% %

| Table 2. Biochemical composition of maize bran PEARL-11 and FSH-810. |
|---------------------------------------------------------------|
| **Maize Bran Samples**                                       |
| **Biochemical composition (%)**                              |
| **PEARL-11**                     | FSH-810                          |
| Moisture content       | 10.5 ± 0.4\textsuperscript{a}   | 9.5 ± 0.2\textsuperscript{b}   |
| Crude Fat             | 6.4 ± 0.2\textsuperscript{b}    | 7.3 ± 0.4\textsuperscript{a}   |
| Crude Protein         | 11.8 ± 0.7\textsuperscript{a}   | 9.8 ± 0.4\textsuperscript{b}   |
| Crude Fiber           | 8.2 ± 0.4\textsuperscript{a}    | 7.8 ± 0.3\textsuperscript{b}   |
| Ash                  | 1.8 ± 0.01\textsuperscript{b}   | 2.2 ± 0.02\textsuperscript{a}   |
| NFE                  | 60.9 ± 1.8\textsuperscript{a}   | 62.8 ± 2.3\textsuperscript{b}   |
| Total dietary fiber   | 46.1 ± 0.9\textsuperscript{a}   | 44.3 ± 1.1\textsuperscript{b}   |
| Insoluble dietary fiber | 42.1 ± 0.8\textsuperscript{a} | 41.3 ± 0.8\textsuperscript{b} |
| Soluble dietary fiber | 4.1 ± 0.04\textsuperscript{a}   | 3.1 ± 0.06\textsuperscript{b}   |
in different maize bran varieties. Maize bran is a stable cereal byproduct. It has high nutritional significance enriched with an abundant quantity of macronutrients. It is high in fiber and may be utilized in various food products for technological purposes. It is used in home cooking to improve the fiber content of many food products as well as to improve texture. [21]

**Dietary fibers in maize bran**

Maize bran is known as a good source of several bioactive compounds as well as dietary fiber. [26] It has high total dietary fiber content as compared to other cereal brans. Its dietary fiber has beneficial effects on human health and nutrition. Dietary fiber helps against cardiovascular diseases, obesity, cancer and diabetes type II. [27] Dietary fiber has many nutraceuticals and functional properties like fiber retain the digestive system healthy. It also has many beneficial physiological effects including intestinal function improvement, reduces cholesterol level and increased microbial mass. [21] Total dietary fiber contents in maize bran samples PEARL-11 and FSH-810 were 46.1 ± 0.9 and 44.3 ± 1.1% respectively.

In maize bran, the dietary fiber content was mostly insoluble and the main constituent of bran sample. Insoluble content in maize bran samples PEARL-11 and FSH-810 was determined 42.1 ± 0.8% and 41.3 ± 0.8% respectively. The insoluble fibers have many nutraceuticals and functional properties. It comprises mostly cellulose, hemicellulose and lignin which helps to prevent constipation. [21] Maize bran soluble dietary fiber content was observed in PEARL-11 (4.1 ± 0.04%) and FSH-810 (3.1 ± 0.06%). The previous research of Herrera-Balandrano, et al. [28] reported that the maize bran has total dietary fiber content was 59.31% from which 54.92% was the insoluble dietary fiber and 4.40% was soluble dietary fiber.

**Total phenolics and total flavonoid**

Phenolic acids are among the major groups of bioactive components in cereal grains. Cereal bran has the maximum amount of phenolic compounds. The total phenolic content of maize bran samples PEARL-11 and FSH-810 was 472.1 ± 4.3mg GAE/100 g and 418.6 ± 6.2mg GAE/100 g (Figure 2). Furthermore, a significant increasing trend in the total phenolic content was observed in the PEARL-11 maize bran sample. The total flavonoid content of maize brans sample PEARL-11 and FSH-810 was 111 ± 1.3 mg RE/100 g and 93 ± 1.4mg RE/100 g respectively. In a previous study, Zhang et al. [29] reported that the range of total phenolic content in different maize brans was 30.35 to 47.76 mg GAE/100 g.

**Antioxidant potential of maize bran samples**

The antioxidant capacity of maize bran varieties was shown in Figure 3. The DPPH values of maize varieties PEARL-11 and FSH-810 were 86 and 78% respectively. In previous research Smuda et al., [30] explored DPPH values of corn bran ranged from 60.5–71.3%. The FRAP values of maize samples PEARL-11 and FSH-810 were 26.8 ± 0.07 and 22.6 ± 0.04 mg TE/100 g. The current results showed that PEARL-11 has a higher antioxidant capacity than FSH-810. The present research was closely related to the results of Zhang et al. [29] who reported that The FRAP values of eight different corn kernel varieties ranged from 19.16 to 32.42 mgTE/100 g.

**Ferulic acid extraction based on optimized factor**

In the 1st stage of optimization, the concentration of NaOH was tested. The weight of the maize bran sample (25 g) and examined the concentration of NaOH were 0.5 and 1 M for each sample variety. The next optimized parameter was determined by the optimal hydrolysis temperature. The temperatures investigated were 40°C and 60°C for each maize bran sample. Table 3 showed the results of ferulic acid extraction and the results peaks of each sample were shown in Figure 4. The maximum average yields
were attained by using 0.5 M NaOH (3.7 ± 0.07 mg/g). Moreover, increased concentration of NaOH did not cause statistically significant changes in ferulic acid yield. This fact is portentous to possible industrial ferulic acid production. Low concentrations of NaOH can be used, which is economically beneficial.

The optimal hydrolysis temperature was fixed during the last observation. The initial weight was based on previous studies, maize bran and NaOH concentration were chosen (25 g: 0.5 M). The hydrolyzate at 40°C had the lightest color, while the hydrolyzate at 60°C was the darkest. Ferulic acid extracted quantity was shown in Table 3. When extraction temperature 60°C was used then the highest yield (3.9 ± 0.07 mg/g) of ferulic acid was obtained. Further changes in the temperature of hydrolysis did not cause statistically significant changes in the ferulic acid yield. High temperatures can, however, have a positive effect on the ferulic acid yields obtained through the use of hot water extraction of maize bran. The results of NaOH concentration 0.5 M (3.7 ± 0.07 mg/40°C, 3.9 ± 0.07 mg/60°C) and (3.5 ± 0.06 mg/40°C, 3.6 ± 0.04 mg/60°C) in PEARL-11 and FSH-810 respectively. The results of the 1 M concentration of NaOH were (3.0 ± 0.03 mg/g and 2.9 ± 0.01 mg/g: 40°C), (3.2 ± 0.02 mg/g and 3.1 ± 0.03) in PEARL-11 and FSH-810, respectively.

Saulnier et al.\(^5\) showed the ferulic acid content in maize bran cell wall i.e., 2.9%. Another research result of Saulnier et al.\(^{3132}\) showed 3% ferulic acid extract from maize bran through an enzymatic reaction. Rose and Inglett\(^7\) investigated that ferulic acid content in maize bran 2.95 ± 0.03/100 g. Torre et al.\(^{32}\) determined the ferulic acid content 204 mg/L in maize cob.

Figure 2. Total Phenolic content & total flavonoid content of maize bran samples.
Quantification of other selective phenolic acids

Other selected phenolic compounds (diferulic acid and p-coumaric acid) were analyzed and quantified in the purified hydrolyzate. Diferulic and p-coumaric acids were quantified during the optimization of NaOH concentration and the temperature of hydrolysis. The quantification of the selected phenolic acids was shown in Table 3. The findings result of HPLC (Figure 4) explored that the maize bran contains certain phenolic acids. But, the quantity of these selective acids was lower than ferulic acid. Diferulic acid was the most abundant of these selected phenolic compounds, with a maximum average yield of 0.8 ± 0.01 mg/g. The p-coumaric acid was found in trace amounts i.e., 0.4 ± 0.02 mg/g.

**Table 3. Optimization of concentration of NaOH and hydrolysis temperature for yield of ferulic acid, diferulic acid and p-coumaric acid.**

| Maize bran Samples | Concentration of NaOH (M) | Temperature of hydrolysis (°C) | Yield of Ferulic acid mg/g | Yield of diferulic acid mg/g | Yield of p-coumaric acid mg/g |
|--------------------|---------------------------|-------------------------------|-----------------------------|-----------------------------|-------------------------------|
| Pearl-11           | 0.5                       | 40                            | 3.7 ± 0.07<sup>a</sup>      | 0.7 ± 0.01<sup>a</sup>     | 0.4 ± 0.00<sup>a</sup>       |
|                    | 1                         | 40                            | 3.0 ± 0.03<sup>c</sup>      | 0.6 ± 0.01<sup>b</sup>     | 0.3 ± 0.00<sup>ab</sup>      |
|                    | 0.5                       | 60                            | 3.9 ± 0.07<sup>a</sup>      | 0.7 ± 0.01<sup>b</sup>      | 0.4 ± 0.00<sup>a</sup>       |
|                    | 1                         | 60                            | 3.2 ± 0.02<sup>c</sup>      | 0.6 ± 0.01<sup>b</sup>      | 0.3 ± 0.00<sup>b</sup>       |
| FSH-810            | 0.5                       | 40                            | 3.5 ± 0.06<sup>c</sup>      | 0.5 ± 0.01<sup>b</sup>      | 0.3 ± 0.00<sup>b</sup>       |
|                    | 1                         | 40                            | 2.9 ± 0.01<sup>c</sup>      | 0.4 ± 0.01<sup>c</sup>      | 0.3 ± 0.00<sup>b</sup>       |
|                    | 0.5                       | 60                            | 3.6 ± 0.04<sup>a</sup>      | 0.6 ± 0.01<sup>a</sup>      | 0.4 ± 0.00<sup>a</sup>       |
|                    | 1                         | 60                            | 3.1 ± 0.03<sup>b</sup>      | 0.4 ± 0.01<sup>c</sup>      | 0.3 ± 0.00<sup>b</sup>       |

**Quantification of other selective phenolic acids**

Other selected phenolic compounds (diferulic acid and p-coumaric acid) were analyzed and quantified in the purified hydrolyzate. Diferulic and p-coumaric acids were quantified during the optimization of NaOH concentration and the temperature of hydrolysis. The quantification of the selected phenolic acids was shown in Table 3. The findings result of HPLC (Figure 4) explored that the maize bran contains certain phenolic acids. But, the quantity of these selective acids was lower than ferulic acid. Diferulic acid was the most abundant of these selected phenolic compounds, with a maximum average yield of 0.8 ± 0.01 mg/g. The p-coumaric acid was found in trace amounts i.e., 0.4 ± 0.02 mg/g.
CONCLUSION

From current research work, it is concluded that maize bran (PEARL-11) has high nutritional properties and antioxidant activity than FSH-810. The yield of ferulic acid was significantly affected by the changing temperature and concentration of NaOH. Moreover, the maximum amount of ferulic acid was obtained from PEARL-11 at 0.5 M NaOH concentration and 60°C. Additionally, diferulic acid and p-coumaric acid were found significantly lower than ferulic acid yields. The optimization of the alkaline hydrolysis extraction process has high industrial usage potential, and there is a dire need to explore its full potential.

ORCID

Farhan Saeed http://orcid.org/0000-0001-5340-4015

References

[1] Dowswell, C.: *Maize in the Third World*; CRC Press: New York, 2019.
[2] Saeed, F.; Hussain, M.; Arshad, M. S.; Afzaal, M.; Munir, H.; Imran, M.; Tufail, T.; Anjum, F. M.; et al. Functional and Nutraceutical Properties of Maize Bran Cell Wall Non-starch Polysaccharides. *Int. J. Food Prop*. 2021, 24(1), 233–248. DOI: 10.1080/10942912.2020.1858864.
[3] Rose, D. J.; Inglett, G. E.; Liu, S. X. Utilisation of Corn (Zea Mays) Bran and Corn Fiber in the Production of Food Components. *J. Sci. Food Agr.* 2010, 90, 915–924.
[4] Singh, M.; Liu, S. X.; Vaughn, S. F. Effect of Corn Bran as Dietary Fiber Addition on Baking and Sensory Quality. *Biocatal. Agric. Biotechnol.* 2012, 1(4), 348–352.
[5] Saulnier, L.; Marot, C.; Chanlaud, E.; Thibault, J. F. Cell Wall Polysaccharide Interactions in Maize Bran. *Carbohydr. Polym.* 1995, 26, 279–287. DOI: 10.1016/0144-8617(95)00020-8.
[6] Carvajal-Millan, E.; Rascón-Chu, A.; Márquez-Escalante, J. A.; Micard, V.; de León, N. P.; Gardea, A. Maize Bran Gum: Extraction, Characterization and Functional Properties. *Carbohydr. Polym.* 2007, 69, 280–285. DOI:10.1016/j.carbpol.2006.10.006.
[7] Rose, D. J.; Inglett, G. E. Production of Feruloylated Arabinoxyl-o-oligosaccharides from Maize (Zea Mays) Bran by Microwave-assisted Autohydrolysis. *Food Chem.* 2010, 119, 1613–1618. DOI: 10.1016/j.foodchem.2009.09.053.
[8] Chaudhary, D. P.; Kumar, S.; Yadav, O. P. Nutritive value of maize: Improvements, applications and constraints. In *Maize: Nutrition dynamics and novel uses* (pp. 3-17). 2014, Springer, New Delhi.
Alkaline Torre, Release Saulnier, Smuda, Zhang, Cui, Noblet, Mutayoba, Fermented milling Biochemical Hussain, 131-141. Feedstuffs Munguti, Extraction Stavova, Improved Zhao, Power: Benzie, J. Yen, Chem AACC. Agric. Promising Eight Feruloyl Antioxidant L.; Milling L.; Hussain, J.; Yen, J.; Hussain, L.; Liti, N.; Afzaal, R.; Stursa, R.; Lin, F.; Bonnin, M.; Rivas, B.; Carvajal-Millán, G.; Converti, A.; Soui-Waterhouse, M.; Choudhary, K.; Cereals Science. Wohlers, M.; Sweet Dierenfeld, C.; S.; Elgorriaga, M.; Auger, F.; and Dierenfeld, M.; 2019, 11, 1478–1483. AACC. Approved Methods of American Association of Cereal Chemists, 10th Ed.; American Association Cereal Chemists: St. Paul, MN, 2000.

Chaovanalikit, A.; Wrolstad, R. E. Total Anthocyanins and Total Phenolics of Fresh and Processed Cherries and Their Antioxidant Properties. Food Chem. Toxicol. 2004, 69, 67–72.

Sultana, B.; Anwar, F.; Przybylski, R. Antioxidant Activity of Phenolic Components Present in Barks of Azadirachta Indica, Terminalia Arjuna, Acacia Nilotica, and Eugenia Jambolana Lam. Trees. Food Chem. 2007, 104(3), 1106–1114.

Yen, G. C.; Chen, H. Y. Antioxidant Activity of Various Tea Extracts in Relation to Their Antimutagenicity. J. Agric. Food Chem. 1995, 43(1), 27–32. DOI: 10.1021/jf00049a007.

Benzie, I. F.; Strain, J. J. The Ferric Reducing Ability of Plasma (FRAP) as a Measure of “Antioxidant Power”: The FRAP Assay. Anal. Biochem. 1996, 239(1), 70–76. DOI: 10.1006/abio.1996.0292.

Zhao, S.; Yao, S.; Ou, S.; Lin, J.; Wang, Y.; Peng, X.; Li, A.; Yu, B. Preparation of Ferulic Acid from Corn Bran: Its Improved Extraction and Purification by Membrane Separation. Food Bioprod. Process. 2014, 92(3), 309–313. https://doi.org/10.1057/jfns000011.

Steel, R. G. D.; Torrie, J. H.; Dicky, D. A. Principles and Procedures of Statistics, A Biometrical Approach. 3rd Edition, McGraw Hill, Inc. Book Co., 1997. New York, 352-358.

Munguti, J. M.; Liti, D. M.; Waidbacher, H.; Straif, M.; Zollitsch, W. Proximate Composition of Selected Potential Feedstuffs for Nile Tilapia (Oreochromis Niloticus Linnaeus) Production in Kenya; Die Bodenkultur, 57 (3) 2006, 131-141.

Hussain, M.; Saeed, F.; Niaz, B.; Afzaal, M.; Ikram, A.; Hussain, S.; Mohamed, A. A.; Alamri, M. S.; Anjum, F. M. Biochemical and Nutritional Profile of Maize Bran-enriched Flour in Relation to Its End-use Quality. Food Sci. Nutr. 2021, 9(6), 3336–3345.

Hespell, R. B. Extraction and Characterization of Hemicellulose from the Corn Fiber Produced by Corn Wet-milling Processes. J. Agric. Food Chem. 1988, 46(7), 2615–2619. DOI: 10.1021/jf971040y.

Asuk, A. A.; Kisambira, A.; Lubowa, M.; Senabulya, D. Evaluation of the Effect of Gurus, a Ugandan Cereal-Based Fermented Food on Serum Lipid Profile and Body Weight of Wistar Albino Rats. Food Nutr. Sci. 2016, 7(4), 284.

Mutayoba, S. K.; Dierenfeld, E.; Mercedes, V. A.; Frances, Y.; Knight, C. D. Determination of Chemical Composition and Anti-nutritive Components for Tanzanian Locally Available Poultry Feed Ingredients. Int. J. Poultry Sci. 2011, 10(5), 350–357.

Noblet, J.; Le Goff, G. Effect of Dietary Fibre on the Energy Value of Feeds for Pigs. Anim. Feed Sci. Technol. 2001, 90(1–2), 35–52. DOI: 10.1016/S0377-8401(01)00195-X.

Sharma, S. K.; Bansal, S.; Mangal, M.; Dixit, A. K.; Gupta, R. K.; Mangal, A. Utilization of Food Processing By-products as Dietary, Functional, and Novel Fiber: A Review. Crit. Rev. Food Sci. Nutr. 2016, 56(10), 1647–1661. https://doi.org/10.1080/10408398.2015.1031474.

Cui, Z.; Kennedy, L.; Weili, L. The effects of intake of bread with treated corn bran inclusion on postprandial glycemic response. Journal of Food & Nutritional Sciences, 2019, 12(2), 73–81. https://doi.org/10.1057/jfns000011.

Herrera-Balandrano, D. D.; Báez-González, J. G.; Carvajal-Millán, E.; Muy-Rangel, D.; Urias-Orona, V.; Martínez-López, A. L.; Márquez-Escalante, J. A.; Heredia, J. B.; Beta, T.; Niño-Medina, G.; et al. Alkali-extracted Feruloylated Arabinoxylans from Nixtamalized Maize Bran Byproduct: A Synonymous with Soluble Antioxidant Dietary Fiber. Waste Biomass Valorization. 2020, 11, 403–409. DOI: 10.1007/s12649-018-0462-z.

Zhang, R.; Huang, L.; Deng, Y.; Chi, J.; Zhang, Y.; Wei, Z.; Zhang, M. Phenolic Content and Antioxidant Activity of Eight Representative Sweet Corn Varieties Grown in South China. Int. J. Food Prop. 2017, 20(12), 3043–3055.

Smuda, S. S.; Mohsen, S. M.; Olsen, K.; Aly, M. H. Bioactive Compounds and Antioxidant Activities of Some Cereal Milling By-products. J. Food Sci. Technol. 2018, 55(3), 1134–1142.

Saulnier, I.; Marot, C.; Elgorriaga, M.; Bonnin, E.; Thibault, J. F. Thermal and Enzymatic Treatments for the Release of Free Ferulic Acid from Maize Bran. Carbohydr. Polym. 2001, 45(3), 269–275.

Torre, P.; Alakbarian, B.; Rivas, B.; Dominguez, J. M.; Conventi, A. Release of Ferulic Acid from Corn Cobs by Alkaline Hydrolysis. Biochem. Eng. J. 2008, 40(3), 500–506.