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# Peanut (*Arachis hypogaea* L.) flour and oilcake flour: Exploring the influence of roasting and varietal differences on proximal composition, elemental profiling, antimicrobial and antioxidant properties

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## ARTICLE INFO

*Keywords:* Peanut flour Oilcake flour By-products Phenolic content Green products Sustainable yield Biological activity ABSTRACT

Peanuts are highly valued for their abundance of essential nutrients and health-promoting phenolic compounds. Peanut press cake, an inexpensive and underutilized agro-industrial by-product of oil production, is typically discarded or used as animal feed. This study investigated the influence of thermal processing and varietal disparities on the nutritional composition, phenolic content, and biological properties of peanut flour and oilcake flour, aiming to enhance their value as food ingredients. The findings showed that roasting significantly increased the oil (9.98  $\pm$  0.11–44.13  $\pm$  0.10 %), ash (1.28  $\pm$  0.01–5.45  $\pm$  0.05 %), carbohydrate contents (0.90  $\pm$  0.01–28.09  $\pm$  0.28 %), and energy value (406.69  $\pm$  0.09–609.13  $\pm$  1.08 kcal/100 g), along with the total polyphenol content (28.64  $\pm$  0.19–62.79  $\pm$  1.18 mg GAE/g), total flavonoid content (4.20  $\pm$  0.07–18.35  $\pm$  0.06 mg QE/g) and antioxidant activity in both peanut flour and its oilcake. Conversely, it led to a reduction in the moisture (1.48  $\pm$  0.09–6.25  $\pm$  0.15 %) and protein content (49.50  $\pm$  0.05–54.24  $\pm$  0.01 %). Notable variations were found between the two peanut varieties in terms of these nutritional parameters. Elemental analysis unveiled significant discrepancies among peanut varieties and with roasting, with potassium (12,237.56  $\pm$ 101.36–14,513.34  $\pm$  168.62 mg/kg) emerging as the predominant macro-element followed by phosphorus  $(6156.86 \pm 36.19 - 8815.22 \pm 130.70$  mg/kg) and magnesium (3037.92  $\pm$  13.87-4096.44  $\pm$  8.54 mg/kg), while zinc (53.98  $\pm$  0.61–81.77  $\pm$  0.44 mg/kg) predominated among the microelements. Moreover, peanut and oilcake flours demonstrated antibacterial activity against several bacteria. It can be inferred that roasted peanut and oilcake flours offer substantial nutritional value, making them promising candidates for addressing proteinenergy malnutrition and serving as valuable ingredients in developing new food products.

#### **1. Introduction**

Peanut (*Arachis hypogaea* L.), an annual legume of the Leguminosae

family, stands as one of the foremost valuable oilseed crops, extensively cultivated across more than 29.62 million hectares worldwide in 2023 ([USDA, 2024a](#page-11-0)). This crop represents a natural resource with immense

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<span id="page-1-0"></span>economic potential for utilization in human and animal nutrition. In May 2023, the global production yields of peanut grain, oil, and meal were 50.46, 6.14, and 7.55 million metric tons, respectively ([USDA,](#page-11-0)  [2024b\)](#page-11-0). It is mainly grown in China, India, Nigeria, and the USA, which are distributed in Asia, Africa, and America. These nations collectively account for 63.5 % of the world's peanut grain, 75.3 % of peanut oil, and 75.8 % of peanut meal/cake production ([USDA, 2024b](#page-11-0)). Peanuts are categorized into four types: Runner and Virginia, which belong to the *hypogaea* subspecies, and Spanish and Valencia, which are classified under the *fastigiata* subspecies [\(Idrissi et al., 2022](#page-11-0)). Peanuts are commonly consumed raw, boiled, or roasted and serve as a key ingredient in a wide range of packaged foods, including peanut butter, candies, confections, and snack products ([Arya et al., 2016\)](#page-10-0).

Peanuts have garnered significant attention as a functional food, offering a rich nutritional profile comprising oil (44–56 %), dietary fiber, carbohydrates, proteins, and essential minerals such as phosphorus, calcium, magnesium, and potassium, alongside vitamins E, K, and various B group vitamins ([El Idrissi et al., 2023](#page-11-0); [Settaluri et al.,](#page-11-0)  [2012\)](#page-11-0). Additionally, recent studies have unveiled peanuts as a remarkable source of bioactive compounds like resveratrol, phenolic acids (hydroxybenzoic acid, ferulic acid, p-coumaric acid), and flavonoids (catechin, procyanidins, quercetin, and kaempferol) (Ciftci  $\&$  Suna, [2022;](#page-11-0) [Toomer, 2018\)](#page-11-0). Alongside its favorable fatty acids composition, the health benefits attributed to peanut consumption predominantly stem from these bioactive compounds. Epidemiological studies indicate that including peanuts and peanut-based products in the diet may help mitigate the risk of coronary heart disease by reducing low-density lipoprotein cholesterol levels, lowering the risk of developing type II diabetes, and aiding in weight management [\(Arya et al., 2016;](#page-10-0) [Çiftçi](#page-11-0) & [Suna, 2022](#page-11-0)).

The peanut cake, a by-product obtained after extracting oil from peanuts through screw pressing, presents a rich source of protein, dietary fiber, minerals, vitamins, and bioactive compounds ([Zhao et al.,](#page-11-0)  [2012\)](#page-11-0). Also, this cake holds significant potential as a valuable food ingredient for producing low-cost, nutritious foods [\(Duhan et al., 2021](#page-11-0)). The removal of oil enhances the protein, minerals, and fiber content in peanut cake [\(Kalpana Devi et al., 2013\)](#page-11-0). However, despite its potential, peanut cake remains underutilized as a dietary ingredient, and there is a notable gap in the literature regarding a comparative analysis of the chemical composition of peanut flour and peanut press cake flour. Both flours offer immense potential in food product development, given their nutrient and antioxidant-rich profiles. Therefore, a detailed investigation and comparison of peanut flour and peanut oilcake flour are required to promote their utilization as an ingredient in food products.

Roasting stands as a pivotal stage in the peanut processing industry, crucial for enhancing the flavor, color, texture, and overall palatability of peanuts (Çiftçi & [Suna, 2022;](#page-11-0) [El Idrissi, El Guezzane, et al., 2024](#page-11-0)). [Ayoola and Adeyeye \(2010\)](#page-10-0) analyzed groundnut seeds− raw, sun-dried, and roasted− for their proximate composition and nutritional mineral content, revealing that roasted groundnuts are rich in valuable minerals, while raw ones offer high protein content with significant nutritional value. Numerous researchers have extensively studied the aforementioned aspects, focusing on specific varieties of groundnut seeds or conducting comparative studies to evaluate nutritional compositions such as total lipids, fatty acid profile, and proximate properties using various quantitative methodologies (Belete & [Bayissa, 2020;](#page-10-0) [Kamuhu](#page-11-0)  [et al., 2019; Kumar et al., 2013; Mada et al., 2012](#page-11-0)). It is noteworthy that during the roasting process, certain chemical changes occur wherein may condense with free amino acids, peptides, or proteins, resulting in the formation of brown Maillard reaction products such as pyrroles and furans, which exhibit potential antioxidant properties (Kaur et al., [2024\)](#page-11-0). These products have been observed to augment the total phenolic compounds present in roasted peanuts. According to [Yunusa et al.](#page-11-0)  [\(2023\),](#page-11-0) peanuts roasted at 140 ◦C for 10 min displayed the highest total phenolic content (67.26  $\pm$  1.77 mg GAE/g) and exhibited the most significant DPPH radical scavenging activity, with an  $IC_{50}$  value of

417.44 μg/mL. Conversely, those roasted at 150 ◦C for 5 min demonstrated the highest total flavonoid content (12.91  $\pm$  0.56 mg QE/g). Moreover, [Win, Abdul-Hamid, Baharin, Anwar, and Saari \(2011\)](#page-11-0) reported that roasting has significantly affected the antioxidant attributes and the phenolic composition of two different forms of peanut flour (with and without skin).

Numerous studies have explored the impact of roasting on phenolic content and its relation with the antioxidant capacity of blanched peanuts ([Asen et al., 2021;](#page-10-0) [Chukwumah et al., 2007](#page-11-0); [Win, Abdul-Hamid,](#page-11-0)  [Baharin, Anwar, Sabu,](#page-11-0) & Pak-Dek, 2011). However, research on whole peanuts and their press oilcake is scarce. Further, no reports were documented in the literature about the microbial activity of peanuts. Due to the differences among the species and/or varieties of peanuts grown in different areas of the world, two specific peanut varieties (Virginia and Valencia) grown in Morocco were selected for investigation. Hence, this study aimed to explore the impact of thermal processing and varietal differences on proximate composition, elemental profiling, phenolic content, antioxidant activity as well as antibacterial activity of peanut flour and oilcake flour.

#### **2. Material and methods**

#### *2.1. Chemicals and reagents*

All analytical-grade reagents and solvents used in this study were supplied by Sigma-Aldrich (St. Louis, MO, USA). These include Folin–Ciocalteu's phenol reagent, sodium carbonate (Na2CO3; 99.5 %), aluminum chloride (AlCl<sub>3</sub>; 98 %), sodium nitrite (NaNO<sub>2</sub>; 97.0 %), sodium hydroxide (NaOH; 97.0 %), 2,2-Diphenyl-1-picrylhydrazyl (DPPH; ≥90 %), 2,2′-Azinobis-(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS; ≥98 %), L-ascorbic acid (≥99 %), quercetin (≥95 %), Trolox (≥97 %), potassium persulfate (K2S2O<sup>a</sup>; 98 %), absolute methanol ( $\geq$ 99.9 %), and ethanol (99.5 %).

#### *2.2. Plant material*

Virginia and Valencia peanut varieties grown in Morocco were used in this study. These were sourced from a local farm market in Kenitra, Morocco (34◦ 15′ 0" N 6◦ 34' 59.999" W). The samples were harvested at the full maturity stage, and then cleaned and sorted to remove damaged seeds and unusual particles (dust, sand, etc.). The designations of the peanut samples are provided in Table 1.

#### *2.3. Sample processing*

*Conventional roasting*: Peanuts were roasted in a stainless-steel drum for 15 min at a temperature of 175  $\pm$  5 °C, utilizing sand as the heat source, and a diesel burner to power the roaster ([El Idrissi, Amakhmakh,](#page-11-0)  [et al., 2024](#page-11-0)). To preserve the desired flavor, the roasting process was carefully controlled to prevent overheating. The kernels and sand were vigorously mixed to ensure uniform heating.

Following roasting, the peanuts were cooled at room temperature and then divided into two equal portions for further processing into peanut flour and peanut oilcake flour. For the first portion, both

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Identification and labeling of the examined peanuts.



unroasted and roasted peanuts were delipidated with hexane using a Soxhlet apparatus to obtain defatted peanut powder ([Idrissi et al., 2022](#page-11-0)). This powder was then milled and sieved through a 35-μm mesh to obtain peanut flour. For the second portion, the residual peanut oilcake remaining after the cold-press oil extraction was pulverized and passed through a 35-μm mesh sieve to obtain peanut cake flour. All samples were stored in polyethylene bags at −4 °C until analysis.

#### *2.4. Nutrient composition*

Peanut flour and peanut cake flour were subjected to analyses comprising of oil content (OC), moisture content (MC), ash content (AC), protein content (PC), carbohydrates content (CC), energy value (EV), and mineral profiling. Each analysis was conducted in triplicate, and the average results were reported. All component contents were calculated on a dry matter basis.

## *2.4.1. Proximate analysis*

*2.4.1.1. Oil content (OC).* The extraction of crude oil from peanuts and oilcake was conducted using a Soxhlet apparatus (R-200, Buchi, Zurich, Switzerland). A 30 g sample of powder was extracted for 8 h with 300 mL of n-hexane. The solvent was then removed from the mixture using a vacuum rotary evaporator (model VV2000, Heidolph, Schwabach, Germany) at 50 ◦C ([Idrissi et al., 2022](#page-11-0)). The oil content was estimated gravimetrically.

*2.4.1.2. Moisture content (MC).* Moisture content (MC) was determined according to [ISO 665 \(2020\)](#page-11-0). Briefly, 5 g of peanut flour and oilcake flour were placed in a ventilated oven (Precision Scientific Co., USA) at  $103 \pm 2$  °C, until a consistent weight was attained. The MC was calculated using the following formula, expressed as a percentage by mass:

$$
MC (%) = \frac{W_1 - W_2}{W_1 - W_0} \times 100
$$
 (1)

where  $W_0$  represents the weight of the container with its lid,  $W_1$  denotes the weight of the container with its lid and the sample prior to drying, and  $W_2$  indicates the weight of the container with its lid and the sample after drying.

*2.4.1.3. Ash content (AC).* Ash content was determined following the [ISO 749 \(1977\)](#page-11-0) method. For each sample, 5 g was placed into crucibles and subsequently calcined in a muffle furnace at  $550 \pm 5$  °C for 5 h until white ash remained. After cooling in a desiccator, the crucibles were weighed.

*2.4.1.4. Protein content (PC).* Protein content (PC) was measured based on the detection of nitrogen through combustion using a LECO FP628 (LECO Corp., MI, USA) nitrogen elemental analyzer, following the Dumas approach as specified in [ISO 16634-1 \(2008\)](#page-11-0) method. The measured total nitrogen content was converted to PC by multiplying by a conversion factor of 6.25.

*2.4.1.5. Total carbohydrates content (CC) and energy value (EV).* Total carbohydrate content was estimated for each sample by subtracting the sum of moisture, crude protein, ash, and fat from 100 %, using the following formula ([Oubannin et al., 2022\)](#page-11-0):

Carbohydrate content (CC, %) = 
$$
100\%-(\%MC + \%PC + \%AC + \%OC)
$$
 (2)

From the data on the content of protein, carbs, and lipids, energy values were computed by applying factors of 4, 4, and 9, respectively, and reported as kcal/100 g dry basis.

EnergyvalueEVkcal/100g) =  $(2.62 \times %PC) + (8.37 \times %OC) + (4.2 \times %CC)$ (3)

## *2.4.2. Mineral profiling (MP)*

Mineral profiling was determined using the method outlined by [Ibourki et al. \(2022\).](#page-11-0) Five macro-minerals⸺potassium (K), magnesium (Mg), sodium (Na), calcium (Ca), and phosphorus (P), as well as five trace minerals⸺zinc (Zn), iron (Fe), boron (B), manganese (Mn), and copper (Cu) were quantified.

Peanut flour and oilcake flour were digested using a microwaveassisted digestion Speed Wave system (Berghof Analytik, Germany). Briefly, 0.3 g of each sample was weighed into Teflon vessels, to which 8 mL of 65 % nitric acid ( $HNO<sub>3</sub>$ ) and 2 mL of 30 % oxygenated water  $(H<sub>2</sub>O<sub>2</sub>)$  were added. The vessels were placed in the mineralizer, and the digestion program was initiated. Once completed, the resulting clear solution was diluted to 25 mL with deionized water and injected in an ICP–AES spectrometer (Perkin Elmer, Model Optima 8000 DV, Waltham, USA) equipped with an autosampler ASX-520 (Teledyne CETAC Technologies, Omaha, USA) and a CCD detector. The operating conditions were as follows: power range of 1.15–1.2 kW, plasma flow gas range of 12–14 L/min, auxiliary gas flow of 1.5 L/min, and nebulizer gas flow of 0.2 L/min. The measures' accuracy was confirmed using standard samples, and the data were expressed in mg/kg.

### *2.5. Extraction and determination of phenolic content*

#### *2.5.1. Extract preparation*

Delipidated peanut kernels and delipidated cold-pressed oilcake, both unroasted and roasted, were studied in this section. Following the methodology established by [Idrissi et al. \(2022\)](#page-11-0), the methanolic extracts were prepared using two conventional methods: Soxhlet extraction for 8 h at 60 °C and cold maceration for 48 h in the dark. The extracts were then concentrated by removing methanol using a rotary vacuum evaporator (model VV2000, Heidolph, Schwabach, Germany), and freezedried to obtain phenolic extracts in powder form. These extracts were stored in flasks at 4 ◦C until analysis.

#### *2.5.2. Polyphenol content*

The total polyphenol content (TPC) was determined using the Folin-Ciocalteu method, as redescribed by [Eddahhaoui et al. \(2022\)](#page-11-0). A 0.5 mL aliquot of a stock solution (1 mg/mL) prepared in methanol was mixed with 2.5 mL of a Folin-Ciocalteu (FC) solution diluted in deionized water  $(1:10, v/v)$ , followed by the addition of 2 mL of aqueous solution of sodium carbonate (7.5 %, *w*/*v*). The mixture was incubated in a water bath at 45 ◦C for 30 min. After the incubation, the absorbance was measured at 765 nm utilizing a UV-5800PC spectrophotometer (Shanghai Metash Instruments Co., Ltd., Shanghai, China) against a blank comprising the same solution, but with methanol and distilled water (70:30) instead of extract. A standard curve of gallic acid was prepared under the same conditions, using concentrations ranging from 2 to 100 μg/mL. The TPC was reported as mg of gallic acid equivalent per gram of extract (mg GAE/g).

#### *2.5.3. Flavonoids content*

The total flavonoid content (TFC) was determined using the aluminum trichloride colorimetric method as previously outlined by [Eddahhaoui et al. \(2022\)](#page-11-0). In a test tube, 0.25 mL of extract solution was combined with 0.075 mL of an aqueous solution of NaNO<sub>2</sub> (5 %,  $w/v$ ) and 1.25 mL of distilled water. After 5 min, 0.15 mL of  $AlCl<sub>3</sub>$  solution (10 %, *w*/*v*) was added, and 6 min later, the mixture was treated with 0.5 mL of 1 M NaOH. After 30 min of incubation, the absorbance of the reaction mixture was measured at 510 nm against a blank. A standard curve of quercetin was prepared under the same conditions, using concentrations ranging from 2 to 100 μg/mL The TFC was reported in milligrams of quercetin equivalent per gram of extract (mg QE/g).

## *2.6. Antioxidant activity*

#### *2.6.1. DPPH radical scavenging activity*

The free radical scavenging activity of the methanolic extracts was evaluated using the DPPH assay, as described by [Boujemaa et al. \(2020\)](#page-10-0). Briefly, 0.2 mM solution of DPPH in methanol was prepared; afterward, 0.5 mL of this solution was mixed with 2.5 mL of extract solutions at different concentrations. The mixtures were vortexed and incubated in the dark at room temperature for 30 min. The absorbance was measured at 517 nm using a UV-5800PC spectrophotometer, against a blank sample. The results were reported as  $IC_{50}$  values in  $\mu$ g/mL, representing the extract concentration required to inhibit 50 % of the initial DPPH free radical. These values were compared with those of ascorbic acid  $(1-20 \mu g/mL)$ .

## *2.6.2. ABTS radical scavenging activity*

The ABTS radical scavenging activity was determined following the procedure outlined by El Moudden et al.  $(2022)$ . The ABTS<sup>+</sup> cationic radical was generated by reacting 10 mL of 2 mM ABTS solution in distilled water with 100 μL of 70 mM potassium persulfate  $(K_2S_2O_8)$ solution. This mixture was kept at room temperature in the dark for 24 h. The resulting solution was then adjusted with methanol to achieve an absorbance of  $0.70 \pm 0.02$  at 734 nm. After combining 2 mL of the ABTS solution with 200 μL of methanolic extracts of different concentrations, the absorbance was measured at 734 nm using a UV-5800PC spectrophotometer after 30 min of incubation. The antioxidant activity of the extracts was expressed as  $IC_{50}$  values, representing the concentration required to reduce the ABTS free radical by 50 % (μg/mL). These results were compared to the Trolox standard (5–60 μg/mL).

## *2.7. Antibacterial activity*

Methanolic peanut extracts, with a mass of 30 mg, were solubilized in 1 mL dimethyl sulfoxide (DMSO). Prior to use, the extract solution was filtered through 0.22 μm Millipore filters for purification.

#### *2.7.1. Microorganisms*

The antibacterial activity of peanut methanolic extracts was evaluated against two Gram-positive bacteria (*Staphylococcus aureus* and *Staphylococcus epidermidis*) and four Gram-negative bacteria (*Klebsiella pneumoniae*, *Acinetobacter baumannii*, *Escherichia coli*, and *Enterobacter cloacae*). These strains were selected for their high resistance to antibiotics and were maintained through regular subculturing on nutrient agar.

## *2.7.2. Disc diffusion method*

The antibacterial activity was assessed using the disk diffusion method, following the protocol outlined by [Oubihi et al. \(2020\)](#page-11-0). A microbial suspension, adjusted to an optical density of 1 McFarland, was uniformly spread onto an agar medium in Petri dishes. Whatman absorbent paper discs (6 mm in diameter) were sterilized by autoclaving at 121 ◦C for 20 min, then soaked with 15 μL of the extract solution. These impregnated discs were placed on the inoculated agar surface and incubated for 24 h at 37 ◦C. After incubation, the extract diffused into the agar, creating a circular zone of inhibition (measured in mm) around the discs, indicating the extract's efficacy against the microorganisms. A larger zone of inhibition suggests a higher sensitivity of the microorganisms to the extract. Penicillin (5 μg/disc) and amoxicillin (25 μg/ disc) were used separately as positive controls. All tests were conducted in triplicate for reliability.

## *2.7.3. Minimum inhibitory concentration*

The minimum inhibitory concentration (MIC) was assessed using the macro-dilution method, following the methodology described by [Nasri](#page-11-0)  [et al. \(2022\).](#page-11-0) Extracts were emulsified in a 0.2 % agar solution to ensure consistent distribution throughout the medium. Serial dilutions of 100 μL/mL, 40 μL/mL, 20 μL/mL, 10 μL/mL, 5 μL/mL, 3.3 μL/mL, and 2 μL/ mL were prepared in the agar solution. Subsequently, 1.5 mL of each dilution was aseptically added to test tubes containing 13.5 mL of Muller Hinton solid medium, resulting in final concentrations of 10 μL/mL, 4 μL/mL, 2 μL/mL, 1 μL/mL, 0.5 μL/mL, 0.33 μL/mL, and 0.2 μL/mL. After shaking each tube for 15 s, the contents were immediately poured into sterile Petri dishes. Control samples, containing culture medium and 0.2 % agar solution, were also prepared. Inoculation was done using a calibrated platinum loop to maintain a consistent volume of inoculum. The Petri dishes were then incubated at 37 °C for 24 h. The MIC ( $\mu$ L/mL) was defined as the lowest concentration at which no bacterial growth occurred. Each experiment was repeated three times to ensure accuracy and reliability.

## *2.8. Statistical data analysis*

The analytical experiments were performed in duplicate or triplicate, with results reported as the mean  $\pm$  standard deviation. Data analysis included analysis of variance (ANOVA) and Tukey's test at a 95 % confidence level, conducted using IBM SPSS Statistics Version 21 (SPSS Inc., Chicago, IL, USA). Statistical significance was determined at a *p*value of less than 0.05. Pearson's correlation test was also applied to assess the relationships between variables.

The proximate composition (OC, MC, PC, AC, CC), EV, macromineral elements (K, Mg, Na, Ca, P), phenolic compound content (TPC and TFC), and antioxidant activity (DPPH and ABTS) were subjected to principal components analysis (PCA) to ascertain any discrimination between peanut samples. Additionally, hierarchical cluster analysis (HCA) was employed to explore the interrelationships among peanut samples. The dendrogram was constructed using Ward's method and squared Euclidean distance as similarity measures. Both PCA and HCA were conducted using the XLSTAT 2014 software (Microsoft®, Redmond, Washington, USA).

## **3. Results and discussion**

#### *3.1. Proximate composition*

The assessment of the proximate composition of peanut flour and oilcake flour samples ([Table 1\)](#page-1-0) is of great interest as it provides consumers with essential insights into the nutritional value and product quality. In this study, the contents of oil, moisture, protein, ash, total carbohydrates, and calorific value were determined. The resulting data based on the roasting treatment were summarized in [Table 2](#page-4-0), while the data based on variety were shown in Table 1S.

#### *3.1.1. Oil content (OC)*

OC is a primary criterion for evaluating oil production effectiveness from oilseeds. Understanding the OC of seeds is pivotal to their value, as it directly influences monetary valuation in the economic oilseed trade ([Gagour et al., 2022](#page-11-0)).

As shown in [Table 2,](#page-4-0) peanut roasting resulted in a notable alteration of the oil extraction yield (*p <* 0.05). An increase in OC was observed, for instance, passing from  $43.31 \pm 0.12$  and  $5.47 \pm 0.13$  % for UVI and UVIp to  $44.13 \pm 0.10$  and  $9.98 \pm 0.11$  % for RVI and RVIp, respectively, which contrasts with the published data of [Ayoola and Adeyeye \(2010\)](#page-10-0). The increase in OC of roasted peanuts may be attributed to the breakdown of bonds between fat and matrix, facilitating a more efficient release or mobilization of oil from disrupted cells into the flour ([Kaur](#page-11-0)  [et al., 2024\)](#page-11-0).

Results also revealed significant variation in OC among the studied varieties (p *<* 0.05) (Table 1S), with the Virginia type exhibiting the highest OC, ranging from  $5.47 \pm 0.13$  to  $44.13 \pm 0.13$  % compared to the Valencia type, varying from  $4.24 \pm 0.07$  to  $43.54 \pm 0.09$  %. Nevertheless, there is a significant disparity in the OC of various peanut varieties as documented in the literature, often ascribed to genetic

#### <span id="page-4-0"></span>**Table 2**

and energy value (EV, kcal/100 g) of peanut flour and oilcake flour from two varieties, based on roasting treatment. Variety Sample OC MC PC AC CC EV *Virginia* Unroasted UVI  $43.31 \pm 0.12^a$   $2.23 \pm 0.07^a$   $53.38 \pm 0.01^a$   $0.60 \pm 0.06^a$   $0.47 \pm 0.12^a$   $605.23 \pm 0.55^a$  $\textrm{UVD} \qquad \qquad 5.47 \pm 0.13^{\textrm{b}} \qquad \qquad 5.70 \pm 0.10^{\textrm{be}} \qquad \qquad 58.30 \pm 0.03^{\textrm{b}} \qquad \qquad 4.70 \pm 0.10^{\textrm{b}} \qquad \qquad 25.82 \pm 0.36^{\textrm{b}} \qquad \qquad 385.78 \pm 0.13^{\textrm{b}}$ Roasted RVI  $44.13 \pm 0.10^{\circ}$   $1.59 \pm 0.13^{\circ}$   $51.18 \pm 0.03^{\circ}$   $1.28 \pm 0.01^{\circ}$   $1.81 \pm 0.01^{\circ}$   $609.13 \pm 1.08^{\circ}$  $\rm RVIp \quad \rm 9.98\pm0.11^d \quad \rm 5.30\pm0.10^b \quad \rm 54.24\pm0.01^d \quad \rm 4.85\pm0.05^b \quad \rm 25.62\pm0.28^b \quad \rm 409.33\pm0.03^d$ *Valencia*

Unroasted UVA  $41.35 \pm 0.13^{\circ}$   $1.85 \pm 0.02^{\circ}$   $54.93 \pm 0.01^{\circ}$   $1.08 \pm 0.02^{\circ}$   $0.78 \pm 0.11^{\circ}$   $595.01 \pm 0.65^{\circ}$ 

Roasted RVA  $43.54 \pm 0.09^{ac}$   $1.48 \pm 0.09^c$   $52.61 \pm 0.03^g$   $1.46 \pm 0.04^c$   $0.90 \pm 0.01^{ac}$   $605.91 \pm 1.00^{ac}$ 

UVAp  $4.24 \pm 0.07^{\rm f}$   $6.75 \pm 0.15^{\rm d}$   $63.37 \pm 0.01^{\rm f}$   $7.35 \pm 0.15^{\rm d}$   $18.28 \pm 0.06^{\rm d}$   $364.81 \pm 0.38^{\rm i}$ 

 $\rm RVAp \hspace{1.5 cm} 10.70\pm0.14^{8} \hspace{1.5 cm} 6.25\pm0.15^{de} \hspace{1.5 cm} 49.50\pm0.05^{h} \hspace{1.5 cm} 5.45\pm0.05^{e} \hspace{1.5 cm} 28.09\pm0.28^{e} \hspace{1.5 cm} 406.69\pm0.09^{e} \hspace{1.5 cm} 40.05^{e} \hspace{1.5 cm} 40.05^{e} \hspace{1.5 cm} 40.05^{e} \hspace{1.5 cm} 40.05^{e}$ 

Means values of oil content (OC, % DM), moisture content (MC, % DM), crude protein content (PC, % DM), ash content (AC, % DM) carbohydrates content (CC, % DM),

Results are presented as means ± standard deviation ( $n = 2$ ). Values with different letters within each column are significantly different at a significance level of  $p \leq$ 0.05. UVI and UVIp: Unroasted Virginia flour and oilcake, respectively; RVI and RVIp: Roasted Virginia flour and oilcake, respectively; UVA and UVAp: Unroasted Valencia flour and oilcake, respectively; RVA and RVAp: Roasted Valencia flour and oilcake, respectively; DM: Dry matter.

diversity and climate conditions (Campos-Mondragón et al., 2009; [Eshun et al., 2013](#page-11-0)). In addition, each variety's oil press cake  $(4.24 \pm 1)$ 0.07–10.70  $\pm$  0.14 %) showed lower OC compared to the kernels (41.35  $\pm$  0.13–44.13  $\pm$  0.10 %). The findings align with those documented by [Kaur et al. \(2024\)](#page-11-0) for flaxseed flour and cake flour. These agri-food byproducts have the potential to be used as raw materials for producing high-quality residual oil, which presents exciting opportunities for valorization.

### *3.1.2. Moisture content (MC)*

MC plays a crucial role in providing insights into the stability of oilseeds and oilcakes, particularly during storage. Additionally, it serves as a significant factor influencing the quality of the extracted oil, as high water content leads to seed deterioration and the production of rancid, low-quality oil ([Nounah et al., 2021](#page-11-0)).

Based on the recorded data (Table 2), roasting significantly affected peanut and oilcake flour's MC. In peanut flour, the MC decreased from  $2.23\pm0.07$  and  $1.85\pm0.02$  % for UVI and UVA to  $1.59\pm0.13$  and  $1.48$  $\pm$  0.09 % for RVI and RVA, respectively, while for oilcake flour, it decreased from 5.70  $\pm$  0.10 and 6.75  $\pm$  0.15 % for UVIp and UVAp to 5.30  $\pm$  0.10 and 6.25  $\pm$  0.15 % for RVIp and RVAp, respectively. The obtained results for MC were closer to the values reported by [Ayoola and](#page-10-0)  [Adeyeye \(2010\)](#page-10-0) for Nigerian groundnuts (1.07–7.48 %).

In addition, the oilcake flour (5.30  $\pm$  0.10–6.75  $\pm$  0.15 %) showed a higher MC than the peanut flour  $(1.48 \pm 0.09 - 2.23 \pm 0.07 \%)$ (Table 1S), which is in agreement with similar reported findings for argan kernel and black cumin seed and their respective press cake ([Oubannin et al., 2022\)](#page-11-0). Nonetheless, the oilcake's high MC is due to its OC, which prevents water from reaching the surface of the material, thereby affecting moisture evacuation. In contrast to our findings, [Riaz](#page-11-0)  [and Cheewapramong \(2009\)](#page-11-0) revealed that the whole peanut (7.95 %) exhibited higher MC than the press cake (6.13 %). Indeed, an MC below 9 % is deemed safe for storage as it inhibits the rapid proliferation of fungi. In this regard, implementing good hygienic practices is essential to prevent microbial colonization.

#### *3.1.3. Protein content (PC)*

Proteins, vital biological macromolecules present in every living cell, are abundant sources of essential amino acids and crucial macronutrients. Classified as legumes, peanuts share a nutritional profile closer to chickpeas and soybeans in terms of PC, rather than resembling almonds, walnuts, and other oilseeds [\(Toomer, 2018](#page-11-0)).

As evident in Table 2, the PC exhibited a decrease as a result of roasting. For instance, PC decreased from 53.38  $\pm$  0.00 and 54.93  $\pm$ 0.01 % for UVI and UVA to 51.18  $\pm$  0.03 and 52.61  $\pm$  0.03 % for RVI and RVA, respectively. Results are comparable to data published previously (Ayoola & [Adeyeye, 2010;](#page-10-0) [Kaur et al., 2024](#page-11-0)). The observed decrease in PC might be attributed to the denaturation process and the

contribution of proteins in Maillard reactions with reducing sugars during roasting [\(Kaur et al., 2024](#page-11-0)). Furthermore, it has been established in the literature that the process of heating and roasting peanuts leads to a reduction in protein solubility by approximately 50 % (Riaz & [Chee](#page-11-0)[wapramong, 2009\)](#page-11-0).

There were significant differences ( $p < 0.05$ ) among the samples with respect to PC (Table 1S). Nevertheless, both varieties are proteinrich, with values exceeding 50 %, except for RVAp. Our results for PC were higher than those reported by [Alhassan et al. \(2017\)](#page-10-0) in new accessions of groundnut from Ghana (20.7–25.3 %), but closer to the values found by [Kain and Chen \(2010\)](#page-11-0) in samples from China (49.8–52.1 %).

The highest PC was noted in UVAp and UVIp with values of 63.37  $\pm$ 0.01 and 58.30  $\pm$  0.03 %, respectively. These results agreed with those reported by [Kaur et al. \(2024\)](#page-11-0) for flaxseed flour and flaxseed cake flour. In another study, [Oubannin et al. \(2022\)](#page-11-0) stated that argan oilcake (45.93 %) and black cumin cake (23.96 %) had greater PC compared to argan kernels (10.81 %) and black cumin seed (21.66 %). The observed rise in PC of peanut oilcake flour might potentially be ascribed to the process of oil-lipid separation during seed screw pressing, as well as the subsequent accumulation of polypeptides within the seed tissues ([Kaur](#page-11-0)  [et al., 2024\)](#page-11-0).

#### *3.1.4. Ash content (AC)*

Ash is the inorganic residue that remains after the combustion or acid-facilitated complete oxidation of organic compounds present in food [\(Gagour et al., 2022\)](#page-11-0). As seen in Table 2, a significantly greater impact of roasting was observed on the ash content (AC). The latter demonstrated a notable increase in all samples, except for RVAp, wherein the AC decreased from 7.35  $\pm$  0.15 % (UVAp) to 5.45  $\pm$  0.05 %. Our results concur with those of [Kaur et al. \(2024\),](#page-11-0) who stated that roasting led to an increase in AC in flaxseed cake flour and a decrease in AC in flaxseed flour.

A noticeable variation was outlined among varieties, with the Virginia type recording the lowest AC (0.60  $\pm$  0.06–4.85  $\pm$  0.05 %) while Valencia exhibited the highest AC (1.08  $\pm$  0.06–7.35  $\pm$  0.15 %) (Table 1S). [Eshun et al. \(2013\)](#page-11-0) reported an AC varying from 2.45 to 2.96 % in four varieties of peanut flour (Sinkarzie, F-mix, JL 24, and Man-ipintar), which falls within the range observed in our study, while [Kal](#page-11-0)[pana Devi et al. \(2013\)](#page-11-0) found higher AC in defatted groundnut oilcake (9–11.2 %). Moreover, AC was found to be higher in oilcake flour, ranging from 4.70  $\pm$  0.10 to 7.35  $\pm$  0.15 % in comparison to peanut flour (0.60  $\pm$  0.06–1.46  $\pm$  0.04 %), owing to the retention of minerals in the cake during pressing. As discussed in the work of [Kaur et al. \(2024\)](#page-11-0), the lower AC observed in flaxseed cake flour (4.49 %) might be attributed to the release of minerals into oil during screw pressing of seeds. Thus, the higher mineral content reflects higher AC observed in flour samples.

## *3.1.5. Carbohydrate content (CC)*

Peanuts are a rich source of dietary fiber. Sucrose and starch comprise the majority while reducing sugars constitute a smaller proportion of peanut carbohydrates [\(Arya et al., 2016](#page-10-0)). This characteristic may contribute to the low glycemic index and glycemic load associated with peanuts.

Results showed a slight increase in the CC when the peanut was subjected to roasting treatment ([Table 2\)](#page-4-0). Similar reports have indicated relatively higher CC in the roasted groundnut samples (Belete & Bayissa, [2020;](#page-10-0) [Kumar et al., 2013](#page-11-0); [Mada et al., 2012\)](#page-11-0). In contrast, [Kamuhu et al.](#page-11-0)  [\(2019\)](#page-11-0) stated that roasted Red Valencia showed a lower CC (6.63 %) compared to raw Red Valencia (12.37 %). However, this study also revealed a significant variation in the carbohydrate content (CC) among the samples studied, as depicted in Table 1S. These findings were in good agreement with other studies, which previously observed differences in CC as a result of various factors such as cultivar, maturation, and geographic location ([Alhassan et al., 2017;](#page-10-0) [Eshun et al., 2013](#page-11-0)).

Hence, the oilcake of both varieties displayed the highest CC, ranging from  $18.28 \pm 0.06$  % to  $28.09 \pm 0.28$  %, whereas the lowest CC was recorded in peanut flour varying from  $0.47 \pm 0.12$  % to  $1.81 \pm 0.01$  %. This observation aligns with the reported results for argan oilcake (35.12 %) and black cumin cake (51.31 %) ([Oubannin et al., 2022](#page-11-0)). Our findings were lower than those reported by [Riaz and Cheewapramong](#page-11-0)  [\(2009\),](#page-11-0) for whole peanuts (23.72 %) and peanut press cake (41.23 %). However, the CC in our oilcake findings was comparable to the range reported by [Kain and Chen \(2010\)](#page-11-0) of 23.5–24.8 %.

#### *3.1.6. Energy value (EV)*

Foods contain a range of organic substrates that can be oxidized to release energy. The optimal substrates for cellular metabolism are proteins, carbohydrates, and fats, which closely resemble the components found in the human body [\(Ibourki et al., 2022](#page-11-0)).

As indicated in [Table 2](#page-4-0), the energy values (EVs) associated with peanut flour and oilcake flour increased when roasted. According to [Kumar et al. \(2013\),](#page-11-0) 100 g of raw groundnuts contribute 551.9 cal of energy, whereas the same quantity of roasted groundnut seeds contributes 564.5 cal. Indeed, the studied samples, whether raw or roasted, were found to retain high EVs. The corresponding EVs for peanut flour are slightly above that reported by [Alhassan et al. \(2017\)](#page-10-0) in ten new accessions of groundnut and surpasses the EV recorded by [Eshun et al.](#page-11-0)  [\(2013\)](#page-11-0) for the four cultivars.

A remarkable variation was outlined among varieties, with the Virginia type recording greater EVs compared to the Valencia type (Table 1S). In addition, EVs of peanut oilcake seemed to be lower, varying from  $364.81 \pm 0.38$  to  $409.33 \pm 0.03$  kcal/100 g in comparison to the peanut flour, ranging from 595.01  $\pm$  0.65 to 609.13  $\pm$  1.08 kcal/ 100 g. The current results correspond with [Oubannin et al. \(2022\)](#page-11-0), who found lower EVs for argan oilcake and black cumin cake, with 389.45 and 421.89 kcal/100 g values, respectively, compared to the seeds. This is interpreted through the formula used to calculate the energy value, where the OC is multiplied by the highest coefficient (8.37). Based on the aforementioned results, both peanut flour and oilcake flour hold significant potential for utilization. They can be effectively incorporated into processed foods to enhance nutritional value, catering to individuals with high energy requirements, or, alternatively, they can be used as cattle feed.

#### *3.2. Elemental profiling*

Minerals are essential for the metabolic and physiological processes of living organisms, requiring well-defined quantities for proper function. Insufficient or excessive intake of minerals and trace elements can disrupt the immune system, potentially causing clinical symptoms ([Arya](#page-10-0)  [et al., 2016](#page-10-0); [Toomer, 2018\)](#page-11-0). Thus, maintaining an optimal intake level is critical for supporting a healthy immune system. Numerous previous studies have shown that peanuts serve as a beneficial dietary source of macro minerals, essential nutrients needed in daily amounts exceeding 100 mg, and trace elements, required in smaller quantities [\(Toomer,](#page-11-0)  [2018\)](#page-11-0).

The concentration of five macro-minerals (K, Mg, Na, Ca, P) and five trace minerals (Zn, Fe, B, Mn, Cu) was examined in peanut flour and oilcake flour. As shown in [Table 3,](#page-6-0) the mineral analysis revealed notable variations among the samples as a result of roasting, with certain minerals increasing and others decreasing, aligning with the observations of [Ayoola and Adeyeye \(2010\)](#page-10-0) as well as [Belete and Bayissa \(2020\)](#page-10-0). The rise in mineral levels after roasting might be due to the heat destruction of compounds that formed complexes with minerals seeds, resulting in the release of minerals in peanut flour and oilcake flour [\(Kaur et al.,](#page-11-0)  [2024\)](#page-11-0). These outcomes were consistent with results reported by [Kaur](#page-11-0)  [et al. \(2024\)](#page-11-0) for flaxseed flour and cake flour during dry-air roasting.

K was the most abundant macro-mineral in all samples, with concentrations ranging from 12,237.56  $\pm$  101.36 mg/kg in UVIp to 14,513.34  $\pm$  168.62 mg/kg in RVAp. These values were found to be much higher than those reported by [Kumar et al. \(2013\)](#page-11-0) (988.30–1034.30 mg/100 g). In addition, the K content increased with roasting in all samples except for RVI, where it decreased.

P emerged as the second most abundant element, with concentrations ranging from  $6156.86 \pm 36.19$  mg/kg in RVI to  $8815.22 \pm 130.7$ mg/kg in RVAp. The P levels in the studied samples were also higher than those reported for raw and roasted groundnuts (340.20 and 363.40 mg/100 g, respectively) ([Kumar et al., 2013\)](#page-11-0). In fact, P, along with Ca, is crucial for maintaining healthy bones and for protein synthesis during tissue development and repair [\(Toomer, 2018\)](#page-11-0). In both peanut varieties, P content increased in peanut flour with roasting, while it decreased in oilcake flour.

Mg was the third most abundant mineral, with concentrations ranging from 3037.92  $\pm$  13.87 mg/kg in RVI to 4096.44  $\pm$  8.54 mg/kg in RVIp. Ca ranked fourth, exhibiting levels ranging from  $978.40 \pm 3.27$ mg/kg in UVA to  $1618.06 \pm 0.62$  mg/kg in RVIp. Mg plays a vital role in protein synthesis, energy release, and enamel strength by aiding Ca retention, muscle relaxation, bone density, and blood pressure regulation, and it also acts as a neurotransmitter [\(Arya et al., 2016](#page-10-0); [Toomer,](#page-11-0)  [2018\)](#page-11-0). Ca is essential for teeth and bone structure and facilitates blood clotting processes. As seen in [Table 3](#page-6-0), roasting increased Ca levels across all samples except for RVAp, while Mg levels rose only in the oilcake of both varieties. [Kumar et al. \(2013\)](#page-11-0) claimed lower concentrations of Mg and Ca compared to those observed in this study. Also, they found that Mg and Ca levels decreased with peanut roasting from 210.30 and 89.30 mg/100 to 209.50 and 78.90 mg/100 g, respectively.

Na occupied the fifth position, with concentrations ranging from  $276.24 \pm 0.00$  mg/kg in UVI to  $1232.95 \pm 6.34$  mg/kg in RVIp. The Na contents varied significantly among the samples and were generally higher in roasted peanut flour and oilcake flour for both varieties, except for RVAp.

The remaining minerals were found in small quantities, below 100 mg/kg, as follows: Zn (53.98  $\pm$  0.61 to 81.77  $\pm$  0.44 mg/kg), Fe (31.56  $\pm$  0.33 to 56.80  $\pm$  0.17 mg/kg), B (32.83  $\pm$  0.21 to 45.44  $\pm$  0.36 mg/ kg), Mn (17.17  $\pm$  0.03 to 31.16  $\pm$  0.01 mg/kg), and Cu (7.26  $\pm$  0.01 to  $15.46 \pm 0.04$  mg/kg) [\(Table 3\)](#page-6-0).

As indicated in Table 2S, significant varietal differences were observed for nearly all minerals, particularly Na, Ca, Zn, B, and Mn. The Valencia type exhibited higher levels of Na, Zn, Fe, B, and Mn, whereas the Virginia type displayed higher levels of Ca. Similar findings were reported by [Eshun et al. \(2013\),](#page-11-0) who found substantial differences in the elemental profile among four peanut varieties from Ghana. It can be inferred that both peanut flour and oilcake flour are rich in minerals, making them valuable ingredients for developing new food products.

## *3.3. Analysis of the extract*

*3.3.1. Total polyphenol (TPC) and flavonoid (TFC) content*

Phenolic compounds play a crucial role in preventing lipid

#### <span id="page-6-0"></span>**Table 3**

Mineral profiling (mg/kg) of peanut flour and oilcake flour from two varieties, based on roasting treatment.



Results are presented as means  $\pm$  standard deviation (n = 2). Values with different letters within each column are significantly different at a significance level of p < 0.05. UVI and UVIp: Unroasted Virginia flour and oilcake, respectively; RVI and RVIp: Roasted Virginia flour and oilcake, respectively; UVA and UVAp: Unroasted Valencia flour and oilcake, respectively; RVA and RVAp: Roasted Valencia flour and oilcake, respectively.

peroxidation and inhibiting oxidizing enzymes, thereby mitigating oxidative stress and related health problems ([Asen et al., 2021](#page-10-0)). Results of the TPC and TFC of the methanolic peanut extracts demonstrate an

#### **Table 4**

Content of secondary metabolites and antioxidant activity in peanut flour and oilcake flour extracts from two varieties, based on roasting treatment.



Results are presented as means  $\pm$  standard deviation ( $n = 3$ ). Values with different letters within each column are significantly different at a significance level of  $p \le 0.05$ . TPC: total polyphenolic content; TFC: total flavonoid content. UVIs and UVIm: Unroasted Virginia flour extracted by Soxhlet and maceration, respectively; RVIs and RVIm: Roasted Virginia flour extracted by Soxhlet and maceration, respectively; UVIp and RVIp: Unroasted and roasted Virginia oilcake extracted by Soxhlet, respectively; UVAs and UVAm: Unroasted Valencia flour extracted by Soxhlet and maceration, respectively; RVAs and RVAm: Roasted Valencia flour extracted by Soxhlet and maceration, respectively; UVAp and RVAp: Unroasted and roasted Valencia oilcake extracted by Soxhlet, respectively.

observable effect of processing, as illustrated in Table 4.

For both varieties, there was a significant increase in TPC from 24.56  $\pm$  0.32–54.56  $\pm$  1.11 mg GAE/g (unroasted) to 28.64  $\pm$  0.19–62.79  $\pm$ 1.18 mg GAE/g (roasted). Similarly, TFC showed an increase from 3.41  $\pm$  0.06–14.72  $\pm$  0.06 mg QE/g (unroasted) to 4.20  $\pm$  0.07–18.35  $\pm$ 0.06 mg QE/g (roasted). These values closely align with those reported by [Yunusa et al. \(2023\)](#page-11-0) for peanut kernel, with TPC ranging from 39.34 (unroasted) to 53.89–67.26 (roasted) mg GAE/g, and TFC varying from 4.12 (unroasted) to 10.22–12.91(roasted) mg QE/g. Additionally, [Win,](#page-11-0)  [Abdul-Hamid, Baharin, Anwar, Sabu, and Pak-Dek \(2011\)](#page-11-0) stated that roasted peanut flour exhibited a higher TPC of 1.17 mg GAE/g compared to raw kernel (0.92 mg GAE/g).

Another study by [Win, Abdul-Hamid, Baharin, Anwar, and Saari](#page-11-0)  [\(2011\)](#page-11-0) suggested that whether raw or roasted, peanut kernel flour with skin contained significantly higher TPC than peanut flour without skin, possibly due to the presence of certain phenolics such as proanthocyanidins (condensed tannins) in the skin. However, the reported values from both studies were lower than those observed in our study. It is anticipated that heat treatment likely liberated bound phenolics into simpler free forms in peanut samples, thereby enhancing the overall TPC ([Chukwumah et al., 2007](#page-11-0)). Moreover, the rise in TPC could also be ascribed to Maillard reaction products formed during roasting, potentially contributing to higher absorbance readings as measured by the Folin assay [\(Win, Abdul-Hamid, Baharin, Anwar,](#page-11-0) & Saari, 2011).

Based on variety, Virginia had the greatest TPC and TFC among both unroasted and roasted samples, ranging from 37.46  $\pm$  0.72 to 62.79  $\pm$ 1.18 mg GAE/g and from 10.23  $\pm$  0.13 to 18.35  $\pm$  0.06 mg QE/g, respectively (Table 3S). Notably, the values for RVIp and UVIp were the highest recorded. In a similar study conducted by [Kaur et al. \(2024\)](#page-11-0), both raw and roasted flaxseed cake flour exhibited higher TPC and TFC compared to flaxseed flour. This might be attributed to the accumulation of these compounds in the cake resulting from the cold pressing extraction of oil.

In the present study, phenolic compounds were extracted from defatted peanut flour using cold (maceration) and hot (soxhlet) extraction methods. As illustrated in Table 3S, the extracts obtained through maceration displayed higher TPC and TFC compared to those extracted by the Soxhlet method. A similar trend was noted by [Eddahhaoui et al.](#page-11-0)  [\(2022\)](#page-11-0) in the two parts of the fruit of *Chamaerops humilis* (pulp and seeds). The TFC and TPC results for each variety are presented in the following order: For Virginia: RVIm*>*UVIm*>*RVIs*>*UVIs, and for Valencia: RVAm*>*UVAm*>*RVAs*>*UVAs. The observed discrepancies in TPC and TFC could be explained by the difference in thermal stabilities of certain phenolic compounds during the extraction and thermal processing of seeds [\(Kaur et al., 2024\)](#page-11-0).

#### *3.3.2. Antioxidant assays (DPPH and ABTS)*

Free radicals, known as reactive oxygen species, induce oxidative damage to macromolecules. In turn, phenolic components in plants serve as potent antioxidants by donating hydrogen or electrons and forming stable radical intermediates [\(Asen et al., 2021\)](#page-10-0). The current work used two complimentary assays (DPPH and ABTS) to provide a thorough understanding of the antioxidant capabilities of peanut flour and oilcake flour methanolic extracts. The findings of these assays are presented in [Table 4.](#page-6-0)

It can be clearly seen that the process of roasting significantly enhanced the antioxidant activity of the samples under investigation, leading to a notable decrease in the IC<sub>50</sub> value. For each variety, among all extracts, the methanolic extract from roasted oilcake exhibited strong activity against DPPH with IC<sub>50</sub> values of 30.81  $\pm$  0.32 and 120.89  $\pm$ 0.75 μg/mL for RVIp and RVAp, respectively. Conversely, the weakest scavenging property was observed for UVIs and UVAs, with  $IC_{50}$  values of  $134.78 \pm 0.80$  and  $166.61 \pm 0.60$  µg/mL, respectively. Additionally, moderate DPPH scavenging property was noted in RVIm  $(63.53 \pm 0.78)$ μg/mL) and RVAm (127.32  $\pm$  1.07 μg/mL).

Among the varieties, the extracts from Virginia demonstrated a significant anti-free radical capability, with their DPPH radical scavenging activity ranked in the following order: RVIp*>*RVIm*>*R-VIs*>*UVIp*>*UVIm*>*UVIs (Table 3S). Likewise, Valencia extracts exhibited a comparable ranking in antioxidant activity. A similar behavior was observed towards the ABTS radical. Accordingly, the results of the ABTS assay indicated that, for both varieties, the inhibitory action of the extracts improved following roasting, with RVIp and RVIm displaying the strongest antioxidant activity against ABTS and recording the lowest IC<sub>50</sub> values of 92.44  $\pm$  0.97 and 112.04  $\pm$  0.89, respectively. This might be attributed to the cake's enrichment with phenolic compounds, as evidenced by TPC and TFC analysis.

In agreement with our results, [Yunusa et al. \(2023\)](#page-11-0) revealed that the highest DPPH scavenging property was seen in the roasted peanut samples with IC<sub>50</sub> ranging from  $442.41 \pm 3.23$  to  $471.44 \pm 3.47$  µg/mL compared to unroasted peanut (697.96  $\pm$  31.84). The reported values were extremely higher than those obtained in our study, likely due to the differences in cultivar, growth conditions, processing, and extraction

method used. A similar trend in roasting's impact on the antioxidant capacity of peanut kernel flour was previously documented ([Win, Abdul-](#page-11-0)[Hamid, Baharin, Anwar,](#page-11-0) & Saari, 2011; [Win, Abdul-Hamid, Baharin,](#page-11-0)  Anwar, Sabu, & [Pak-Dek, 2011](#page-11-0)). In the same vein, [Kaur et al. \(2024\)](#page-11-0) revealed an increase in the free, bound, and total antioxidant capacity of both raw and roasted flaxseed cake flour compared to flaxseed flour.

#### *3.3.3. Antibacterial activity*

Antibiotic resistance stands as a critical global challenge, intricately linked with the unintended or excessive use of drugs and has spurred significant interest in exploring novel antimicrobial agents derived from plants [\(Nasri et al., 2022](#page-11-0)). Preliminarily, the antibacterial activity of peanut extracts was qualitatively and quantitatively evaluated by disc diffusion method and agar dilution method against a panel of bacteria, including two Gram-positive strains (*Staphylococcus aureus* and *Staphylococcus epidermidis*) and four Gram-negative strains (*Klebsiella pneumonia*, *Acinetobacter baumannii*, *Escherichia coli,* and *Enterobacter cloacae*). Penicillin and amoxicillin served as reference antibiotics for comparative analysis.

The findings regarding the antibacterial potential of the investigated peanut extracts are summarized in Table 5. Notably, all extracts demonstrated considerable effectiveness against gram-negative bacteria, exhibiting inhibition zone diameters ranging from  $6.3 \pm 0.0$  to 7.0  $\pm$  0.1 mm. Conversely, there was no sensitivity observed for grampositive strains, except for RVAm. The control wells, containing only solvent, did not exhibit any zone of inhibition, suggesting that the solvents themselves did not hinder bacterial growth.

The aromatogram test results unveil a variability in the inhibitory efficacy of the studied extracts. In fact, RVAm showed various inhibitory effects against four bacterial species. The largest zone of inhibition was observed against *S. epidermidis* and *K. pneumonia* (7 ± 0.0 mm), followed by *A. baumannii*  $(6.5 \pm 0.0 \text{ mm})$  and *E. coli*  $(6.4 \pm 0.1 \text{ mm})$ . Additionally, the RVAm extract displayed the highest effectiveness against *K. pneumoniae*, producing a zone of inhibition of  $7 \pm 0.0$  mm, compared to UVIp, UVAp, and RVIm  $(6.5 \pm 0.0 \text{ mm})$ . Moreover, the largest zone of inhibition against *A. baumannii* was observed with RVAp, while the smallest zone was obtained with UVIm. Furthermore, only UVAp exhibited effectiveness against *E. cloacae* (6.5 mm). However, the remaining extracts showed no discernible effect on any of the tested strains.

Our findings unveiled that the antibacterial efficacy of the studied

#### **Table 5**

Antibacterial activity of peanut flour and oilcake flour extracts assessed using the disc diffusion method, with the inhibition zone size measured in millimeters.

Inhibition zone diameter (mm)			Gram-positive bacteria		Gram-negative bacteria			
		S. aureus	S. epidermidis	K. pneumoniae	E. coli	A. baumannii	E. cloacae	
Unroasted								
Virginia	<b>UVIs</b>	$\overline{\phantom{0}}$	$\hspace{0.1mm}-\hspace{0.1mm}$	$\overline{\phantom{0}}$	$\hspace{0.1mm}-\hspace{0.1mm}$	$\qquad \qquad -$	$\qquad \qquad -$	
	<b>UVIm</b>	$\overline{\phantom{0}}$	$\hspace{0.1mm}-\hspace{0.1mm}$		$\qquad \qquad$	$6.3 \pm 0.0^a$	$6.5 \pm 0.1^{\rm a}$	
	UVIp	$\overline{\phantom{0}}$	$\hspace{0.1mm}-\hspace{0.1mm}$	$6.5 \pm 0.0^a$	$\overline{\phantom{0}}$	-	-	
Valencia	<b>UVAs</b>	-	$\overline{\phantom{a}}$	$\overline{\phantom{0}}$	$\hspace{0.1mm}-\hspace{0.1mm}$	$\overline{\phantom{0}}$	-	
	<b>UVAm</b>	-	$\overline{\phantom{a}}$	$\overline{\phantom{0}}$	$\hspace{0.1mm}-\hspace{0.1mm}$	-	-	
	<b>UVAp</b>	$\overline{\phantom{0}}$	$\overline{\phantom{a}}$	$6.5 \pm 0.0^a$	$\overline{\phantom{0}}$	$6.6 \pm 0.1^{\rm b}$	$\overline{\phantom{0}}$	
Roasted								
Virginia	<b>RVIs</b>	-			$\overline{\phantom{a}}$	-		
	RVIm	-	$\overline{\phantom{a}}$	$6.5 \pm 0.0^a$	$\overline{\phantom{0}}$	-	-	
	RVIp	$\overline{\phantom{a}}$			$\hspace{0.1mm}-\hspace{0.1mm}$	$\overline{\phantom{a}}$		
Valencia	<b>RVAs</b>	-	$\overline{\phantom{a}}$	$\overline{\phantom{0}}$	$\overline{\phantom{0}}$	$\qquad \qquad -$	-	
	<b>RVAm</b>	-	$7.0\pm0.0^{\rm a}$	$7.0 \pm 0.0^{\rm b}$	$6.4 \pm 0.1^a$	$6.5 \pm 0.0^{\circ}$	$\overline{\phantom{0}}$	
	<b>RVAp</b>					$7.0 \pm 0.0$ <sup>d</sup>		
Control								
Penicillin		-		$\overline{\phantom{0}}$	$\qquad \qquad$	-		
Amoxicillin		$13.0 \pm 0.0^a$	$\overline{\phantom{a}}$	-	$\hspace{0.1mm}-\hspace{0.1mm}$	-	-	

The size of the inhibition zone is measured in millimeters (mm). Results are presented as means  $\pm$  standard deviation (n = 3). Values with different letters within each column are significantly different at a significance level of  $p \le 0.05$ . UVIs and UVIm: Unroasted Virginia flour extracted by Soxhlet and maceration, respectively; RVIs and RVIm: Roasted Virginia flour extracted by Soxhlet and maceration, respectively; UVIp and RVIp: Unroasted and roasted Virginia oilcake extracted by Soxhlet, respectively; UVAs and UVAm: Unroasted Valencia flour extracted by Soxhlet and maceration, respectively; RVAs and RVAm: Roasted Valencia flour extracted by Soxhlet and maceration, respectively; UVAp and RVAp: Unroasted and roasted Valencia oilcake extracted by Soxhlet, respectively.

peanut extracts was more effective than that of the reference antibiotics. Specifically, penicillin (5 μg) exhibited inactivity against all studied bacteria, whereas amoxicillin (25 μg) demonstrated activity solely against *S. aureus* ( $13 \pm 0.0$  mm). The relatively modest inhibition zones observed may be attributed to several factors, including agro-climatic conditions, the handling of extracts, and the composition of phytochemical compounds within the extracts.

No prior research has explored the antibacterial activity of peanut flour and oilcake flour extracts. Nonetheless, studies have been conducted on the skin and hull of the peanut. Previously, [AL-Azawi and](#page-10-0)  [Hassan \(2017\)](#page-10-0) reported that methanolic peanut skin extract exhibited notable effects against *S. aureus*, with inhibition zones of  $10.67 \pm 0.67$ ,  $13.00 \pm 1.00$ , and  $14.67 \pm 0.88$  at concentrations of 25, 50, and 100 mg/mL respectively. In the same vein, [Prakash et al. \(2018\)](#page-11-0) highlighted that groundnut hull ethanolic extract inhibited *E. coli* (14 mm), *Salmonella enterica* (14 mm), *S. aureus* (17 mm), and *Streptococcus pyogenes* (20 mm), while the groundnut hull methanolic extract effectively inhibited the growth of *Shigella flexneri* (14 mm) only. An investigation conducted on *Nigella sativa* and flaxseed extracts revealed that both extracts shared the same minimum inhibitory concentration of 12.5 mg/mL against *Streptococcus pyogenes* ([Alahmad et al., 2020](#page-10-0)).

## *3.4. Correlation matrix*

Table 6 presents the Pearson correlation coefficients for proximate composition (OC, MC, PC, AC, CC), EV, and macro-mineral elements (K, Mg, Na, Ca, P). Similarly, Table 7 displays the Pearson correlation coefficients for phenolic compound content (TPC and TFC) and antioxidant activity (DPPH and ABTS).

The correlation analysis revealed that OC was positively and significantly linked to EV ( $r^2 = 0.999$ ,  $p < 0.0001$ ), but negatively associated with MC ( $r^2 = -0.982$ ), AC ( $r^2 = -0.946$ ), and CC ( $r^2 = -0.950$ ). Conversely, MC exhibited a highly significant positive correlation with AC ( $r^2 = 0.956$ ,  $p < 0.05$ ) and CC ( $r^2 = 0.930$ ,  $p < 0.05$ ). Additionally, a strong positive correlation was observed between AC and CC ( $r^2$  = 0.857,  $p < 0.05$ ), as well as between AC and the mineral elements Mg ( $r^2$  $= 0.738$ ,  $p < 0.05$ ) and P ( $r<sup>2</sup> = 0.788$ ,  $p < 0.05$ ).

As shown in Table 7, there is a significant positive association (p *<* 0.0001) between TPC and TFC ( $r^2 = 0.967$ ), DPPH ( $r^2 = 0.763$ ), and ABTS ( $r^2$  = 0.891). Additionally, TFC demonstrated a highly positive correlation with both DPPH ( $r^2 = 0.720$ ,  $p < 0.05$ ) and ABTS ( $r^2 =$ 0.856). Furthermore, a strong positive correlation exists between the DPPH and ABTS assays ( $r^2 = 0.935$ ), indicating that the bioactive molecules present in the examined extracts are responsible for the scavenging power of these two free radicals. Similar trends of correlations were highlighted by other authors ([Gagour et al., 2022](#page-11-0); [Ibourki et al.,](#page-11-0)  [2022; Oubannin et al., 2022\)](#page-11-0).

#### **Table 7**

Pearson's correlation matrix coefficients between the variables: Phenolic compound content (TPC and TFC) and antioxidant activity (DPPH and ABTS) of unroasted and roasted peanut flour and oilcake flour extracts.



#### *3.5. Principal component analysis*

Principal component analysis (PCA), an efficient discriminative approach, was used as a multivariate statistical method to discriminate among peanut flour and peanut cake flour varieties based on the studied parameters (dependent variables).

[Fig. 1](#page-9-0)A illustrates the PCA projection of various components, including proximate composition (OC, MC, PC, AC, and CC), EV, and macro-minerals (K, Mg, Na, Ca, and P), on the factorial planes F1–F2. The first principal component (F1) accounts for 64.54 % of the total variance, while the second principal component (F2) explains 18.79 %. Together, these two principal components capture 83.33 % of the total variance, indicating that their linear combination sufficiently represents the variables, surpassing the 50 % threshold. Thus, the first two axes are appropriate for representing the data. Additionally, [Fig. 1A](#page-9-0) illustrates the distribution of eight samples into three distinct groups.

Group I, which includes RVA, RVI, UVA, and UVI, exhibits strong contents of EV and OC as well as moderate values of Na, PC, and Ca. Moreover, they show lower values of K, P, CC, MC, Mg, and AC compared to groups II and III. Group II comprises UVAp and RVAp, characterized by high contents of Na, PC, and Ca, and average values of K, P, CC, MC, Mg, and AC, while displaying lower values of EV and OC relative to group I. Group III consists of UVAp and RVAp, distinguished by high contents of K, P, CC, MC, Mg, and AC, moderate values of Na, PC, and Ca, and lower EV and OC values compared to Group I.

[Fig. 1](#page-9-0)B presents the PCA projection of TPC, TFC, DPPH, and ABTS on the F1 and F2 factorial planes. F1 accounts for 89.22 % of the total variance, while F2 explains 9.19 %, with a cumulative percentage of 98.39 % for the first two principal components. Consequently, the twelve examined extracts are distributed into three distinct groups. Group I, represented by UVIs, UVAp, RVIs, and RVAp, is associated with lower phenolic content (TPC and TFC) and antioxidant activity (DPPH and ABTS). Group II, comprising RVIm, RVIp, UVIm, and RVIp, is distinguished by high levels of TPC, TFC, DPPH, and ABTS. Group III encompasses four individuals (RVAs, RVAm, UVAm, UVAs), exhibiting

**Table 6** 







<span id="page-9-0"></span>

**Fig. 1.** Principal component analysis describing the relationship among the investigated parameters of peanut flour and peanut cake flour samples. **A**: Proximate composition (OC, MC, PC, AC, and CC), energy value (EV), and mineral elements (K, Mg, Na, Ca, and P). **B**: Phenolic compound content (TPC and TFC) and antioxidant activity (DPPH and ABTS). GI: Group I; GII: Group II; and GIII: Group III.

medium levels of TPC, TFC, DPPH, and ABTS compared to Group II. These findings are in accordance with the correlation analysis results.

## *3.6. Hierarchical clustering analysis (HCA)*

Hierarchical clustering analysis (HCA) was employed to assess correlation and visualize similarity among the peanut samples based on the same variables used in the earlier PCA analysis. The samples were classified using Ward's method and the Euclidean square to evaluate similarity. The resulting dendrogram, depicted in [Fig. 2A](#page-10-0), indicates that the eight samples were grouped into three distinct clusters.

Cluster I, comprising RVA, RVI, UVA, and UVI, represents 50 % of the total samples. These individuals are characterized by high average values of OC (43.08 %), and EV (603.82 kcal/100 g), alongside moderate mean values of PC (53.02 %), and K (12,808.75 mg/kg). Conversely, they display lower average values of MC (1.79 %), AC (1.25 %), CC (0.99 %), Mg (2934.29 mg/kg), Na (467.12 mg/kg), Ca (1230.61 mg/kg) and P (6637.8 mg/kg).

Cluster II contains two samples (RVIp and UVIp), accounting for 25 % of the total samples. These samples exhibit higher average values of MC (6.23 %), PC (60.84 %), AC (6.03 %), and Ca (1478.75 mg/kg). This cluster also shows moderate mean values of CC (22.05 %), Mg (3838.84 mg/kg), Na (838.25 mg/kg), and P (7687.41 mg/kg), with lower mean values of OC (4.86 %), EV (375.29 kcal/100 g), and K (12,421.11 mg/ kg).

Cluster III consists of RVAp, and UVAp samples, representing 25 % of the total samples. These samples are distinguished by elevated mean values of CC (26.85 %), K (14,249.59 mg/kg), Mg (4043.92 mg/kg), Na (840.82 mg/kg), and P (8470.96 mg/kg). Additionally, this cluster demonstrates moderate mean values of OC (10.34 %), MC (5.77 %), and PC (51.88 %).

[Fig. 2B](#page-10-0) depicts the dendrogram showcasing the clustering of twelve peanut extracts into three distinct groups. Cluster I, which includes UVIs, UVAp, RVIs, and RVAp, constitutes 33.33 % of the total samples. This cluster exhibits moderate average values of TPC (36.50 mg GAE/g) and TFC (9.69 mg QE/g). Cluster II, comprising RVIm, RVIp, UVIm, and UVIp, represents another 33.33 % of the total samples, characterized by higher mean values of TPC (53.45 mg GAE/g) and TFC (14.45 mg QE/g). Consequently, their antioxidant activity is notably stronger compared to both Cluster III and Cluster I. Cluster III, which includes RVAs, RVAm, UVAm, and UVAs, also represents 33.33 % of the total samples. These samples are distinguished by lower mean values of TPC (28.92 mg GAE/ g) and TFC (4.373 mg QE/g), resulting in comparatively lower antioxidant activity. These findings align with the PCA data, where the distribution of all peanut extracts on the score plot reflects a similar pattern.

## **4. Conclusion**

In this study, the impact of roasting and varietal differences on

<span id="page-10-0"></span>

**Fig. 2.** Dendrogram of the studied peanut flour and oilcake flour varieties analyzed by cluster analysis using Ward's method and Euclidean distance. **A**: Proximate composition (OC, MC, PC, AC, and CC), energy value (EV), and mineral elements (K, Mg, Na, Ca, and P). **B**: Phenolic compound content (TPC and TFC) and antioxidant activity (DPPH and ABTS). CI: Cluster I; CII: Cluster II; CIII: Cluster III.

nutritional composition, phenolic content, and biological properties of peanut flour and cake flour were evaluated. Proximate composition and elemental profiling revealed significant disparities among peanut varieties and with roasting. The Virginia variety showed the highest OC and EV. Both varieties demonstrated protein-rich profiles, exceeding 50 %, with the highest levels found in UVAp and UVIp. Additionally, oilcake flour from both varieties, whether raw or roasted, exhibited higher MC, AC, and CC compared to peanut flour. Significant variations were noted in mineral concentrations, with the Valencia type showing higher levels of K, P, Zn, Fe, and B, and the Virginia type displaying higher Na, Ca, P, and Mn levels. Roasting influenced mineral content, with certain minerals increasing and others decreasing. TPC and TFC notably increased as a result of roasting, with the Virginia variety exhibiting the highest values. Furthermore, roasting enhanced antioxidant activity, as evidenced by a decrease in the  $IC_{50}$  value. The methanolic extract from roasted oilcake flour, particularly from the Virginia variety, displayed the strongest antioxidant activity against DPPH and ABTS radicals. All extracts exhibited efficacy against gram-negative bacteria, with varying inhibitory effects observed against four bacterial species. Conversely, gram-positive strains showed no sensitivity, except for RVAm, which exhibited inhibitory effects against various bacterial species. Based on the results of the present study, peanut flour, regardless of the variety, is of great interest from a nutritional perspective as a source of several nutritional elements. Peanut oilcake flour, a less valued by-product recovered from peanut oil processing, has presented a nutritional composition that cannot be disregarded.

### **CRediT authorship contribution statement**

**Zineb Lakhlifi El Idrissi:** Writing – original draft, Formal analysis, Data curation. **Asmaa Oubihi:** Methodology, Investigation, Formal analysis. **Mourad El Youssfi:** Writing – review & editing, Investigation, Data curation. **Said Gharby:** Supervision, Methodology, Investigation. **Chakir El Guezzane:** Software, Formal analysis. **Riaz Ullah:** Writing – review & editing, Investigation, Funding acquisition. **Zafar Iqbal:**  Writing – review  $\&$  editing, Validation, Methodology, Funding acquisition, Formal analysis. **Khang Wen Goh:** Visualization, Validation, Investigation, Funding acquisition, Data curation. **Monica Gallo:**  Investigation, Visualization, Writing – review & editing. **Abdelhakim Bouyahya:** Writing – review & editing, Supervision, Formal analysis, Data curation. **Hicham Harhar:** Writing – review & editing, Supervision, Project administration, Methodology, Conceptualization. **Mohamed Tabyaoui:** Writing – review & editing, Project administration, Methodology, Conceptualization.

### **Declaration of competing interest**

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

## **Data availability**

Data will be made available on request.

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#### **Appendix A. Supplementary data**

Supplementary data to this article can be found online at [https://doi.](https://doi.org/10.1016/j.fochx.2024.101791)  [org/10.1016/j.fochx.2024.101791](https://doi.org/10.1016/j.fochx.2024.101791).

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