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Metabolomic and biochemical insights into bioactive compounds and antioxidant properties of black oilseed testa and peeled seeds

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ABSTRACT

Black oilseed crops are rich in diverse phenolic compounds and have excellent antioxidant activities, as reported in traditional Chinese medicine. Testa (seed coat) and peeled seeds (cotyledon, embryo, and other structures) are the seed's crucial components, contributing to the variation in phytonutrient, phenol content, bioactive component, and protective and pharmacological effects. However, comprehensive and comparative information on total phenol, flavonoid, antioxidant, and metabolic profiles in black seed testa and peeled sesame, soybean, peanut, and rapeseed seeds is rare. Here, we investigated the metabolic profiles, phenolic contents, and antioxidant activities of four black oilseed crop testas and peeled seeds. This study revealed that testa has higher total phenol, flavonoid, and antioxidant activities than peeled seeds. A total of 1847 metabolites were identified across all samples and categorized into 17 major classes: flavonoids (20.02%), phenolic acids (15.15%), lipids (11.47%), amino acids and derivatives (9.36%), alkaloids (7.47%), organic acids (5.79%), terpenoids (5.68%), lignans (5.57%), saccharides (4.27%), and nucleotides and derivatives (4.17%) among the top ten. Primary class metabolites such as amino acids, saccharides, and vitamins were higher in the peeled seeds than in the testa, signifying the role of energy reservoirs and nutritive potential. However, flavonoids, phenolic acids, coumarins, chromones, lignans, terpenoids, tannins, organic acids, and lipids were abundant in the testa. Interestingly, the diversity and content of secondary metabolites were more abundant in the testa than in the peeled seeds of each crop, explaining their potential for phenol content, bioactivity, antioxidant activity, and pharmacological potential. The bioactivity of peeled seeds and testas may be associated with the phytochemical composition and content of flavonoids, phenolic acids, terpenoids, alkaloids, lipids, terpenoids, lignans, amino acids, and saccharides. Therefore, according to our results, peeled seeds offer higher nutritional value, and the testa has medicinal and protective properties. This study provides insights into the variations in phytochemical composition, phenolic content, and antioxidant activity of testa and peeled black sesame, soybean, peanut, and rapeseed seeds for further application of oilseeds in food products and to maximize nutritional benefits.

1. Background

Oilseeds are a significant source of nutritional, agricultural, economic, and pharmacological functions, with noticeable health benefits (Hadidi et al., 2024; Wei et al., 2022). Sesame, soybean, rapeseed, and peanut are the top oilseed crops produced and consumed worldwide as vegetable oils (Kefale et al., 2023; Wei et al., 2022; Tokel and Erkencioglu, 2021). Asian countries, including China, India, Japan, Korea, and

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Pakistan, produce oilseed crops for direct use (food), cooking oil, and other foods (Ye and Liu, 2023; Jati, 2020). These important crops have gained more attention in omics because of their rich composition of bioactive compounds in seeds and other plant parts (Zhang et al., 2023; Shahidi et al., 2006). The nutritional and bioactive components in oilseeds are vitamins, amino acids, organic acids, flavonoids, fatty acids, phenolic acids, terpenoids, alkaloids, lignans, coumarins, phytosterols, and quinones (Kefale et al., 2023; Ye and Liu, 2023; Zhang et al., 2023). Additionally, most oilseeds are rich in mineral nutrients, such as potassium, iron, zinc, magnesium, vitamins, plant-based proteins, and dietary fiber, which are essential for overall health, making them vital elements of a balanced diet (Sanni et al., 2024; W. Zhang et al., 2024). Similarly, lignans in sesame, glucosinolates in rapeseed, isoflavones in soybean, and resveratrol in peanuts have been identified as potential nutraceuticals and pharmaceuticals (Brigante et al., 2022; Zhang et al., 2022; Shen et al., 2021).

Today's society is increasingly concerned about the high rate of chronic illnesses such as diabetes, cancer, obesity, and cardiovascular diseases (Hadidi et al., 2023). Various bioactive metabolites have shown positive health-promoting effects in different disorders (Tao et al., 2023). However, comprehensive and comparative studies on global profiling and evaluation of biochemical and antioxidant activities between the testa and peeled parts of black seeds are limited. Environmental and genetic factors influence the composition and content of bioactive metabolites in plants and plant parts (El Hanafi et al., 2023; Choi et al., 2020). The colors of seeds and plant parts are genetically controlled factors that influence the variability, composition, and distribution of bioactive components. Testa (seedcoat) color is an important quantitative trait that determines the biochemistry of seeds, contributes to their nutritional and therapeutic value, and determines consumer preferences (Jiang et al., 2024; Dossou et al., 2022). For over a hundred years, traditional Chinese medicine has extensively used black seeds from various crops as herbal remedies and healthy foods. Compared to light-colored oil seeds, black seeds contain more phenolic chemicals, which have desirable nutritional and functional qualities for human health (Dossou et al., 2024; Wang et al., 2018). The phenolic and antioxidant capacities of black oilseed crops have led to their traditional use in herbal medicine owing to their potential health advantages. Moreover, black oilseed crops are employed in various culinary applications or cuisines because of their distinct flavor, texture, and color. Whole seeds and other parts of most oilseeds can be incorporated into multiple dishes, including stir-fries, salads, baked goods, and desserts. Therefore, seed color is a determining factor for consumption and market price, showing that black seeds of oil crops are preferred and more expensive than light/white seeds (Dossou et al., 2022). Previous studies have shown that black sesame has higher total phenolic content and antioxidant activity than white sesame (Wang et al., 2018; Shahidi et al., 2006). Similarly, black and brown soybean lines showed higher antioxidant activity and anthocyanin and isoflavone contents than light-yellow soybeans (Lim et al., 2021). Black ground nut (peanut) species have higher total phenol and antioxidant capacities than red, brown, and mixed samples (Adedayo et al., 2021).

The development of oilseed crops with higher nutritional quality and antioxidant activity is the ultimate goal of the current novel breeding of oilseed crops. The dietary qualities of oilseed crops are mainly associated with testa and color. However, despite being a reservoir for various bioactive and nutritional components, the testa of most oilseed crops is removed during industrial food processing. Moreover, recent trends in human nutrition have experienced a significant shift, and processed foods have assumed the role of traditional foods (El Hanafi et al., 2023). As a result, the testa of most crops is often removed (dehulled) during food processing and cooking to obtain peeled seeds, which are then processed into foodstuffs such as butter and oil (Kuang et al., 2017). Dehulling reduces the pharmacological and nutritional quality, antioxidant activity, and desirable characteristics of food and food products. This could be because the most important bioactive components contained in the testa could be removed during dehulling, resulting in the produce's desirability to decline. Previous studies have shown that peeled sesame, rapeseed, peanut, and soybean seeds contain bioactive compounds that have considerable nutraceutical, cosmetic, and medicinal applications in the era of metabolomics (Zhang et al., 2022; Lim et al., 2021; Shahidi et al., 2006). Further, various studies have indicated that seeds without seed coat (peeled seeds) have lower total phenolic, flavonoid, and antioxidant activities in soybean, peanut, sesame, and common beans (Lim et al., 2021; Chávez-Mendoza et al., 2019; Attree et al., 2015; Xu and Chang, 2008). The bioactive components, primarily flavonoids, phenolic acids, coumarins, amino acids, fatty acids, vitamins, lignans, terpenoids, and other polyphenols composition and content in the testa and peeled/hulled seed/results variations in bioactivity, antioxidant activity, nutritional quality, and other end-use products quality (Zhang et al., 2022; Lim et al., 2021). The removal of the seed coat led to a notable decrease in the levels of tannins, phytic acids, catechins, and quercetin (Pal et al., 2017). Consequently, there was a reduction in the Ferric Reducing Antioxidant Power (FRAP), absorbance (ABS), and total antioxidant activity (TAA) (Pal et al., 2017). These findings underscore the critical role of phenolic components in the potential bioactivity of the seeds. Consumption of seeds and seed products rich in those bioactive compounds can significantly enhance human health benefits such as preventing chronic diseases (cardiovascular and cancer), protecting cells from oxidative stress, and improving metabolic processes (Hou et al., 2022; Zhang et al., 2022). Therefore, assessing how different industrial processes affect food biochemistry and antioxidant performance is highly encouraged.

Advancements in omics technologies have allowed the investigation of metabolite composition and distribution in biological samples, particularly in the study of small molecules present in seeds and their links to medicinal and nutritional attributes (Lolli and Caligiani, 2024). Given the increasing public concern for food authenticity, targeted metabolomics profiling has been widely employed to efficiently identify diverse compounds in the global metabolome of plant organs (Zhang et al., 2023; Kefale et al., 2023; Manickam et al., 2023). Therefore, we performed widely targeted metabolomic profiling using ultra-high-performance liquid chromatography coupled with tandem mass spectrometry (UHPLC-MS/MS) to uncover the variation in metabolic profiles between the testa and peeled seeds of black sesame, soybean, peanut, and rapeseed. Our study revealed significant variations in the metabolic profiles and phenolic and antioxidant contents between testa and peeled seeds, which can provide valuable scientific information for nutritional and therapeutic purposes and guide future research on the bioactive ingredients of these seeds.

2. Material and method

2.1. Preparation of plant materials

Black sesame (SM), soybean (SB), peanut (PN), and rapeseed (RS) seeds offered by the Oil Crops Research Institute, Chinese Academy of Agricultural Sciences (OCRI-CAAS), Wuhan, China, were used to investigate metabolite variations among the samples. Except for rapeseed, the other seeds were cultivated under identical environmental conditions from June to September 2022 at the OCRI experimental field station (N 30.57°, E 114.30°, 27 m altitude) in Wuhan, China. Rapeseeds were grown at the same location from October 2022–May to 2023. Seed samples were harvested from ten representative plants of each genotype. After harvesting, seeds were maintained at a moisture content of 10%. The testa was then peeled and separated from the seed to obtain the testa and peeled seeds using forceps. Finally, 24 dried samples were prepared in triplicate and stored at −80 °C until samples from both the peeled seeds and testa of each species were used for metabolic profiling, total phenolic content, total flavonoid content, and antioxidant analysis (Table S1 and Fig. 1).

Fig. 1. Morphological appearances of peeled seeds and testa of the samples. The sample labels are described in Table S1.

2.2. Materials and reagents

HPLC-grade solvents, such as pure methanol (CH_3OH), acetonitrile, and acetic acid, were obtained from Merck, Germany. Formic acid from Aladdin and ethanol were purchased from Sigma-Aldrich (St. Louis, MO, USA). Additionally, 2-chlorophenyl alanine (98%, w/w) was purchased from Bailingwei Technology Co., Ltd, China.

2.3. Sample extraction process

Freeze-dried biological samples were prepared and subjected to a mixer mill (MM 400, Retsch, Haan, Germany) for (30 Hz, 1.5 min) until it becomes powdery. Then, 50 mg of sample powder was extracted at 4 °C (overnight) with 1200 μL of 70% methanol (v/v), vortexed once every 30 min, each time lasting 30 s, a total of 6 vortex times. After centrifugation (12,000×*g* for 20 min at 4 °C), the supernatants were collected and filtered with a microporous membrane (0.22 μm pore size, SCAA-104, ANPEL, Shanghai, China), and stored in a vial at −20 °C until UHPLC-ESI-QqQLIT-MS/MS analysis at Metware Biotechnology Co., Ltd., (MWDB), Wuhan, China. Equal volumes of all sample seed extracts were mixed to constitute QC (quality control) samples.

2.4. The UHPLC-MS/MS conditions

The data acquisition instrument system includes ultra-high-performance liquid chromatography (UHPLC) (ExionLC™ AD, https://sciex.com.cn/) and tandem mass spectrometry (MS/MS). The liquid phase was adjusted to Agilent SB-C18, 1.8 μm, 2.1 mm * 100 mm column, and ultrapure water (with 0.1% formic acid) for the mobile phase A and acetonitrile (with 0.1% formic acid) was used for phase B. The elution gradient consisting of the proportion of phase B 5% in 0.00 min, the proportion of phase B increased linearly to 95% in 9.00 min, and remained at 95% for 1 min, the proportion of phase B decreased to 5% in 10.00–11.10 min, and equilibrated at 5% to 14 min. The flow rate was 0.35 mL/min, the column temperature was 40 °C, and the injection volume was 2 μL. An alternative connection for the effluent was made to the ESI-triple quadrupole linear ion trap (QTRAP-MS). Metabolomic analysis was carried out following previously outlined approaches (Dossou et al., 2024; Dossou et al., 2022). The MS conditions for the liquid phase are listed in Table S2.

2.5. Identification and quantification of metabolites

Spectrum information, mass spectra, and retention times were integrated to qualitatively identify phenolic compounds. Specifically, the values of Q1 (precursor ions) and Q3 (product ion), retention times, fragmentation patterns, collision energy, and de-clustering potential were allied with standards when available (Sigma-Aldrich, St. Louis, MO, USA). When no standards were available, the compounds were structurally confirmed using the MWDB self-build database and verified in open databases (KNApSAcK, MassBank, MoTo DB, HMDB, and METLIN) (W. Chen et al., 2013). After collecting the metabolite profile data from several samples, the peak areas of all chromatographic peaks of all species were integrated, and the mass spectrometry peaks of the same metabolite in different samples were integrated and corrected (Fraga et al., 2010). The contents of the identified compounds were calculated via triple quadrupole (QqQ) MS analysis (MRM modes) using integrated SCIEX-OS software (version 1.4).

2.6. Determination of total phenolic (TPC) and total Flavonoid contents (TFC)

The extraction and determination of the total phenolic and flavonoid content of the samples were performed according to the method described by Choi et al. (2023; Dossou et al., 2023). Briefly, seeds (0.5 g) were mixed with 5 mL of 80% ethanol (v/v), followed by extraction

(shaken in darkness) for 4 h and centrifugation at 5000×*g* for 15 min. The supernatants were collected and stored at −20 °C for further analyses for one week. Total phenol content was determined according to the method described by (Choi et al., 2023). An aliquot (100 μL) of each extract was mixed with 400 μL of distilled water and 100 μL of the Folin-Ciocalteu reagent. After 6 min, 1 mL 7% (m/v) Na_2CO_3 and 0.8 mL of distilled water were added. The mixture was incubated at room temperature for 90 min. Absorbance was recorded against a blank (80% ethanol (v/v)) at 760 nm using a spectrophotometer (UV5200, Shanghai Metash Instruments Co., Ltd., Shanghai, China). TPC was determined from a standard gallic acid curve ($y = 1.971x - 0.0068$, $R^2 = 0.99$) and expressed as milligrams of gallic acid equivalents (mg GAE/g) per gram of dry weight (DW). The total flavonoid content was measured as described by Choi et al. (2023; Dossou et al., 2023). The sample extract (1 mL) was mixed with 150 μL $NaNO_2$ and allowed to stand for 6 min. Next, 300 μL of $AlCl_3 \cdot 6H_2O$ was added, and the mixture was incubated for 6 min. Finally, 1 mL of 1 M NaOH and 1.05 mL of distilled water were added and mixed, and the absorbance was immediately read at 510 nm. The TFC was calculated from a standard catechin curve ($y = 3.253x + 0.1447$ ($R^2 = 0.9702$) and expressed as catechin equivalent (CAE) mg per gram of sample seeds (mg CAE/g).

2.7. Antioxidant activity

The extraction and determination of antioxidant activities was performed as described by the method in our newly published paper (Kefale et al., 2023) modified from Kim et al. (2014). We performed antioxidant evaluation of peeled seeds and testa (seedcoat) samples of black sesame, soybean, peanut, and rapeseed using the two most commonly used antioxidant assays, 2,2-Diphenyl-1-picrylhydrazyl free radical scavenging activity (DPPH) and ferric-reducing antioxidant potential (FRAP).

Briefly, 0.5 g of ground powder from each crop sample was extracted with 5 mL of 80 % ethanol (v/v) in a shaking incubator for 4 h at room temperature. The supernatants were filtered and carefully collected individually after centrifugation of 5000 rpm for 15 min. Then, the DPPH and FRAP assays were performed as follows;

DPPH Assay: The 1,1-diphenyl-1-picrylhydrazino radical (DPPH) assay was performed with a slight modification of the method reported by (Kim et al., 2014). Sample extracts of each crop seed (10 μL), 1, 1-diphnyl-1-picrylhydrazino radical (DPPH, 0.2 mM) (190 μL), and absolute ethanol solution in 96-well plates were incubated for 30 min at room temperature in darkness. The absorbance of the experiment was determined at 515 nm wavelength using a microplate reader. The blank consists of 10 μL of 80 % ethanol (v/v) and 190 μL DPPH, whereas the control consists of absolute ethanol 190 μL and sample extracts of 10 μL. Finally, the scavenging activity result was expressed as a percentage using the following formula:

$$\text{DPPH radical scavenging effect (\%)} = [1 - (AS - AC) \div AB] \times 100.$$

Where, AS = absorbance of sample, AC = Absorbance of control, AB = absorbance of blank.

FRAP assay: The ferric ion-reducing antioxidant power (FRAP) assay was performed according to the method reported by Ahmed & Tavaszi-sarosi (2019) with little modification. To prepare an acetate buffer, 3.1 g of sodium acetate, 16 mL of acetic acid, and 11 mL of distilled water were combined in a measuring cylinder. Additionally, a solution of 2,4,6-tripyridyl-S-triazine (TPTZ) was prepared by dissolving 0.03123 g of TPTZ in 10 mL of distilled water and adding 33.6 μL of hydrochloric acid. Then, iron chloride ($FeCL_3$) 0.054 g was diluted with 10 mL of DW. Finally, the FRAP solution was prepared by mixing 50 mL acetate buffer, TPTZ solution (5 mL), and iron chloride (5 mL) solutions. To establish the standard curve, a solution of ascorbic acid (0.017613 mg) was prepared by dissolving it in 10 mL of deionized water (DW). Subsequently, 100 μL of this solution was diluted with an additional 900 μL of DW. A series of dilutions from an ascorbic acid stock solution were then created, comprising 10 μL, 20 μL, 30 μL, 40 μL, 50 μL, and 100 μL, which served as standard references. Additionally, a blank solution was prepared, containing 6 μL of 80% ethanol (v/v), 180 μL of FRAP solution, and 18 μL of DW. For the experimental analysis, 6 μL of the sample extract, 180 μL of FRAP solution, and 18 μL of DW were combined in test tubes. The absorbance of the samples was subsequently measured using a microplate reader (Thermo Evolution 201 spectrophotometer) at a wavelength of 593 nm. The FRAP values were calculated from the standard curve equation ($X = (Y + 0.0038)/0.005$). They revealed a good regression quotient ($R^2 = 0.999$) close to 1, where Y and X represent the absorbance and ascorbic acid concentration, respectively. The result was presented as mg ascorbic acid equivalent (mg AAE/g DM). The measurements were performed in triplicate.

2.8. Statistical analysis

Multivariate analyses were performed following z-score normalization and data quality assessment. With the help of the statistics packages pheatmap, prcomp, and MetaboAnalystR (www.r-project.org.), the PCA (principal component analysis), HCA (hierarchical clustering analysis), and OPLS-DA (orthogonal partial least squares discriminant analysis) were performed in R (version 3.5.0). Differentially accumulated metabolites or significant DAMs were identified by their fold-change (FC ≥ 2 or ≤ 0.5), VIP ≥1, and p-value ≤0.05. The values of variable importance in projection (VIPs) were considered from the OPLS-DA analysis. Ultimately, mapping to the Kyoto Encyclopedia of Genes and Genomes (KEGG) database and subsequent significant enrichment studies utilizing metabolite set enrichment analysis (MSEA) were used to carry out the functional annotation of DAMs. The *p*-values from the hypergeometric test were used to filter the considerably enriched pathways. To draw bar graphs and Venn diagrams, GraphPad prism (v9.0.0121, GraphPad 159 Software Inc., La Jolla, CA, USA), SRplot, Chip plot, and TB-tools software (v.1.9) were applied (C. Chen et al., 2020). The analysis of variance (ANOVA) test was used to compare groups, and statistical differences were determined at $P < 0.05$. Both technical and biological replicates were used for statistical analyses.

3. Results

3.1. Total phenols (TPC) and total Flavonoid (TFC) contents in the testa and peeled seeds

To reveal the potential of phenolic components, we evaluated the total phenol and flavonoid contents in the testa and peeled black sesame, soybean, peanut, and rapeseed seeds. We found significant variations ($P < 0.01$) in total phenol and total flavonoid contents between the testa and peeled seeds of the four oilseeds (Fig. s 2A and B). The TPC in the testa ranged from 24.9 to 36.31 mg GAE/g in TRS and TPN, while the peeled seeds ranged from 5.9 to 7.2 mg GAE/g in CPN and CRS, respectively (Table S3). As shown in Table S3, higher (36 mg CAE/g) and lower (17.2 mg CAE/g) TFC were observed in TSB and TRS, respectively. Similarly, CSB (22.6 mg CAE/g) and CPN (11.9 mg CAE/g) showed higher and lower TFC, respectively.

3.2. Antioxidant activities in the testa and peeled seeds of sesame, soybean, peanut and rapeseed

To validate the performance of phenolics and bioactive compounds, we evaluated the antioxidant activity potential of peeled and testa of black oilseeds using the most frequently used DPPH and FRAP assays (Fig. s 2C and D). A comparison of the peeled seeds and testa of each crop revealed highly significant differences ($P < 0.01$). TSB, TPN, and CSB showed high free radical scavenging activity (DPPH) and ferric-reducing antioxidant potential (FRAP) (Fig. s 2C and D). The maximum DPPH scavenging activity was recorded for TPN (107.5%)

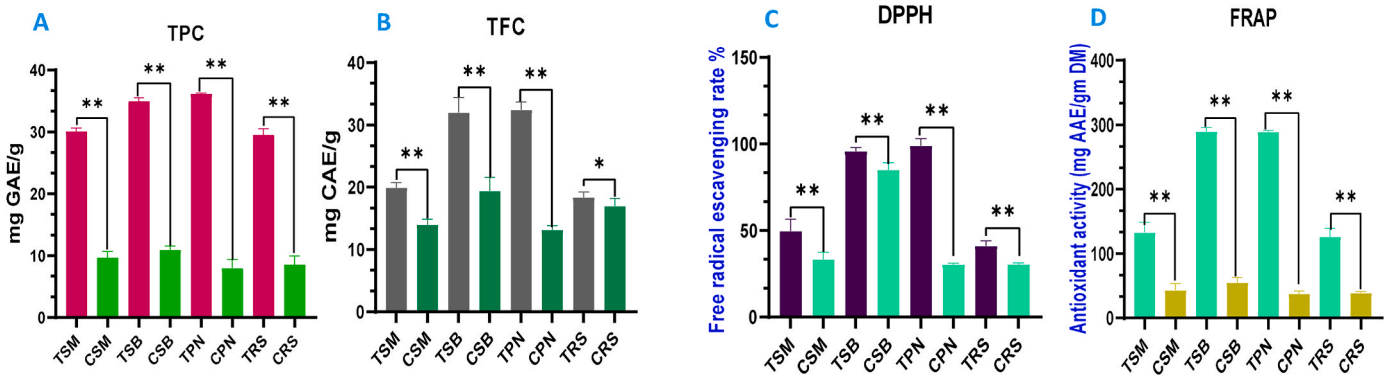

Fig. 2. Total phenol content (TPC), total flavonoid contents (TFC), antioxidant activity (free radical scavenging activity (DPPH assay) and ferric reducing antioxidant potential (FRAP assay) in the testa and peeled seeds of black sesame, soybean, peanut and rapeseed crops. (A) TPC and (B) TFC, (C) DPPH and (D) FRAP.

and CSB (96.2%), whereas the minimum was observed for TRS (37.5%) and CSM (28.64%) (Table S4). Likewise, elevated FRAP was recorded from TSB (299.36 mg AAE/g DM) and reduced in TRS (108.76 mg AAE/g DM) %) (Table S4). At the same time, a maximum of 62.16 mg AAE/g and 31.36 mg AAE/g was observed from CSB and CPN, respectively.

3.3. Metabolic profiles of peeled seeds and testa of four oilseeds

To understand the diversity and distribution of metabolites in peeled seeds and testa of black oilseeds, 24 samples were subjected to UHPLC-MS/MS. Upon initial examination of the raw data presented as base peak chromatograms and total ion currents (TICs), distinct qualitative and quantitative variations were observed (Fig. S1). A total of 1847 metabolites from negative (895) and positive (952) ionizations were detected and structurally identified (Fig. 3A and Table S5). Flavonoids (20.02%), phenolic acids (15.15%), lipids (11.47%), amino acids and derivatives (9.36%), alkaloids (7.47%), organic acids (5.79%), terpenoids (5.68%), lignans (5.57%), saccharides (4.27%), and nucleotides and derivatives (4.17%) were top ten dominant major classes of metabolites (Fig. 3A). However, coumarins, chromones, tannins, quinones, vitamins, phytohormones, and other compounds accounted for 12.01% of the total (Fig. 3A). Most (>61%) of the identified metabolites were secondary metabolites. Flavonoids and phenolic acids were the first top two classes. The upset plot revealed 865 overlapping metabolites when comparing CSM, TSM, CSB, TSB, CPN, TPN, CRS, and TRS (Fig. 3B). A total of 103 metabolites were unique/crop-specific (Table S6 and

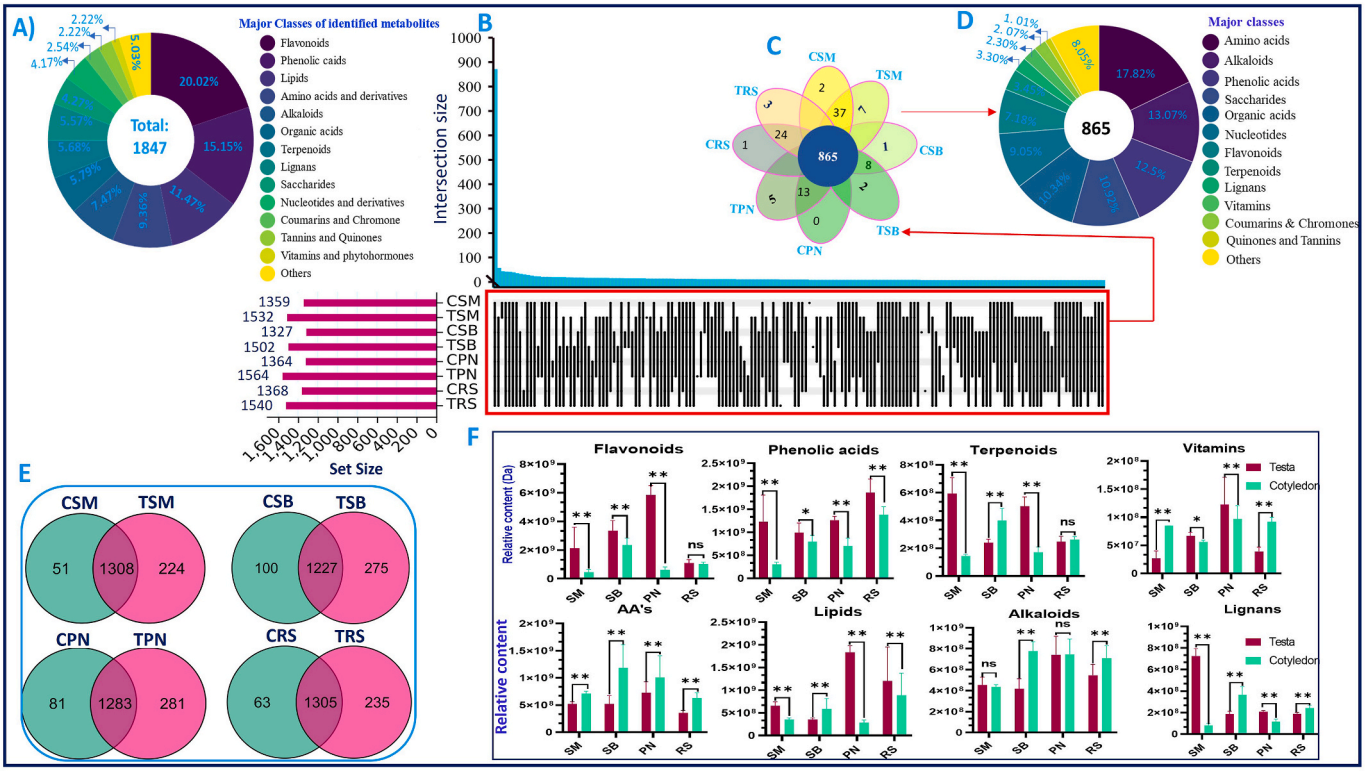

Fig. 3. Classification and variation of metabolites. (A) Classification of the 1847 identified metabolites in testa and peeled part samples of oilseeds. (B) Upset plot showing the number of metabolites identified in each crop seed components (CSM, TSM, CSB, TSB, CPN, TPN, CRS, and TRS) and shared metabolites. (C) Venn diagram showing the numbers of common and specific metabolites accumulated in the testa and peeled parts of oilseeds, (D) Classification of shared metabolites among testa and peeled parts of oilseeds, (E) Venn diagram showing the numbers of shared and unique metabolites accumulated in the CSM. vs.TSM, CSB. vs.TSB, CPN. vs.TPN, and CRS. vs.TRS. (F) Relative content of major classes of metabolites in the testa and peeled seeds of SM, SB, PN, and RS. C_: peeled seeds and T_: testa of sesame, soybean, peanut, and rapeseed. AA's: amino acids, and OA's: organic acids. The sample labels are described in Table S1.

Fig. 3C). As depicted in Fig. 3C and Table S6, a higher number of metabolites were specific to sesame (46), followed by rapeseed (28) and peanut (18), of which 21 were specific to the testa and peeled parts of some species. Furthermore, we classified the shared metabolites (865) into 13 primary classes with compositions and distributions different from those of the total metabolites (Fig. 3D). Amino acids, alkaloids, phenolic acids, saccharides, and nucleotides were the top five major classes shared among all compared oilseeds testa and peeled seeds.

3.4. The distribution characteristics of metabolites in the peeled seeds and testa of black sesame, soybean, peanut, and rapeseed

We performed a Venn diagram to compare the metabolic profiles and distribution characteristics of metabolites in the peeled seeds and testa of sesame, soybean, peanut, and rapeseed (Fig. 3E). The metabolite composition and distribution patterns of peeled seeds and testas showed greater variability. In the comparison between peeled seeds and testa of each crop, a higher number of metabolites were unique to the testa, TSM (224), TSB (275), TPN (281), and TRS (235), than peeled seeds of each seed (Fig. 3E). Similarly, 51, 100, 81, and 63 metabolites were unique to CSM, CSB, CPN, and CRS, respectively (Fig. 3E). Among the unique metabolite classes in the testa, AAs, flavonoids, OAs, phenolic acids, terpenoids, and lignans were dominant (Tables S7–10).

Furthermore, we compared the relative intensities of significant classes of metabolites, and higher variability ($P < 0.05$) was observed between the testa and peeled seeds of each crop (Fig. 4F and Fig. S2). A higher flavonoid content was observed in TPN, TSB, TSM, and CSB (Fig. 3F). AAs and vitamins were higher in the CSB, CPN, CSM), and TPN groups (Fig. 3F). CRS and TRS revealed elevated phenolic acid contents. Terpenoids and lignans were more abundant in TSM than in the other samples. Except for TPN, the others showed reduced vitamin content compared to the peeled seeds. TRS exhibited a higher content of coumarins, chromones, and other unclassified compounds (Fig. S2). Overall, increased amino acid, vitamin, and saccharide contents were observed in most peeled seeds. In contrast, higher contents of flavonoids, terpenoids, phenolic acids, coumarins, quinones, chromones, tannins, and other compounds were found in the testa (Fig. 3F and Fig. S2).

3.5. Multivariate analysis

To accurately evaluate the variability of metabolites between and within the groups, we performed multivariate analysis (Fig. 4A and B). Principal component analysis (PCA) and hierarchical cluster analysis (HCA) revealed that the metabolite profiles of peeled seeds and testas of black sesame (CSM, TSM), soybean (CSB, TSB), peanut (CPN, TPN), and rapeseed (CRS, TRS) were categorized into different clusters (Fig. 4A and B). The PCA plot clearly separated each sample into eight groups, without outliers (Fig. 4A). PC1 and PC2 explained the total variations by 46.5%. This was further confirmed by the HCA results, which showed distinct metabolic accumulation patterns in the eight groups (Fig. 4B). Additionally, the testa samples exhibited a higher accumulation pattern than the peeled seeds, indicating the richness of the testa with many metabolites (Figs. S3A–D). TSM, TPN, and TSB revealed higher accumulation patterns of most metabolites than the other groups. Furthermore, to validate the existing variability, we performed OPLS-DA analysis, and the results were highly predictive and had significant goodness of fit (Fig. 4C–F).

3.6. Differentially accumulated metabolites (DAMs) and pathways

Three thresholds, VIP ≥1.0, FC ≥ 1.2, or FC < 0.833, and *P*-value ≤0.05, were applied to filter all DAMs from the pairwise comparison across all comparison groups. Volcano plots revealed significantly accumulated differential metabolites (DAMs) (Fig. 5A and B, Figs. S4A and B). The pairwise comparisons between the testa and peeled seeds of each crop (TSM.vs.CSM, TSB. vs.CSB, TPN. vs.CPN, and TRS. vs.CRS) showed a total of 1004 (741 up in TSM), 923 (520 up in TSB), 957 (704

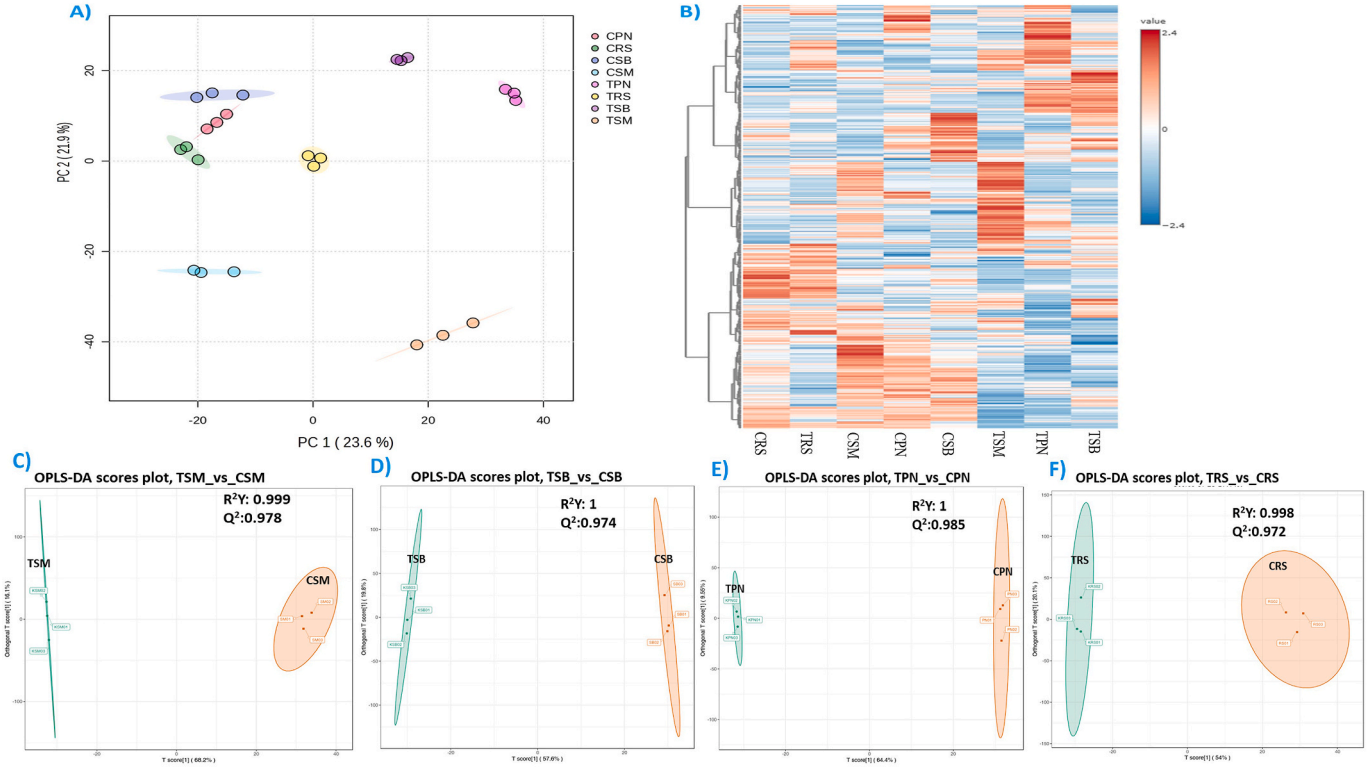

Fig. 4. Multivariate analysis showing: A) principal component analysis (PCA), B) hierarchical clustering analysis (HCA), (C–F), OPLS-DA scores plot in a pairwise comparison between the testa and peeled seeds of each crop.

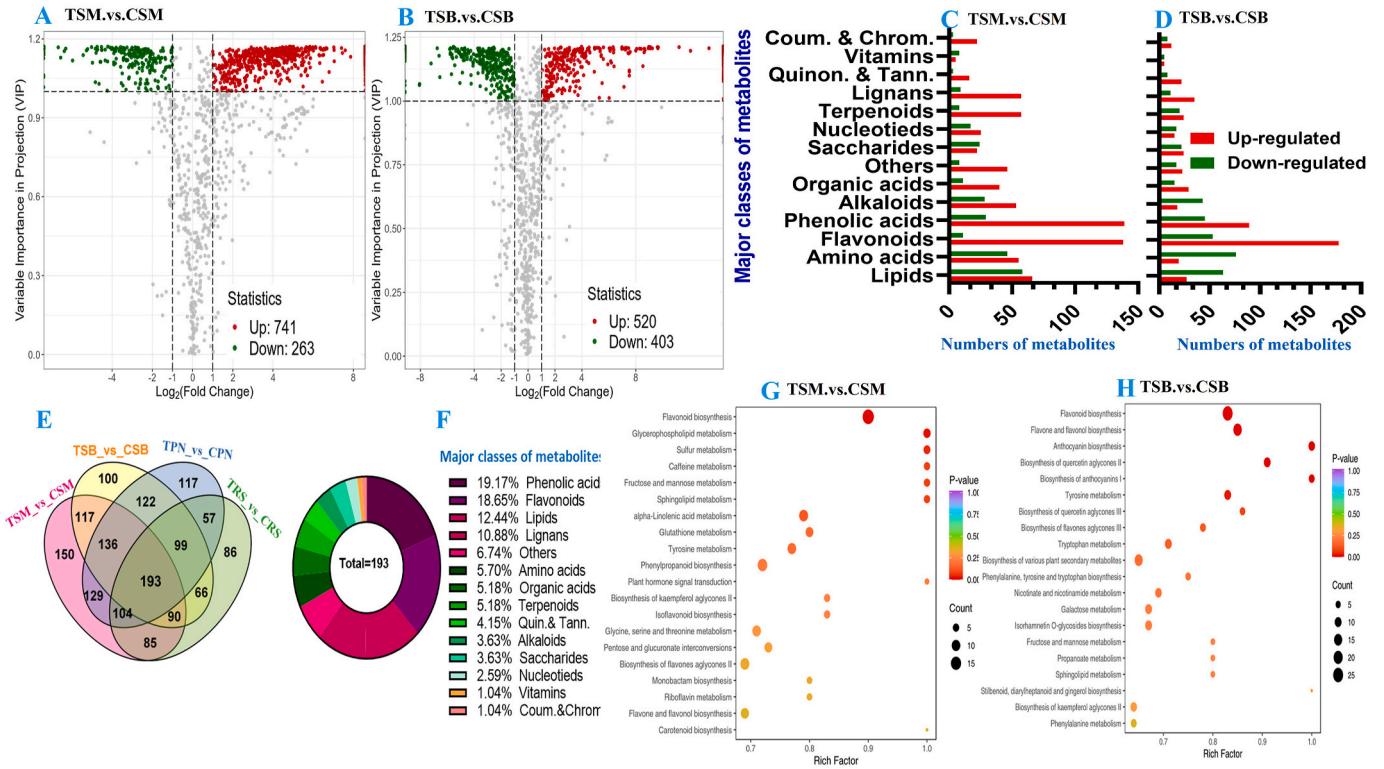

Fig. 5. Differentially accumulated metabolites and KEEG enrichment in the pairwise comparison between testa and peeled black sesame, soybean, peanut, and rapeseed seeds. (A&B) volcano plots showing the number of up and down-regulated metabolites, (C&D) classification of up and down-regulated metabolites in each pairwise comparison, (E) Venn diagram showing the number of key DAMs in each pairwise comparison, (F) Classification of 193 overlapping DAMs and (G&H) the KEGG enrichment result showing significant pathways altered in the pairwise comparison. TSM: sesame testa, CSM: sesame peeled seeds, TSB: soybean testa, CSB: soybean peeled seeds, TPN, peanut testa; CPN, peanut peeled seeds; TRS, rapeseed testa; CRS, rapeseed peeled seeds.

up in TPN), and 780 (522 up in TRS) DAMs, respectively. The classification of DAMs between testa and peeled seeds revealed that flavonoids, lipids, amino acids, phenolic acids, lignans, terpenoids, organic acids, alkaloids, coumarins, and chromones were the dominant components (Fig. 5C and D, Figs. S4C and D). Accordingly, except alkaloids, most secondary metabolites, mainly terpenoids, lignans, phenolic acids, coumarins, quinones, and tannins, were upregulated in testa compared to peeled seeds. However, primary metabolites such as saccharides, amino acids, and vitamins were upregulated in the peeled seeds. The Venn diagram revealed 193 overlapping DAMs in the pairwise comparison between each crop testa and peeled seeds (Fig. 5E). Phenolic acids (19.17%), flavonoids (18.65%), lipids (12.44%), lignans (10.88%), amino acids (5.7%), organic acids (5.18), and terpenoids (5.18%) accounted for the majority of the overlapping metabolite compositions (Fig. 5F).

As illustrated in Fig. 5G, in between TSM and CSM, mainly flavonoid biosynthesis (flavonoid and isoflavonoids), phenylpropanoid, fatty acid (glycerophospholipid, sphingolipid, alpha-linoleic acid), amino acid (glutathione, tyrosine, glycine, serine, and threonine) biosynthesis and sugar (pentose & glucuronate, and fructose) biosynthesis pathways were altered. Flavonoid biosynthesis (flavones, flavanols, anthocyanins, flavonols), amino acid biosynthesis (tyrosine, tryptophan, and phenylalanine), vitamins (nicotinate and nicotinamide), and other secondary metabolism pathways were enriched between TSB. vs.CSB vs. CSB (Fig. 5H). Similarly, flavonoids (flavones, flavanols, anthocyanin, and flavonols), fatty acids (linoleic and alpha-linoleic acid, UFA, sphingolipid), and ascorbate and aldarate biosynthesis pathways were enriched (Fig. S4E). Furthermore, flavonoid (flavone and flavonols), TCA, stilbenoid, and pentose phosphate (pentose and glucuronate) biosynthesis pathways were enriched between TRS. vs. CRS (Fig. S4F). Therefore, four main pathways, including flavonoid, fatty acid, amino acid, and pentose phosphate biosynthesis, were differentially regulated between testa and peeled seeds of the compared crops.

3.7. *Characteristics of differentially accumulated bioactive components in peeled seeds and testa*

Various phytocompounds, such as flavonoids, organic acids, lignans, phenolic acids, alkaloids, terpenoids, coumarins, and vitamins, have beneficial effects against long-term illnesses, such as diabetes, cancer, oxidative stress, sleeplessness, memory loss, angiogenesis, osteoclast genesis, neurodegeneration, and inflammation (Wei et al., 2022; Ye and Liu, 2023). We examined the relative content of 402 DAMs, including 22 AAs, 29 OAs, 56 flavonoids, 14 coumarins, 10 vitamins, 12 chromones, 10 tannins, 11 quinones, 48 alkaloids, 51 lignans, 58 phenolic acids, and 49 terpenoids, to investigate the variation of bioactive compounds in the testa and peeled seeds of oilseeds.

3.7.1. *Fatty acids and lignans*

Fatty acids and lignans are major components of oilseeds. Erucic acid, lauric acid, eicosenoic acid, phytosphingosine, arachidonic acid, and myristoleic acid were higher in TSM (Fig. 6A). Most fatty acids exhibited the highest relative content in the TPN. Only hexadecyl sphingosine exhibited a higher content in CSB. A total of ten fatty acids, five in CRS and five in TRS, had higher contents. Overall, the testa showed a higher content of bioactive fatty acids than peeled seeds (Fig. 6A). As depicted in Fig. 6B, most lignans exhibited higher contents in the CSM and TSM. Except for sesamolinol, matairesinoside, sesamin, asarinin, and hinokinin, in CSM, most lignans were higher in TSM. Two lignans (horsfieldin glucoside and sesamol) in CSB and four lignans (trachelogenin, kadangustin I, lirioresinol A, and nectandrin B) in TPN showed higher content. Accordingly, fraxiresinol, piperitol,

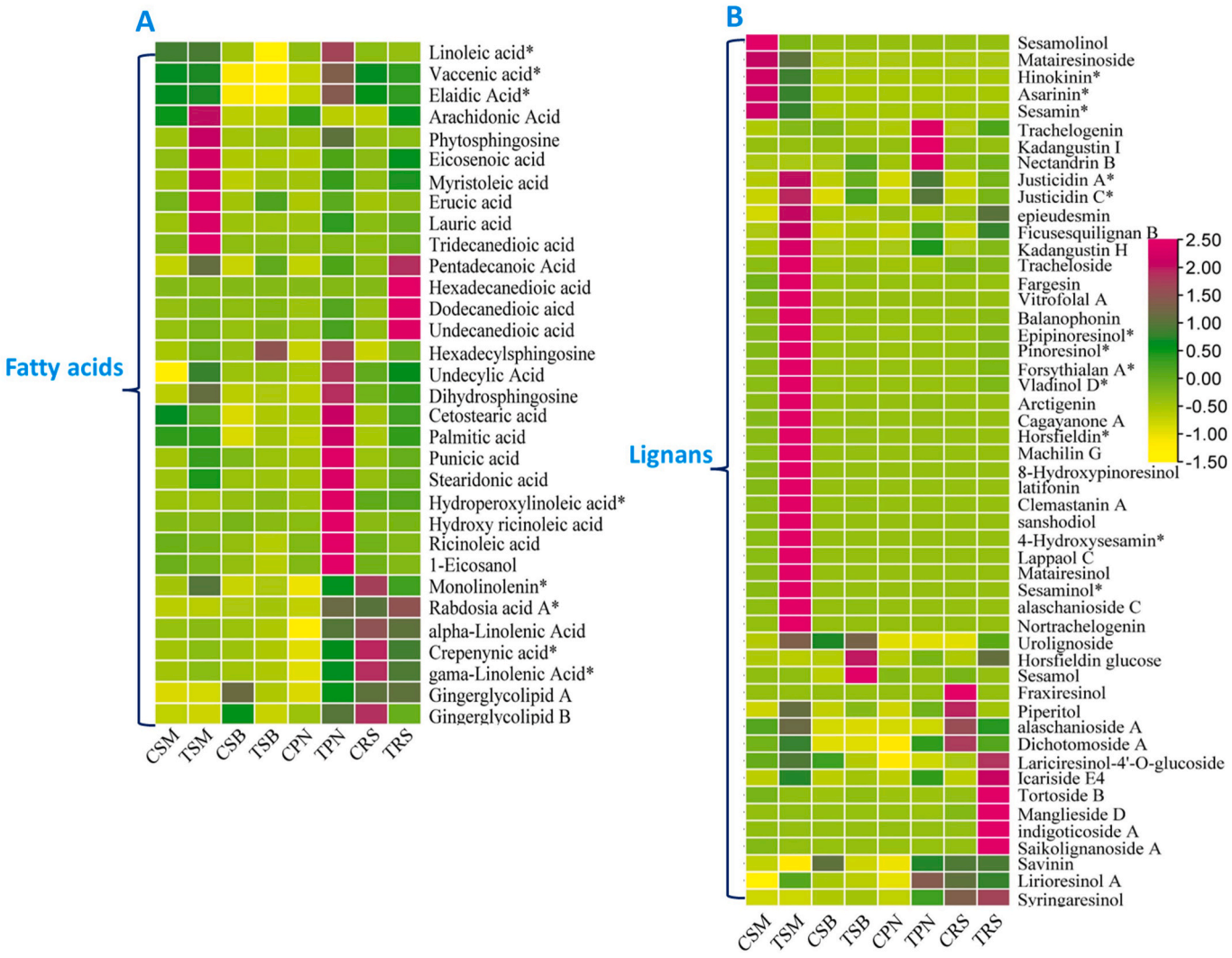

Fig. 6. Major bioactive metabolites' contents include (A) fatty acids and B) lignans.

alaschinioside A, and dichotomoside A in CRS and eight other lignans in TRS showed elevated contents, signifying rapeseed richness in bioactive lignans next to sesame (Fig. 6B). Nearly 53% of differential lignans were higher in the testa of sesame. Furthermore, the distribution pattern shows that the testa of oilseeds is richer in bioactive lignans than in peeled seeds.

3.7.2. *Flavonoids*

A subclass of flavonoids, such as anthocyanins, flavanols, flavanones, flavanonols, flavones, flavonols, isoflavones, and chalcones, was identified in peeled seeds and testa with higher diversity and variation in relative intensity. Interestingly, sesame testa is rich in flavones (rehderianin I, jaceosidin, tricin 7-O-glucoside, salvigenin, altsin, baicalin, mikanin, retusin, and flindulatins), flavanon (abyssinone II), and flavonol (flavoyadorinin A) (Fig. 7A and B). Among all samples, CSB demonstrated higher contents of isoflavones (daidzein, genistein, glycitin, and their derivatives), flavone (apigenin-7-O-glucoside), flavanones (butin 7-O-glucoside, choerospondin, and naringenin), and flavonols (kaempferol 7-O-glucoside) (Fig. 7A and B). Furthermore, most of the flavonoid class metabolites, including flavones, flavanols, flavonols, and isoflavones, exhibited higher contents in TPN than in other samples (Fig. 7A and B). However, the levels of only two flavonoids, epiafzelechin and kaempferol triglucoside, were higher in the TRS and CRS groups, respectively. Except in CSB, most of the flavonoids exhibited elevated contents in the testa of each crop compared to their peeled seeds, signifying that the testa is a hub for flavonoids (Fig. 7A and B).

3.7.3. *Organic acids (AO) and amino acids (AA)*

The distribution characteristics and contents of bioactive DAMs across 29 organic acids and 22 amino acids revealed higher differences between the testa and peeled seeds of tested seeds (Fig. 7C). The contents of two organic acids (phytic acid and allantoin) in CSM and feruloyl lactate, shikimic acid, oxalic acid, and muconic acid in TSM had higher contents (Fig. 7C). Meanwhile, a total of eleven OAs, five in CSB and six OAs were higher in TSB. L-tartaric acid and L-pipecolic acid in CPN, and L-tartaric acid, tianshic acid, and decumbic acid were elevated in TPN. Furthermore, fumaric acid, L-malic acid, iminodiacetic acid, and creatine were higher in CRS, whereas six other OAs were higher in TRS.

Amino acids, including lysine, valine, asparagine, ornithine, and aspartic acid in CSM and glutamine, phenylalanine, methionine, tryptophan, proline, serine, and threonine, were higher in TSM (Fig. 7C). Methionine, tryptophan, histidine, arginine, valine, isoleucine, and leucine were more abundant in the TSB, whereas glutamine and alanine were more abundant in the CSB. Likewise, alanine, arginine, and glutamic acid in CPN, and phenylalanine, cystine, proline, glycine, serine, threonine, valine, isoleucine, leucine, and lysine in TPN showed higher contents. Furthermore, lysine and aspartic acid in CRS and glycine in TRS exhibited higher concentrations. Although there was a higher accumulation of amino acids in the peeled seeds than in the testa of the

Fig. 7. Major bioactive metabolites' contents include flavonoids, organic acids, and amino acids. (A) Anthocyanins, flavanols, and flavones; (B) flavanones, flavonols, and isoflavones; (C) amino acids, and organic acids. The colors indicate the degree of concentration or content; pink shows higher, green-lower, and grey-medium.

targeted samples, the peanut testa (TPN) showed higher amounts of amino acids than its peeled seeds. Overall, except for TPN, the contents of amino acids showed higher contents in the peeled seeds, whereas organic acids in testa (Fig. 7C).

3.7.4. Phenolic acids

We investigated 60 differential bioactive components of phenolic acids between the testa and peeled seeds of sesame, soybean, peanut, and rapeseed (Fig. S5A). Several phenolic acids in TSM and leonoside in CSM had higher content (Fig. S5A). Likewise, ten components (awsoniaside B, hydrageifolin I, and moracin Y in CSB, and kaoburaside, digallic acid, gallic acid, terephthalic acid, neochlorogenic acid, protocatechuic acid, 2-3-hydroxybenzoic acid, and gentisic acid in TSB) exhibited higher contents. CPN had higher levels of 2-hydroxycinnamic acid, chicoric acid, and regaloside G. In contrast, orobanchoside, calccolarioside A, salidroside, arbutin, leonuriside A, cimidahurinine, protocatechuic acid, 2-3-hydroxybenzoic acid, and gentisic acid levels were higher in TPN. Furthermore, 23 phenolic components in rapeseed revealed higher contents in the testa and peeled seeds, indicating its richness in phenolic acids (Fig. S5A). However, caffeic acid and syringin exhibited higher contents in the testa and peeled rapeseed seeds. Although the testa of all crops revealed higher phenolic acid contents, rapeseed had higher contents of several phenolic acids in the testa and peeled seeds.

3.7.5. Terpenoids and alkaloids

As indicated in Fig. 5B, the content of several terpenoids in the TSM and peeled CSM was higher. Except for loganin, asperulosidic acid, mudanpioside A, and kankanoside in CSM, many terpenoids had higher contents in TSM (Fig. S5B). However, lamiophlomiol and syringopicaroside exhibited higher contents in both testa and peeled sesame seeds. Interestingly, soyasaponins (abrisaponin Ca, soyasaponin βb, soyasaponin A-glucuronic glucoside soyasaponin B-glucuronic glucoside, and soyasaponin γg) and ajugoside in CSB and majoroside in TSB were higher, signifying the richness of soybean in saponins. TPN showed higher contents of many terpenoids. Further, micheliolide and vomifoliol (blumenol A) were higher in the CRS and TRS, respectively. Saponins (soyasaponins) are dominantly found in soybean-peeled seeds. Overall, the testa revealed magnificent diversity and content of most bioactive terpenoids compared to the peeled seeds of each crop, except for soybean peeled seeds (Fig. S5B).

Alkaloids, mainly hydroxymenisdaurin D and p-coumaroyl spermidine in CSM and many others in TSM were higher. In soybeans, there were many alkaloids in CSB and two alkaloids (guanidinoacetate and 3-hydroxy pyridine) in TSB at higher concentrations. However, the levels of three alkaloids, phenylethanolamine, histidinol, and stachydine, were higher in TSB and CSB. As shown in Fig. S6A, benzamine, O-phosphocholine, pipecolic acid, 4-hydroxy pipecolic acid, and hypaphorine in CPN, while several alkaloids in TPN had higher contents. Furthermore, seven alkaloids in CRS and two (aurantiamide acetate, and 10-hydroxy methyllycaconitine) in TRS were higher. However, anthriscifolicine A and sinapine 4-O-glucoside were higher in both the TRS and CRS groups (Fig. S6A).

3.7.6. Coumarins, chromones, tannins and vitamins

Coumarins, chromones, tannins, and vitamins are excellent sources with nutritional, pharmacological, and antioxidative properties. To reveal the variation between the testa and peeled seeds of sesame,

soybean, peanut, and rapeseed, we evaluated coumarins, chromones, tannins, and vitamins (Figs. S6B–F). As shown in Fig. S6B, several coumarins in TSM, dicumarol, and nitesoside A in CSM revealed higher contents, signifying sesame richness in coumarins than in other seed samples. Moreover, nodakenin, esculetin, and methylillicinone E levels were higher in the CSB, CPN, and TPN groups, respectively. However, in rapeseed, CRS had higher mandshurin, isofraxidin, skimmin, cichoriin, and daphnin contents. Four chromones in the TSM had elevated contents (Fig. S6C). Furthermore, tababiphenyl C and aloeresin in CRS and tabaisocoumarin D in TRS were higher. The content of most coumarins in TSM and TPN indicates that the testa is richer than the peeled seeds of these crops.

Quinones dominantly accumulated in the TSM, CSB, and TPN. Five quinones, including 2–5, dimethoxybenzoquinone in CSM, and citreosein, hydroxysesamone, auranthio, obtusin-6-O-glucoside, and xanthorin in TSM, showed elevated levels (Fig. S6D). Likewise, 2–5, dimethoxybenzoquinone, aloeemodin-8-O-glucoside, aloeemodin 1-O-glucoside, and rehein 8-O-glucoside had higher contents in CSB. Moreover, soranjidiol, 5,6-dihydroxylucidin, pseudopurpurin, and xanthorin had higher contents in TPN. Tannins are concentrated in the testa of soybeans and peanuts. Accordingly, nine tannins, including procyanidin C1, C2, A6, B2, and B3 in TSB, whereas procyanidin B1 and A2, cinnamtannin B1, and cinnamtannin D1 in TPN had higher contents, indicating that the testa is rich in tannins (Fig. S6E). Vitamin B complex components, including Vit. B2, Vit. B3, Vit. B5, Vit. B6, Vit. B13, choline, and biotin revealed higher distribution and variability in the compared samples (Fig. S6F). Accordingly, nicotinic acid (Vit.B3) and dehydroascorbic acid in CSM and erythorbic acid (isoascorbic acid) in TSM had higher contents. Meanwhile, pantothenic acid (Vit.B5) in CSB, while nicotinamide, pyridoxine (Vit.B6), and orotic acid in the testa and peeled seeds exhibited elevated contents. Pantothenic acid, biotin, and choline were higher in CPN, whereas riboflavin and nicotinamide were higher in TPN. Moreover, biotin and choline in CRS and erythorbic acid in TRS had higher contents. Therefore, peeled seeds had higher levels of many vitamins, implying that peeled seeds are likely to have more vitamins than the testa. However, coumarins, chromones, quinones, and tannins were higher in the testa of each sample, indicating potential differences in the chemical composition and nutrition between the two parts of the seed.

3.8. Correlation between bioactive compounds and antioxidant activities

To gain insight into the relationship between bioactive compounds and the potential health benefits of consuming the testa or peeled seeds of oilseeds, we performed a correlation analysis between bioactive components, total phenol, flavonoid content, and antioxidant activities (Figs. S7A and B). Accordingly, DPPH and FRAP were notably associated with the bioactive component, total phenol, and flavonoid contents. As indicated in Fig. S7A, the overall evaluation results revealed that DPPH had a positive and strong association with TFC, TPC, flavonoids, tannins, quinones, and organic acids, suggesting that their higher content contributed to the potential antioxidant role in a sample. Meanwhile, FRAP exhibited a strong, positive, and significant ($P < 0.01$) association with TFC, TPC, flavonoids, amino acids, quinone, tannin, vitamins, and organic acids, indicating their contribution to the antioxidant capacity. This result is consistent with the contents of TFC, TPC, flavonoids, organic acids, tannins, quinones, and other phenolic compounds in the testa of the tested seeds, which had higher bioactive and antioxidant potential. Unexpectedly, coumarins and lignans with FRAP and DPPH and terpenoids with FRAP had negative and significant ($P < 0.01$) associations, indicating a lower impact on the overall antioxidative potential. This may be due to the crop-specific accumulation effects of the antioxidant components. Because terpenoids and lignans were higher in sesame, we extended the correlation analysis to examine the association of lignans, terpenoids, and other phenolic compounds with DPPH and FRAP in sesame (Fig. S7B). Interestingly, the results revealed that lignans, terpenoids, phenolic acids, flavonoids, amino acids, organic acids, flavonoids, coumarins, alkaloids, tannins, TPC, and TFC were strongly and significantly ($P < 0.01$) positively associated with DPPH and FRAP, signifying their contribution to antioxidative potentials in sesame.

4. Discussion

Black seeds of various crops have been shown to have significant antioxidant, nutritive, pharmacological, and cosmetic potential (Liu et al., 2024; Jati, 2020; Xu and Chang, 2008). This is because black seeds contain elevated levels of total phenols, total flavonoids, and various bioactive compounds (Lim et al., 2021; Dossou et al., 2022). Peeled seeds and testa are the two most crucial parts of the seed, with different physiological and structural functions, and are composed of various nutritional and potent phytochemicals beneficial for human health. However, regardless of their relevance to plants, animals, and humans, most food-processing industries stripped off testa from seeds. This process might reduce the nutritional and other desirable characteristics obtained from the seed testa. Therefore, to provide scientific evidence on how dehulling could impact the nutritional composition, antioxidant activity, and other desirable features, we performed total phenol and flavonoid tests, antioxidant activity, and comparative metabolome profiling of testa and peeled seeds of black sesame, soybean, peanut, and rapeseed using widely targeted ultra-high performance liquid chromatography coupled with tandem mass spectrometry (UHPLC-MS/MS). This study revealed differences in total phenol content, total flavonoid content, metabolic profiles, and antioxidant activity between peeled seeds and testa of oilseeds. In addition, this study emphasized the contents and distribution characteristics of differentially accumulated bioactive metabolites between testa and peeled seeds of the four oilseed crops.

4.1. TPC, TFC, and antioxidant activity in the testa and peeled parts of seeds

The peeled seeds and testa of different samples revealed notable differences in the total phenol and flavonoid contents. The mean of total phenol content varied from 24.9 to 36.31 mg GAE/g in the testa and 5.9–7.2 mg GAE/g in the peeled seeds. Likewise, total flavonoid contents ranged from 17.2 to 32.4 mg CAE/g and 11.9–22.6 mg CAE/g in the peeled seeds and testa, respectively. The higher total phenol and flavonoid content in the testa of each crop indicates that this seed portion harbours many phenolic compounds and may possess stronger bioactive properties and antioxidant activity. Lim et al. (2021) reported higher total phenol and flavonoid contents in the testa of soybeans. Likewise, Adedayo et al. (2021) demonstrated that seeds without a seed coat (dehulled seeds) had lower total phenol content and reduced antioxidant activity. Further, higher contents of TPC, TFC, condensed tannin content (CTC), monomeric anthocyanin content (MAC), and antioxidant activity (DPPH and FRAP) values in the seed coat than cotyledon (seeds without seed coat or peeled seeds) of peanut, explaining the significance of testa in phenolic content and antioxidant activity (Attree et al., 2015). Zhang et al. (2011) demonstrated that the total phenol and anthocyanin contents are strongly associated with antioxidative properties. Previous studies have indicated that phenolic components, such as flavonoids and other phenols, are antioxidants and have notable health benefits such as anti-inflammatory, antimicrobial, anticancer, and cardioprotective properties (El Hanafi et al., 2023; Ye and Liu, 2023). Our study found a strong correlation between the antioxidant properties of seeds and their bioactive compounds, phenol, and flavonoid contents, emphasizing the importance of the testa in determining antioxidant qualities. A strong positive correlation between antioxidant activity and bioactive components has been reported in different beans (soybeans, mung beans, peas, and common beans) (Zhang et al., 2024). The results highlight the importance of considering

the testa when consuming and assessing the seed's potential nutritional and health-related properties. Variations in TPC and TFC between testa and peeled seeds indicate variations in their phytochemical profiles, antioxidants, and potential health-promoting qualities.

4.2. Metabolic profiles and the dynamics of metabolic components in testa and peeled black oil seeds

Developing new and high-quality varieties of oil seeds is crucial for enhancing nutrition and health, driving market demand, boosting economic output, and propelling the oil crop sector expansion. Widely targeted metabolomics analysis is widely used to investigate metabolites' composition and distribution characteristics in a biological sample. In this study, we identified 1847 metabolites in the testa and peeled parts of the samples. By comparing each oilseed sample's testa and peeled seeds, we identified unique (274, 375, 362, and 298) and overlapping (1308, 1227, 1283, and 1305) metabolites, respectively. Among the unique metabolites identified between the testa and peeled seeds, more than 73.3% of identified metabolites belonged to the testa, showing their richness with more compounds than peeled seeds. Likewise, we observed higher levels of saccharides, vitamins, and AAs in peeled seeds, explaining the main function of peeled seeds in storing nutrients and energy that support the development of seedlings during germination. Peeled seeds may contain higher amounts of vitamins, proteins, and carbohydrates/saccharides, but they often do not need the same quantity of defense-related or protective substances as the testa (Xu and Chang, 2008). Supportive findings indicates that the cotyledon (peeled seed) is rich in total soluble sugars, starch, and lipids (Pal et al., 2017). However, flavonoids, phenolic acids, lipids, lignans, quinones, and tannins exhibited higher content in the testa of each sample, which might be linked to the total phenol content, antioxidant activity, protection, and pharmacological significance. For example, in the previous studies, significantly reduced contents of phytic acid, tannins, gallic acid, catechins, and quercetin were observed in dehulled seeds (peeled seeds) than in seeds with seed coat (testa), signifying the richness of testa with phenolic components (Pal et al., 2017). Certain compounds tend to accumulate more specifically in the testa or peeled seeds, which might be due to the ecological, nutritional, protection, and other physiological functions of the metabolites in different parts of the seed. The testa protects the delicate tissues of peeled seeds and embryos, whereas peeled seeds store nutrients and energy to supplement the developing embryo. This necessitates tissue-specific accumulation of metabolites in seeds. In our result, we found crop and plant-part specific accumulations of certain metabolites. Supportively, tissue and seed color-specific accumulation of metabolites has been reported in sesame (Dossou et al., 2022a,b; Dossou et al., 2021). In line with this result, we have reported crop-specific metabolites in sesame, soybean, and perilla, showing the variation in metabolic profiles due to differences in the regulation of metabolic processes during seed development (Kefale et al., 2023).

Food processing (transformation) is a common activity in modern food industries that reduces the quality, organic nature, and antioxidative potential of food and food products (Liu et al., 2024). Among these, removing the testa (dehulling) is an example of oilseeds to produce various food items, such as creamy paste "tahini" in sesame, "tofu, doenjang, and dajiang" in soybean, and "traditional peanut butter" in peanuts (Lee et al., 2024; El Hanafi et al., 2023; Abib et al., 2023). Dehulling reduces the content of phenolic compounds (gallic acid, catechin dihydrate, and quercetin), antinutritive components (phytic acid and tannins), and antioxidant activities in lentils (Pal et al., 2017). Likewise, the dehulling process results in a loss of lignans, which changes sesame quality traits and antioxidant activity (Abib et al., 2023). Consumption of whole seeds, which have a higher fiber content than refined (dehulled) seeds, can reduce starch accessibility to alpha-amylase, stimulate satiety signals, decrease glucose diffusion, influence the synthesis of hormones involved in energy homeostasis and body weight regulation, control glucose, and reduce the risk of type 2 diabetes, mainly because of the high fiber and phenolic content of the testa (Blahova et al., 2021).

The PCA, HCA, and OPLS-DA results of the multivariate analysis separated the samples into eight groups, suggesting distinct accumulation patterns. Additional analysis revealed that over 580 DAMs were observed in the pairwise comparison between the testa and the peeled seeds of each crop. The KEGG enrichment analysis results and classification revealed that three key pathways, including flavonoid, amino acid, and pentose phosphate biosynthesis, were differentially regulated between the testa and peeled seeds of the four crops. The variation in metabolic profiles between the testa and peeled seeds of the compared crops may be attributed to physiological function, genetic control, developmental stage, and tissue specificity of the seed portions. The diversity and content of differential bioactive metabolites were notably higher in testa and peeled seeds. This might determine the biochemical composition and nutritional, pharmacological, and antioxidant activities of oilseeds and their products.

Moreover, regardless of their TPC, TFC, antioxidant activity, and metabolic profile variation, the seed parts (peeled seeds and testa) comprised diverse bioactive components. Therefore, it is better to complement metabolomics with genomic research to identify master genes that regulate flavonoid, amino acid, and other phenolic component biosynthesis, resulting in improved traits of oilseed and oilseed products, such as enhanced nutritional, pharmacological, and antioxidative potentials. Our results imply that concentrating on the seed coat/testa may offer information on the nutritional, antioxidant, and other bioactive characteristics, which could culminate in the creation of functional meals or dietary supplements with improved health advantages. Furthermore, advancements in metabolomic technology and awareness of food crops and their products have escalated the growing interest in high-quality breeding for crop nutrient constituents (Shi et al., 2024). This necessitates providing science-based and clear access to the nutritional composition and antioxidant potential of food crops and their parts. Therefore, our investigation will increase the science-driven information on the phytochemical composition, phenol content, and antioxidant activity of oilseed crop testas and peeled seeds for specific or comprehensive use.

5. Conclusion

Our study revealed metabolic, antioxidant, total phenol, and total flavonoid content variations among oilseed parts (testa and peeled seeds). A total of 1847 metabolites were identified from the testa and peeled parts of black sesame, soybean, peanut, and rapeseed using a widely targeted metabolomics analysis approach. This study revealed seed part-specific accumulation of metabolites. The samples from each crop testa revealed higher TPC, TPC, antioxidant activity, and better secondary metabolite profiles than peeled seeds. Similarly, the peeled seeds/cotyledons revealed a good index of primary metabolites. The distribution and relative content characteristics of differentially accumulated bioactive metabolites revealed differences in the composition of biochemical and potential nutritional and health benefits between testa and peeled seeds. The bioactivity of seed parts (peeled seeds and testa) may be associated with the phytochemical contents and compositions such as flavonoids, phenolic acids, terpenoids, alkaloids, lipids, terpenoids, lignans, amino acids, saccharides, chromones, and coumarins. Further, biological samples' nutritional quality, pharmaceutical, cosmetic, and antioxidant potentials can be influenced by the crop type, plant part, phytochemical composition, relative content, and diversity of metabolic components. Therefore, according to our results, it can be inferred that peeled seeds (cotyledons) may offer higher nutritional value, whereas the testa may possess medicinal and protective properties. The increased levels of flavonoids, total phenols, and antioxidant activity in oilseed testa have significant applications in dietary habits, agricultural strategies, health promotion, and the development of

functional foods. By highlighting these advantages, we may promote healthy eating practices and raise public awareness of the value of whole oilseeds in a balanced diet. Further, this study is limited to global metabolic profiling and evaluation for total phenol, flavonoid, and antioxidant activities. Therefore, further investigation is needed on the identified metabolites and the mechanisms of actions of bioactive components.

CRediT authorship contribution statement

Habtamu Kefale: Writing – original draft, Investigation, sample preparation, review, Methodology, Writing – review & editing, Formal analysis. **Rong Zhou:** prepared materials and original data. **Zishu Luo:** involved in experiment design. **Senouwa Segla Koffi Dossou:** review, Writing – review & editing. **Muez Berhe:** review, Writing – review & editing. **Lei Wang:** prepared materials and original data. **Ahmed A. Abbas:** review, Writing – review & editing. **Yanxin Zhang:** prepared materials and original data. **Ting Zhou:** involved in experiment design. **Jun You:** design the experiment, Resources, Supervision, funding, review, Validation. **Linhai Wang:** design the experiment, Resources, Supervision, funding, review, Validation.

Ethics approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

Declaration of competing interest

All authors declare that they have no personal, financial, or other conflicts of interest.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.crfs.2024.100939.

Data availability

Data will be made available on request.

References

Abib, B., Afifi, S.M., El-Din, M.G.S., Farag, M.A., 2023. How do cultivar origin and stepwise industrial processing impact Sesamum indicum seeds' metabolome and its paste and in relation to their antioxidant effects? A case study from the sesame industry. Food Chem. 420, 136134. https://doi.org/10.1016/j.foodchem.2023.136134.

Adedayo, B.C., Anyasi, T.A., Taylor, M.J.C., Rautenbauch, F., Le Roes-Hill, M., Jideani, V. A., 2021. Phytochemical composition and antioxidant properties of methanolic extracts of whole and dehulled Bambara groundnut (*Vigna subterranea*) seeds. Sci. Rep. 11 (1), 1–11. https://doi.org/10.1038/s41598-021-93525-w.

Ahmed, H.M., Tavaszi-sarosi, S., 2019. Identification and quantification of essential oil content and composition, total polyphenols and antioxidant capacity of *Perilla frutescens* (L.) Britt. Food Chem. 275, 730–738. https://doi.org/10.1016/j.foodchem.2018.09.155.

Attree, R., Du, B., Xu, B., 2015. Distribution of phenolic compounds in seed coat and cotyledon, and their contribution to antioxidant capacities of red and black seed coat peanuts (*Arachis hypogaea* L.). Ind. Crop. Prod. 67, 448–456. https://doi.org/10.1016/j.indcrop.2015.01.080.

Blahova, J., Martiniakova, M., Babikova, M., Kovacova, V., Mondockova, V., Omelka, R., 2021. Pharmaceutical drugs and natural therapeutic products for the treatment of type 2 diabetes mellitus. Pharmaceuticals 14, 1–32.

Brigante, F.I., García, M.E., Radcenco, A.L., Moyna, G., Wunderlin, D.A., Baroni, M.V., 2022. Identification of chia, flax and sesame seeds authenticity markers by NMR-based untargeted metabolomics and their validation in bakery products containing them. Food Chem. 387, 1–10. https://doi.org/10.1016/j.foodchem.2022.132925.

Chávez-Mendoza, C., Hernández-Fig ueroa, K.I., Sánchez, E., 2019. Antioxidant capacity and phytonutrient content in the seed coat and cotyledon of common beans (Phaseolus vulgaris L.) from various regions in Mexico. Antioxidants 8 (1). https://doi.org/10.3390/antiox8010005.

Chen, C., Chen, H., Zhang, Y., Thomas, H.R., Frank, M.H., He, Y., Xia, R., 2020. TBtools: an integrative toolkit developed for interactive analyses of big biological data. Mol. Plant 13 (8), 1194–1202. https://doi.org/10.1016/j.molp.2020.06.009.

Chen, W., Gong, L., Guo, Z., Wang, W., Zhang, H., Liu, X., Yu, S., Xiong, L., Luoa, J., 2013. A novel integrated method for large-scale detection, identification, and quantification of widely targeted metabolites: application in the study of rice metabolomics. Mol. Plant 6 (6), 1769–1780. https://doi.org/10.1093/mp/sst080.

Choi, G.Y., Han, Y.S., Sim, K.H., Kim, M.H., 2023. Phenolic compounds, antioxidant capacity, and α-amylase and α-glucosidase inhibitory activity of ethanol extracts of perilla seed meal. Food Sci. Nutr. 11 (8), 4596–4606. https://doi.org/10.1002/fsn3.3419.

Choi, Y.M., Yoon, H., Lee, S., Ko, H.C., Shin, M.J., Lee, M.C., Hur, O.S., Ro, N.Y., Desta, K. T., 2020. Isoflavones, anthocyanins, phenolic content, and antioxidant activities of black soybeans (*Glycine max* (L .) Merrill) as affected by seed weight. Sci. Rep. 10, 1–13. https://doi.org/10.1038/s41598-020-76985-4.

Dossou, S.S.K., Deng, Q., Li, F., Jiang, N., Zhou, R., Wang, L., Li, D., Tan, M., You, J., Wang, L., 2023. Comparative metabolomics analysis of different perilla varieties provides insights into variation in seed metabolite profiles and antioxidant activities. Foods 12 (23), 12234370. https://doi.org/10.3390/foods12234370.

Dossou, S.S.K., Luo, Z., Deng, Q., Zhou, R., Zhang, Y., Li, D., Li, H., Tozo, K., You, J., Wang, L., 2024. Biochemical and molecular insights into variation in sesame seed antioxidant capability as revealed by metabolomics and transcriptomics analysis. Antioxidants 13 (5), 514. https://doi.org/10.3390/antiox13050514.

Dossou, S.S.K., Luo, Z., Wang, Z., Zhou, W., Zhou, R., Zhang, Y., Li, D., Liu, A., Dossa, K., You, J., Wang, L., 2022a. The dark pigment in the sesame (*Sesamum indicum* L.) seed coat: isolation, characterization, and its potential precursors. Front. Nutr. 9 (February), 858673. https://doi.org/10.3389/fnut.2022.858673.

Dossou, S.S.K., Xu, F., Cui, X., Sheng, C., Zhou, R., You, J., Tozo, K., Wang, L., 2021. Comparative metabolomics analysis of different sesame (*Sesamum indicum* L.) tissues reveals a tissue - specific accumulation of metabolites. BMC Plant Biol. 21, 1–14. https://doi.org/10.1186/s12870-021-03132-0.

Dossou, S.S.K., Xu, F., You, J., Zhou, R., Li, D., Wang, L., 2022b. Widely targeted metabolome profiling of different colored sesame (*Sesamum indicum* L.) seeds provides new insight into their antioxidant activities. Food Res. Int. 151, 110850. https://doi.org/10.1016/j.foodres.2021.110850.

El Hanafi, L., Mssillou, I., Nekhla, H., Bessi, A., Bakour, M., Laaroussi, H., Ben Khadda, Z., Slimani, C., Giesy, J.P., Greche, H., Ali, G.A.M., Aboul-Soud, M.A.M., 2023. Effects of dehulling and roasting on the phytochemical composition and biological activities of *Sesamum indicum* L. Seeds. J. Chem., 5394315 https://doi.org/10.1155/2023/5394315, 2023.

Fraga, C.G., Clowers, B.H., Moore, R.J., Zink, E.M., 2010. Signature-discovery approach for sample matching of a nerve-agent precursor using liquid chromatography-mass spectrometry, XCMS, and chemometrics. Anal. Chem. 82 (10), 4165–4173. https://doi.org/10.1021/ac1003568.

Hadidi, M., Tan, C., Assadpour, E., Jafari, S.M., 2024. Oilseed meal proteins: from novel extraction methods to nanocarriers of bioactive compounds. Food Chem. 438, 137971. https://doi.org/10.1016/j.foodchem.2023.137971.

Hadidi, M., Tan, C., Assadpour, E., Kharazmi, M.S., Jafari, S.M., 2023. Emerging plant proteins as nanocarriers of bioactive compounds. J. Contr. Release 355 (February), 327–342. https://doi.org/10.1016/j.jconrel.2023.01.069.

Jati, I.R.A.P., 2020. Black soybean seed: black soybean seed antioxidant capacity. In: Nuts and Seeds in Health and Disease Prevention. Elsevier Inc. https://doi.org/10.1016/B978-0-12-818553-7.00012-7.

Jiang, L., Yang, X., Gao, X., Yang, H., Huang, S., Zhu, J., Zhou, H., Li, X., Gu, X., Zhou, H., Liang, Z., Yang, A., Huang, Y., Xiao, M., 2024. Multiomics analyses reveal the dual role of flavonoids in pigmentation and abiotic stress tolerance of soybean seeds. J. Agric. Food Chem. xxxx (xxx), 1–13. https://doi.org/10.1021/acs.jafc.3c08202.

Kefale, H., Dossou, S.S.K., Li, F., Jiang, N., Zhou, R., Wang, L., Zhang, Y., Li, D., You, J., Wang, L., 2023. Widely targeted metabolic profiling provides insights into variations in bioactive compounds and antioxidant activity of sesame, soybean, peanut, and perilla. Food Res. Int. 174 (P1), 113586. https://doi.org/10.1016/j.foodres.2023.113586.

Kim, J.H., Seo, W.D., Lee, S.K., Lee, Y.B., Park, C.H., Ryu, H.W., Lee, J.H., 2014. Comparative assessment of compositional components, antioxidant effects, and lignan extractions from Korean white and black sesame (*Sesamum indicum* L.) seeds for different crop years. J. Funct.Foods 7, 495–505. https://doi.org/10.1016/j.jff.2014.01.006.

Kuang, Q., Yu, Y., Attree, R., Xu, B., 2017. A comparative study on anthocyanin, saponin, and oil profiles of black and red seed coat peanut (Arachis hypogacea) grown in China. Int. J. Food Prop. 20 (1), S131–S140. https://doi.org/10.1080/10942912.2017.1291676.

Lee, H.J., Jo, E., Song, J.H., Min, J., Song, Y., Lee, H., Choe, Y., Cha, J., Lee, H., 2024. Correlation between monosaccharide, oligosaccharide, and microbial community profile changes in traditional soybean brick (meju) fermentation. Food Res. Int. 184 (January). https://doi.org/10.1016/j.foodres.2024.114233.

Lim, Y.J., Kwon, S.J., Qu, S., Kim, D.G., Eom, S.H., 2021. Antioxidant contributors in seed, seed coat, and cotyledon of γ-ray-induced soybean mutant lines with different seed coat colors. Antioxidants 10 (3), 353. https://doi.org/10.3390/antiox10030353.

Liu, Y., Xiao, X., Wang, Z., Shan, X., Liu, G., Wei, B., 2024. Metabolomic analysis of black sesame seeds: effects of processing and active compounds in antioxidant and anti-inflammatory properties. Food Res. Int. 176, 113789. https://doi.org/10.1016/j.foodres.2023.113789.

Lolli, V., Caligiani, A., 2024. How nuclear magnetic resonance contributes to food authentication: current trends and perspectives. Curr. Opin. Food Sci. 58, 101200. https://doi.org/10.1016/j.cofs.2024.101200.

Manickam, S., Rajagopalan, V.R., Kambale, R., Rajasekaran, R., Kanagarajan, S., Muthurajan, R., 2023. Plant metabolomics: current initiatives and future prospects. Curr. Issues Mol. Biol. 45 (11), 8894–8906. https://doi.org/10.3390/cimb45110558.

Pal, R.S., Bhartiya, A., Yadav, P., Kant, L., Mishra, K.K., Aditya, J.P., Pattanayak, A., 2017. Effect of dehulling, germination and cooking on nutrients, anti-nutrients, fatty acid composition and antioxidant properties in lentil (Lens culinaris). J. Food Sci. Technol. 54 (4), 909–920. https://doi.org/10.1007/s13197-016-2351-4.

Sanni, G.B.T.A., Ezin, V., Chabi, I.B., Missihoun, A.A., Florent, Q., Hamissou, Z., Niang, M., Ahanchede, A., 2024. Production and achievements of Sesamum indicum industry in the world: past and current state. Oil Crop Science 9 (3), 187–197. https://doi.org/10.1016/j.ocsci.2024.06.006.

Shahidi, F., Liyana-pathirana, C.M., Wall, D.S., 2006. Food Chemistry Antioxidant activity of white and black sesame seeds and their hull fractions. Food Chem. 99 (2006), 478–483. https://doi.org/10.1016/j.foodchem.2005.08.009.

Shen, S., Tang, Y., Zhang, C., Yin, N., Mao, Y., Sun, F., Chen, S., Hu, R., Liu, X., Shang, G., Liu, L., Lu, K., Li, J., Qu, C., 2021. Metabolite profiling and transcriptome analysis provide insight into seed coat color in brassica juncea. Int. J. Mol. Sci. 22 (13), 22137215. https://doi.org/10.3390/ijms22137215.

Shi, H., Zhu, Y., Wu, X., Jiang, T., Li, X., Liu, J., Di, Y., Chen, F., Gao, J., Xu, X., Xiao, N., Feng, X., Zhang, P., Wu, Y., La, Q., Li, A., Chen, P., Li, X., 2024. CropMetabolome: a comprehensive metabolome database for major crops cross eight categories. Plant J. 119, 1613–1626. https://doi.org/10.1111/tpj.16858.

Tao, H., Zhao, Y., Li, L., He, Y., Zhang, X., Zhu, Y., Hong, G., 2023. Comparative metabolomics of flavonoids in twenty vegetables reveal their nutritional diversity and potential health benefits. Food Res. Int. 164 (December 2022), 112384. https://doi.org/10.1016/j.foodres.2022.112384.

Tokel, D., Erkencioglu, B.N., 2021. Production and trade of oil crops, and their contribution to the world economy. Oil Crop Genomics 415–427. https://doi.org/10.1007/978-3-030-70420-9_20.

Wang, D., Zhang, L., Huang, X., Wang, X., Yang, R., Mao, J., Wang, X., Wang, X., Zhang, Q., Li, P., 2018. Identification of nutritional components in black sesame determined by widely targeted metabolomics and traditional Chinese medicines. Molecules 23 (5), 1–12. https://doi.org/10.3390/molecules23051180.

Wei, P., Zhao, F., Wang, Z., Wang, Q., Chai, X., Hou, G., Meng, Q., 2022. Sesame (Sesamum indicum L.): a comprehensive review of nutritional value phytochemical composition, health benefits, development of food, and industrial applications. Nutrients 14, 1–26.

Xu, B., Chang, S.K.C., 2008. Antioxidant capacity of seed coat, dehulled bean, and whole black soybeans in relation to their distributions of total phenolics, phenolic acids, anthocyanins, and isoflavones. J. Agric. Food Chem. 56, 8365–8373.

Ye, Z., Liu, Y., 2023. Polyphenolic compounds from rapeseeds (Brassica napus L.): the major types, biofunctional roles, bioavailability, and the influences of rapeseed oil processing technologies on the content. Food Res. Int. 163 (December 2022), 112282. https://doi.org/10.1016/j.foodres.2022.112282.

Zhang, D., Shen, D., Cao, Y., Duan, X., Sun, H., 2023. Widely targeted metabolomic approach reveals dynamic changes in non-volatile and volatile metabolites of peanuts during roasting. Food Chem. 412 (January), 135577.

Zhang, K., Ma, J., Gangurde, S.S., Hou, L., Xia, H., Li, N., Pan, J., Tian, R., Huang, H., Wang, X., Zhang, Y., Zhao, C., 2022a. Targeted metabolome analysis reveals accumulation of metabolites in testa of four peanut germplasms. Front. Plant Sci. 13 (September), 992124. https://doi.org/10.3389/fpls.2022.992124.

Zhang, R.F., Zhang, F.X., Zhang, M.W., Wei, Z.C., Yang, C.Y., Zhang, Y., Tang, X.J., Deng, Y.Y., Chi, J.W., 2011. Phenolic composition and antioxidant activity in seed coats of 60 Chinese black soybean (Glycine max L. Merr.) varieties. J. Agric. Food Chem. 59 (11), 5935–5944. https://doi.org/10.1021/jf201593n.

Zhang, W., Zhao, Y., Yang, H., Liu, Y., Zhang, Y., Zhang, Z., Li, Y., Wang, X., Xu, Z., Deng, J., 2024. Comparison analysis of bioactive metabolites in soybean, pea, mung bean, and common beans: reveal the potential variations of their antioxidant property. Food Chem. 457, 140137. https://doi.org/10.1016/j.foodchem.2024.140137.

Zhang, Y., Xiao, H., Lv, X., Wang, D., Chen, H., Wei, F., 2022b. Comprehensive review of composition distribution and advances in profiling of phenolic compounds in oilseeds. Front. Nutr. 9 (October), 1–16. https://doi.org/10.3389/fnut.2022.1044871.