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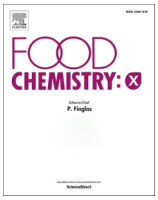

Metabolomics of black beans (*Phaseolus vulgaris* L.) during atmospheric pressure steaming and in vitro simulated digestion

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ABSTRACT

In the paper, metabolomics techniques based on UHPLC-QE-MS were used to study raw black beans, steaming black beans, and their in vitro digestion products. The results show that the three groups of raw black beans, atmospheric pressure-steamed black beans, and their in vitro digests comprised 922, 945, and 878 characteristic metabolites, respectively, dominated by amino acids, organic acids, polyphenols, and sugars. After screening the differential metabolites, content comparison, the content of amino acids, sugars, and phenolics in black beans was found to be increased after atmospheric steaming. During in vitro digestion, the amino acid content increased and the phenolic content decreased, with amino acid synthesis, phenolic degradation, and conversion predominating. This study provides data to support the changes in black beans metabolites during atmospheric steam processing and in vitro digestion.

1. Introduction

Black beans are known as the “king of beans” and are one of the main cash crops in Heilongjiang, Shanxi, and Hebei, China. Black beans have a high nutritional value and are rich in protein (36 %–40 %), fat (15 %), crude fibre (4 %), phenolic compounds (total phenolic content of 18.03 mg/g), and other nutritional and bioactive components (Cho et al., 2013; Yu et al., 2021), which play an important role in the daily dietary system of the population. Black beans are generally cooked before ingestion by humans by boiling, steaming, or microwave heating to enhance the palatability and flavour of black beans, and to help improve the efficiency of nutrient absorption by the human intestinal tract. Previous studies have shown that steaming is more effective in retaining active substances (e.g., phenolics and vitamin C) (Lafarga et al., 2018) and improving protein solubility than other cooking methods (Habiba, 2002). However, at the same time, steaming can also cause the loss or transformation of various components of black beans during processing, thus adversely affecting their nutrition, palatability, and functional activity. The pattern of metabolite changes at this stage remains unclear. Xu et al. (Habiba, 2002; Xu and Chang, 2008) found that the total phenol, total flavonoid, condensed tannin, and monomeric anthocyanin contents of black beans decreased after steaming and processing, and they found that the black bean isoflavone glycoside element content increased significantly and was accompanied by a decrease in antioxidant capacity. Audu et al. (Audu et al., 2013) found significant differences ($P < 0.05$) in the fatty acid composition of black beans after different processing treatments such as boiling, roasting, sprouting, and fermentation. There have been more studies on black bean metabolites, but most of them have focused on specific metabolites (e.g., phenolic compounds and anthocyanins), and no in-depth studies have been conducted on the whole metabolome. Second, to assess the complete nutritional value and physiological efficacy of food consumed by humans, the bioaccessibility of its components in the gastrointestinal tract must be fully considered. However, considering only the amount of each component in the food would greatly overestimate the true intake (Shi et al., 2022). Because of the complexity of the human digestive process, it is difficult to effectively monitor the processes of substance release and absorption at this stage, especially for black beans, which are rich in various nutrients. However, a paucity of research has been reported in this area, which poses difficulties in understanding the processes of degradation, transformation, and absorption of the entire spectrum of substances in black beans at this stage and prevents a

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comprehensive assessment of the bioaccessibility.

Metabolomics is a new discipline that mainly studies the changes in the types and quantities of small-molecule metabolites in biological samples in biological systems, with high throughput, high sensitivity, and a wide detection range, and it has been widely used in the study of changes in metabolites during food processing (Utpott et al., 2022). For example, Caprioli et al. (Caprioli et al., 2018) found up to 458 $mg{\cdot}kg^{-1}$ of polyphenols in black beans after the analysis of 31 bean varieties using metabolomics techniques. Yu et al. (Yu et al., 2021) used UHPLC-QE-MS metabolomics to identify 92 polyphenolic metabolites in raw/cooked black beans, and they screened 47 differential metabolites, mainly flavonoids, isoflavone derivatives, and phenolic acids. The changes in the composition, content, and types of substances in the black bean fraction after processing, digestion, and absorption are not yet clear. In light of this, the present study used a UHPLC-QE-MS non-targeted metabolomics technique to analyse the metabolomic profile of atmospheric pressure-steamed black beans, as well as the in vitro simulated digestion products of atmospheric pressure steamed black beans, in order to provide theoretical support for future enhancement of the black bean maturation process and nutritional changes after digestion and absorption of mixed grain legumes.

2. Materials and methods

2.1. Materials and reagents

Black beans (full grain, uniform colour, and no pest damage) were obtained from Daqing City, Heilongjiang Province, China. Methanol and acetonitrile were purchased from CNW Technologies, and acetic acid (Fisher Chemical), ammonium acetate (Sigma-Aldrich), and the above reagents were all LC-MS grade. α-amylase (enzyme activity of 1500 U/mL), pepsin (enzyme activity of 1500 U/mL), trypsin (enzyme activity of 800 U/mL), oral simulated digestive fluid, gastric digestion fluid, simulated intestinal fluid were provided from Shanghai Yuanye Biotechnology Co. (Shanghai, China).

2.2. Processing of raw black bean

The black bean seeds were washed with distilled water and immediately placed in an oven for drying. The final moisture content was kept at 10–11 %. The dried black bean samples were crushed using a grinder and made to pass through a sieve with a sieve size of 0.180 mm, and stored at − 20 °C for further use.

2.3. Steaming method

A 50 g sample of black beans were taken for pretreatment such as washing and soaking to ensure that they were about 50 % hydrated, placed in CFXB16YA3–36 Midea rice cooker (Midea Life Electric Appliance Manufacturing Co., CN) for steaming (500 mL distilled water for 60 min). The beans were then dried in an oven at low temperature until the sample remained at a constant weight, followed by grinding and storage at −20 °C until further use.

2.4. In vitro simulation of digestion

The pattern of metabolite changes was studied during in vitro digestion of black beans steamed at atmospheric pressure. In vitro simulated digestion experiments, including simulated oral, gastric, and small intestinal digestions, were performed using the method described by Minekus et al. (Minekus et al., 2014).

Oral mock digestion: 3 g of black bean powder after atmospheric pressure steaming was placed in a beaker, 4 mL of oral simulated digestive fluid (SSF, pH = 7) was mixed well and 0.3 mL of α-amylase was added, and the solution was mixed well. The samples were shaken for 5 min in a water bath at 37 °C, protected from light.

Gastric mock digestion: The gastric phase digestion test was initiated by adding 4.5 mL of gastric digestion fluid (SGF, pH = 3) and 0.1 mL of pepsin to the oral digestion products described above. The samples were shaken for 2 h in a water bath at 37 °C, protected from light.

Intestinal mock digestion: 6.6 mL of simulated intestinal fluid (SIF, pH = 7), and 3.0 mL of trypsin were added to the oral-gastric digestion products described above. The samples were shaken for 2 h in a water bath at 37 °C, protected from light.

At the end of the intestinal digestion, the digested samples were centrifuged (×12,000 *g*, 10 min) and the collected supernatants. The next step is to remove most of the inorganic salt ions from the supernatant with a desalination membrane. Finally, the processed digested products were freeze-dried and stored at −80 °C for metabolomics analysis by UHPLC-QE-MS.

2.5. UHPLC-QE-MS detection

The above samples were divided into three groups: a raw black bean group (group r), an atmospheric steam-processed black bean group (group ops), and an atmospheric steam-processed black bean digestion group (group opsd) for metabolomic analysis.

Metabolite extraction: 5 g of sample was added to 20 mL of extract (methanol, containing isotope-labeled internal standard mixture). Then the samples were homogenized at 35 Hz for 4 min and sonicated (A KQ-250E ultrasonicator, Shenzhen Redbone Electronics Co., Shenzhen, CN) for 10 min in an ice-water bath. The homogenization and sonication cycle was repeated 3 times. After completion, the mixed solution was centrifuged (4 °C, 12000 rpm, 15 min). The supernatants were filtrated with a 0.22 μm millipore filter and collected for analysis, and the quality control (QC) sample was prepared by mixing an equal aliquot of the supernatants from all of the samples.

Chromatographic conditions (A Vanquish™ Ultra-high-performance liquid chromatography system, Thermo Fisher Scientific, USA): The chromatographic column was a Waters ACQUITY UPLC HSS T3 (2.1 mm × 100 mm, 1.8 μm). Phase A consisted of 5 mmol/L ammonium acetate and 5 mmol/L acetic acid, phase B consisted of acetonitrile, and the injection volume was 3 μL.

Mass spectrometry conditions (Q Exactive™ HFX high-resolution mass spectrometer, Thermo Fisher Scientific, USA): Mass spectrometry voltage 4.0 kV (positive ions), 3.8 kV (negative ions). Sheath gas flow rate of 30 arb; auxiliary gas flow rate of 10 arb; full MS resolution 60,000; MS/MS Resolution 7500; and ion transfer tube temperature 350 °C.

The ionisation sources of the QE platform electrospray ionisation, positive ion mode (POS) and negative ion mode (NEG), were used in this study. The combination of these two modes ensures high coverage and good detection of sample metabolites.

2.6. Data processing

Metabolites were compared between the r group and the ops group, and between the ops group and the opsd group, denoted as r vs. ops and ops vs. opsd, respectively. The raw data were pre-processed using ProteoWizard software and R package (kernel XCMS) for format transformation and feature peak extraction. The processed data were subsequently subjected to orthogonal partial least squares-discriminant analysis (OPLS-DA) and hierarchical cluster analysis (HCA) using SIMCA software. According to the OPLS-DA model, the *P* value of Students' *t*-test was less than 0.05, and the variable importance in the projection (VIP) parameter of the principal component of the OPLS-DA model was greater than 1 as the set parameter for screening the differential metabolites between the groups.

3. Results and analysis

3.1. Metabolite characterization

The metabolites detected were matched and annotated by HMDB, KEGG, and other databases, and 922, 945, and 878 metabolites were identified in the r, ops, and opsd groups, respectively, including 13 secondary metabolites, such as amino acids and their derivatives, organic acids and their derivatives, lipids, polyphenols, sugars, nucleic acids, nucleotides and their derivatives, alkaloids, and terpenoids (specific data for the metabolite characterization are shown in the Table S1 of Appendix:).

As shown in Table 1, most metabolites in raw black beans were amino acids, organic acids, phenols, lipids, and sugars, accounting for more than 50 % of the total, indicating that these metabolic components are the main constituents of black beans. The ops group had 13 more total metabolite species than the r group, mainly amino acids and their derivatives, lipids, and sugars. In addition, the two groups differed in the number and type of unique metabolites. There were 33 metabolites unique to the r group and 55 metabolites unique to the ops group. The proportion of amino acids and lipids in the unique metabolites of the ops group was significantly higher than that of the r group, probably because of the denaturation of proteins in black beans after autoclavin, which produced free amino acids under the action of endogenous proteases, resulting in an elevated proportion of amino acids among the metabolites. Black bean fats and oils also undergo hydrolysis in the presence of heat and water to produce glycerol and fatty acids (Feng et al., 2022). It has been reported that the synthesis and degradation of metabolites in raw materials during steaming and processing is a dynamic process, and longer steaming may lead to a higher rate of synthesis of amino acids and fatty acids than their degradation and transformation (Li et al., 2019), which is consistent with the results obtained in this study. These results indicate that the atmospheric pressure steaming method promotes the production of amino acids and lipids, and these changes in endogenous components not only enhance the nutritional value of processed black beans but also give them a unique taste.

The opsd group showed a decrease in the number of total metabolites compared to the ops group, with the most significant decrease in phenolic compounds (48.05 % decrease in number), whereas the number of amino acids increased by 9.52 % after in vitro digestion. By comparing the specific metabolites of the two groups, we found that the number of phenolic and terpenoid compounds with antioxidant and anti-inflammatory physiological effects in the opsd group was significantly lower than that in the ops group (9 phenolic and 20 terpenoid compounds in the opsd group, 36 phenolic and 15 terpenoid compounds in the ops group). Numerous studies have confirmed that the phenolic and terpene components of black beans exert substantial hypoglycaemic, anti-inflammatory, and immune-enhancing effects, but they are also at risk of degradation during the digestive stage and are not highly bioaccessible. The chemical structures, molecular sizes, and degrees of polymerisation of phenolic compounds and their combination with macronutrients (proteins, lipids, and carbohydrates) can affect the final bioaccessibility (Iglesias-Carres et al., 2019). The proportion of amino acid components in the unique metabolites of both groups was high (36 in the opsd group and 15 in the ops group), and the number of amino acid substances in the opsd group was much greater than that in the ops group and was dominated by dipeptides or free amino acids, presumably generated by the hydrolysis of black bean proteins by digestive enzymes, which is conducive to meeting the nutritional requirements of the organism. In conclusion, after in vitro digestion of black beans under atmospheric pressure, there is a tendency for degradation and transformation of phenolic compounds, which affects bioaccessibility; however, at the same time, the digestion process helps the production of amino acids (especially short peptides with better digestive and absorptive properties), which facilitates the efficient utilisation of protein-based nutrients in black beans, and ultimately facilitates nutrient supplementation in humans.

Table 1
Statistics of metabolites in each group.

Serial number	Category	Number (as a percentage)		
		r	ops	opsd
1	Amino acids and their derivatives	205 (22.23 %)	210 (22.22 %)	230 (26.20 %)
2	Organic acids and their derivatives	99(10.74 %)	99(10.48 %)	96(10.93 %)
3	Lipids	70(7.59 %)	75(7.94 %)	63(7.18 %)
4	Polyphenols	74(8.03 %)	77(8.15 %)	40(4.56 %)
5	Sugar	51(5.53 %)	55(5.82 %)	52(5.92 %)
6	Nucleic acids, nucleotides and their derivatives	39(4.23 %)	42(4.44 %)	42(4.78 %)
7	Alkaloids	24(2.60 %)	24(2.54 %)	24(2.73 %)
8	Terpenoids	22(2.39 %)	22(2.33 %)	27(3.08 %)
9	Vitamins	12(1.3 %)	13(1.38 %)	10(1.14 %)
10	Steroid	6(0.65 %)	6(0.63 %)	13(1.48 %)
11	Anthocyanins	4(0.43 %)	4(0.42 %)	2(0.23 %)
12	Coumarin	4(0.43 %)	4(0.42 %)	4(0.46 %)
13	Other	312 (33.84 %)	314 (33.23 %)	275 (31.32 %)
Total		922(100 %)	945(100 %)	878(100 %)

3.2. Multivariate statistical analysis

To accurately identify variables with little correlation within the samples, maximise the distinction between groups, and facilitate more efficient and accurate screening of differential metabolites, orthogonal variables in black bean metabolites that were not related to differential variables were filtered using a supervised-mode OPLS-DA multivariate statistical model, and orthogonal and non-orthogonal variables were analysed simultaneously. As shown in Fig. 1, R^2X, R^2Y, and Q^2 of the OPLS-DA model were greater than 0.5 for each group (for the r vs. ops group, $R^2X = 0.625$, $R^2Y = 1$, and $Q^2 = 0.955$ for the ESI+ model, and $R^2X = 0.75$, $R^2Y = 0.997$, and $Q^2 = 0.95$ for the ESI model. For the ops vs. opsd group, $R^2X = 0.912$, $R^2Y = 0.999$, and $Q^2 = 0.996$ in the ESI+ mode and $R^2X = 0.939$, $R^2Y = 1$, and $Q^2 = 0.999$ in the ESI- model), where Q^2 (ESI+ and ESI-) was greater than 0.9, indicating the reliability and predictability effect of the OPLS-DA model developed in this study are good (Li, Lin, et al., 2021). The excellent separation of sample points on both sides of the confidence interval between the two groups for both r vs. ops and ops vs. opsd indicates the feasibility of the model for screening differential metabolites and also confirms that both atmospheric steam processing and in vitro digestion can affect the black bean metabolome.

From Fig. 2A-D, it can be seen that all R^2 and Q^2 points obtained from the OPLS-DA model replacement test (ESI+ and ESI-) for both groups were lower than the most original R^2 and Q^2 on the right. The original model Q^2 is close to 1, the value of the point where the Q^2 regression line intersects the Y-axis is less than 0, and the R^2 and Q^2 of the stochastic model gradually decrease as the amount of permutation retention decreases, indicating that the original model is not overfitted (Zhang et al., 2021). In summary, the model had good predictive performance and can be reliably used for screening differential marker metabolites.

3.3. Differential metabolite screening and HCA

In this study, a total of 210 differential metabolites in the r vs. ops group and 150 differential metabolites in the ops vs. opsd group were

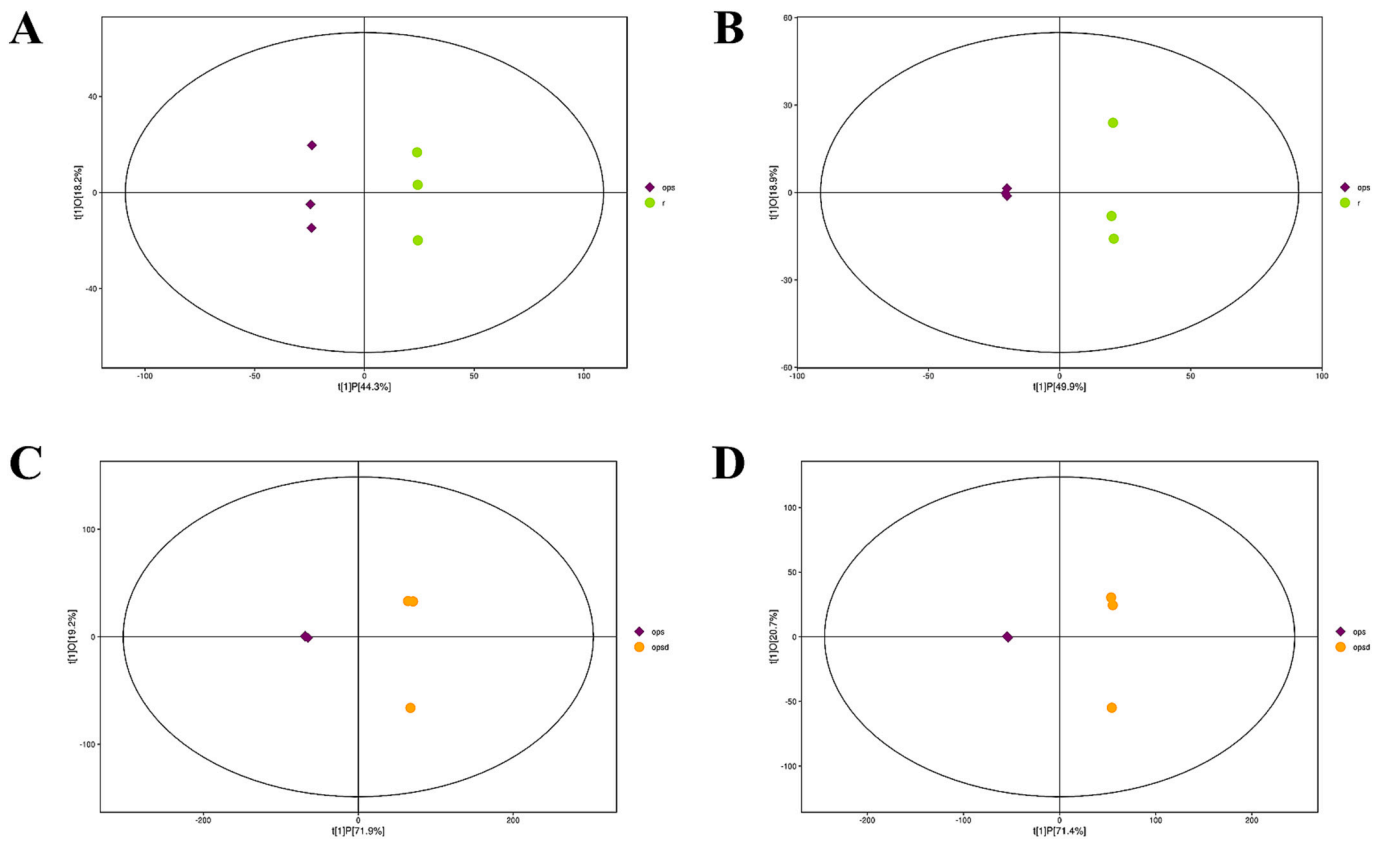

Fig. 1. OPLS-DA results for r vs ops group (A and B) and ops vs opsd group (C and D). A, C represent ESI+ modes. B, D represent ESI- modes.

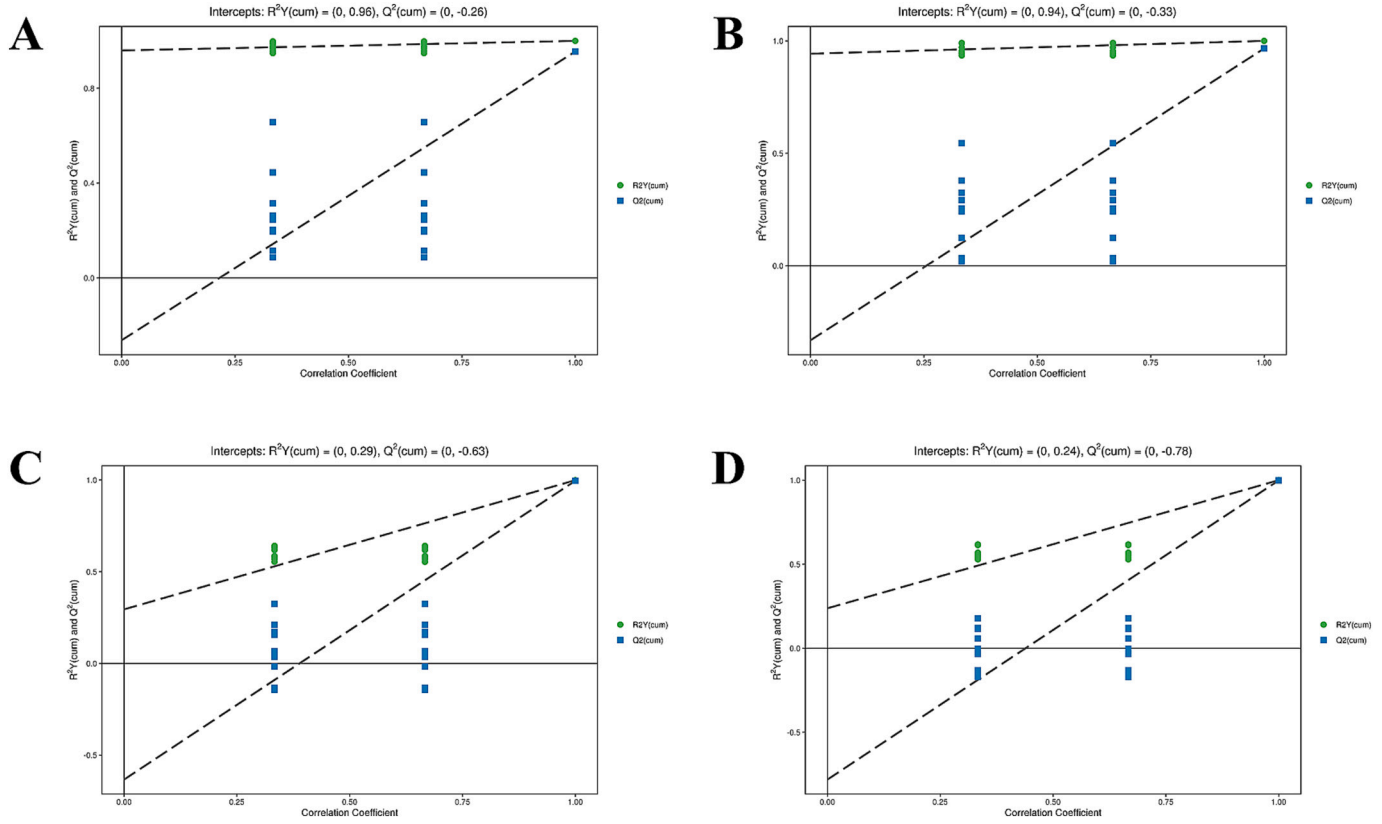

Fig. 2. OPLS-DA model replacement test results for r vs ops group (A and B) and ops vs opsd group (C and D). A, C represent ESI+ modes. B, D represent ESI- modes.

screened by setting in the OPLS-DA discriminant model (VIP > 1, $p < 0.05$), and specific information about the name, VIP value, and fold change (FC) of each metabolite is shown in the Appendix (Table S2 and Table S3). The effect of steam processing treatment and in vitro simulated digestion on the expression of differential metabolites in black beans was demonstrated by a heat map (Ai et al., 2021), with red colour indicating high expression of the substance and blue colour indicating low expression.

3.3.1. r vs. ops group

For the r vs. ops group, 210 differential metabolites were identified, dominated by the three groups of amino acids and their derivatives, phenols, and organic acids and their derivatives, as well as a certain number of lipids, sugars, and substances such as nucleic acids and nucleotides and their derivatives, with ploidy variations ranging from 0.00033 to 289.57. As shown in Table 2, Fig. 3A-B, the expression of 113 metabolites was higher in normal-pressure steamed black beans than in raw black beans, and these mostly comprised amino acids and phenolic compounds, and the amino acids mainly included dipeptides such as Lys-Val, Arg-Phe, His-Phe, and monomers such as histidine and γ-aminobutyric acid. There were 29 phenolic compounds, including naringin and root bark glycosides. In addition, the expression of differential metabolites (97) such as phenolics compounds, lipids, and amino acids was significantly lower in normal-pressure steamed black beans than in raw black beans. On the one hand, the high temperature of the steaming process and the penetration of water between the cells of black beans facilitate changes in the polymerisation state of phenols, proteins, carbohydrates, and lipids, and facilitate the release of each substance (Rinaldi et al., 2021). On the other hand, high temperature, water infiltration, and other conditions enhance the activity of various kinases in black bean cells, and each molecule is more active, catalysing complex biochemical reactions and mutual transformation, which can lead to changes in the content of these substances (Cianciosi et al., 2022). Naringenin and luteolin are bioactive molecules that exert anti-inflammatory, neuroprotective, and antioxidant functions (Caporali et al., 2022; Escribano-Ferrer et al., 2019), and activation of this flavonoid biosynthetic during steaming facilitates the maintenance of adequate and diverse physiological activities of steamed black beans to maintain the health of the organism. It has been shown that steaming treatment adversely affects the total polyphenols of beans. For example, Mba et al. (Mba et al., 2019) found that the total phenolic content of eight common beans decreased to varying degrees after soaking-steaming (where the maximum loss could reach 80 %), but the results of this study showed that there was limited downregulation of the amounts of phenolic compounds in atmospherically steamed black beans compared to raw black beans, which was considered to be due to the inherent characteristics of black beans, such as the cell wall structure, the ratio and degree of binding of compounds, and soaking conditions. In the present study, we also found that some phenolics such as (−)-Epiafzelechin, Luteolin 7-galactoside, and 6″-Malonylgenistin showed a down-regulation of their content after atmospheric pressure cooking, which may be attributed to the de-glycosylation of these phenolic compounds during this process, leading to structural transformation and self-degradation (Hsiao and Hsieh, 2018). The decrease of some other amino acids such as D-Glutamine, Lysinoalanine, D-Aspartic acid, etc. may be due to the high temperature conditions of atmospheric steaming which caused the dissolution and leaching of amino acids, accompanied by transamination and deamination reactions (Chopra and Hira, 1986; Yagoub et al., 2008). In addition, it can be seen from Table S1 that elevated levels of glucose-1-phosphate, a metabolite of starch and sucrose conversion, (G1P) facilitates sugar accumulation, makes the finished product sweeter, and ultimately improving the taste quality of black beans after steaming. Generally, the steaming process can largely preserve the functional activity and flavouring components of food and enhance its sensory and nutritional characteristics (Lafarga et al., 2018), while in this process, various substances (such as black bean proteins and carbohydrates) undergo colour and flavouring reactions such as Merad (Han et al., 2022), which work synergistically and jointly to form the unique flavour characteristics of black beans under normal pressure steaming.

Table 2
Statistics of the differential metabolites screened in the two groups.

Serial number	Category	Significant differences	
		r VS ops(Up/Down)	ops VS ops(Up/Down)
1	Amino acids and their derivatives	33(18/15)	36(29/7)
2	Organic acids and their derivatives	23(12/11)	7(1/6)
3	Lipids	14(2/12)	16(5/11)
4	Polyphenols	49(29/20)	28(2/26)
5	Sugar	11(8/3)	9(3/6)
6	Nucleic acids, nucleotides and their derivatives	17(12/5)	6(1/5)
7	Alkaloids	6(3/3)	4(2/2)
8	Terpenoids	10(5/5)	12(5/7)
9	Vitamins	3(2/1)	0(0/0)
10	Steroid	2(2/0)	1(1/0)
11	Anthocyanins	2(0/2)	2(0/2)
12	Coumarin	3(1/2)	2(1/1)
13	Other	37(18/19)	27(11/16)
Total		210(113/97)	150(61/89)

3.3.2. opd vs. opsd group

As shown in Table 2, Fig. 3C-D, for the ops vs. opsd group, there were 150 differential metabolites, with amino acids and their derivatives and phenolic compounds accounting for a large proportion. After in vitro digestion of atmospherically steamed black beans, amino acids were predominantly upregulated (61 differential metabolites), and most phenolic components showed a downward trend (89 differential metabolites), indicating that these two components were more significantly affected during the process. In in vitro digestion, oral amylase, pepsin, and trypsin have specific cleavage sites for most proteins and long-chain polypeptides in black beans steamed at atmospheric pressure into small molecules (such as short peptides and free amino acids) for efficient absorption by humans. These highly expressed differential amino acid substances contain significant amounts of essential amino acids such as lysine and tryptophan, further enhancing their nutritional properties and physiological activity (Do Evangelho et al., 2017). In addition, when digestive enzymes hydrolyse these food macronutrients, phenolics bound to macromolecules can be released, thereby increasing the bioaccessibility of polyphenols (Perez-Hernandez et al., 2021). However, studies have shown that the bioaccessibility of plant polyphenols also decreases to varying degrees after oral ingestion and digestion, accompanied by a loss of their physiological activities, such as antioxidant activity. The results revealed that the content of three flavonoids, naringenin, epiafzelechin, and kaempferol, decreased after in vitro digestion with fold changes between 13.26 and 8866.50. These active substances showed excellent performances in loss of cell repair, inflammation amelioration, and regulation of oxidative stress responses (Li, Sun, et al., 2021; Zhang et al., 2022), and these results suggest that active substances such as naringenin are poorly tolerated in the environment, leading to a limitation in the exertion of physiological activity. In the meantime, the binding and release of phenolic acids constitute a dynamic process, but it is usually the case that only a few phenolic substances are released and utilized by the organism during the digestion phase. For example, the strong covalent bonding forces between some phenolic acids (e.g., ferulic acid) and cell wall polysaccharides, which cannot be cleaved by trypsin, can lead to insufficient release of some phenolic substances and affect their bioaccessibility (Giusti et al., 2019), as described in the studies by Li et al. (Li et al., 2023) and Giusti et al. (Giusti et al., 2019). This ist also consistent with the decrease in

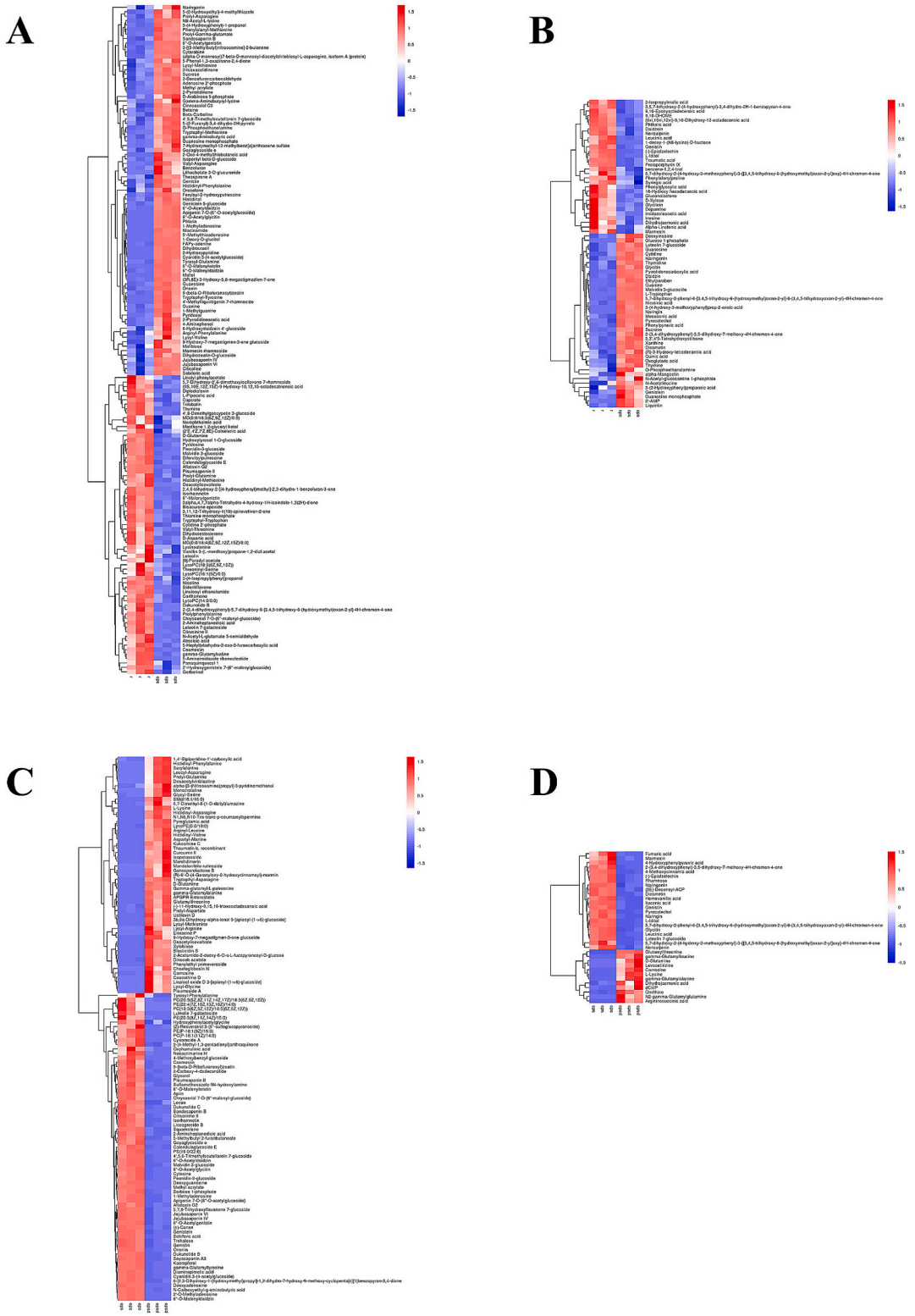

Fig. 3. Differential metabolite HCA plot of r vs ops group (A and B) and ops vs opsd group (C and D). A, C represent ESI+ modes. B, D represent ESI- modes.

phenolic component fractions and content after the in vitro digestion of black beans in this study. In summary, the in vitro digestion process promotes the release of beneficial nutrients such as amino acids and ensures nutrient supply; however, it also affects the full release and activity assurance of some active substances such as phenols, and overall affects the bioaccessibility of some substances in black beans. Thus, it can be seen that in vitro digestion has obvious positive effects on the production and provision of nutrients for the human body by black beans, but it also affects the stable release and physiological activity assurance of its active substances. More in-depth studies can be conducted in the future on the delivery of bioactive substances in black beans.

4. Conclusion

In this study, we used a UHPLC-QE-MS non-targeted metabolomics technique to investigate the effects of atmospheric steam processing and in vitro digestion on the metabolic components of black beans. The results showed that the content of most amino acids, sugars, and phenolic substances increased and that lipids decomposed in raw black beans after autoclave processing, and the changes in the content of these metabolic substances contributed to the formation of the unique taste (texture and flavour) and the good nutritional and physiological functional properties of autoclaved black beans. In vitro digestion increases the amino acid content of normally processed black beans, which helps meet the nutritional requirements of the organism, while decreasing the phenolic content, affecting their bioaccessibility. In this study, we established the metabolic profiles of black beans during atmospheric pressure steaming and in vitro simulated digestion, and we elucidated the metabolite transformation patterns affecting the nutritional, taste, and physiological activity characteristics of black beans in these two processes, thus providing data to support the processing and utilisation of black beans.

CRediT authorship contribution statement

Lu Bai: Writing – original draft. **Zhiming Li:** Formal analysis. **Shu Zhang:** Methodology. **Yuchao Feng:** Investigation. **Miao Yu:** Validation. **Tong Wu:** Methodology. **Changyuan Wang:** Writing – review & editing.

Declaration of competing interest

The authors declare no competing financial 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.fochx.2024.101997.

Data availability

Data will be made available on request.

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