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Metabolomics analysis of Jining Gray and Lubei White muttons through LC-MS technique

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Abstract

To explore variation in the meat metabolites of two goat breeds, metabolomics analysis of the mutton samples of Jining Gray and Lubei White goat breeds was performed using mass spectrometry (LC-MS). The profile revealed a total of 2,599 metabolites in the mutton samples. The significantly higher ($p < 0.05$) metabolites included methionyl asparagine ($p = 0.017$), hydrocarbons, tryptophylhistidine, L-glutamine, D-glutamine, and indole acrylic acid in the longissimus dorsi muscle of Jining Gray goats compared to Lubei White goats. The essential amino acids, such as histidine, phenylalanine, and L-tyrosine were significantly higher in the mutton of the Lubei White goats compared to that of the Jining Gray goats. The contents of 2-hydroxycinnamic acid ($p = 0.027$) and nicotinamide ($p = 0.002$) in Lubei White goat were also significantly high. Taken together, the data showed that the meat profile of the Lubei White and Jining Gray goats significantly varied. The data would be helpful in further studying the meat traits in the two goat breeds.

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Introduction

Meat is full of essential nutrients such as minerals, trace elements, and amino acids required for normal biological functions of the body (Toldrá and Reig, 2011). The meat from livestock species is an important component of the diet, which provides essential nutrients to the human population (Hoffman and Falvo, 2004; Khan *et al*., 2020). The demand for meat is increasing globally, and 25% increase was seen in the past ten years (Esua *et al*., 2021). As the living standards of humans improve, the quality of meat becomes an essential factor in consumers' preferences (Bai *et al*., 2018; Khan *et al*., 2019; Junjvlieke *et al*., 2020). This increasing trend in the consumers' demand for high-quality meat steered researchers to explore meat with health benefits and nutritional quality (Elmasry *et al*., 2012; Taheri-Garavand *et al*., 2019).

Following significant innovation in the determination mechanism of goat meat quality, further exploration of complex interactive factors leads to variation in meat quality based on data-driven "omics" techniques. The omics methods, such as transcriptomics, proteomics, genomics, and metabolomics, are the latest innovations with proven potential in the exploration of meat quality assessment (Herrero *et al*., 2012).

Goat meat is very promising compared to other meats in terms of nutrition and health characteristics. Goat meat contains a low level of cholesterol, which makes it an ideal choice of food in terms of its nutritious and health benefits. Additionally, goat meat also contains various antioxidants and essential minerals (Nassu *et al*., 2003). Goat meat is easily digestible, and very hypoallergenic (Tokysheva *et al*., 2022).

The liquid chromatography-mass spectrometry (LC-MS) technique can analyse and detect various metabolites, especially metabolic mapping that helps researchers explore factors affecting the quality of food commodities including meat and milk (Rocchetti and O'Callaghan, 2021). There are various factors influencing food quality, particularly flavour compounds. Recently, a metabolomics study explored variations in arabitol, citric acid, α-ketoglutaric acid, glyceric acid, *myo*-inositol, and glycine in sheep and goat milk (Caboni *et al*., 2019). Similar variation was found in the metabolomics profiling of different livestock breeds in terms of

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phosphate, creatinine, and lactate metabolites, which contribute to the taste and flavour variation of beef from different geographical origins (Jung *et al*., 2010).

The mutton from goat is considered one of the most preferred meats worldwide, and contributes about 6% of the total meat consumption around the globe (Aepli and Finger, 2013). In China, there are about 58 indigenous goat breeds. Unfortunately, 7% of the known goat breeds are extinct, and 20% are endangered. The Jining Gray and Lubei White goats are two important indigenous caprine breeds of China, mainly found in Shandong Province. These indigenous Chinese breeds possess many traits of economic importance such as extensive adaptability to stressful environments, resistance to infectious diseases, survival in feed scarcity on coarse fodder, and high prolificacy (Gao *et al*., 2023; Ma *et al*., 2024). Recently, Chinese indigenous goat breeds have been endangered by the introduction of exotic goat breeds, which have typically been selected for optimal production of meat, wool, or other products (Liu *et al*., 2019). These short-term profits pose a severe threat to the indigenous goat breeds, and will cause the loss of the genetic pool if crossing and replacement continue with the exotic breeds. Therefore, it is imperative to screen the indigenous goat breeds for traits of economic importance such as meat quality.

Previously, a metabolomics study explored significant variations in the meat flavour metabolites of three different caprine breeds namely Boer, Lubei White, and Jining Gray (Wang *et al*., 2019). However, the differential variation of the health and nutritionally beneficial metabolites in the Jining Gray and Lubei White goat breeds still needs to be explored.

Materials and methods

The experimental animals were farmed at Heze Zhengdao Animal Husbandry Technology Co., Ltd., Shandong Province, China. A total of 12 bucks of ten months, six each from the Lubei White and Jining Grey goats of similar body weight were selected, housed, and managed under the same environmental conditions of humidity and temperature in individual cages. All experimental animals were fed with the uniform standard ration on a TMR (total mixed ration) basis (Table 1). The animals were handled following the guidelines of the ethical committee of the School of Pharmacy, Heze University, China.

Table 1. Ration composition on dry matter basis.

Component	Percentage	Nutritional value	-
Hay (peanut)	58.31	Dry matter ($kg \cdot d^{-1}$)	0.6
Corn	29.3	Digestive energy ($MJ \cdot d^{-1}$)	6.29
Bran	4.36	Protein ($g \cdot d^{-1}$)	64
Soybean meal	2.01	Calcium ($g \cdot d^{-1}$)	4.6
Cottonseed meal	2.05	Phosphorus ($g \cdot d^{-1}$)	3
Calcium bicarbonate	1.1	Sodium ($g \cdot d^{-1}$)	3.1
Sodium bicarbonate	0.37	-	-
Sodium chloride	0.5	-	-
5% Premix (trace elements)	2	-	-

Sample pre-treatment

The animals were kept fasted for 12 h before sampling, then humanely euthanised through electric stunning, and completely bled. The carcass was chilled at 4°C for 24 h, and a sample from the longissimus dorsi muscle was collected and stored at -80°C for further analysis.

Liquid chromatography (LC)

For the preparation of each sample, 30 mg meat tissue was taken in 1.5 mL Eppendorf tube along with two small steel balls. Then 20 µL of 2-chloro-l-phenylalanine in methanol, 0.3 mg/mL (internal standard), and 400 µL methanol/water (4/1, v/v) as extraction solvent were added to each sample, and stored at -80°C for 2 min. After grinding at 60 Hz for 2 min, the samples were ultrasonicated at ambient temperature for 10 min, and stored at -20°C for 30 min. The extract was centrifuged at 13,000 rpm at 4°C for 15 min. Next, 300 mL of supernatant in a glass vial was freeze-dried through centrifugation, and a 300 µL mixture of methanol and water (1/4, v/v) was

added to each sample, vortexed for 30 s, and incubated at 4°C for 2 min. The samples were further centrifuged at 13,000 rpm at 4°C for 5 min, and 150 µL supernatant from the samples was transferred into crystal syringes, filtered with 0.22 µm micro-filters, collected in LC vials, and stored at -80°C until analysed in LC-MS. A pooled sample was prepared by mixing the aliquots of all the samples for quality control (QC).

Mass spectrometry

The metabolic profiling analysis was used in both ESI negative and ESI positive ion modes with heated electrospray ionisation (ESI) source through Dionex Ultimate HPLC system (3000 RS UHPLC) fitted with Q Executive Quadrupole Orbitrap Mass Spectrometer (Thermo Fisher Scientific, Waltham, MA, USA). In both the positive and negative modes, a column of 1.7 µm, 2.1 × 100 mm (ACQUITY UPLC BEH C18 column) was used with the binary gradient elution system, which contained buffer "A" (0.1% formic acid in water, v/v) and buffer "B" (0.1% formic acid in acetonitrile, v/v). The separation was performed through the following gradient of B: 5 - 20% at 0 - 2 min; 20 - 60% at 2 - 4 min; 60 - 100% over 4 - 11 min; the composition was held at 100% for 2 min; 5 - 100% for 13 - 13.5 min; and finally, 13.5 - 14.5 min holding at 5%. The column temperature was 45°C with the flow rate of 0.4 mL per min, and the injection volume was 5 µL.

The mass range was (*m/z*) from 66.70 to 1,000.5. The resolution parameters for the full MS scans were 70,000, and for HCD MS/MS scans was 35,000 at 10, 20, and 40 eV collision energy. The operating parameters of the mass spectrometer were set as follows = sheath gas flow rate: 45 arbitrary units' spray; the voltage: 3000 V (+) and 2500 V (−); auxiliary gas flow rate: 15 arbitrary units; and capillary temperature: 350°C. The QC samples were run after every 10 sample injections to assess the repeatability of the data set.

Statistical and bioinformatics analysis

The collected raw data from the LC-MS were analysed by the Progenesis QI software (Waters Corporation, USA), with RT (retention time) tolerance: 0.02 min; precursor tolerance: 5 ppm; and fragment tolerance: 10 ppm. The internal standard detection parameters and isotopic peaks were de-selected for peak RT alignment. The noise elimination level was set at 10.00, and the minimum intensity was set as 15% of the base peak intensity. The Excel file was obtained based on peak RT and peak intensities; mass-to-charge (*m/z*) ratio; and RT-*m/z* pairs were used as indicators for each ion. The qualitative compounds were screened based on their result scores. The screening criteria were 36 points with a full score of 60 points, and 36 points were considered erroneous and excluded. The negative and positive ions data were collectively put in a data matrix table, which included completed details of the information extracted from the raw data, which served as base for subsequent analysis. The internal standard was used for quality control and data reproducibility. The metabolites were documented through the data processing software Progenesis QI (Waters Corporation, USA) available at http://hmdb.ca, http://lipidmaps.org, and self-built databases. The PCA (principle component analysis), PLS-DA, and OPLS-DA (orthogonal partial least-squares discriminant analysis were performed through R ropls package to visualise the metabolomics variation between the experimental groups. The 95% confidence interval of the model variation was shown as an ellipse in score plots of the models presented through Hotelling's T2 region. The variables with VIP > 1 (variable importance in the projection) were measured as relevant for group discernment. The VIP value (VIP > 1) and *p*-value ($p < 0.05$) from a two-tailed Student's *t*-test on the normalised peak areas were the selection criteria for the differential metabolites in both experimental groups. The annotated peak areas were normalised based on the sample quantity and internal standard levels to extract the relative levels of each metabolite.

Results and discussion

Metabolomics profiling of goat meat

Goat meat (chevon or mutton) contributes to about 6% of red meat consumption in the world, and is known for its delicacy (Aepli and Finger, 2013). Same as lamb, the cooked meat of goat produces a peculiar flavour that is very different from beef. This distinct mutton flavour is a base for consumer preferences. Consequently, efforts are continuing to explore various factors affecting goat meat flavour (Dashdorj *et al*., 2015; Arshad *et al*., 2018). Genetics plays an essential role in meat flavour and other quality attributes (Laborde *et al*., 2001). The variation in meat quality within different goat breeds is due to different metabolites (Gregory *et al*., 1994) and

nutrition such as carbohydrates, amino acids, carbonyls, and lipids (Gregory *et al*., 1994). In the present work, the LC-MS analysis identified 2,599 metabolites in muscle samples of two goat breeds. The PCA analysis exhibited significant ($p < 0.05$) variation between the metabolic profiles of Jining Grey and Lubei White goat breeds (Figure 1).

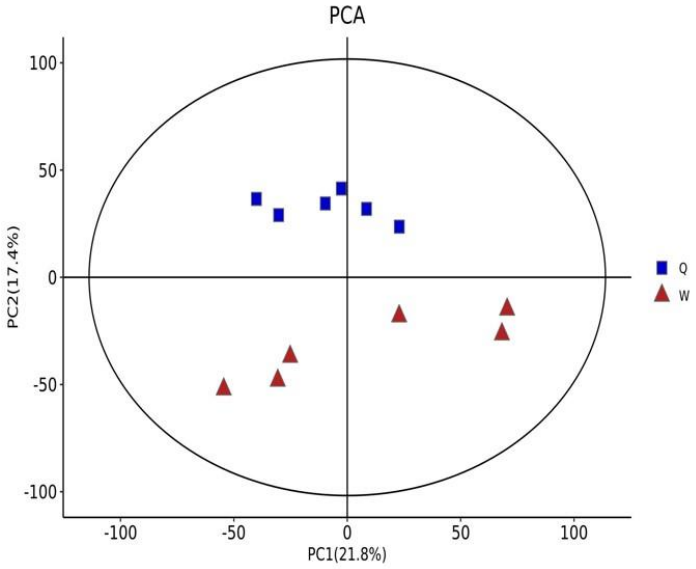

Figure 1. PCA plot of different metabolic patterns in Jining Gray and Lubei White muttons.

Moreover, the PLS-DA score plot revealed no overlap in the qualitative and quantitative data between the mutton samples of the two goat breeds which also indicated significant ($p < 0.05$) metabolite variation (Figure 2).

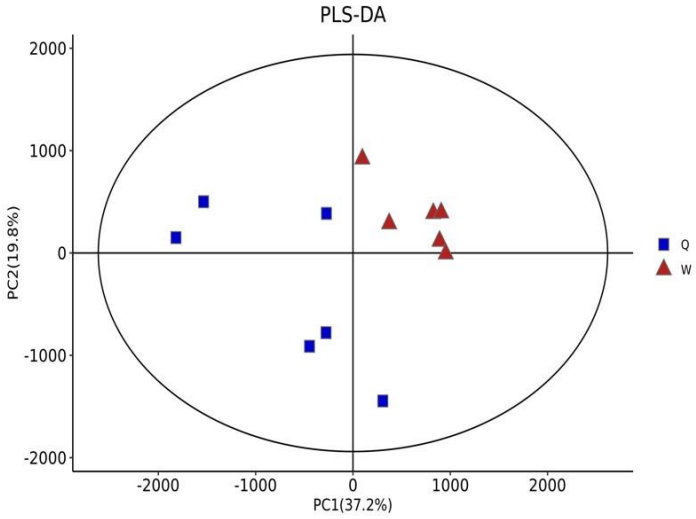

Figure 2. PLS-DA plot of different metabolic patterns in Jining Gray and Lubei White muttons.

A similar trend was exhibited through OPLS-DA plots (Figure 3). In the present work, the identification of various metabolites depicted variation in the mutton of two goat breeds, and we assumed that this could be a base of variation for taste, aroma, nutrition, and health benefit qualities. However, a functional study of these metabolites in meat was not performed in the present work. Therefore, an in-depth functional study is needed to explore the role of these metabolites in meat quality, especially in nutrition and health benefits.

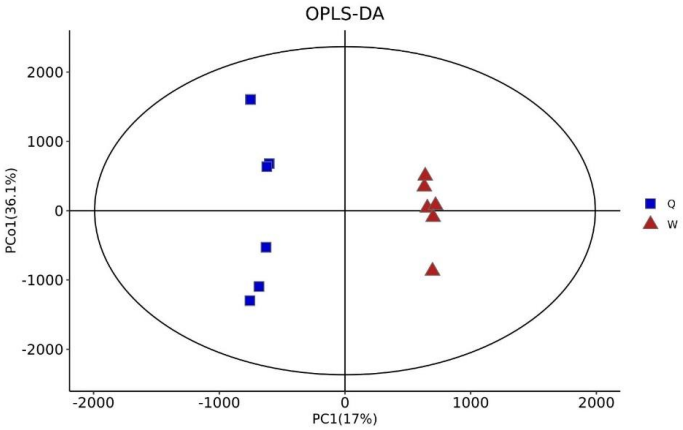

Figure 3. OPLS-DA plot of different metabolic patterns in Jining Gray and Lubei White muttons.

Differential metabolomics profiling of goat meat

In-depth exploration of the biological mechanism behind the metabolite variation in meat quality of different goat breeds increased the interest of researchers. Despite greater development in the metabolomics application, still little information is available regarding differential metabolomics profiling addressing meat quality variation in different breeds. Therefore, the present work was aimed at comparative profiling of the metabolites in the mutton of Jining Gray and Lubei White goat breeds. A significant ($p < 0.05$) variation was found in the metabolic profile between the muttons of the two goat breeds (Figure 4A). The hierarchical clustering analysis also exhibited significantly ($p < 0.05$) diverse metabolic patterns in the meat of both goat breeds (Figure 4B).

The results of differential metabolites profiling showed that 42 metabolites increased, and 38 metabolites decreased in production (Table 2). The most abundant metabolite classes in the muttons of Lubei White and Jining Grey goat breeds were lipid and lipid-like molecules, organic acids and derivatives, organic oxygen compounds, organo-heterocyclic compounds, benzenoids, nucleosides, nucleotides, and analogues, phenylpropanoids and polyketides, and organic nitrogen compounds. These

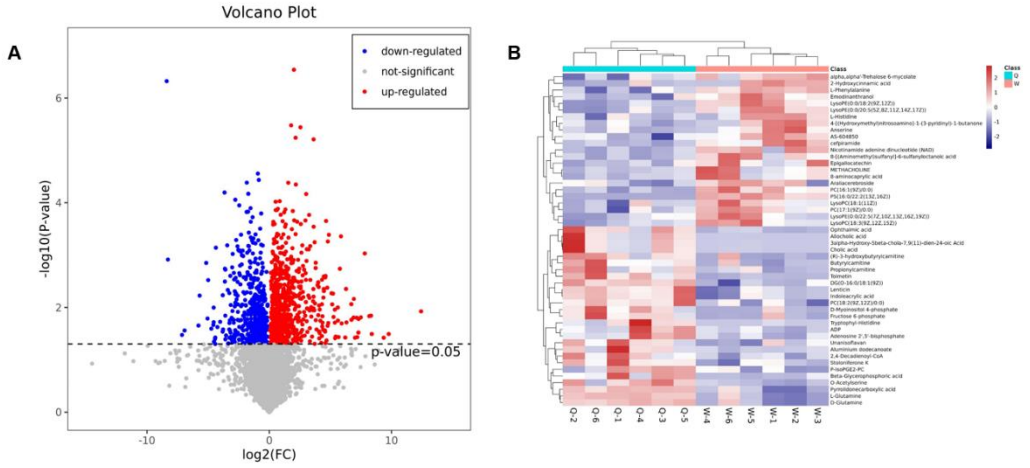

Figure 4. (A) Volcano plot of metabolite distribution based on VIP > 1, $p < 0.05$, and fold change ≥ 2 or ≤ 0.5 screening criteria. Different metabolites were found between Jining Gray and Lubei White goat breeds. **(B)** Hierarchical clustering analysis of different metabolites between Jining Gray and Lubei White muttons.

Table 2. Metabolites detected in Jining Gray and Lubei White muttons.

Metabolite	*m/z*	Rt (min)	Super class	VIP	*p*-value	Avg (JG)	Avg (LW)
Allocholic acid	407.280	9.525	Lipid and lipid-like molecule	2.376	0.032	1144.480	3.410
Cholic acid	426.320	9.530	Lipid and lipid-like molecule	1.835	0.039	710.038	16.966
Beta-Glycerophosphoric acid	171.006	0.799	Lipid and lipid-like molecule	1.727	0.012	709.246	127.420
Secologanin	369.120	0.837	Lipid and lipid-like molecule	1.032	0.006	296.047	94.041
(R)-3-hydroxybutyrylcarnitin	248.150	1.757	Lipid and lipid-like molecule	8.743	0.029	24658.606	8415.176
O-hexanoyl-R-carnitine	260.185	5.204	Lipid and lipid-like molecule	1.159	0.028	423.587	159.349
3-hydroxyisovalerylcarnitine	244.154	4.613	Lipid and lipid-like molecule	1.071	0.025	410.742	169.769
Butyrylcarnitine	232.155	4.267	Lipid and lipid-like molecule	6.272	0.028	15128.943	6916.603
P-IsoPGE2-PC	830.555	13.639	Lipid and lipid-like molecule	1.771	0.044	1334.556	622.430
Propionylcarnitine	218.138	3.064	Lipid and lipid-like molecule	4.230	0.046	10047.609	6064.436
2,4-Decadienoyl-CoA	898.205	1.259	Lipid and lipid-like molecule	2.394	0.015	3585.727	2616.954
Stoloniferone K	447.345	14.884	Lipid and lipid-like molecule	2.017	0.015	2921.704	2183.036
Trisnor-17-methyl-1alpha,25-dihydroxyvitamin D3	396.346	12.590	Lipid and lipid-like molecule	1.196	0.011	971.983	727.211
Unanisoflavan	388.211	11.424	Lipid and lipid-like molecule	2.174	0.033	4228.737	3323.230
DG (O-16:0/18:1(9Z))	598.576	15.416	Lipid and lipid-like molecule	3.591	0.000	9377.466	7533.455
PC (18:2(9Z,12Z)/0:0)	564.332	10.711	Lipid and lipid-like molecule	4.224	0.008	17105.928	13771.380
LysoPC (18:1(11Z))	522.355	11.176	Lipid and lipid-like molecule	9.047	0.032	58154.347	75245.103
PC (17:1(9Z)/0:0)	508.339	10.812	Lipid and lipid-like molecule	1.647	0.005	1327.761	1794.105
LysoPE (7Z,10Z,13Z,16Z,19Z	528.307	10.871	Lipid and lipid-like molecule	1.886	0.042	1337.314	2067.043
PC (16:1(9Z)/0:0)	494.323	10.444	Lipid and lipid-like molecule	2.024	0.001	1091.156	1724.119
LysoPC (18:3(9Z,12Z,15Z))	518.323	10.312	Lipid and lipid-like molecule	1.848	0.011	923.598	1534.474
Araliacerebroside	732.564	14.299	Lipid and lipid-like molecule	13.971	0.043	51913.400	92417.742
LysoPE (0:0/22:5(4Z,7Z,10Z,13Z,16Z))	526.295	10.866	Lipid and lipid-like molecule	1.209	0.036	377.072	677.989
2,4-Dimethylpimelic acid	187.098	5.997	Lipid and lipid-like molecule	1.196	0.021	324.421	584.987
8-aminocaprylic acid	160.133	0.979	Lipid and lipid-like molecule	4.211	0.024	4336.706	7886.702

Dihydrozeatin-O-glucoside	401.213	3.674	Lipid and lipid-like molecule	1.024	0.006	156.504	341.718
alpha,alpha'-Trehalose 6-mycolate	836.613	14.025	Lipid and lipid-like molecule	5.385	0.007	4071.629	9153.620
Schidigeragenin B	446.325	10.458	Lipid and lipid-like molecule	1.020	0.021	157.852	356.265
(+)-Marmasmic acid	280.155	5.044	Lipid and lipid-like molecule	1.066	0.048	195.062	449.880
LysoPE (0:0/20:5(5Z,8Z,11Z,14Z,17Z))	498.263	10.322	Lipid and lipid-like molecule	1.493	0.002	185.643	549.897
LysoPE (0:0/18:2(9Z,12Z))	500.276	10.326	Lipid and lipid-like molecule	2.064	0.000	291.410	927.178
8 (Aminomethyl)sulfanyl]-6-sulfanyloctanoic acid	282.084	2.789	Lipid and lipid-like molecule	1.540	0.004	93.094	477.770
5,6-Dihydroxyprostaglandin F1a	406.279	11.673	Lipid and lipid-like molecule	1.250	0.018	66.143	365.731
PS (16:0/22:2(13Z,16Z)	838.558	10.665	Lipid and lipid-like molecule	1.390	0.000	0.770	258.134
Uridine diphosphate-N-acetylglucosamine	606.074	0.902	Nucleoside, nucleotide and analogue	1.052	0.014	584.310	373.114
ADP	426.022	0.953	Nucleoside, nucleotide, and analogue	2.907	0.046	5919.776	4136.277
8-Oxo-dGMP	362.050	1.259	Nucleoside, nucleotide, and analogue	1.019	0.026	232.991	444.061
N1-(5-Phospho-a-D-ribosyl)-5,6-dimethylbenzimidazole	357.089	4.024	Nucleoside, nucleotide, and analogue	1.059	0.003	32.291	216.430
Methionyl-asparagine	286.083	5.412	Organic acid and derivative	1.048	0.017	210.263	1.984
Tryptophyl-histidine	380.111	3.035	Organic acid and derivative	1.393	0.049	432.070	32.827
Tolmetin	240.101	3.950	Organic acid and derivative	1.779	0.001	546.481	64.934
Ophthalmic acid	288.120	1.586	Organic acid and derivative	2.130	0.004	939.957	174.351
Lenticin	247.144	4.684	Organic acid and derivative	1.985	0.002	1056.088	382.504
O-Acetylserine	146.047	0.825	Organic acid and derivative	1.613	0.003	656.235	242.243
Alanylglycine	129.065	0.878	Organic acid and derivative	1.185	0.008	441.163	178.122
L-Glutamine	147.076	0.806	Organic acid and derivative	3.789	0.000	3603.614	1465.709
L-Glutamic acid	148.060	0.836	Organic acid and derivative	1.320	0.004	490.482	205.657
D-Glutamine	145.062	0.799	Organic acid and derivative	1.910	0.001	988.176	415.141
Pyrrolidonecarboxylic acid	130.050	0.806	Organic acid and derivative	4.421	0.000	5327.322	2392.164
L-Histidine	156.076	0.777	Organic acid and derivative	1.671	0.030	1882.797	2458.128
L-Tyrosine	180.067	2.171	Organic acid and derivative	1.059	0.042	744.654	998.313
Anserine	239.115	0.761	Organic acid and derivative	2.749	0.022	2788.898	4249.825
L-Phenylalanine	164.072	3.626	Organic acid and derivative	1.584	0.001	225.172	599.682
Cefpiramide	595.118	0.759	Organic acid and derivative	1.575	0.003	146.491	554.710
N-Acetylneuraminate 9-phosphate	407.105	0.806	Organic oxygen compound	1.046	0.002	187.442	13.509
N-Acetyl-D-mannosamine 6-phosphate	346.054	1.259	Organic oxygen compound	1.123	0.001	257.054	50.107
D-Myoinositol 4-phosphate	259.023	0.786	Organic oxygen compound	4.638	0.019	5119.464	1003.859
Aminophenazone	270.101	0.864	Organoheterocyclic compound	1.122	0.016	323.442	72.193
Fructose 6-phosphate	298.992	0.792	Organic oxygen compound	1.445	0.032	582.219	142.091
Methacholine	160.133	1.265	Organic nitrogen compound	4.309	0.030	3396.854	7234.831
4(Hydroxymethyl)nitrosoamino]-1-(3-pyridinyl)-1-butanone	241.130	0.777	Organic oxygen compound	7.326	0.025	14209.920	25109.777
Indoleacrylic acid	188.070	4.684	Organoheterocyclic compound	1.360	0.003	518.575	200.188
5-Butyltetrahydro-2-oxo-3-furancarboxylic acid	204.122	4.497	Organoheterocyclic compound	1.085	0.021	532.164	320.795

Topiramate	384.099	2.686	Organoheterocyclic compound	1.127	0.031	254.547	513.773
2-Hydroxycinnamic acid	182.081	2.148	Phenylpropanoids and polyketide	1.386	0.027	875.814	1273.596
Epigallocatechin	611.146	2.482	Phenylpropanoids and polyketide	3.669	0.026	2109.490	4847.781
Citreovirenone	266.138	4.873	Benzenoid	1.079	0.019	393.543	176.700
Emodinanthranol	551.113	0.759	Benzenoid	1.448	0.007	517.110	889.419
Nicotinamide adenine dinucleotide (NAD)	664.117	1.280	Unclassified	2.733	0.002	306.310	1463.237

m/z: mass to charge ratio; RT: retention time; JG: Jining Gray; and LW: Lubei White.

metabolic pathways play vital roles in meat quality characteristics (Cheng *et al*., 2019; Cui *et al*., 2020; Tang *et al*., 2021).

The specific metabolites in these pathways play a key role in the nutritional and biological value of the meat. The nutritional and biological value of meat is dependent on the composition of amino acids such as lysine, histidine, methionine, tryptophan, and phenylalanine, which not only affect protein content, but also the biological significance of the meat. These amino acids are necessary for normal growth and development, and regulate the function of the central nervous system (Brzostowski *et al*., 2008; Migdał *et al*., 2021). In the present work, the mean histidine and phenylalanine concentrations were high in the mutton sample of Lubei White goat breed compared to those of Jining Gray goat breed. In terms of the mutton nutritional quality of the two goat breeds, the comparative evaluation of the metabolites exhibited 17 organic acids and derivatives. The content of asparagine in the longissimus dorsi muscle of Jining Gray goats was significantly higher than in that of Lubei White goats. Previously, the role of asparagine in maintaining proper function of the central nervous system was reported. It also helps to resist physical fatigue, which has a certain positive effect on improving human health. The content of tryptophyl-histidine in the mutton of Jining Gray goats was significantly (p = 0.049) higher than that of Lubei White goats. Tryptophyl-histidine plays a role in preventing vascular injury, cell proliferation, and atherosclerosis, which may be related to the inhibition of the vasoconstriction signalling cascade (Kobayashi *et al*., 2015; Luo *et al*., 2019). The L-glutamine (p < 0.0001) and D-glutamine (p = 0.001) levels were higher in the mutton of Jining Gray goat than in Lubei White goats. Glutamine is one of the essential amino acids needed for different disease conditions. Moreover, it helps in protein and carbohydrate metabolism, and regulates the growth of lymphocytes and fibroblasts (Tapiero *et al*., 2002; Kim, 2011). Similarly, histidine is an essential amino acid that reduces oxidative stress through the regulation of the antioxidant enzyme system, and also regulates RBC production (Holeček, 2020). Phenylalanine (Phe) is an essential amino acid that regulates the endocrine system and hormone synthesis. It also helps in protein biosynthesis (Behiry *et al*., 2018). The aromatic amino acid L-tyrosine is used as a dietary supplement, and a conditionally essential amino acid for the human body (Lütke-Eversloh *et al*., 2007). These are essential amino acids, which means that they cannot be produced within the body, and must be obtained from the diet. Obtaining essential amino acids for life activities is the survival of all mammals, and for the performance of essential metabolic processes (Adams *et al*., 2019; Ennis *et al*., 2020). The concentrations of essential amino acids such as histidine, phenylalanine, and L-tyrosine were high in the mutton of Lubei White goat compared to those in Jining Gray goat. Therefore, regular consumption of mutton of Lubei White goat can supplement the levels of histidine, phenylalanine, and L-tyrosine in the body up to a certain extent.

Among the metabolites, the content of hydroxycinnamic acid in Lubei White goat was relatively high. Studies have shown that hydroxycinnamic acid exert anti-tumour effects through various mechanisms, including reducing MAPK (mitogen-activated protein kinase) signalling, inhibiting NF-κB (nuclear transcription factor) activation, inhibiting nitric oxide synthase, and scavenging oxygen free radicals, *etc*., thus affecting tumour cell apoptosis, invasion, and metastasis (Mancuso and Santangelo, 2014; Adams *et al*., 2019). In the present work, the mutton of Lubei White goat exhibited a significantly high level of nicotinamide concentration compared to that of Jining Gray goat.

Nicotinamide is an important part of nicotinamide adenine dinucleotide and nicotinamide adenine dinucleotide phosphate. It is often used as a coenzyme of dehydrogenase, and participates in the body's metabolic activities. Niacinamide not only has antioxidant and anti-inflammatory biological functions, but also plays an important role in nutritional regulation. For example, nicotinamide enhances lipid synthesis in adipose tissue and cells, and reduces lipolysis (Bäckesjö *et al*., 2009). The concentration of indole acrylic acid in the mutton of Jining Gray goat was also high compared to that in the mutton of Lubei White goat. Indoleacrylic acid has certain benefits to human physical health, such as enhancing the function of the intestinal epithelial barrier, and mitigating inflammatory responses by immune cells (Wlodarska *et al*., 2017).

Conclusion

The metabolite classes within the muttons of two goat breeds were lipid and lipid-like molecules, organic acid and derivatives, organoheterocyclic compounds, benzenoids, nucleosides, nucleotides, and analogues, phenylpropanoids and polyketides, and organic nitrogen compounds. Moreover, the contents of methionyl-asparagine, hydrocarbons, tryptophyl-histidine, L-glutamine, and indole acrylic acid in the longissimus dorsi muscle of Jining Gray mutton were significantly higher than that of Lubei White. Similarly, the concentrations of essential amino acids such as histidine, phenylalanine, and L-tyrosine were significantly higher in the mutton of Lubei White compared to that of Jining Gray. Additionally, the contents of hydroxycinnamic acid and nicotinamide in Lubei White goat were also significantly higher than in Jining Gray goat meat. The findings of the present work will deepen our understanding regarding metabolomics variation in the meat of the two goat breeds, and will help in the goat breed improvement program for meat quality traits.

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