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Original Research Article

Dietary inositol supplementation improves meat quality by modulating amino acid metabolism and gut microbiota composition of finishing pigs

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ABSTRACT

Intramuscular fat (IMF) content influences various meat quality traits, including tenderness, flavor, juiciness and nutritional value. This study aimed to investigate the effects of dietary inositol supplementation on meat quality, metabolic profiles, and gut microbiota composition of finishing pigs. A total of 144 finishing pigs (initial body weight 70.41 ± 0.78 kg) were randomly divided into control, 0.075%, 0.15%, and 0.3% inositol groups. The data showed that inositol increased backfat thickness at the 6th to 7th rib and 10th rib, IMF content, and improved tenderness ($P \leq 0.05$, $n = 8$). Paralleling an increase in fat deposition, 0.3% inositol also increased the protein level of PPARγ in the subcutaneous fat ($P \leq 0.05$) and longissimus thoracis (LT) muscle ($P = 0.062$). Inositol elevated the content of amino acids in LT muscle and enhanced amino acid metabolism of finishing pigs, including lysine degradation, tyrosine metabolism, and arginine and proline metabolism. The 16S ribosomal RNA (rRNA) sequencing showed that 0.3% inositol supplementation altered the profiles of microbes in the colon, particularly decreasing the abundance of Firmicutes ($P < 0.01$) and increasing the abundance of Bacteroidota ($P \leq 0.05$). Correlation analysis showed that differential microbes had strong correlation with differential metabolites in serum, including amino acids. In conclusion, this study demonstrated that dietary inositol supplementation could effectively improve IMF content and tenderness of pork, enhance amino acid metabolism, and regulate gut microbiota composition of finishing pigs.

1. Introduction

In recent years, with the rising demand for pork worldwide, meat quality has been playing a crucial part in influencing consumer choice and meat processing. Meat quality is complex and is affected by various physicochemical properties, such as meat color, pH, composition of fatty acids, drip loss, sensory quality, tenderness and intramuscular fat (IMF) (Matarneh et al., 2001). IMF content influences various aspects of meat quality, including tenderness, flavor, juiciness and nutritional value (Wood et al., 2008). Nevertheless, IMF content has dramatically declined over the past 40 years with the progress made in animal nutrition, genetics, management, as well as changes in processing technology (Scollan et al., 2017). Our previous study evaluated the pork quality in China from 2021 to 2022 and found that IMF content had a large coefficient of variation (42.9%) (Wang et al., 2023). Therefore, increasing IMF content is an effective avenue for improving meat quality.

Fat deposition in pigs is regulated by genetic, environmental, and nutritional factors. For example, Taoyuan black pigs, a Chinese indigenous pig breed, have lower carcass weight and lean percentage but have higher IMF content compared with the Duroc pigs (Song et al., 2023). Similarly, compared with Large White pigs, pH

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values, redness and IMF content of Qinling Black pigs were obviously outperformed (Yu et al., 2023). The wingless/integrated (Wnt) signaling pathway, phosphatidylinositol 3-kinase (PI3K)-protein kinase B (Akt) signaling pathway, Ras-associated protein 1 (Rap1) signaling pathway, and Ras signaling pathway were connected with the meat quality traits of these two pig breeds (Yu et al., 2023). Impacts of dietary fatty acids and amino acids on fat deposition in skeletal muscle have also been reviewed extensively in our previous work (Yan et al., 2023). Importantly, given the tight relationship between host metabolism and gut microbiota, the contribution of gut microbiota in regulating fat deposition of pigs cannot be overlooked. The gut microbiota of obese pigs could promote IMF accumulation in lean commercial pigs and antibiotic-treated mice through increasing the expression of lipogenesis-associated genes and decreasing the production of short-chain fatty acids (Wu et al., 2021; Xie et al., 2021).

Phosphoinositides, phosphorylated forms of phosphatidylinositol (PI), play essential roles in cellular signaling, lipid transport, and human diseases (Hammond and Burke, 2020). A previous work also revealed the contribution of faster PI turnover to hepatic triglyceride accumulation by supplying diacylalycerol (Tanaka et al., 2021). Inositol is a precursor for the production of phosphoinositides and PI. Hsu and co-workers demonstrated that inositol could restrict adenosine 5'-monophosphate (AMP)-activated protein kinase (AMPK) activation through competing with AMP for AMPKγ binding (Hsu et al., 2021). Inositol is also the precursor of inositol (1,4,5) triphosphates (IP3), and our study found that the deletion of adipocyte IP3 receptor 1 (IP3R1) could enhance lipolysis and the AMPK signaling pathway in the visceral fat (Zhang et al., 2023). As briefly mentioned above, inositol may be implicated in fat deposition.

Previous work in weaning piglets showed that dietary supplementation of inositol increased the average daily gain (Moran et al., 2019). Its beneficial effect on gut barrier integrity was also observed in intestinal porcine epithelial cells (Ogunribido et al., 2022). However, the effect of inositol on pork quality, especially IMF accumulation, is less well explored. Therefore, this research aimed to assess the effect of inositol on meat quality in finishing pigs. Metabolomics and gut microbiota composition analysis were performed to explore the underlying mechanisms. This study provides the first evidence that inositol is a promising candidate for promoting fat deposition and improving pork quality by modulating the metabolism of amino acid and gut microbiota.

2. Materials and methods

2.1. Ethics statement

All procedures were approved by the Institutional Animal Care and Use Committee of China Agricultural University (approval number: SKLAB-2011–04-03).

2.2. Experimental design and sample collection

A total of 144 Duroc × Landrace × Yorkshire crossbred castrated male finishing pigs (70.41 ± 0.78 kg) were divided randomly into four groups on the basis of the initial body weight (BW). The basal diet was formulated to satisfy the nutritional demands concerning finishing pigs (75 to 100 kg) based on the National Research Council (NRC, 2012). The four dietary treatments consisted of a basal diet (control) and basal diet supplemented with 0.075%, 0.15%, or 0.3% inositol, respectively. Inositol was purchased from Jilin Fuli Biotechnology Development Co., Ltd., China, and purity exceeded 98%. The composition and nutrient levels of the basal diet are shown in Table 1. Crude protein of basal diets was analyzed according the procedures of AOAC (AOAC, 2006; method 984.13). The contents of amino acids in basal diet were measured as described previously (Heinrikson and Meredith, 1984). Levels of digestible energy and metabolizable energy were calculated according to NRC (2012). The standardized ileal digestible (SID) amino acid content was calculated by multiplying the SID coefficients provided by NRC (2012) by the amino acid content of feed ingredients. All pigs had free access to feed and clean drinking water. The experiment lasted 49 days. At the beginning and end of the experiment, the BW of all 144 pigs was measured, and the growth performance was analyzed using each pig ($n = 36$).

Table 1
Ingredients and nutrient levels of basal diets (%, as fed basis).

Ingredients	Content	Nutrient levels	Content
Corn	81.00	Crude protein	12.29
Soybean meal	11.50	Lysine	0.83
Wheat bran	3.20	Methionine + Cysteine	0.42
Soybean oil	1.20	Threonine	0.51
Limestone	0.70	Tryptophan	0.14
Dicalcium phosphate	0.90	Isoleucine	0.43
Salt	0.34	Leucine	1.10
L-Lysine • HCl	0.39	Valine	0.55
L-Threonine	0.11	Calculated nutrient levels	
L-Tryptophan	0.03	Digestible energy, MJ/kg	14.35
L-Valine	0.02	Metabolizable energy, MJ/kg	13.99
DL-Methionine	0.03	Standardized ileal digestible amino acids	
50% Choline chloride	0.08	Lysine	0.84
Premix [1]	0.50	Methionine + cysteine	0.42
Total	100.00	Threonine	0.46
		Leucine	1.03
		Valine	0.48

[1] The premix provided the following per kilogram of diets: vitamin A, 6000 IU; vitamin D_3, 2400 IU; vitamin E, 20 IU; vitamin K_3, 2 mg; vitamin B_1, 0.96 mg; vitamin B_2, 4 mg; vitamin B_6, 2 mg; vitamin B_{12}, 0.012 mg; biotin, 0.04 mg; folic acid 0.40 mg; pantothenic acid 11.2 mg; nicotinic acid 22 mg; Cu, 120 mg; Fe, 76 mg; Mn, 12 mg; Zn, 76 mg; I, 0.24 mg; Se, 0.40 mg.

At the end of the experiment, eight pigs with the average BW of each group were picked up ($n = 8$). The slaughter weight of finishing pigs was approximately 115 kg. Blood samples were collected from the precaval vein after overnight starvation of 16 h, centrifuged at 3000 × g for 15 min at 4 °C to obtain serum, and stored at −80 °C for further analysis. After the collection of blood samples, pigs were transported to a local abattoir, rested for at least 4 h, slaughtered by electrical stunning, exsanguinated, and eviscerated. Then, to evaluate the meat quality, about 100 g longissimus thoracis (LT) muscle was isolated from the carcass on the left side between the 10th and 12th ribs and stored at 4 °C. A piece of LT muscle (approximately 100 g) was also collected and kept at −80 °C for protein extraction and the measurement of IMF content, and the composition of amino acids and fatty acids. An approximate 5 g backfat sample was isolated at the 10th rib and kept at −80 °C for protein extraction. In addition, approximately 2 g fresh digesta was rapidly collected from the distal colon in a 2-mL sterile centrifuge tube and stored at −80 °C for analysis of the gut microbiota.

2.3. Carcass traits

At the slaughter line, the carcass weight of each pig was tracked, and the dressing percentage was represented by the ratio of carcass weight to live BW. Backfat depth values opposite to the thickest shoulder, the last rib, the last lumbar vertebra, the 6th to 7th rib, and 10th rib, were recorded. The average backfat depth was expressed as the mean of shoulder backfat, the last rib backfat, and lumbosacral backfat. Moreover, the height and width of the lion eye at 10th rib were measured. The lion eye area and fat-free lean index were calculated as the following (NRC, 1998):

Lion eye area (cm^2) = lion eye height (cm) × width (cm) × 0.7;

Fat-free lean index = 50.767 + [0.035 × hot carcass weight (lb)] - [8.979 × last rib fat depth (in)].

2.4. Meat quality

Fresh LT muscle samples were used for meat quality evaluation. To assess the flesh color score and marbling score of fresh meat, standards by National Pork Producer Council (NPPC) were used. In terms of the score for meat color, 6.0 means dark purplish red and 1.0 means very pale, white. At 45 min and 24 h postmortem, pH values of LT muscle were measured through a SPK pH meter (pH-star, DK2730, Herlev, Denmark). The meat color parameters, such as lightness (L*), yellowness (b*) and redness (a*), were also detected at 45 min and 24 h after slaughter using a tristimulus colorimeter (Minolta Chroma Meter Measuring Head CR-410 Minolta, Osaka, Japan) according to the standard method of CIE Lab system. The colorimeter was calibrated on a white tile in accordance with the manufacturer's instructions.

Drip loss was measured at 4 °C for 24 h and calculated using the following equation: drip loss (%) = [(initial weight − final weight)/initial weight] × 100. The muscle samples were then weighed and cooked in a water bath at 70 °C for 30 min in one cooking batch. After cooling to room temperature, the weight of muscle samples was measured again and the cooking loss was determined by calculating the percentage weight change. Shear force was then tested using a digital display muscle tenderness meter (C-LM3B, Tenovo, Harbin, China) as described previously (Luo et al., 2018). For each muscle sample, measurements were repeated at least ten times.

2.5. Intramuscular fat content measurement

About 10 g of each LT muscle sample was sliced up and weighed in aluminum boxes, then put into a vacuum frozen dryer (Freezone 4.5™, Labconco Corp., Kansas City, MO, USA) for 48 h and weighted again. The difference between the initial weight and the dried muscle sample weight was the percentage of moisture. Freeze-dried muscle was ground into powder. IMF was extracted with petroleum ether using a Soxhlet petroleum-ether extraction apparatus (Budwi Extraction System B-11; Budwi, Lausanne, Switzerland), and its content was converted to percentage of fresh meat weight.

2.6. Texture characteristics

LT samples were cooked in a water bath at 70 °C for 30 min, and then left at room temperature for 2 h. The samples were cut into uniform squares of approximately 1 cm^3. Texture parameters including hardness, adhesiveness, cohesiveness, springiness, gumminess, and chewiness were measured using a Texture Analyzer (TMS-Touch, Food Technology Corp., USA) with a probe P 0.5. Parameters were consistent with our previous study (Zhang et al., 2022).

2.7. Amino acid composition

About 200 mg freeze-dried muscle samples and 50 μL serum samples were used to identify the amino acid composition according to the standard methods in AOAC (2008). Briefly, after hydrolysis with 6 mol/L HCl at 110 °C for 24 h, the concentrations of amino acids were determined by an amino acid analyzer except tryptophan methionine, and cysteine (Hitachi L-8900, Tokyo, Japan). The concentrations of methionine and cysteine were determined using an amino acid analyzer (Hitachi L-8900, Tokyo, Japan), followed by acid oxidation and hydrolysis with 7.5 mol/L HCl at 110 °C for 24 h. Tryptophan was determined by high performance liquid chromatography (Agilent 1200 Series, Santa Clara, CA, USA) after alkaline hydrolysis (LiOH) at 110 °C for 22 h.

2.8. Fatty acid composition

Targeted metabolomics was conducted to analyze the fatty acid composition in meat. About 50 mg muscle samples were weighted and homogenized with 1 mL of dichloromethane:methanol (1:1, v:v) which contained mixed internal standards in 2 mL tubes. After centrifugation at 13,000 × *g* for 10 min, the supernatant was dried with nitrogen and dissolved with 0.5 mL sodium hydroxide methanol solution (0.5 mol/L). One hundred microliters of supernatant was put into a sample bottle for gas chromatography (GC)-mass spectrometry (MS) analysis. The GC–MS analysis was performed using an Agilent 8890B gas chromatography coupled to an Agilent 5977B/7000D mass selective detector with an inert electron impact (EI) ionization source and ionization voltage was 70 eV (Agilent, USA). Metabolites in samples were identified and quantified by Mass hunter (v10.0.707.0, Agilent, USA). A quality control sample was used to evaluate the stability of the analytical system.

2.9. Western blotting

The extraction of proteins in LT muscle and backfat samples were performed using RIPA lysis buffer (Huaxingbio, Beijing, China) with a protease inhibitor cocktail and a phosphatase inhibitor cocktail (Huaxingbio, Beijing, China). A BCA protein assay kit (HX18651, Huaxingbio, Beijing, China) was used to determine the protein content. After separation by 12% SDS-PAGE electrophoresis, the proteins were transferred to a polyvinylidene difluoride membrane. The membrane was blocked and incubated with the corresponding primary antibodies overnight. The primary antibodies were anti-PPARγ (Cell Signaling Technology, 95128S), anti-FABP4 (abcam, ab92501), and β-Tubulin (Cell Signaling Technology, 2146S). After incubating with DyLight 800-labelled secondary antibodies and detection with Odyssey Clx (LiCor Biosciences, USA), gray analysis of protein bands was conducted using ImageJ (National Institutes of Health, Bethesda, USA) software. Beta-Tubulin protein was used as an internal control.

2.10. Untargeted metabolomics analysis

Samples of serum were used for untargeted metabolomics analysis. Approximately 100 μL serum samples were used and eight replicates were prepared for each group. Metabolites were extracted using 400 μL of methanol:water (4:1, vol:vol). L-2-chlorophenylalanine was used as an internal standard. After centrifugation at 13,000 × *g* for 15 min at 4 °C, the supernatant was transferred to sample vials for next analysis.

A Thermo UHPLC-Q Exactive HF-X system equipped with an ACQUITY HSS T3 column (2.1 mm × 100 mm, 1.8 μm) was used to conduct the LC-MS/MS analysis of sample at 40 °C with a flow rate of 0.4 mL/min. A Q Exactive HF-X Mass Spectrometer with an electrospray ionization source was used. The following conditions were used: heater temperature, 425 °C; capillary temperature, 325 °C; sheath gas flow rate, 50 arb; aux gas flow rate, 13 arb; spray voltage floating, −3500 and 3500 V in negative and positive mode, respectively. The full MS resolution was 60,000, and the MS/MS resolution was 7500. The mode of Data Dependent Acquisition was applied for data collection. The detected mass range was 70 to 1050 *m/z*.

After data preprocessing and annotation by Human Metabolome Database (HMDB, http://www.hmdb.ca/) and Metlin

(https://metlin.scripps.edu/), the obtained data were subjected to principal component analysis (PCA) and partial least squares discriminant analysis (PLS-DA). The criteria of variable importance projection (VIP) > 1.0 and $P < 0.05$ was considered significantly different. Further, metabolic pathway analysis was performed according to the Kyoto Encyclopedia of Genes and Genomes (KEGG, http://www.genome.jp/kegg/) to reveal the biological importance of the metabolites.

2.11. DNA extraction and 16S rRNA sequencing

Microbial genomic DNA was extracted from colonic digesta using the Mag-Bind® Soil DNA kit (M5636, Omega, Norcross, USA). After the determination of the quality and concentration of DNA, PCR amplification was performed by primer pairs (forward: 5'-ACTCCTACGGGGAGGCAGCAG-3', reverse: 5'-GGACTACHVGGTWTCTAAT-3') using TransStart Fastpfu DNA polymerase (TransGen Biotech, Beijing, China). The PCR products (approximately 500 bp) were extracted and purified. Purified amplicons were subjected to paired-end sequencing on an Illumina MiSeq PE300/NovaSeq PE250 platform (Illumina, San Diego, USA) according to the procedures of Majorbio Bio-Pharm Technology Co. Ltd. (Shanghai, China).

All raw sequences were filtered, denoised, merged, and non-chimeric removed by the DADA2 plug-in of the QIIME2 software (https://qiime2.org/) to form operational taxonomic units (OTUs). The representative sequences of OTUs were compared with the Silva Release 138 database for species annotation information. At each taxonomic level, α-diversity index, β-diversity index, and species abundances were analyzed. Various α-diversity indexes, including Shannon and Simpson index, Chao1 richness estimator, and abundance-based coverage estimator (ACE) metric were calculated using QIIME (version 1.9.1). Bacterial taxa (phylum to genus) that were significantly abundant in different groups were identified by linear discriminant analysis (LDA) effect sizes (http://huttenhower.sph.harvard.edu/LEfSe) (LDA scores >2, $P < 0.05$).

2.12. Statistical analysis

Data are expressed as means and SEM and statistically analyzed using the mixed linear model in SAS (v.9.2, SAS Institute, USA) as follows:

$$Y_{ijk} = \mu + T_i + P_j + S_k + e_{ijk},$$

where Y_{ijk} is the dependent variable; μ is the overall mean; T_i is the fixed treatment effect; P_j is the period effect; S_k is the steer effect; e_{ijk} is random error.

Unpaired two-tailed Student's *t*-test or one-way ANOVA procedures of SAS were used when comparing differences between two or four groups, respectively. Linear and quadratic contrasts were analyzed using regression analysis procedure of SAS to assess the effect of expository doses. Spearman's correlation was used to assess the relationship between differential microbiota and differential metabolites. $P \leq 0.05$ was considered significant, and $0.05 < P \leq 0.10$ was considered to have a trend.

3. Results

3.1. Growth performance, carcass traits, and meat quality

The growth performance of finishing pigs is shown in Table 2. There were no differences in initial BW and final BW. The average daily gain (ADG) linearly decreased with inositol supplementation ($P = 0.033$, Table 2). Furthermore, dietary inositol supplementation linearly increased the backfat thickness at the 6th to 7th rib and 10th rib ($P \leq 0.05$, Table 3). Inositol administration also tended to decrease fat-free lean index in a dose-dependent manner ($P = 0.091$, Table 3).

The pork quality traits are shown in Table 4. Dietary inositol supplementation markedly decreased shear force and increased IMF content in a dose-dependent manner ($P \leq 0.05$). Cooking loss was also markedly decreased by 0.075% inositol compared to the control ($P \leq 0.05$). There were no significant differences in other meat characteristics, including the pH_{45min}, pH_{24h}, meat color (flesh color score, L*, a*, and b*), marbling score, and drip loss. To further

Table 2
Effects of dietary inositol supplementation on growth performance of finishing pigs ($n = 36$).

Items	Inositol levels, %				SEM	*P*-value		
	0	0.075	0.15	0.3		ANOVA	Linear	Quadratic
Initial body weight, kg	70.43	70.34	70.42	70.46	0.782	0.992	0.958	0.950
Final body weight, kg	109.21	110.71	109.29	107.53	1.347	0.414	0.218	0.374
Average daily gain, g/d	812	815	807	762	18.6	0.140	0.033	0.342
Average daily intake, kg/d	2.97	3.01	2.93	2.97	–	–	–	–

Table 3
Effects of dietary inositol supplementation on carcass traits of finishing pigs ($n = 8$).

Items	Inositol levels, %				SEM	*P*-value		
	0	0.075	0.15	0.3		ANOVA	Linear	Quadratic
Live body weight, kg	116.64[a]	115.95[a]	116.99[a]	112.09[b]	1.316	0.050	0.023	0.118
Hot carcass weight, kg	82.31	82.32	83.79	80.11	0.963	0.082	0.117	0.073
Dressing percentage, %	70.64	70.97	71.60	71.49	0.453	0.424	0.168	0.413
Back fat depth, mm								
Shoulder fat thickness	37.20	36.68	40.73	39.02	1.687	0.334	0.287	0.418
The 6th to 7th rib fat thickness	25.14[ab]	24.87[b]	28.03[ab]	28.32[a]	0.861	0.013	<0.001	0.567
The 10th rib fat thickness	22.09	23.44	24.73	25.81	1.121	0.126	0.024	0.568
The last rib fat thickness	22.52	20.32	23.15	23.51	1.056	0.173	0.214	0.515
Lumbosacral fat thickness	16.99	14.95	16.91	16.40	1.013	0.460	0.945	0.649
Average backfat depth	25.40	24.12	27.02	26.41	0.887	0.141	0.180	0.800
Loin eye area, cm^2	40.27	42.88	43.54	37.97	1.724	0.105	0.232	0.033
Fat-free lean index, %	49.15	49.96	49.08	48.63	0.342	0.067	0.091	0.224

[a,b] Within a row, means without a common superscript differ at $P \leq 0.05$.

Table 4
Effects of dietary inositol supplementation on meat quality of finishing pigs ($n = 8$).

Items	Inositol levels, %				SEM	*P*-value		
	0	0.075	0.15	0.3		ANOVA	Linear	Quadratic
$pH_{45\ min}$	6.22	6.07	6.15	6.03	0.063	0.162	0.078	0.771
$pH_{24\ h}$	5.64	5.69	5.66	5.65	0.021	0.349	0.903	0.214
Flesh color score	2.21	2.08	2.14	2.14	0.068	0.600	0.646	0.383
Marbling score	2.06	2.16	2.32	2.12	0.113	0.372	0.685	0.107
Meat color, 45 min								
L*	42.96	44.45	43.96	45.16	0.741	0.232	0.081	0.754
a*	5.01	5.19	5.48	5.63	0.279	0.406	0.113	0.704
b*	3.85	4.13	3.86	4.32	0.193	0.280	0.145	0.638
Meat color, 24 h								
L*	48.79	48.22	49.76	48.98	0.702	0.482	0.600	0.612
a*	6.17	5.98	5.71	6.47	0.363	0.503	0.508	0.192
b*	6.21	5.34	6.16	6.47	0.308	0.091	0.188	0.204
Drip loss, %	1.71	1.37	1.41	1.61	0.188	0.543	0.949	0.173
Cooking loss, %	29.44^{a}	25.36^{b}	27.31^{ab}	26.92^{ab}	0.834	0.021	0.217	0.058
Shear force, N	74.81^{ab}	69.63^{bc}	81.99^{a}	62.66^{c}	3.533	0.006	0.043	0.046
IMF content, %	1.74^{b}	2.45^{a}	2.12^{ab}	2.45^{a}	0.165	0.017	0.030	0.268

IMF = intramuscular fat.
[a,b] Within a row, means without a common superscript differ at $P \leq 0.05$.

verify the fat deposition, PPARγ and FABP4 protein levels were measured in the LT muscle and backfat. Supplementation with 0.3% inositol tended to increase PPARγ level in the LT muscle ($P = 0.062$, Fig. 1A) and significantly increased PPARγ level in the backfat ($P \leq 0.05$, Fig. 1B). However, the FABP4 level was not changed.

3.2. Texture characteristics

The texture characteristics of pig meat are shown in Table 5. Inositol supplementation linearly decreased hardness ($P = 0.003$) and gumminess ($P = 0.014$) of meat, while 0.15% inositol significantly improved the springiness compared with the control group ($P \leq 0.05$). There were no significant differences in adhesiveness, cohesiveness, and chewiness.

3.3. The composition of amino acids and fatty acids in the muscle of finishing pigs

The amino acid profile of LT muscle is shown in Table 6. We found that inositol supplementation significantly increased the contents of methionine, valine, arginine, phenylalanine, tyrosine, and asparagine in a dose-dependent manner. Inositol also tended to increase the contents of threonine, histidine, and glycine in the LT muscle. Meanwhile, the total content of essential amino acids was significantly higher after inositol supplementation ($P \leq 0.05$).

Considering that inositol supplementation linearly increased IMF content, the fatty acid composition in the LT muscle was further detected in the control and 0.3% inositol group. In accordance with the increased IMF content, inositol also significantly increased C14:1

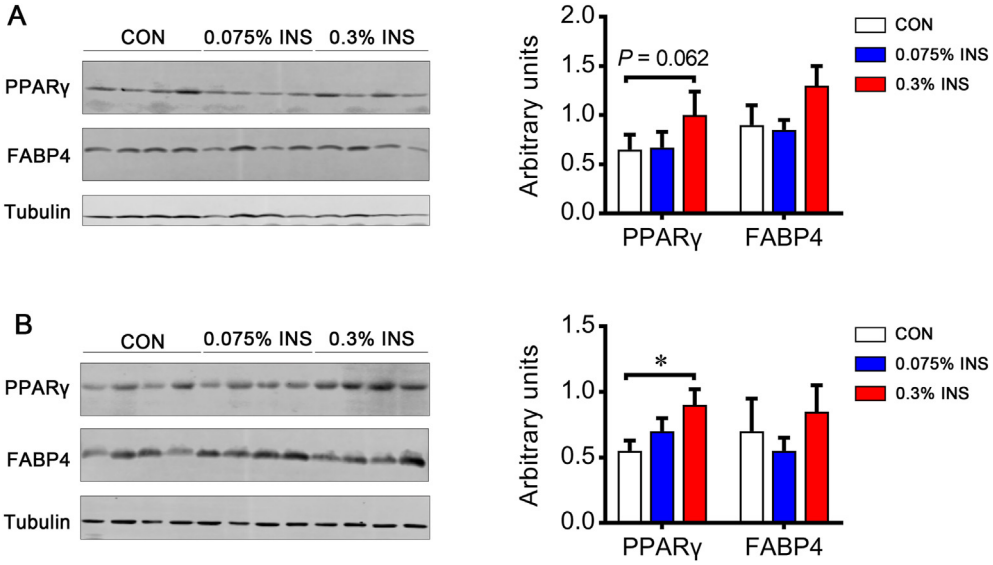

Fig. 1. Effects of dietary inositol (INS) supplementation on the expression of PPARγ and FABP4 in the skeletal muscle and subcutaneous fat of finishing pigs. The expression of PPARγ and FABP4 was detected by western blotting in longissimus thoracis muscle (A) and backfat at the 10th rib (B). Relative expression level was calculated. Beta-tubulin was used as internal control. Data are presented as means ± SEM ($n = 8$). Statistics were performed with Student's *t*-test. *$P \leq 0.05$. PPARγ = peroxisome proliferator-activated receptor γ; FABP4 = fatty acid binding protein 4.

Table 5
Effects of dietary inositol supplementation on texture characteristics of finishing pigs ($n = 8$).

Items	Inositol levels, %				SEM	*P*-value		
	0	0.075	0.15	0.3		ANOVA	Linear	Quadratic
Hardness, N	40.41[a]	42.77[a]	37.10[ab]	32.89[b]	1.707	0.006	0.003	0.343
Adhesiveness, mJ	0.16	0.18	0.14	0.12	0.021	0.182	0.087	0.503
Cohesiveness ratio[1]	0.45	0.45	0.45	0.47	0.008	0.586	0.352	0.358
Springiness, mm	3.53[b]	3.86[ab]	3.90[a]	3.56[ab]	0.114	0.048	0.793	0.005
Gumminess, N	18.37[ab]	19.20[a]	16.82[ab]	15.48[b]	0.943	0.032	0.014	0.648
Chewiness, mJ	65.78	73.70	65.11	54.99	5.204	0.117	0.064	0.230

[a,b] Within a row, means without a common superscript differ at $P \leq 0.05$.
[1] Cohesiveness is the relative resistance after the first compression deformation to that after the second compression deformation, and cohesiveness ratio is the ratio of the positive work done in the two compressions.

Table 6
Effects of dietary inositol supplementation on amino acid composition in longissimus thoracis muscle of finishing pigs ($n = 8$, mg/100 g fresh meat weight).

Items	Inositol levels, %				SEM	*P*-value		
	0	0.075	0.15	0.3		ANOVA	Linear	Quadratic
Lysine (Lys)	2.59[ab]	2.06[b]	3.37[a]	2.44[ab]	0.303	0.035	0.798	0.216
Methionine (Met)	0.50	0.46	0.59	0.62	0.048	0.079	0.034	0.979
Tryptophan (Trp)	1.53	1.54	1.91	1.71	0.128	0.155	0.218	0.192
Threonine (Thr)	0.37	0.44	0.68	0.57	0.078	0.054	0.060	0.093
Valine (Val)	2.34	2.14	2.75	2.94	0.222	0.066	0.025	0.841
Isoleucine (Ile)	1.75	1.48	1.78	1.82	0.122	0.215	0.333	0.463
Leucine (Leu)	2.06	1.90	2.20	2.28	0.149	0.305	0.163	0.732
Arginine (Arg)	0.60	0.65	0.75	0.80	0.071	0.222	0.048	0.662
Histidine (His)	0.95	0.94	1.06	1.25	0.122	0.262	0.061	0.672
Phenylalanine (Phe)	1.87	1.85	1.96	2.08	0.076	0.155	0.035	0.670
Alanine (Ala)	2.79	2.55	3.46	2.70	0.331	0.249	0.888	0.264
Aspartic acid (Asp)	0.12	0.12	0.11	0.08	0.022	0.700	0.275	0.706
Glutamic acid (Glu)	2.19	1.78	1.91	1.81	0.164	0.333	0.224	0.346
Glycine (Gly)	7.17	7.37	9.19	8.48	0.578	0.072	0.072	0.166
Proline (Pro)	1.37	1.41	1.75	1.54	0.114	0.115	0.226	0.111
Serine (Ser)	1.64	1.58	1.95	1.63	0.138	0.245	0.806	0.215
Tyrosine (Tyr)	2.85[b]	3.07[b]	3.27[ab]	3.80[a]	0.173	0.005	<0.001	0.829
Asparagine (Asn)	1.21	1.22	1.42	1.53	0.113	0.173	0.039	0.923
Glutamine (Gln)	63.28	54.79	65.06	58.15	5.506	0.549	0.774	0.933
Flavor amino acids [1]	12.87	12.47	15.41	13.87	0.902	0.126	0.258	0.241
Essential amino acids [2]	14.56[ab]	13.44[b]	17.05[a]	16.51[ab]	0.895	0.029	0.041	0.620
Non-essential amino acids [3]	82.62	73.89	88.12	79.72	6.525	0.493	0.950	0.778
Total amino acids	97.18	87.33	105.17	96.23	7.116	0.386	0.748	0.748

[a,b] Within a row, means without a common superscript differ at $P \leq 0.05$.
[1] Flavor amino acids = Glu + Asp + Ala + Arg + Gly.
[2] Essential amino acids = Lys + Met + Trp + Thr + Val + Ile + Leu + Arg + His + Phe.
[3] Non-essential amino acids = Ala + Asp + Glu + Gly + Pro + Ser + Tyr + Asn + Gln.

content ($P \leq 0.05$) and tended to increase the concentration of C16:0, C20:0, C16:1, C18:1n9t, C18:1n9c, and C20:1n9 (Table 7). Moreover, the total concentration of saturated fatty acid (SFA) and monounsaturated fatty acid (MUFA) tended to increase in the 0.3% inositol group ($P = 0.092$ and $P = 0.094$, Table 7).

3.4. The metabolic profiles of finishing pigs

To determine the metabolic profiles altered by inositol, untargeted metabolomic analysis was performed to detect the metabolic profiles in the serum between the control and 0.3% inositol groups. The composition differences were evaluated, and PLS-DA showed a clear separation between the two groups (Fig. 2B). A total of 1,112 metabolites were detected. Among them, 93 metabolites were up-regulated and 55 were down-regulated in the 0.3% inositol group (VIP >1, $P < 0.05$). Based on the annotation of HMDB database, most differential metabolites were "amino acid, peptides, and analogues", "fatty acids and conjugates", and "carbohydrates and carbohydrate conjugates". As shown in Table S1, 60.71% metabolites belonging to "amino acid, peptides, and analogues" and 85% metabolites belonging to "fatty acids and conjugates" were up-regulated in the 0.3% inositol group, while 81.82% metabolites belonging to "carbohydrates and carbohydrate conjugates" were down-regulated.

To further decipher the biological processes specifically affected by inositol, KEGG enrichment analysis was conducted. Notably, there were seven metabolic pathways significantly enriched ($P < 0.05$), including linoleic acid metabolism, arginine biosynthesis, lysine degradation, pyrimidine metabolism, tyrosine metabolism, arginine and proline metabolism, and D-amino acid metabolism (Fig. 2D). Additionally, the enrichment pathways also included pathways associated with cellular processes, environmental information processing, human diseases, and organismal systems (Fig. 2D). A total of 19 differential metabolites were involved in the significantly enriched pathways. Among them, 11 showed higher abundances in the 0.3% inositol group (Fig. 2E).

3.5. Amino acid metabolism in finishing pigs

To clarify the effects of inositol supplementation on amino acid metabolism in finishing pigs, we investigated the amino acid composition of serum using targeted metabolomics. The results showed that 0.3% inositol supplementation significantly decreased the levels of lysine, methionine, threonine, alanine, asparagine,

Table 7
Effects of dietary inositol supplementation on meat fatty acid composition (μg/g, fresh meat) of finishing pigs ($n = 8$).

Items	CON	0.3% Inositol	SEM	*P*-value
C10:0	3.14	4.98	0.923	0.178
C12:0	1.78	3.06	0.645	0.174
C14:0	38.45	63.30	11.632	0.153
C15:0	1.20	1.27	0.138	0.761
C16:0	422.76	787.09	117.322	0.045
C17:0	5.48	5.20	0.874	0.821
C18:0	490.37	754.43	113.148	0.183
C20:0	2.45	4.50	0.752	0.074
C21:0	0.39	0.40	0.014	0.316
C22:0	0.67	0.64	0.035	0.483
C24:0	0.73	0.74	0.012	0.530
C14:1	1.46	1.73	0.076	0.032
C16:1	95.58	143.50	19.110	0.098
C17:1	3.12	3.57	0.534	0.563
C18:1n9t	3.67	5.54	0.648	0.060
C18:1n9c	1439.72	2308.52	345.162	0.097
C20:1n9	9.52	19.36	3.354	0.057
C24:1n9	1.59	1.68	0.055	0.255
C18:2n6c	419.23	481.84	52.678	0.415
C18:3n6	2.38	2.15	0.140	0.118
C18:3n3	5.65	6.14	0.880	0.705
C20:2	6.01	7.44	1.127	0.385
C20:3n6	4.17	4.25	0.283	0.844
C20:4n6	40.36	40.52	2.754	0.967
C20:3n3	2.24	2.62	0.203	0.212
C20:5n3	1.42	1.48	0.074	0.537
C22:6n3	1.43	1.71	0.143	0.183
SFA[1]	967.41	1625.60	258.086	0.092
MUFA[2]	1554.66	2483.89	365.763	0.094
PUFA[3]	482.90	548.15	56.129	0.425
n-3 PUFA[4]	10.75	11.95	1.103	0.452
n-6 PUFA[5]	466.15	528.76	54.028	0.426
n-6/n-3	43.53	43.88	1.744	0.887

SFA = saturated fatty acid; MUFA = monounsaturated fatty acid; PUFA = polyunsaturated fatty acid.
[1] SFA = C10:0+ C12:0+ C14:0+ C15:0+ C16:0+ C17:0+ C18:0+ C20:0+ C21:0+ C22:0+ C24:0.
[2] MUFA = C14:1+ C16:1+ C17:1+ C18:1n9t + C18:1n9c + C20:1n9+ C24:1n9.
[3] PUFA = C18:2n6c + C18:3n6+ C18:3n3+ C20:2+ C20:3n6+ C20:4n6+ C20:3n3+ C20:5n3+ C22:6n3.
[4] n-3 PUFA = C18:3n3+ C20:3n3+ C20:5n3+ C22:6n3.
[5] n-6 PUFA = C18:2n6c + C18:3n6+ C20:3n6+ C20:4n6.

aspartate, glutamine, and serine in serum (Fig. 3A). There was a tendency for the levels of histidine and hydroxyproline to decrease in the 0.3% inositol group ($P = 0.097$ and $P = 0.081$, Fig. 3A). Dietary supplementation of 0.3% inositol also contributed to the decreased levels of total amino acids in serum ($P < 0.01$, Fig. 3B). These results, together with the differential metabolites in Fig. 2E, revealed that 0.3% inositol supplementation increased metabolizing ability of glutamine and proline, which led to the production of more N-acetyl-glutamate, urea, and 5-amino-pentanoate (Fig. 3C and D). Enhanced lysine degradation was also observed in the 0.3% inositol group, as evidenced by the increased abundance of 5-amino-pentanoate, N6-acetyl-L-lysine and 5-aminopentanal (Fig. 3E). Furthermore, inositol treatment induced the capability of tyrosine metabolism, as evidenced by the increased abundance of 3-methoxytyramine and L-normetanephrine (Fig. 3F).

3.6. The microbial profiles of the colon

Herein, 16S rRNA sequencing was used to analyze the microbial composition in the colon. A total of 2318 OTUs were identified from the two groups. The number of common OTUs was 1428, and the unique OTUs in the control and 0.3% inositol group was 346 and 544, respectively (Fig. 4A). The α-diversity was measured by Shannon index, Simpson index, and Chao1 index (Fig. 4B–D). Briefly, Shannon index tended to be increased ($P = 0.081$) and Chao1 index was significantly increased ($P \leq 0.05$) in the 0.3% inositol group. Next, the β-diversity of gut microbiota was assessed using PLS-DA and Nonmetric Multidimensional Scaling (NMDS) analysis, which showed that the overall gut microbial compositions differed between the two groups (Fig. 4E and F). To characterize the key microorganisms for each group, LDA Effect Size (LEfSe) analysis was conducted. As shown in Fig. 4G, Firmicutes and three bacterial genera, including *Turicibacter*, *Anaerovibrio*, and *norank_f_Clostridium_methylpentosum_group* were enriched in the control group, while Bacteroidota and *norank_f_norank_o_RF39* were enriched in the 0.3% inositol group ($\log_{10}$LDA score >3).

At the phylum level, the major phyla were Firmicutes, Bacteroidota, and Spirochaetota (Fig. 4H). Supplementation with 0.3% inositol significantly reduced Firmicutes ($P < 0.01$) but increased Bacteroidota ($P \leq 0.05$) compared to the control (Fig. 4J). At the genus level, the relative abundance of the top 20 genera is shown in Fig. 4I. The relative abundance of *norank_f_norank_o_RF39* was significantly elevated in the 0.3% inositol group, while the relative abundance of *Anaerovibrio* was significantly decreased ($P \leq 0.05$, Fig. 4K).

3.7. Correlations between differential microbiota and differential metabolites

Spearman correlation analysis was performed to show the correlation between differential microbiota and differential metabolites. As shown in Fig. 5A, the level of 48 up-regulated metabolites was positively or negatively associated with the abundance of Firmicutes or Bacteroidota, respectively. Additionally, 19 and 22 up-regulated metabolites were negatively and positively correlated with *Anaerovibrio* and *norank_f_norank_o_RF39*, respectively (Fig. 5A). Similarly, Bacteroidota and *norank_f_norank_o_RF39*, the microbes enriched in the 0.3% inositol group, were negatively correlated with 14 and 7 down-regulated metabolites, respectively (Fig. 5B). Importantly, microbes reduced in the 0.3% group, including Firmicutes and *Anaerovibrio*, were significantly correlated with lysine, methionine, aspartate, sernine, hydroxyproline, and total amino acids (Fig. 5C). The abundance of Bacteroidota in the colon was negatively correlated with the level of lysine, alanine, and methionine in the serum (Fig. 5C). These results indicated the essential role of gut microbiota in the inositol-altered metabolism, including the enhanced catabolism of amino acids.

4. Discussion

Inositol is a 6-carbon sugar alcohol consisting of nine isomers based on the spatial orientation of the hydroxyl groups. Myo-inositol is the most common form and is present in plant cells, animals and foods (Pani et al., 2020). Inositol is usually produced from phytate through a serious of chemical reactions. Recently, the efficient production of inositol from glucose was achieved by metabolic engineering (You et al., 2020). It can also be synthesized de novo endogenously from D-glucose, and this biosynthesis mainly occurs in kidneys. The beneficial effects of inositol on metabolic diseases have been studied previously. A combination of myo-inositol and D-chiro-inositol decreased fasting blood glucose and HbA1c levels in patients with type 2 diabetes (Pintaudi et al., 2016). By evaluating preclinical and clinical evidence, a literature review also observed the positive effect of inositol supplementation on hepatic triglyceride accumulation (Pani et al., 2020). As a water-soluble vitamin B group, inositol is included in the catalogue of feed additive varieties released by the Ministry of Agriculture and Rural Affairs of the People's Republic of China. It is permitted in animal diets and has been widely used in fish. However, no relevant studies

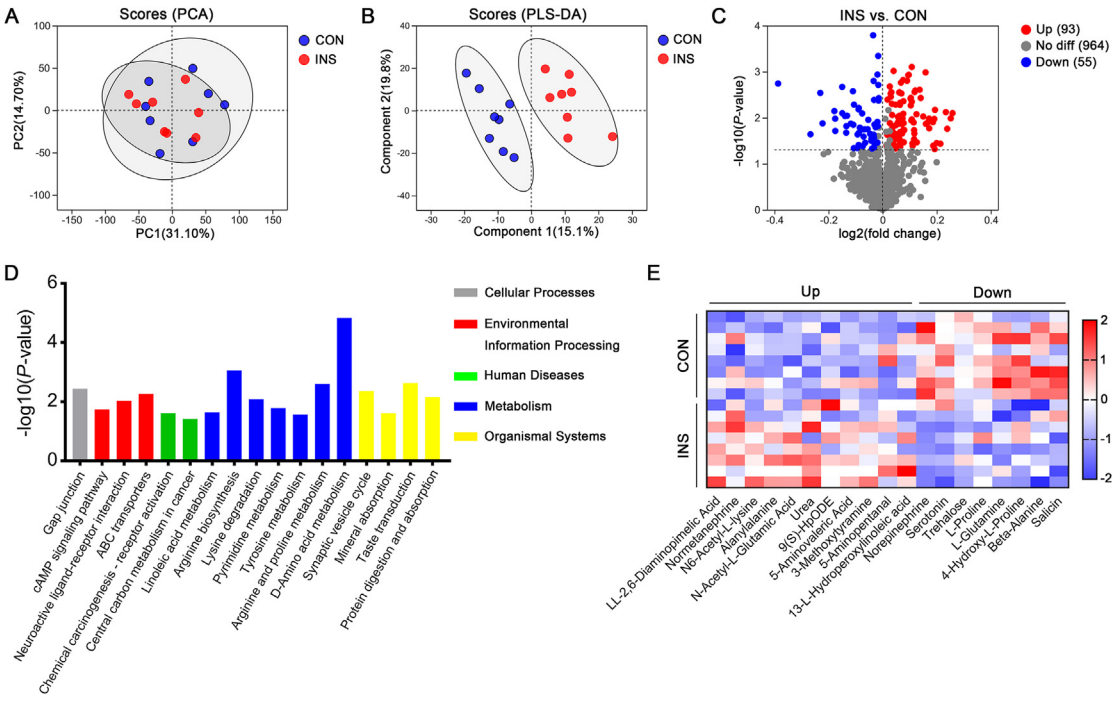

Fig. 2. Effects of dietary 0.3% inositol (INS) supplementation on the serum metabolome of finishing pigs ($n = 8$). (A) Principal component analysis (PCA) and (B) partial least squares discriminant analysis (PLS-DA) showing separation of data depending on 0.3% INS supplementation. (C) Volcano plots of all metabolites. Red and blue modes represent up- and down-regulated metabolites in the 0.3% INS group, respectively. Dotted horizontal line indicates threshold for *P*-value of 0.05. (D) Enrichment analysis of differential metabolites based on the Kyoto Encyclopedia of Genes and Genomes (KEGG) database. (E) Heatmap showing differential metabolites enriched in KEGG pathways in D.

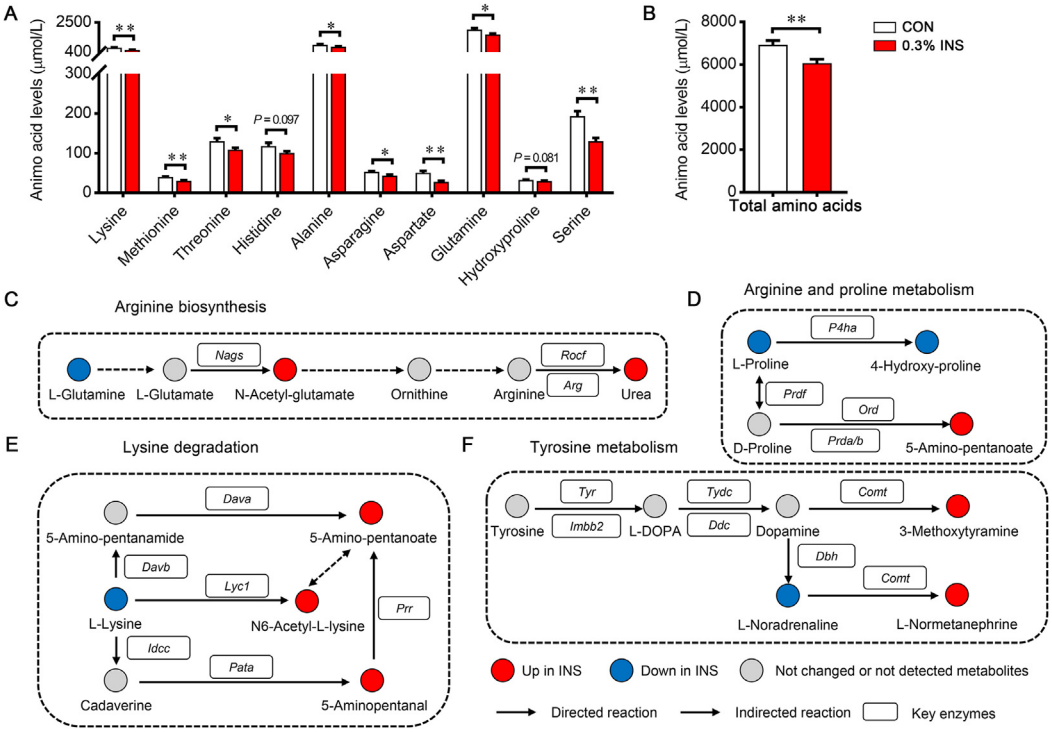

Fig. 3. Effects of dietary 0.3% inositol (INS) supplementation on amino acid metabolism of finishing pigs. (A-B) The concentration of amino acids in serum of finishing pigs. Illustration of metabolite changes in arginine biosynthesis (C), arginine and proline metabolism (D), lysine degradation (E), and tyrosine metabolism (F). Data are presented as means ± SEM ($n = 8$). Statistics were performed with Student's *t*-test. $^*P \leq 0.05$, $^{**}P < 0.01$. Arg = arginine dihydrolase; Comt = catechol O-methyltransferase; Dava = 5-aminopentanamidase; Davb = lysine 2-monooxygenase; Dbh = dopamine beta-monooxygenase; Ddc = L-tryptophan decarboxylase; Lyc1 = lysine N6-acetyltransferase; Nags = N-acetylglutamate synthase; Ord = 2,4-diaminopentanoate dehydrogenase; P4ha = prolyl 4-hydroxylase; Pata = putrescine aminotransferase; Prda/b = D-proline reductase (dithiol); Prdf = proline racemase; Prr = aminobutyraldehyde dehydrogenase; Rocf = arginase; Tydc = tyrosine decarboxylase; Tyr = tyrosinase.

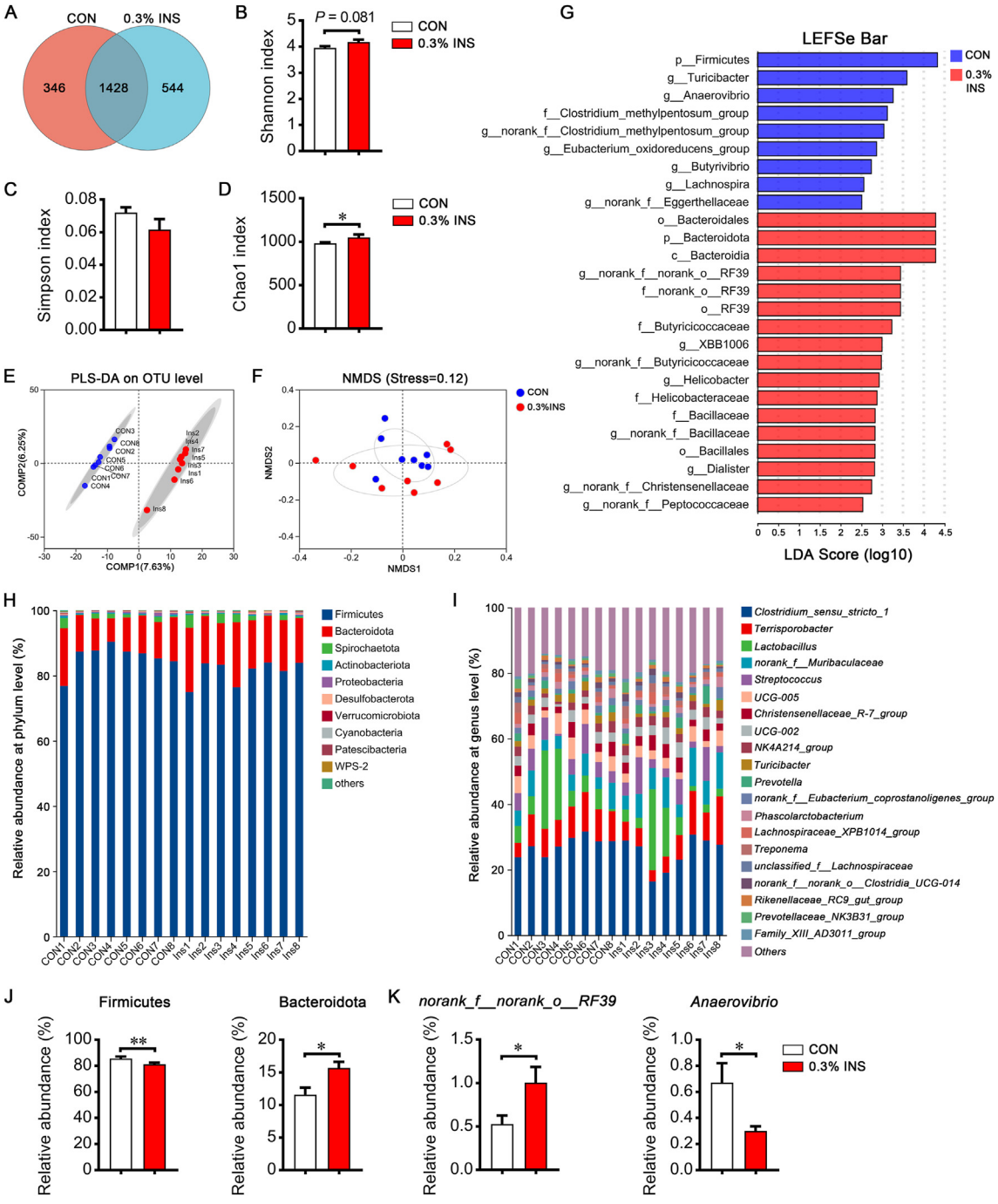

Fig. 4. Effects of dietary 0.3% inositol (INS) supplementation on colonic microbiota of finishing pigs. (A) The common and special OTUs between control and 0.3% inositol groups are shown by Venn diagram. The α diversity of colonic microbiota in finishing pigs, reflected by Shannon index (B), Simpson index (C), and Chao1 index (D). PLS-DA (E) and NMDS analysis (F) was used to distinguish the distribution of samples in different groups. (G) The biomarker species identified by the LEfSe analysis (LDA score ≥3). The relative abundance of bacteria at the phylum (H) and genus level (I). The top 10 and 20 most abundant phyla and genera are shown respectively. Relative abundance of bacteria that differ significantly at the phylum (J) and genus level (K). Data are presented as means ± SEM ($n = 8$). Statistics were performed with Student's t-test. $^{*}P \leq 0.05$, $^{**}P < 0.01$. CON = control group; Ins = 0.3% inositol group; LDA = linear discriminant analysis; LEfSe = LDA effect size; NMDS = nonmetric multidimensional scaling; OTUs = operational taxonomic units; PLS-DA = partial least squares discriminant analysis.

have investigated the effect of dietary inositol supplementation on meat quality of finishing pigs.

In this study, our data revealed that dietary inositol supplementation linearly decreased ADG of finishing pigs but had no adverse effect on hot carcass weight and dressing percentage. Considering that 36 pigs within each treatment were kept in one pen, feed intake data was not used for statistical analysis, and we mainly focused on the effects of inositol supplementation on meat quality in this work. Importantly, inositol supplementation increased backfat thickness and IMF content of finishing pigs. PPARγ is the major regulator of adipogenesis and no factor has been reported to promote adipogenesis in the absence of PPARγ (Rosen and Macdougald, 2006). Skeletal-specific overexpression of PPARγ in pigs could increase IMF content through promoting

Fig. 5. Correlations between differential bacteria and differential metabolites in serum. Spearman analysis of differential bacteria in colon and up-regulated metabolites (A), down-regulated metabolites (B), or significantly changed amino acids (C) in serum. Blue and red represent negative and positive correlations, respectively. $^{*}P \leq 0.05$, $^{**}P < 0.01$.

adipocyte differentiation (Gu et al., 2021). Similarly, lipolysis is reduced in the absence of FABP4 and its deletion could protect obese mice from insulin resistance, type 2 diabetes, and fatty liver disease (Maeda et al., 2005). The increased PPARγ level further corroborated the positive role of inositol in fat deposition in our study. It is worth noting that the backfat thickness at the 6th to 7th rib and 10th rib was linearly increased by inositol, but the fat-free lean index of finishing pigs was not markedly changed. Therefore, inositol had little detrimental effect on carcass quality. Dietary supplementation of inositol also improved meat tenderness, as evidenced by the decreased shear force and hardness. This is in accordance with previous studies, in which the positive correlation between IMF content and tenderness was observed (Aaslyng and Hviid, 2020; Zhang et al., 2022). It should be noted that tenderness is affected by a number of factors, including breed, nutritional status, and postmortem factors (temperature, sarcomere length, proteolysis) (Maltin et al., 2003). Thus, the relationship between IMF content and tenderness is not consistent in many published studies. For instance, IMF accounted for 47% of the differences in shear force in Duroc pork, but their relationship was not significant in Hampshire and Berkshire pork (Laack et al., 2001). Dietary calcium supplementation promoted IMF accumulation of finishing pigs but had no effect on shear force (Zhang et al., 2021). Overall, our work showed that dietary supplementation of inositol improved pork quality, mainly by increasing IMF content and tenderness.

The nutritional value and flavor of meat is partly determined by the composition of amino acids in muscle. For instance, lysine,

arginine, aspartic acid, and glutamic acid in meat are considered to be precursors of flavoring substances and can react with soluble reducing sugars to produce flavoring substances (Idolo Imafidon and Spanier, 1994; Zhang et al., 2010). Methionine, tryptophan, valine, isoleucine, leucine, arginine, histidine, phenylalanine, and tyrosine are bitter, while lysine, threonine, alanine, glycine, proline, serine, and hydroxyproline are sweet (Ma et al., 2020). In this study, the contents of nine amino acids were elevated by inositol treatment in muscle. It can be anticipated that the pork flavor was altered by inositol. Future studies need to address the effect of inositol on taste and aroma profiles of pork by E-nose and E-tongue analysis.

The process of amino acid metabolism and its relationship with metabolic diseases has also been reviewed (Ling et al., 2023). The contents of glycine, histidine, and methionine were lower in the skeletal muscle of obese subjects (Baker et al., 2015). Conversely, elevated circulating levels of branched-chain amino acids, methionine, phenylalanine, and tryptophan were associated with obesity and metabolic disorders in humans (Yang et al., 2018). Contrary to previous studies, amino acid contents were elevated in the LT muscle and decreased in the serum of finishing pigs by inositol administration, paralleling an increase in fat deposition. This may be due to the fact that inositol could reduce triglyceride accumulation in liver (Shimada et al., 2017, 2019), and metabolic disorders were not present in this study. Food absorption, tissue decomposition, and internal synthesis are three ways for animals to obtain amino acids. In this study, dietary composition of amino acids was consistent among four groups. Thus, inositol may expand amino acid pool size by enhancing protein dissociation in the skeletal muscle. It contributes to lipid synthesis through the production of α-ketoacid, as evidenced by the increased degradation of ketogenic amino acids, including lysine and tyrosine. Furthermore, skeletal muscle accounts for approximately 40% of the BW in adult mammals, and its mass is largely determined by the imbalance between protein synthesis and dissociation (Jefferson and Kimball, 2001). Therefore, inositol supplementation may decrease ADG of finishing pigs through enhancing muscle protein dissociation. To date, although stimulation of inositol on muscle glucose uptake was reported in mice (Dang et al., 2010), the effect of inositol on protein metabolism in skeletal muscle is still unclear. To test the above hypothesis, the activation of the mTOR signaling pathway and the expression of amino acid transporters in the skeletal muscle should be further examined.

Gut microbiota and its derived metabolites link to the host metabolism and the development of metabolic diseases (Fan and Pedersen, 2021). Herein, dietary supplementation of 0.3% inositol reduced Firmicutes and enriched Bacteroidota in the colon of finishing pigs. Lower abundance of Firmicutes and higher abundance of Bacteroidota were detected in obese Laiwu pigs. Interestingly, fecal microbiota transplantation (FMT) from Laiwu pigs to lean commercial pigs increased lipid accumulation (Xie et al., 2022). However, Firmicutes was also reported to promote fat phenotype in broilers, mice, and humans (Kang et al., 2022; Pinart et al., 2021; Zhang et al., 2020). This inconsistency is probably due to species and lifestyle-associated factors, including diet. Gut microbiota composition also affects systemic amino acid metabolism. The microbial contribution of essential amino acids to mouse muscle ranged from less than 5% to about 60% in different diets (Newsome et al., 2020). Importantly, altered ratio of Firmicutes/Bacteroidota was associated with host amino acid metabolism, including lysine degradation, arginine biosynthesis, and phenylalanine metabolism (Jiang et al., 2022). Increased abundance of Bacteroidota and enhanced capability of tyrosine and tryptophan were also observed in the improved metabolic health by vitamin D treatment (Zhang et al., 2023). In future studies, FMT should be performed to confirm the contribution of gut microbiota to inositol-altered fat deposition and amino acid metabolism.

5. Conclusions

This study demonstrates for the first time that dietary inositol supplementation promotes fat deposition and effectively improves meat quality in finishing pigs by improving IMF content and tenderness. Inositol also altered the metabolic profiles of finishing pigs, especially amino acid metabolism, such as lysine degradation and tyrosine metabolism. In addition, 0.3% inositol treatment changed the composition of gut microbiota, which contributed to regulating amino acid metabolism and fat deposition. Based on these results, dietary supplementation with 0.3% inositol is suggested in the diets of finishing pigs.

Credit author statement

Enfa Yan: Data curation, Formal analysis, Investigation, Methodology, Validation, Visualization, Writing-original draft. **Haijun Sun:** Data curation, Investigation, Methodology, Validation. **Linjuan He:** Formal analysis, Investigation, Methodology. **Boyang Wan:** Formal analysis, Investigation, Methodology. **Ming Shen:** Formal analysis, Investigation, Methodology. **Qiyuan Miao:** Formal analysis, Investigation, Methodology. **Jingdong Yin:** Conceptualization, Funding acquisition, Methodology, Project administration, Writing-review & editing. **Xin Zhang:** Conceptualization, Funding acquisition, Methodology, Project administration, Resources, Writing-review & editing.

Declaration of competing interest

We declare that we have no financial and personal relationships with other people or organizations that can inappropriately influence our work, and there is no professional or other personal interest of any nature or kind in any product, service and/or company that could be construed as influencing the content of this paper.

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Appendix supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.aninu.2024.05.012.

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