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Effect of dietary *Auricularia cornea* culture supplementation on growth performance, serum biochemistry profile and meat quality in growing-finishing pigs

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ABSTRACT. *Auricularia cornea* culture (ACC) is a dried product containing *Auricularia cornea* (AC) mycelium and various metabolites of AC fermentation. The objective of this study was to investigate the effects of dietary ACC supplementation on growth performance, short-chain fatty acid concentration in faeces, serum biochemical profile and meat quality in growing-finishing pigs. In total, 96 growing pigs with initial body weight 91.94 ± 7.59 kg, were allotted to one of four dietary treatments for 45 days. Treatments were: basal diet and three experimental diets with 0.3, 0.6 and 1.2% ACC addition, respectively. It was shown that pigs fed ACC diets had a greater average daily gain *(P* < 0.05), and also lower glucose content in serum (*P* < 0.05). In comparison with control animals, in pigs fed diets with ACC an increased butyrate content (*P* < 0.05) in faeces and greater monocarboxylate transporter 1 (*MCT1*) mRNA expression (*P* < 0.05) in the colon were noted. There was also observed an increasing trend concerning a* value (*P* = 0.09) and the higher polyunsaturated fatty acid contents in *longissimus dorsi* muscle (*P* = 0.01). In conclusion, the dietary ACC addition could improve the growth and health of animals as well as meat quality to a certain degree. So, a 1.2% ACC supplementation can be recommended for growing-finishing pigs.

Introduction

As a kind of edible and medicinal fungi, *Auricularia* mushrooms have been widely consumed all around the world. As the fourth most produced mushroom genera, *Auricularia* has been collected, cultivated and consumed in many countries, such as China, Japan, Vietnam and New Zealand, for hundreds of years (Bandara et al., 2015). Mounting studies have revealed a wide range of pharmacological functions of *Auricularia* and their derivatives. *Auricularia* polysaccharides (AP) have been considered to be the major bioactive component, and growing evidences have identified the biological functions of AP with molecular weight (MW) ranging from 4.6 to 3400 kDa, including

antioxidant, immunomodulatory, hepatoprotective and antitumour activities (Miao et al., 2020).

Both crude AP and water-soluble AP were reported to be able to increase the superoxide dismutase (SOD) and glutathione peroxidase (GSH-Px) activities and down-regulate levels of malondialdehyde (MDA) induced by D-galactose in aging mice (Wu et al., 2010; Zhang et al., 2011). *Auricularia* polysaccharides with MW of less than 110 kDa could stimulate macrophage to secrete interleukin (IL)-1β and IL-6 (Yu et al., 2009). Two kinds of AP (exopolysaccharides) could promote the release of cytokines (IL-6, IL-10 and tumour necrosis factor (TNF)-α) and nitric oxide (NO) in RAW 264.7 cells line (Zhang et al., 2018). A water-soluble AP extracted by hot water could significantly decrease the levels of low-density lipoprotein cholesterol, triglyceride and total cholesterol in the high-fat diet-induced hyperlipidemic mice (Zeng et al., 2013).

β-glucan is a sort of functional polysaccharide that widely spreads in the cell wall of bacterial, fungi and cereal seeds (rye, oats and barley, etc.). It possesses various biological functions such as immune function promotion, glucose regulation and anti-infection (Luo et al., 2019). β-glucan is an important polysaccharide present in mushrooms. Many studies have reported the biological activities of β-glucan (derived from mushroom), including anti-oxidative, immunomodulatory (Nandi et al., 2014) and anti-inflammatory ones (Ruthes et al., 2013). Most of mushroom β-glucan was insoluble, with occurrence percentage ranging from 54 to 82%, whereas the percentage of soluble β-glucan was between 16–46% (Gern et al., 2008). Several researches indicated that β-glucan could promote the growth of rats (Belobrajdic et al., 2015) and pigs (Lee et al., 2017), which was concerned with the effects of improving immunity and promoting intestinal health.

Currently, no information regarding the use of *Auricularia cornea* culture (ACC) and the associated effects on growth and meat quality are available. In this study, we hypothesized that ACC could be utilized as a feed additive that may benefit the growth and health of growing-finishing pigs. Therefore, the objective of this study was to evaluate the effects of dietary ACC inclusion at different levels (0.3, 0.6 or 1.2%) on growth performance, serum biochemical profile, faecal short-chain fatty acid (SCFA) contents, carcass characteristics and meat quality in finishing pigs.

Material and methods

All procedures involving animal handling received the approval of the Institutional Animal Care and Use Committee of Guangxi Academy of Agricultural Sciences (no. GAAS21011201).

Auricularia cornea culture contained a combination of mycelium and fermentation metabolites produced during the specific fermentation process. The mycelium is mainly composed of protein, chitin, cellulose, etc. (Haneef et al., 2017). Firstly, the *Auricularia cornea* (AC) strain was inoculated into a 1-m^3 stainless-steel fermentation tank under aseptic conditions. After a 7-day liquid fermentation, the inoculum was used for further solid-state fermentation (SSF). During the 15-day SSF, a specific culture media was inoculated with AC strain and allowed to ferment under sterile, temperature-humidity controlled conditions (26 °C). The entire fermented culture media was subsequently dried at a low temperature (55 °C) to preserve the bioactivity. As AC strain fermented proteins and carbohydrates are present in culture media, they could produce a wide variety of metabolic products, including amino acids, peptides and polysaccharides, such as β-glucan (Mukhopadhyay and Guha, 2015; Osińska-Jaroszuk et al., 2021), and several undefined metabolites that may have beneficial effects for pigs. The nutrient content of ACC is shown in Table 1.

Table 1. Analysed nutrient content and available energy concentration (MJ/kg) in *Auricularia cornea* culture (ACC), %

Indices	ACC
Dry matter	89.83
Gross energy, MJ/kg	17.36
Crude protein	16.37
Ether extract	4.31
Ash	4.23
Crude fibre	6.30
Neutral detergent fibre	26.54
Acid detergent fibre	9.42
Total phosphorus	1.02
Calcium	0.20

values are the means of six observations, n = 6

Animals, experimental design and sample collection

In total, 96 growing-finishing pigs (Duroc × Landrace × Yorkshire, 91.94 ± 7.59 kg of body weight) were randomly allotted to 4 treatments with 6 replicated pens per treatment (4 pigs per each

Table 2. Ingredients and calculated nutrient levels of experimental diets, %, as-fed basis

Indices	Grower groups				Finisher groups			
	Control	0.3% ACC	0.6% ACC	1.2% ACC	Control	0.3% ACC	0.6% ACC	1.2% ACC
Ingredients								
maize	77.61	77.59	77.59	77.59	77.91	77.91	77.91	77.87
soybean meal	15.00	15.00	15.00	15.00	13.00	13.00	13.00	13.00
wheat bran	4.00	3.70	3.40	2.80	6.00	5.70	5.40	4.80
soybean oil	0.70	0.72	0.72	0.72	0.80	0.80	0.80	0.82
ACC	0	0.30	0.60	1.20	–	0.30	0.60	1.20
dicalcium phosphate	0.60	0.60	0.60	0.62	0.42	0.42	0.42	0.42
limestone	0.90	0.90	0.90	0.88	0.84	0.84	0.84	0.84
sodium chloride	0.35	0.35	0.35	0.35	0.35	0.35	0.35	0.35
L-Lys	0.27	0.27	0.27	0.27	0.16	0.16	0.16	0.17
L-Thr	0.06	0.06	0.06	0.06	0.02	0.02	0.02	0.02
L-Trp	0.01	0.01	0.01	0.01	–	–	–	0.01
vitamin-mineral premix[1]	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50
Analysed nutrient levels								
dry matter	88.31	87.92	88.59	89.24	89.45	88.89	88.20	89.07
crude protein	13.40	13.35	13.52	13.21	13.12	13.24	13.09	13.34
gross energy, MJ/kg	16.24	16.05	16.39	16.50	15.91	15.87	16.19	16.22
ash	3.94	4.12	4.16	4.33	4.25	4.10	4.30	4.24
Calculated nutrient level								
DE, MJ/kg	14.23	14.23	14.23	14.23	14.23	14.23	14.23	14.23
SID Lys	0.73	0.73	0.73	0.73	0.61	0.61	0.61	0.61
SID Met	0.21	0.21	0.21	0.21	0.20	0.20	0.20	0.20
SID Thr	0.46	0.46	0.46	0.46	0.40	0.40	0.40	0.40
SID Trp	0.13	0.13	0.13	0.13	0.12	0.12	0.12	0.12

ACC – *Auricularia cornea* culture, DE – digestible energy, SID – standardized ileal digestible, Lys – lysine, Met – methionine, Thr – threonine, Trp – tryptophan; [1] provided the following per kg of complete diet: IU: vitamin A 5 512, vitamin E 30, vitamin D_3 2200; µg: vitamin B_{12} 27.6, biotin 44; mg: vitamin K_3 2.2, D-pantothenic acid 14, pyridoxine 3, riboflavin 4, folic acid 0.7, thiamine 1.5, Mn 40, Cu 100, Fe 75, Zn 75, I 0.35, Se 0.3

pen), including (1) maize-soybean meal basal diet (control diet); (2) control diet supplemented with 0.3% ACC; (3) control diet supplemented with 0.6% ACC; and (4) control diet supplemented with 1.2% ACC (Table 2). The experiment lasted for 45 days, including 2 phases: the grower phase (days 1–23, 75–100 kg) and the finisher phase (days 24–45, 100–135 kg). During the 45-day feeding period, all pigs were housed in a temperature-controlled room (22–26 °C). Water and feed were available *ad libitum*. All treatment diets were formulated to meet the nutrient requirements of NRC (2012). Body weight (BW) and feed were weighed at days 0, 23 and 45 to determine average daily feed intake (ADFI), average daily gain (ADG) and feed to gain ratio (F/G ratio). On days 23 and 45, twenty-four fresh faecal samples (one sample per pen) were acquired by rectal palpation for further SCFA analysis (Zhao et al., 2018). The blood samples were collected from pigs with BW close to the average BW in each pen using precaval vein puncture into the 10-ml vacuette tubes on day 45; then the serum was obtained by centrifugation at 3000 r/s for 15 min and stored at −20 °C until further analysis.

At the end of the trial, a total of 24 pigs (one pig per pen) were selected with BW close to the average BW of each pen. After 18-h fasting, selected pigs were euthanized by exsanguination. About 200 g of *longissimus dorsi* muscle (LDM) on the right half of each carcass between the 10th and 12th ribs were acquired for further assessment. The tissue samples from the colon were taken and immediately stored at −80 °C until further mRNA expression analysis.

Chemical analysis

The ACC powder, diets and faeces were analysed for crude protein (CP), dry matter (DM), ash, ether extract (EE), calcium and total phosphorus (AOAC International, 2006). The gross energy (GE) was detected by an Automatic Isoperibol Oxygen Bomb Calorimeter (C2000, IKA, Staufen, Germany). The concentrations of acid detergent fibre (ADF) and neutral detergent fibre (NDF) were measured by a fibre analyser equipment (2010, FOSS, Hillerød, Denmark) (Van Soest et al., 1991).

The SCFA contents were measured *via* an ion chromatography (883, Metrohm, Herisau, Switzerland) according to the procedure described by

Wu et al. (2017). Antioxidant parameters, including total antioxidant capacity (T-AOC), superoxide dismutase (SOD) activity, glutathione peroxidase (GSH-Px) activity and malondialdehyde (MDA) content were analysed by assay kits purchased from Jiancheng Bioengineering Ltd. (Nanjing, China). The concentrations of IL-1β, IL-2, IL-6, TNF-α were measured using ELISA kits (Jiancheng Bioengineering Ltd., Nanjing, China) with an ELISA Reader (Multiskan, Thermo Fisher Scientific, Waltham, MA, USA). The contents of low-density lipoprotein cholesterol (LDL-C), high-density lipoprotein cholesterol (HDL-C), triglyceride (TG), total cholesterol (TC), glucose (GLU) in serum were analysed by an automatic biochemical analyser (3100, Hitachi, Tokyo, Japan) using corresponding kits (Jiancheng Bioengineering Ltd., Nanjing, China). Fatty acids contents were analysed by gas chromatography (6890, Agilent Technologies, Inc., Santa Clara, CA, USA) according to Sukhija and Palmquist (1988).

Carcass traits and meat quality determination

After slaughtering, pigs were weighed individually to calculate the hot carcass weight, then were chilled at 4 °C to measure the carcass length, backfat thickness, loin eye area and marbling. Meat quality assessment, including pH value, meat colour, cooling loss, shear force and drip loss, were subsequently proceeded.

The dressing percentage was calculated according to the following equation: dressing percentage (%) = 100 × carcass weight / live body weight. Carcass length was calculated between the 1st rib and the public bone (Latorre et al., 2009). The backfat thickness and loin eye area were determined at the 10th rib according to the procedure described by the Chinese Guidelines on Performance Measurement Technology and Regulations for pig (Ministry of Agriculture of the People's Republic of China, 2014). Three points of the 1st rib, last rib and last lumbar vertebra were recorded to determine the backfat thickness by a vemier caliper. The loin eye area was calculated according to the equation: loin eye area (cm^2) = loin eye width (cm) × loin eye height (cm) × 0.7. Marbling was evaluated according to the National Pork Producers Council (NPPC) of the United States guidelines (NPPC, 1999).

After making an incision on the LDM, the muscle pH value was measured using a glass penetration pH electrode (IS400, SP, AL, USA) at 45 min *post-mortem* and recorded as $pH_{45\ min}$. After storing in a chilling room at 4 °C for 24 h, the pH value was measured as $pH_{24\ h}$. The meat colour, including a* (redness), b* (yellowness) and L* (lightness), was also determined at 45 min and 24 h *post-mortem* using a tristimulus colorimeter (NR, Mingao, Nanjing, China). To determine cooking loss, the steaks were weighed individually in their raw state and then immediately weighed after they had reached their final cooking temperature (80 °C) (Aaslyng et al., 2003). The shear force was measured using a muscle tenderness meter (C-LM3, Mingao, Nanjing, China), after the samples were previously cooked in a water bath at 70 °C for 20 min (Ciobanu et al., 2004). The drip loss was measured according to Aaslyng et al. (2003). Briefly, the slice was hung in a plastic bag (4 °C) for 24 h. Then, the drip loss was calculated as: drip loss (%) = the amount of drip (g) / initial meat weight (g).

Relative quantification of *MCT1* mRNA expression

Analysis of monocarboxylate transporter 1 (*MCT1*) mRNA expression was performed as described previously (Metzler-Zebeli et al., 2012; Tudela et al., 2015). The frozen colon sections were pulverized under liquid nitrogen using a pestle and mortar. The total RNA from colonic tissue was extracted using an Invitrogen TRIzol reagent (Thermo Fisher Scientific, Waltham, MA, USA). The RNA quality and quantity were determined on a spectrophotometer (ND-1000, Thermo Fisher Scientific, Waltham, MA, USA). Subsequently, the total RNA was reverse-transcribed into complementary DNA (cDNA) using the Superscript II transcriptase (Invitrogen, Thermo Fisher Scientific, Waltham, MA, USA). Primers for *MCT1* were designed based on published sequences (Tudela et al., 2015) and primer information was summarized in Table 3.

Table 3. Primer sequences for Real-Time polymerase chain reaction analysis

Gene	Primer	Sequence (5'→3')
MCT1	Forward	GGAGACCAGTATAGACGCTGC
	Reverse	CTCCTCCTCTTTGGGGCTTC
ACTB	Forward	TGCGGGACATCAAGGAGAAGC
	Reverse	ACAGCACCGTGTTGGCGTAGAG

MCT1 – monocarboxylate transporter 1; *ACTB* – β-actin

The Real-Time polymerase chain reaction (PCR) was performed in a total volume of 10 µl, which contained 0.2 µl each of forward and reverse primers, 1 µl cDNA template, 0.2 µl ROX Reference Dye and 5 µl SYBR Green mix. Then, the Real-Time quantitative PCR was performed with general cycling

conditions as follows: pre-denaturation at 95 °C for 10 s; 40 cycles of amplification at 95 °C for 5 s and 60 °C for 20 s; melting curve construction from 60 °C to 99 °C with a heating rate of 0.1 °C/s and fluorescence measurements (Applied Biosystems™ 7500 Real-Time PCR System; Thermo Fisher Scientific, Waltham, MA, USA).

Statistical analysis

The data for growth performance, biochemical indices, carcass traits and meat quality were analysed by analysis of variance (ANOVA) using the PROC GLM procedure of SAS (version 9.2, SAS Institute Inc., Cary, NC, USA) followed by Student-Newman-Keuls multiple range tests. The significances were estimated at a probability level of 0.05.

Results

As shown in Table 4, there was a trend (P = 0.07) concerning final BW of pigs fed ACC diets. No effects were observed on ADG, ADFI and F/G ratio during days 1–23. Pigs fed ACC diets showed a higher ADG ($P \leq 0.01$), whereas ADFI and F/G ratio were not influenced during days 24–45 and the overall period.

Table 4. Effects of dietary *Auricularia cornea* culture (ACC) supplementation on growth performance in growing-finishing pigs

Indices	Treatments				SEM	*P*-value
	Control	0.3% ACC	0.6% ACC	1.2% ACC		
IBW	92.02	91.90	91.90	91.97	0.08	0.67
FBW	131.03	131.21	131.48	131.66	0.16	0.07
Days 1 to 23						
ADG, g	904	906	908	910	4.24	0.73
ADFI, g	2450	2392	2437	2460	53.51	0.82
F/G	2.71	2.64	2.68	2.70	0.06	0.84
Days 24 to 45						
ADG, g	828^{b}	840^{ab}	849^{a}	853^{a}	4.35	<0.01
ADFI, g	2525	2514	2558	2578	26.07	0.32
F/G	3.05	2.99	3.01	3.02	0.03	0.69
Days 1 to 45						
ADG, g	867^{b}	874^{ab}	880^{a}	882^{a}	3.00	0.01
ADFI, g	2486	2452	2496	2517	34.02	0.60
F/G	2.87	2.81	2.84	2.85	0.04	0.72

IBW – initial body weight, FBW – final body weight, ADG – average daily gain, ADFI – average daily feed intake, F/G – feed to gain ratio, SEM – standard error of the mean; ^{ab} – means with different superscripts are significantly different at $P < 0.05$

According to obtained data (Table 5), dietary ACC supplementation showed no effects on SCFAs contents on day 23. Dietary 1.2% ACC supplementation in diets increased the butyrate content (P = 0.03) in pig faeces on day 45.

Table 5. Effects of dietary *Auricularia cornea* culture (ACC) supplementation on short-chain fatty acid content measured in fresh pig faeces, mg/kg

Indices	Treatments				SEM	*P*-value
	Control	0.3% ACC	0.6% ACC	1.2% ACC		
Day 23						
acetate	4256.34	4178.04	4226.06	4469.30	222.47	0.80
propionate	3319.74	3370.82	3507.96	3242.03	125.71	0.52
butyrate	488.09	476.59	496.56	484.38	19.70	0.91
total	8064.16	8025.45	8230.58	8195.72	255.92	0.93
Day 45						
acetate	4895.11	4823.64	4993.70	4924.99	108.61	0.74
propionate	3748.29	3615.31	3691.85	3666.96	140.13	0.92
butyrate	601.65^{b}	672.18^{ab}	647.25^{ab}	706.36^{a}	22.53	0.03
total	9245.05	9111.13	9332.79	9298.31	172.54	0.81

n = 6; SEM – standard error of the mean; ^{ab} – means with different superscripts are significantly different at $P < 0.05$

In pigs fed 1.2% ACC a greater *MCT1* mRNA expression (P = 0.01) in colon was observed (Figure 1).

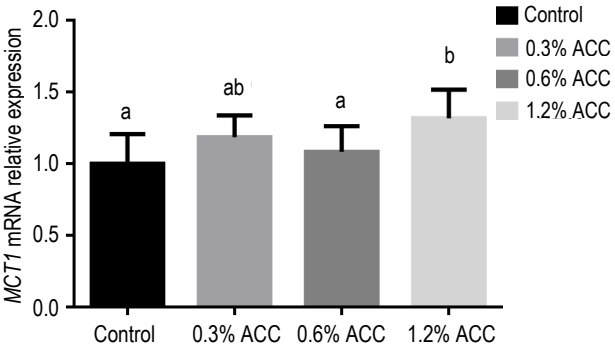

Figure 1. Relative mRNA expression of *MCT1* in colon of finishing pigs fed control diet or diets supplemented with 0.3, 0.6 and 1.2% *Auricularia cornea* culture (ACC); n = 6; *MCT1* – monocarboxylate transporter 1

As shown in Table 6, in comparison with control animals, pigs fed ACC diets revealed reduced glucose content (P = 0.03), and there was no significant effect on GSH-Px, T-AOC, SOD and the other serum indices.

In terms of meat quality (Table 7), there was an increased trend on a* value (P = 0.09). No differences in carcass weight, loin eye area, marbling, shear force, drip loss, and pH values were observed in pigs fed ACC diets in comparison with control ones.

As shown in Table 8, in pigs fed 0.6 and 1.2% ACC diets greater linoleic acid (P = 0.02) and arachidonic acid ($P < 0.01$) contents were noted in comparison with control ones. Diets supplemented with ACC significantly increase the sums of polyunsaturated fatty acids (PUFA) (P = 0.01) and sums of n-6 PUFA ($P < 0.01$) in the LDM.

Table 6. Effects of dietary *Auricularia cornea* culture (ACC) supplementation on serum profile of pigs

Indices	Treatments				SEM	*P*-value
	Control	0.3% ACC	0.6% ACC	1.2% ACC		
Day 45						
GSH-Px, U/ml	1109.25	1070.67	1053.27	1122.16	41.86	0.63
T-AOC, U/ml	13.43	14.66	14.72	14.94	0.80	0.55
SOD, U/ml	75.06	82.21	79.28	80.62	4.79	0.75
MDA, nmol/ml	2.65	2.39	2.76	2.58	0.24	0.73
HDL-C, mmol/l	1.02	1.07	1.16	1.14	0.09	0.65
LDL-C, mmol/l	1.31	1.10	1.24	1.21	0.14	0.76
TC, mmol/l	2.49	2.67	2.41	2.52	0.19	0.80
TG, mmol/l	0.56	0.61	0.64	0.55	0.08	0.83
GLU, mmol/l	2.96^{a}	2.81^{ab}	2.89^{ab}	2.69^{b}	0.06	0.03
IL-1β, µg/l	27.21	27.48	26.84	26.52	1.81	0.98
IL-2, ng/l	488.20	476.24	493.39	496.77	12.87	0.70
IL-6, µg/l	90.94	88.70	93.22	89.38	4.69	0.91
TNF-α, ng/l	206.56	216.00	202.58	218.54	9.52	0.60

n = 6; SEM – standard error of the mean; GSH-Px – glutathione peroxidase, T-AOC – total antioxidant capacity, SOD – superoxide dismutase, MDA – malondialdehyde, HDL-C – high-density lipoprotein cholesterol, LDL-C – low-density lipoprotein cholesterol, TC – total cholesterol, TG – triglyceride, GLU – glucose, IL-1β – interleukin-1β, IL-2 – interleukin-2, IL-6 – interleukin-6, TNF-α – tumour necrosis factor-α; ^{ab} – means with different superscripts are significantly different at $P < 0.05$

Table 7. Effects of dietary *Auricularia cornea* culture (ACC) supplementation on carcass traits and meat quality of pigs

Indices	Treatments				SEM	*P*-value
	Control	0.3% ACC	0.6% ACC	1.2% ACC		
Carcass traits						
carcass length, cm	106.20	105.43	106.08	106.18	1.07	0.95
hot carcass weight, kg	98.55	97.35	99.28	98.15	1.81	0.90
dressing percentage, %	74.78	74.14	75.73	74.99	0.56	0.24
loin eye area, cm^2	42.97	43.77	46.34	46.97	2.12	0.49
backfat thickness	14.83	14.5	14.17	14.67	0.64	0.86
marbling	1.75	1.67	1.92	2.00	0.23	0.74
Meat quality						
cooling loss, %	21.50	21.53	22.89	21.82	0.89	0.66
shear force, kg	33.86	32.35	33.72	32.53	1.42	0.80
drip loss, %	2.25	1.54	1.76	1.93	0.2	0.12
L* (45 min)	35.62	36.15	37.06	37.38	0.57	0.15
a* (45 min)	3.83	3.96	4.13	4.25	0.12	0.11
b* (45 min)	3.55	3.76	3.46	3.52	0.17	0.62
L* (24 h)	55.58	55.16	55.2	55.99	1.18	0.95
a* (24 h)	10.27	10.87	11.56	11.21	0.34	0.09
b* (24 h)	7.51	7.36	7.82	7.92	0.37	0.69
$pH_{45\ min}$	6.72	6.46	6.27	6.31	0.14	0.13
$pH_{24\ h}$	5.91	5.87	5.78	5.82	0.09	0.74

pH_{45min} – pH value 45 min after *post-mortem*, pH_{24h} – pH value 24 h after *post-mortem*, L*, a*, b* (45 min) – meat colour values 45 min after *post-mortem*, L*, a*, b* (24 h) – meat colour values 24 h after *post-mortem*, SEM – standard error of the mean; values are means with pooled SEM, n = 6

Table 8. Effects of dietary *Auricularia cornea* culture (ACC) supplementation on fatty acids profile in the *longissimus dorsi* muscle of finishing pigs, mg/g of fresh meat

Indices	Treatments				SEM	*P*-value
	Control	0.3% ACC	0.6% ACC	1.2% ACC		
SFA						
caproic acid	0.16	0.15	0.15	0.15	0.01	0.61
capric acid	0.16	0.16	0.15	0.15	0.01	0.37
lauric acid	0.09	0.09	0.08	0.08	0.01	0.76
myristic acid	1.26	1.32	1.17	1.13	0.10	0.57
palmitic acid	22.50	22.11	21.16	20.43	1.18	0.61
heptadecanoic acid	0.30	0.29	0.14	0.13	0.09	0.43
stearic acid	11.44	11.14	11.10	11.58	0.95	0.98
icosanoic acid	0.24	0.22	0.20	0.19	0.02	0.37
heneicosanoic acid	0.36	0.35	0.32	0.31	0.02	0.12
MUFA						
palmitoleic acid	2.59	2.64	3.18	3.07	0.19	0.10
oleic acid	48.28	47.69	46.68	47.10	2.06	0.95
eicosenoic acid	0.67	0.72	0.73	0.71	0.10	0.97
PUFA						
linoleic acid	4.27^{b}	5.10^{ab}	5.39^{a}	5.66^{a}	0.28	0.02
α-linolenic acid	0.33	0.31	0.34	0.35	0.06	0.98
dihomo-γ-linolenic	0.19	0.22	0.20	0.21	0.01	0.08
arachidonic acid	1.21^{b}	1.39^{ab}	1.59^{a}	1.62^{a}	0.07	<0.01
eicosatrienoic acid	0.10	0.09	0.10	0.09	0.01	0.90
eicosapentanoic acid	0.05	0.05	0.05	0.05	<0.01	0.55
docosadienoic acid	0.04	0.04	0.05	0.04	<0.01	0.52
docosahexaenoic acid	0.08	0.10	0.09	0.11	0.01	0.38
∑SFA	36.50	35.83	34.48	34.16	2.10	0.84
∑MUFA	51.55	51.05	50.58	50.88	2.10	0.99
∑PUFA	6.26^{b}	7.30^{a}	7.81^{a}	8.12^{a}	0.34	0.01
PUFA/SFA	0.18	0.22	0.24	0.25	0.02	0.09
∑n-6 PUFA	5.67^{b}	6.71^{a}	7.19^{a}	7.50^{a}	0.30	<0.01
∑n-3 PUFA	0.55	0.56	0.58	0.59	0.07	0.96
n-6/n-3	11.04	12.83	13.21	12.89	1.16	0.56

SEM – standard error of the mean, SFA – saturated fatty acid, MUFA – monounsaturated fatty acid, PUFA – polyunsaturated fatty acid; values are means with pooled SEM, n = 6; ^{ab} – means with different superscripts are significantly different at $P < 0.05$

The concentrations of saturated fatty acids (SFA) and monounsaturated fatty acids (MUFA) were not influenced by dietary ACC supplementation. The dihomo-γ-linolenic content ($P = 0.08$) and PUFA/SFA ratio value ($P = 0.09$) tended to improve in pigs fed ACC diets.

Discussion

Polysaccharides are the main component (about 90%) of the AC cell wall, and the β-glucan is the central core of the cell wall (Osińska-Jaroszuk et al., 2021). The β-glucan concentration in mushrooms ranged from 0.21 to 0.53 g/100 g (dry weight

basis) (Rop et al., 2009). It was revealed that the ACC supplementation effectively improve the ADG in pigs. The ameliorated effect might derive from the nutraceutical compounds in ACC. The increased ADG possibly due to the intestinal improvement, as Metzler-Zebeli et al. (2012) indicated that dietary β-glucan could modulate the morphology of piglets and improve intestinal structure. This result is consistent with the previous studies, as Luo et al. (2019) showed that diet supplemented with β-glucan (100 mg/kg) might significantly increase ADG and feed conversion ratio, and 0.025% β-glucan addition has been shown to augment ADG and ADFI in piglets (Dritz et al., 1995). Li et al. (2006) and Vetvicka et al. (2014) also concluded that supplementing diets with β-glucan could improve pig health and growth performance.

Pigs fed ACC revealed a higher butyrate level in the current study. The reason was highly due to the β-glucan proportion, which could act as a unique substrate for the SCFA-generated microbes and modify the relative composition of intestinal microbiota. The mammalian genome does not encode most enzymes required to degrade β-glucan, and β-glucan could escape digestion in the foregut and be fermented in the hindgut, resulting in improving SCFA production that altered the microbial ecology in the gastrointestinal tract of pigs. Pieper et al. (2012) reported that feeding β-glucan enhanced the contents of lactate and propionate in the colon of piglets. Metzler-Zebeli et al. (2012) also showed that oat β-glucan enhanced caecal and colonic butyrate contents, which could be favourable for intestine development in weaned piglets.

Butyrate is the principal energy source for colonic epithelial cells, which plays a vital role in the epithelial maintenance of intestinal barrier function. The butyrate transportation across the colonocyte luminal membrane is mediated by the *MCT1* (Cuff et al., 2002; Plöger et al., 2012). The increased butyrate from the fermented carbohydrates by probiotics has improved *MCT1* expression in pigs (Metzler-Zebeli et al., 2012). The carrier-mediated butyrate absorption from the hindgut lumen could be also through *MCT1* expression (Tudela et al., 2015). Borthakur et al. (2008) reported that butyrate could activate the *MCT1* expression that might occur through a butyrate response element in the *MCT1* gene promoter region. To further explore the mechanisms involved in the improving effect, we determined the colonic *MCT1* mRNA expression, as the generation of butyrate has been positively related to the MCT1 pathway (Cuff et al., 2002). The result showed that 1.2% ACC supplementation could up-regulate the expression of *MCT1* in the colon. The *MCT1* mRNA expression and activity were regulated by butyrate, as Cuff et al. (2002) indicated that butyrate presented a concentration- and time-dependent relationship of both MCT1 protein and mRNA. The result is in accordance with the study of Metzler-Zebeli et al. (2012), who showed that dietary β-glucan could up-regulate *MCT1* expression in the caecum. Tudela et al. (2015) investigated the influence of bacterial metabolites on *MCT1* expression, and showed that *MCT1* expression was higher after incubation with Na-butyrate.

The ACC contains multifarious nutritive materials, especially the polysaccharide from AC, which show various pharmacological activities (Wang et al., 2019). Based on the experimental result, the ACC exhibited a hypoglycemic effect, which was consistent with the study of Wang et al. (2019). In the opinion of Wang et al. (2019), AC appeares to impact glucose metabolism mainly by altering phosphoenolpyruvate carboxykinase (PEPCK) and glucose-6-phosphatase (G-6-Pase) levels in mice's liver. The inhibition of PEPCK and G-6-Pase may effectively regulate blood glucose improvement (Cui et al., 2018).

Meat colour was a vital indicator for the assessment of pork appearance. It was influenced by several factors, such as intramuscular fat content, postmortem glycolysis rate, pigment level and oxidative status of pigment (Van Oeckel et al., 1999). We have revealed that under ACC supplementation a* value had a trend towards increase, which was in accordance with the study in which yeast polysaccharide improved the meat colour (redness had a trend to increase) (Ma et al., 2017). Researches indicated that β-glucan supplementation could increase a* value of pork and chicken meat (Cho et al., 2013; Luo et al., 2019). The underlying reason for the redder meat colour of β-glucan-treated pigs might be attributed to an increased mean fibre cross-sectional area or the increased fast twitch muscle fibres (Petersen et al., 1998; Luo et al., 2019).

The fatty acid profile is a valuable parameter of meat quality. The fatty acid profile is the important physical base of meat flavour and has been a primary area of consumer concern (Luo et al., 2019). The present study showed that the proportions of linoleic acid, arachidonic acid, total PUFA and n-6 PUFA in pigs fed 1.2% ACC diet were increased, in comparison with control animals. Fatty acids are the main components of adipose tissue, the PUFA could only be acquired from diets, as n-3 and n-6 PUFA could not be converted into each other (Liu et al., 2020). To our knowledge, there is limited available literature about dietary AC addition and it's relation to the pork fatty acid profile. The result was

probably analogical with previous studies that showed two types of fungi, *Aspergillus* and *Saccharomyces*, could improve the UFA/SFA ratio and linolenic acid content in broiler meat through its beneficial effects on the intestinal microbiota (Endo and Nakano, 1999; Saleh et al., 2013). Polyunsaturated fatty acids compose more than one unsaturation and are beneficial to health. The n-3 and n-6 PUFAs are necessary for the body and have specific functions, namely anti-inflammatory, vasodilatory and chemotactic effects (Kus-Yamashita et al., 2016). Linoleic acid plays a positive role in reducing blood cholesterol and slowing the development of atherosclerosis (Jandacek, 2017). Arachidonic acid is a fundamental constituent of cell structure and is an essential fatty acid for animals. It impacts the function of specific membrane proteins and plays a vital role in maintaining the integrity of cells and organelle (Tallima and Ridi, 2017). Thus, dietary ACC supplementation could improve PUFA contents in LDM, which is beneficial for human consumption and health. However, the reasons for the changes in the fatty acid composition affected by ACC supplementation are unclear in the current study, so further research is necessary to determine the possible mechanism of such situation.

Conclusions

The supplementation of *Auricularia cornea* culture (ACC) to pig diets could improve growth performance, faecal short-chain fatty acid content, polyunsaturated fatty acid contents in *longissimus dorsi* muscle, and showed a hypoglycemic effect to a certain degree. So, the optimal supplementation of 1.2% ACC into growing-finishing pig diets may be recommended.

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Conflict of interest

The authors declared that there is no conflict of interest.

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