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Effects of dietary herbal powder supplementation on inflammatory cytokines, cecal microbiota, and serum metabolites in finishing Kele pigs

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Abstract

A study was designed to test the efficacy of herbal powder supplement (HPS) on serum inflammatory cytokines, cecal microbial communities, and serum metabolites in finishing Kele pigs. Forty healthy finishing Kele pigs with an initial body weight of 42.67 ± 1.73 kg were randomly assigned to four groups of 10 pigs each. Pigs received a basal diet without HPS (group A) or supplemented with 0.1% (group B), 0.3% (group C), or 0.5% HPS (group D) for 35 days. Among the inflammatory cytokines, group B showed significantly higher serum TGF-β1 and lower IFN-γ levels than the other groups ($P < 0.05$). HPS supplementation did not affect alpha-diversity indices of the cecal microbiota. At the genus level, the relative abundances of *Faecalibacterium* and *Blautia* were higher in group B than in group A ($P < 0.01$). The relative abundances of *Bacteroides* and *Lachnospiraceae_AC2044_group* were higher in group C than in group A ($P < 0.05$), whereas *Treponema* was more abundant in group D ($P < 0.01$). Differential metabolites were significantly enriched in the primary bile acid biosynthesis pathway. Glycocholic acid was positively correlated with TGF-β1 and negatively correlated with IFN-γ ($P < 0.05$). These findings indicate that dietary supplementation with 0.1% HPS may improve the inflammatory cytokine profile of TGF-β1 and IFN-γ in finishing Kele pigs. This effect was associated with specific cecal bacteria, including *Faecalibacterium*, and serum metabolites such as glycocholic acid.

Introduction

With the rapid development of intensive pig farming, the incidence of bacterial diseases has significantly increased, and the overuse of antibiotics poses a serious threat to both animal and human health (Li et al. 2022; Du et al. 2022). Developing novel alternatives to antibiotics to promote pig health has become an important approach to addressing the current challenges and advancing the pig industry (Li et al. 2021; Xu et al. 2021). Compounds commonly found in plant-derived materials, such as flavonoids, polyphenols, polysaccharides, and terpenoids, possess antioxidant, anti-inflammatory, and immunomodulatory properties (Lillehoj et al. 2018; HU et al. 2025). Against the background of reduced antibiotic use in animal feed, Fermented plant-based feed additives and plant extracts have attracted increasing attention in pig nutrition because they help maintain intestinal integrity and microbial balance (ZHANG et al. 2023; LUO et al. 2026). The gut microbiota, often referred to as the “second genome” or the “forgotten functional organ,” plays a pivotal crucial in animal health, growth, development and metabolic functions (O'Hara et al. 2006; Santo et al. 2021). Maintaining the balance of the gut microbiota is essential for effective nutrient absorption and overall host health (Bischoff et al. 2011). Herbal supplements have broad application prospects due to their ability to reduce the development of bacterial resistance and improve gut microbiota. *Acer truncatum* changed tyrosine and lipid metabolism, while Clostridium butyricum enhanced immunity and altered gut microbiota (Sun et al. 2025). Dietary supplementation with 1.5% *Xiasangju* processing by-products improved egg production rate, egg weight, and yolk color. These effects may involve COX2-mediated suppression of intestinal inflammation and modulation of gut microbiota composition (Jin et al. 2026).

Previous studies have shown that *Granati Pericarpium* extract can alleviate DSS-induced colitis in mice through the IL-6/STAT3/SOCS3 pathway (Cheng et al. 2024) and exhibits good antibacterial activity against *Staphylococcus caprae* (Luo et al. 2025). *Fraxini Cortex* extract has a certain therapeutic effect on diarrhea caused by *Escherichia coli* in chickens. It can significantly increase the abundance of Bacteroidota and enhance cysteine and methionine metabolism (Wang et al. 2025). It can also alter the gut microbiota by reducing the ratio of *Akkermansiaceae* to *Lachnospiraceae*, and promote the expression of IL-10 and IL-22, thereby alleviating DSS-induced colitis in mice (Hu et al. 2025). *Sophorae Flavescentis Radix* extract suppresses LPS-induced pro-inflammatory responses in macrophages by regulating CFHR2 expression (Wu et al. 2024). Low-dose treatment significantly reduces pro-inflammatory cytokine levels. Network pharmacology and transcriptomic analyses suggest its therapeutic effect in IBD involves FoxO4, NOD-like receptor, apoptosis, and MAPK signaling pathways (Chen et al. 2025).

The HPS evaluated in this study comprised extracts of *Granati Pericarpium*, *Fraxini Cortex*, and *Sophorae Flavescentis Radix*. Previous studies have used an isolated rabbit ileum model to conduct pharmacological experiments, systematically evaluating the compound's anti-diarrheal activity. Their anti-inflammatory activity was evaluated using a xylene-induced mouse ear swelling model, while their analgesic effects were assessed using the hot-plate test and writhing test. A rat model of diarrhea was established by intragastric administration of a senna leaf decoction (0.3 g/mL). The results showed that the herbal powder supplements exhibited significant antidiarrheal, anti-inflammatory, and analgesic effects in gastrointestinal dysfunction (Yu et al. 2019). Kele pigs, an indigenous high-quality breed native to Southwest China, exhibit remarkable tolerance to humidity and environmental stress, enabling them to adapt well to the complex rearing conditions of the subtropical high-altitude agro-ecological environment of the Yunnan-Guizhou Plateau (Zhu et al. 2025). Their coats are predominantly black, although brown, mouse-gray, and “six-white or incomplete six-white” coat patterns also occur. Owing to their medium-to-large body size and high-quality meat, Kele pigs are commonly used as a raw material for ham production and represent an important industrial resource supporting local economic development (Tan et al. 2017; Zhang et al. 2025). However, the effects of HPS on the intestinal microbiota and metabolism of Kele pigs remain unclear. We therefore compared basal diets supplemented with 0.1%, 0.3%, or 0.5% HPS in finishing Kele pigs. Serum inflammatory cytokines were measured, cecal microbial communities were characterized by 16S rRNA gene sequencing, and serum metabolites were profiled using untargeted liquid chromatography-mass spectrometry (LC-MS). This study aimed to characterize physiological and metabolic responses to HPS and provide a theoretical basis for its use in finishing Kele pig production.

Materials and methods

Ethics statement

All experimental procedures including animal care and sample collection were approved and carried out in accordance with the relevant guidelines set by the Ethics Committee of the Institute of Animal

Husbandry and Veterinary, Guizhou Academy of Agricultural Sciences (Approval No. AWE-GZSXMSY-2025-24). All procedures were performed in accordance with local legislation and institutional requirements.

Experimental design and sample collection

The experiment was conducted at the Qianxi Agricultural Science and Technology Park of the Guizhou Academy of Agricultural Science (Qianxi City, Guizhou Province, China). Forty finishing Kele pigs with an initial body weight of 42.67 ± 1.73 kg were randomly assigned to four treatment groups, with 10 pigs per group. Initial body weight did not differ significantly among groups. The HPS was prepared from aqueous extracts of *Granati Pericarpium*, *Fraxini Cortex*, and *Sophorae Flavescentis Radix* at a mass ratio of 3:3:2, followed by spray drying. Dietary composition and nutrient levels are presented in Table 1. Group A received the basal diet and served as the control. Groups B, C, and D received the basal diet supplemented with 0.1%, 0.3%, and 0.5% HPS, respectively. The 35-day experiment comprised a 7-day adaptation period and a 28-day experimental period. Pigs were housed under the same conditions and had ad libitum access to feed. Vaccination and disinfection procedures followed standard farm management practices.

Table 1
Dietary composition and nutrient levels (air-dry basis)

Ingredient	Content (%)	Nutrient level	Content (%)
Corn	65.00	Digestible energy (MJ/kg)	14.83
Soybean meal	15.50	Crude protein	15.18
Wheat bran	16.00	Crude fat	3.71
Limestone	1.00	Crude fiber	3.52
Dicalcium phosphate	0.60	Calcium	0.63
Salt	0.40	Total phosphorus	0.51
L-Lysine hydrochloride	0.31	Lysine	0.98
DL-Methionine	0.06	Methionine	0.31
L-Threonine	0.08	Threonine	0.63
L-Tryptophan	0.05	Tryptophan	0.20
Premix[1]	1.00		

[1] Each kilogram of premix contained: vitamin A (VA), 8,500 IU; vitamin B_1 (VB_1), 3.5 mg; vitamin B_2 (VB_2), 5 mg; vitamin B_6 (VB_6), 6.25 mg; vitamin B_{12} (VB_{12}), 0.02 mg; vitamin D_3 (VD_3), 1,700 IU; vitamin E (VE), 40

mg; vitamin K_3 (VK_3), 1.1 mg; folic acid, 0.5 mg; niacin, 25 mg; pantothenic acid, 15 mg; biotin, 0.25 mg; Cu, 10 mg; Fe, 70 mg; Mn, 15 mg; Zn, 60 mg; Se, 0.3 mg; and I, 0.25 mg.

Major active compounds and their targets

The Traditional Chinese Medicine Systems Pharmacology database (TCMSP; https://www.tcmsp-e.com/load_intro.php?id=43) was used to identify the major active compounds in HPS and their predicted targets. Compounds were screened using oral bioavailability (OB) ≥ 30% and drug-likeness (DL) ≥ 0.18 as selection criteria.

Serum inflammatory cytokines

At the end of the experiment, blood samples were collected from the ear vein of each pig using a vacuum tube. After standing at room temperature for 30 min, the sample was centrifuged at 3000 rpm at 4 ℃ for 15 min. The supernatant was then transferred to a polypropylene centrifuge tube and stored in liquid nitrogen. The concentrations of porcine interleukin-1β (IL-1β), interleukin-4 (IL-4), interleukin-10 (IL-10), interferon-γ (IFN-γ), and transforming growth factor-β1 (TGF-β1) were measured using assay kits purchased from Nanjing Jiancheng Bioengineering Institute, according to the manufacturer's instructions.

Cecal microbial communities

After blood collection, three pigs from each group were slaughtered, and cecal fecal samples were collected. Approximately 200 mg of cecal contents was transferred into a sterile centrifuge tube, and fecal microbial DNA was extracted and used as the template. The V4 region of the 16S rRNA gene was amplified by PCR using the primers 515F: GTGCCAGCMGCCGCGGTAA, and 806R: GGACTACHVGGGTWTCTAAT. The PCR products were recovered and purified from the gel, followed by fluorescence quantification and library construction. After quantitative analysis using Qubit and qPCR, the libraries were sequenced on the NovaSeq sequencing platform by Beijing Novogene Technology Co., Ltd. The raw reads were merged using FLASH V1.2.7. All Effective Tags from all samples were clustered using the UPARSE algorithm. QIIME V1.9.1 was used to evaluate alpha diversity, beta diversity, and the taxonomy of the cecum microbiota.

Serum metabolomics analysis

Serum metabolomics analysis was performed using LC-MS. Serum sample preparation, LC-MS analysis, mass spectrometric conditions, and raw data processing were completed by Beijing Novogene Technology Co., Ltd. The raw mass spectrometry data were converted into a common data format using ProteoWizard and subsequently subjected to peak extraction, alignment, and retention-time correction using the XCMS software package. Pattern recognition was performed using multivariate statistical

analyses, including orthogonal partial least-squares discriminant analysis (OPLS-DA), to distinguish the overall differences in metabolic profiles among groups. Differential metabolites were screened according to the following criteria: VIP > 1.0, FC > 1.5 or FC < 0.667, and *P*-value < 0.05.

Data processing and analysis

Serum inflammatory cytokine levels were analyzed using one-way analysis of variance (one-way ANOVA) in SPSS 18.0, followed by Duncan's multiple range test for multiple comparisons. Pearson correlation analysis was performed between significantly differential metabolites and inflammatory cytokine levels using the online bioinformatics platform (https://www.bioinformatics.com.cn/), and a correlation matrix heatmap was generated. All results are expressed as the mean±standard deviation.

Results

Major active compounds and their targets

Major active compounds and their targets of HPS are shown in Table 2. The principal active compounds identified in the HPS included quercetin, kaempferol, luteolin, β-sitosterol, and fraxin. Key predicted targets included PTGS1, PTGS2, and IL6.

Table 2
Information on the active compounds and their targets

Component	Active constituents	Target information
Granati Pericarpium	Ellagic acid, β-sitosterol, kaempferol, catechin, luteolin, and quercetin	SLC2A4, CYP1A1, CYP1A2, ESR1, PTGS1, PTGS2, etc.
Fraxini Cortex	β-Sitosterol and fraxin	PGR, NCOA2, PTGS1, PTGS2, PRSS1, etc.
Sophorae Flavescentis Radix	Kaempferol, sophocarpine, maackiain, wighteone, formononetin, dihydroquercetin, (R)-liquiritigenin, matrine, luteolin, glyceollin, and quercetin	PTGS1, PTGS2, TNF, IL6, RELA, PPARG, etc.

Serum inflammatory cytokines

The serum IL-4 level of finishing pigs in group D was significantly higher than those in groups A ($P < 0.05$). The serum IL-10 levels of finishing pigs in groups C and D were significantly higher than those in groups A and B ($P < 0.05$). The serum TGF-β1 level in group B was significantly higher than that in the other groups ($P < 0.05$), whereas no significant difference was observed between groups C and D ($P > 0.05$). Regarding pro-inflammatory cytokines, the serum IFN-γ level in group B was significantly lower than that in the other groups ($P < 0.05$) (Table 3).

Table 3
Effects of HPS supplementation on serum inflammatory cytokines in finishing Kele pigs

Group	IL-1β(pg/mL)	IL-4(pg/mL)	IL-10(pg/mL)	TGF-β1(pg/mL)	IFN-γ(pg/mL)
Group A	58.10 ± 7.91	38.29 ± 4.99^{a}	97.86 ± 15.75^{a}	147.02 ± 30.00^{a}	31.31 ± 4.53^{c}
Group B	55.10 ± 8.72	41.12 ± 6.55^{ab}	103.40 ± 16.24^{a}	227.39 ± 27.16^{c}	20.21 ± 2.83^{a}
Group C	58.94 ± 10.64	43.27 ± 3.35^{ab}	128.46 ± 14.17^{b}	185.73 ± 20.75^{b}	26.69 ± 3.70^{b}
Group D	57.27 ± 4.35	49.12 ± 8.56^{b}	135.97 ± 18.90^{b}	180.32 ± 21.53^{b}	24.44 ± 4.16^{b}

$^{a–c}$ Different superscripts in the same column indicate a significant difference between groups ($P < 0.05$)

Cecal microbial diversity

The cecal content samples were subjected to 16S rRNA gene sequencing. After quality control and chimera removal, an average of 62,840 Effective Tags was obtained from each sample. Operational taxonomic unit (OTU) clustering was performed at a 97% sequence similarity threshold. Venn diagram analysis showed that 863 OTUs were shared between the different groups (Fig. 1). As shown in Table 4, no significant differences were observed in the Chao1, Observed species, Shannon, or Simpson indices among the groups, indicating that the overall diversity, richness, and evenness of the gut microbial communities remained stable.

Table 4
Alpha diversity indices of the cecal microbiota

Group	Chao1 index	Observed species	Shannon index	simpson index
Group A	969.08 ± 41.86	883.00 ± 41.76	5.87 ± 0.41	0.94 ± 0.02
Group B	1197.86 ± 183.57	1116.33 ± 172.37	5.99 ± 0.29	0.91 ± 0.03
Group C	1001.77 ± 54.78	945.00 ± 53.78	5.96 ± 0.59	0.92 ± 0.05
Group D	999.50 ± 36.52	953.33 ± 37.07	6.53 ± 0.25	0.96 ± 0.01

Cecal microbial composition

At the phylum level, the dominant bacterial phyla in groups A, B, C, and D were Firmicutes, Bacteroidota, Spirochaetota, Fusobacteriota, and Campylobacterota (Fig. 2A). At the genus level, the dominant bacterial genera in all groups were *Streptococcus*, *Treponema*, *Fusobacterium*, and *Lactobacillus* (Fig. 2B). These results indicate that supplementation with different levels of HPS did not alter the composition of the core cecal microbial taxa in finishing pigs at the phylum and genus levels.

To further characterize the differences among groups, a t-test was performed at the genus level. The relative abundances of *Faecalibacterium* and *Blautia* in group B were highly significantly higher than

those in group A ($P < 0.01$) (Fig. 3A). The relative abundances of *Bacteroides* and *Lachnospiraceae_AC2044_group* in group C were significantly higher than those in group A ($P < 0.05$) (Fig. 3B). The relative abundance of *Treponema* in group D was highly significantly higher than that in group A ($P < 0.01$) (Fig. 3C).

Serum metabolomic profiles

To investigate the functional outcomes associated with changes in serum inflammatory cytokines, untargeted metabolomics was performed on serum samples. Orthogonal partial least-squares discriminant analysis (OPLS-DA) showed clear separation patterns between the groups supplemented at different levels and the control group. The samples were completely separated in the score plots, while samples within the same group clustered closely (Fig. 4A, B, and C). Metabolomic analysis revealed changes in serum metabolic profiles following supplementation with different levels of HPS. The three most abundant chemical classes were Lipids and lipid-like molecules (53.92%), Organic acids and derivatives (17.16%), and Organoheterocyclic compounds (10.78%) (Fig. 4D). Based on the metabolite clustering heatmap, group D exhibited the most distinct metabolic profile and was clearly separated from the other groups (Fig. 4E). Compared with the control group, 112 differential metabolites were identified in group B, of which 58 were upregulated and 54 were downregulated; 134 differential metabolites were identified in group C, of which 56 were upregulated and 78 were downregulated; and 147 differential metabolites were identified in group D, of which 44 were upregulated and 103 were downregulated (Fig. 4F, G and H).

To elucidate the biological significance of the changes in the serum metabolome, Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway analysis was performed on the identified differential metabolites. Compared with group A, group B showed significant enrichment in the primary bile acid biosynthesis pathway, and the key metabolites identified in this group were glycocholic acid and taurine (Fig. 5A). No significantly enriched metabolic pathways were identified in groups C or D (Fig. 5B, C). Correlation analysis between the key metabolites and serum inflammatory cytokines showed that glycocholic acid was significantly negatively correlated with IFN-γ ($P < 0.05$) and significantly positively correlated with TGF-β1 ($P < 0.05$) (Fig. 6).

Discussion

Inflammatory cytokines

IL-4 is produced by activated T cells, mast cells, and basophils. It can effectively downregulate IFN-γ gene expression and interfere with IFN-γ-driven immunoglobulin class switching to IgG2a in B cells (Junttila et al. 2018). IL-10, an important immunosuppressive cytokine, is widely produced by regulatory T cells, B cells, monocytes/macrophages, and dendritic cells and plays a central role in controlling the intensity of inflammation, preventing excessive immune activation, and maintaining immune homeostasis (Eddie et al. 2017). IFN-γ is a Th1-type cytokine that can inhibit Th2 cell responses and has

therefore been used to treat IgE-mediated allergic diseases (Dong et al. 2020). In addition, the serum IFN-γ/IL-4 ratio is commonly used to evaluate the balance between Th1- and Th2-type immune responses (Kucuksezer et al. 2020). TGF-β1 is an evolutionarily highly conserved signaling molecule involved in regulating embryogenesis, tissue regeneration, homeostasis, and the induction of immune tolerance. It also plays a key role in pathological mechanisms such as fibrosis progression, tumor microenvironment remodeling, and immune evasion (Li et al. 2022). In this study, HPS supplementation altered serum cytokine concentrations, although the response differed by dose. Group B had higher TGF-β1 and lower IFN-γ concentrations than group A, whereas increases in IL-4 and IL-10 were most evident in groups D, respectively. These findings suggest that HPS may modulate anti-inflammatory signaling and the Th1/Th2 balance in finishing Kele pigs. However, the observed cytokine changes alone do not establish an improvement in whole-animal inflammatory resistance.

Cecal microbiota

The gut microbiota performs multiple functions, including regulating animal growth and development and immune stress responses. Firmicutes and Bacteroidota are the dominant bacterial phyla in the animal gut. Firmicutes possess abundant metabolic enzymes and can effectively degrade cellulose and other forms of crude fiber, whereas Bacteroidota plays important roles in degrading complex carbohydrates, producing short-chain fatty acids (SCFAs), and maintaining intestinal microbial balance (Zhou et al. 2025). In this study, Firmicutes and Bacteroidota were the dominant bacterial phyla in the cecum of both pigs supplemented with herbal supplement and control pigs, which is consistent with previous findings. *Faecalibacterium* is a beneficial, butyrate-producing genus that supplies energy to intestinal epithelial cells and supports barrier function and immune regulation (Ferreira et al. 2017). Huang et al. (2016) reported that *Faecalibacterium* supernatant ameliorated colitis in mice by suppressing IL-17A secretion from T helper 17 cells and reducing IL-6. Feng et al. (2015) found that dietary monosodium L-glutamate and fat promoted *Faecalibacterium* colonization in the porcine colon. In our study, HPS supplementation increased the relative abundance of *Faecalibacterium*, which may support intestinal barrier function and immune homeostasis. *Blautia* is an important producer of butyrate and acetate, and its reduced abundance has been associated with intestinal inflammation in children with obesity (Benítez et al. 2020). Supplementation with *Blautia* can prevent intestinal inflammation by increasing Treg differentiation and promoting SCFA production (Yang et al. 2020). *Bacteroides* primarily produces acetate and propionate and participates in the conversion of primary to secondary bile acids. HPS may stimulate *Bacteroides* growth by modulating bile acid feedback, potentially improving host lipid metabolism and anti-inflammatory immune responses (Chen et al. 2021). In this study, the relative abundances of *Blautia* and *Bacteroides* increased after HPS supplementation. These changes may increase cecal SCFA production, provide additional energy substrates, and support epithelial tight junctions and barrier function. *Treponema* was more abundant in the cecum of pigs fed 0.5% HPS. This change may reflect the combined actions of matrine and quercetin. Matrine has broad-spectrum antibacterial activity but more strongly inhibits some Gram-negative pathogens, including *Escherichia coli* (Wu et al. 2021). Such unequal inhibition could reduce competing populations and increase the ecological and nutritional niches available to *Treponema*. Polyphenols are poorly absorbed in the small

intestine and therefore reach the hindgut in substantial amounts (Pérez et al. 2013). *Treponema* species contain glycoside hydrolases and polyphenol-metabolizing enzymes that may enable them to use quercetin and its primary metabolites as carbon and energy sources (Han et al. 2025). These proposed mechanisms require direct experimental verification.

Serum metabolites

Primary bile acid biosynthesis converts cholesterol to bile acids in the liver, thereby supporting lipid digestion and absorption and maintaining cholesterol homeostasis. Free bile acids are conjugated with glycine in the cytosol to form more water-soluble conjugated bile acids, including glycocholic acid (GCA). Ge et al. (2023) found that GCA is a highly effective and safe natural anti-inflammatory substance that can significantly inhibit lipopolysaccharide-induced macrophage infiltration and the secretion of pro-inflammatory cytokines and chemokines. GCA can also upregulate farnesoid X receptor (FXR) expression at both the mRNA and protein levels, suggesting that FXR activation is a key mechanism underlying its anti-inflammatory effects. FXR was the first bile acid nuclear receptor to be identified (Makishima et al. 1999) and is mainly expressed in the liver, intestine, kidneys, adrenal glands, white adipose tissue, and immune cells (Lefebvre et al. 2009). FXR can bind to the IL-6 promoter region and downregulate IL-6 transcription (Cao et al. 2021). In addition, FXR can induce the expression of small heterodimer partner (SHP), an atypical nuclear receptor whose expression is upregulated during macrophage activation and which may participate in the development and progression of inflammatory diseases (Choi et al. 2004). The G protein-coupled bile acid receptor 5 (TGR5) is another key bile acid receptor. Bile acid activation of the TGR5-cAMP-protein kinase A axis induces phosphorylation and ubiquitination of NLRP3 inflammasome components. These modifications inhibit inflammasome activation and attenuate NLRP3-dependent inflammation (Guo et al. 2016). In this study, differential metabolites associated with HPS supplementation were enriched in primary bile acid biosynthesis. GCA is a conjugated primary bile acid that may activate FXR or TGR5 signaling and thereby modulate inflammatory cytokine expression. Yan et al. (2021) reported that Yiqi Jiemin decoction increased TGF-β1 expression in the nasal mucosa, restored the Th1/Th2 balance, reduced serum pro-inflammatory mediators, and partially reversed abnormalities in metabolites such as GCA in allergic rhinitis. Guanyu Zhixie powder contains nine active compounds, including quercetin and gallic acid. It reduced serum IL-6, TNF-α, and PGE2 and increased IL-10, IL-4, and EGF in rats. These effects were associated with changes in serum metabolites, including GCA and epinephrine, and with bile secretion and HIF-1 signaling pathways (Ma et al. 2024). In our study, serum GCA was positively correlated with TGF-β1 and negatively correlated with IFN-γ after HPS supplementation. These associations are consistent with a potential anti-inflammatory role for GCA but do not establish causality or a specific receptor-mediated mechanism.

Conclusions

Dietary supplementation with 0.1% herbal supplement can improve serum inflammatory cytokine profiles of TGF-β1 and IFN-γ in finishing Kele pigs and maintain intestinal health by increasing the abundance of

beneficial bacterial genera in the gut. These effects are associated with the modulation of specific gut bacteria, such as *Faecalibacterium*, and serum metabolites, such as glycocholic acid.

Declarations

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Authors' contributions

Xiong Zhang: Investigation, Writing-original draft, Writing editing, Funding acquisition. Xuan Wu: Data curation, Formal analysis. Chunping Zhao: Validation, Visualization. Yuxi Lu: Conceptualization, Methodology.

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Data Availability

The serum metabolites sequencing data presented in this study are available on request from the corresponding author. The intestinal microbial sequencing data are available at: https://www.ncbi.nlm.nih.gov/bioproject/PRJNA1519707.

Consent for publication

Not applicable.

Competing interests

The authors declare no competing interests.

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Figures

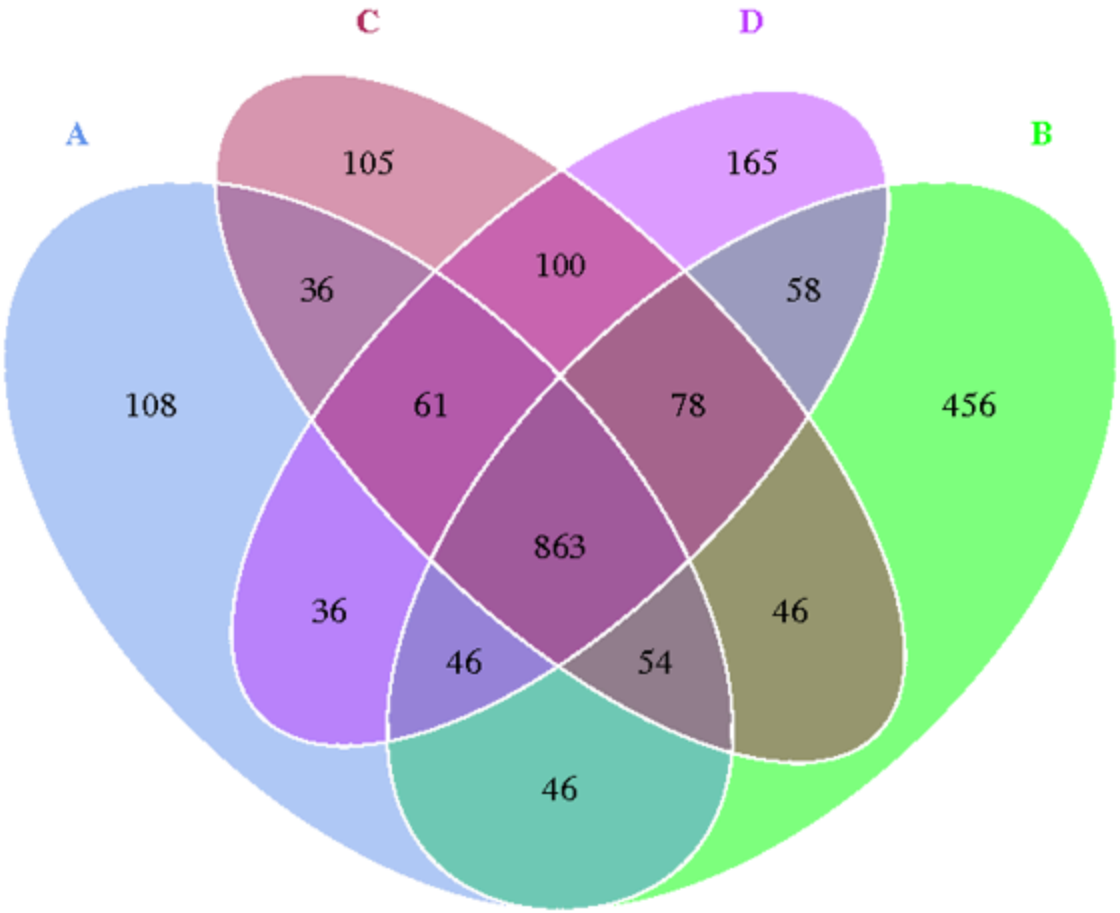


Figure 1

Venn diagram of cecal microbial OTUs

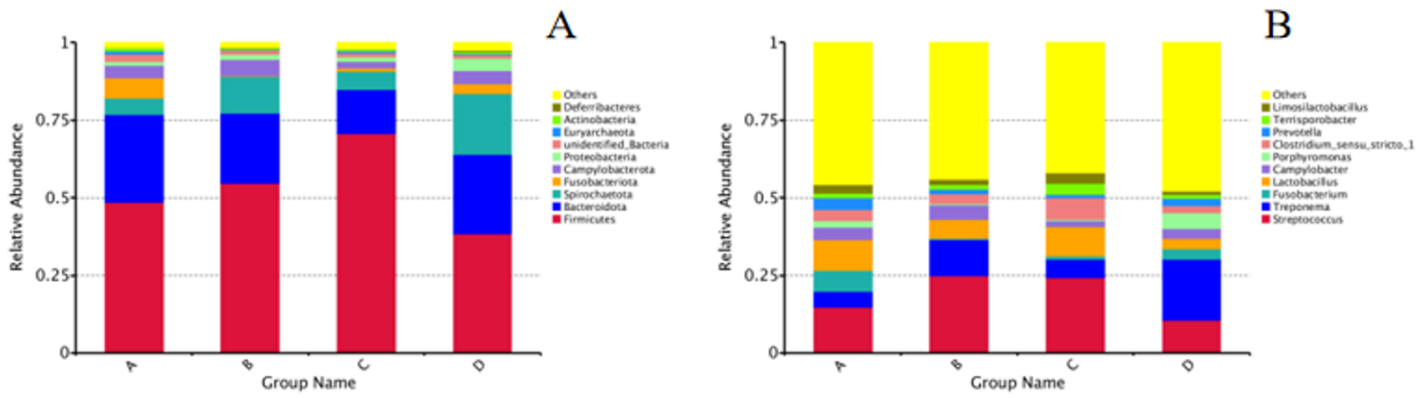


Figure 2

Cecal Microbial Composition**.** (**A**) at the phylum levels. (**B**) at the genus levels

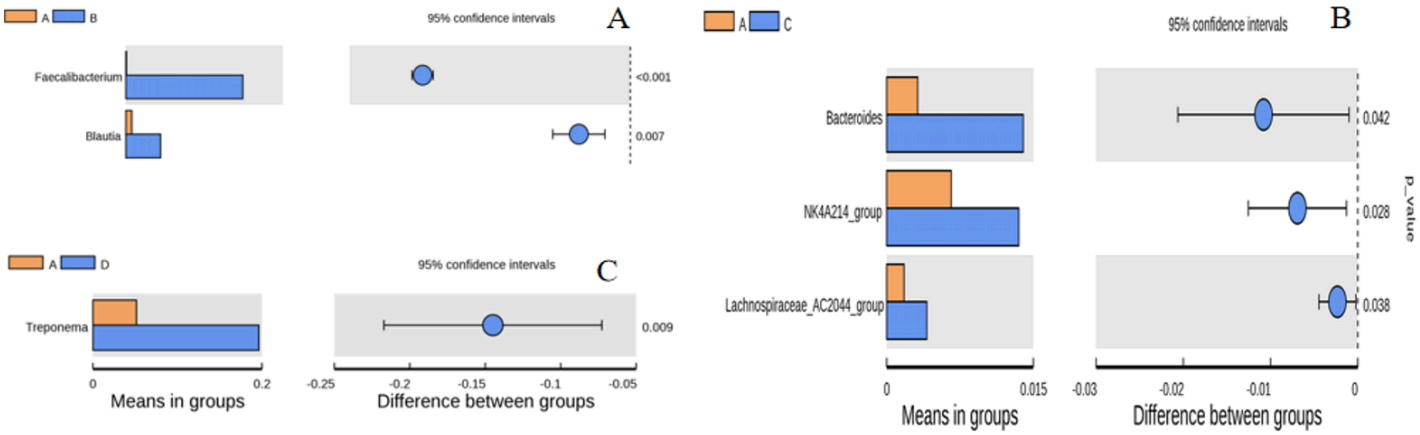


Figure 3

Differentially abundant cecal bacterial genera. (**A**) group A vs. group B t-test. (**B**) group A vs. group C t-test. (**C**) group A vs. group D t-test.

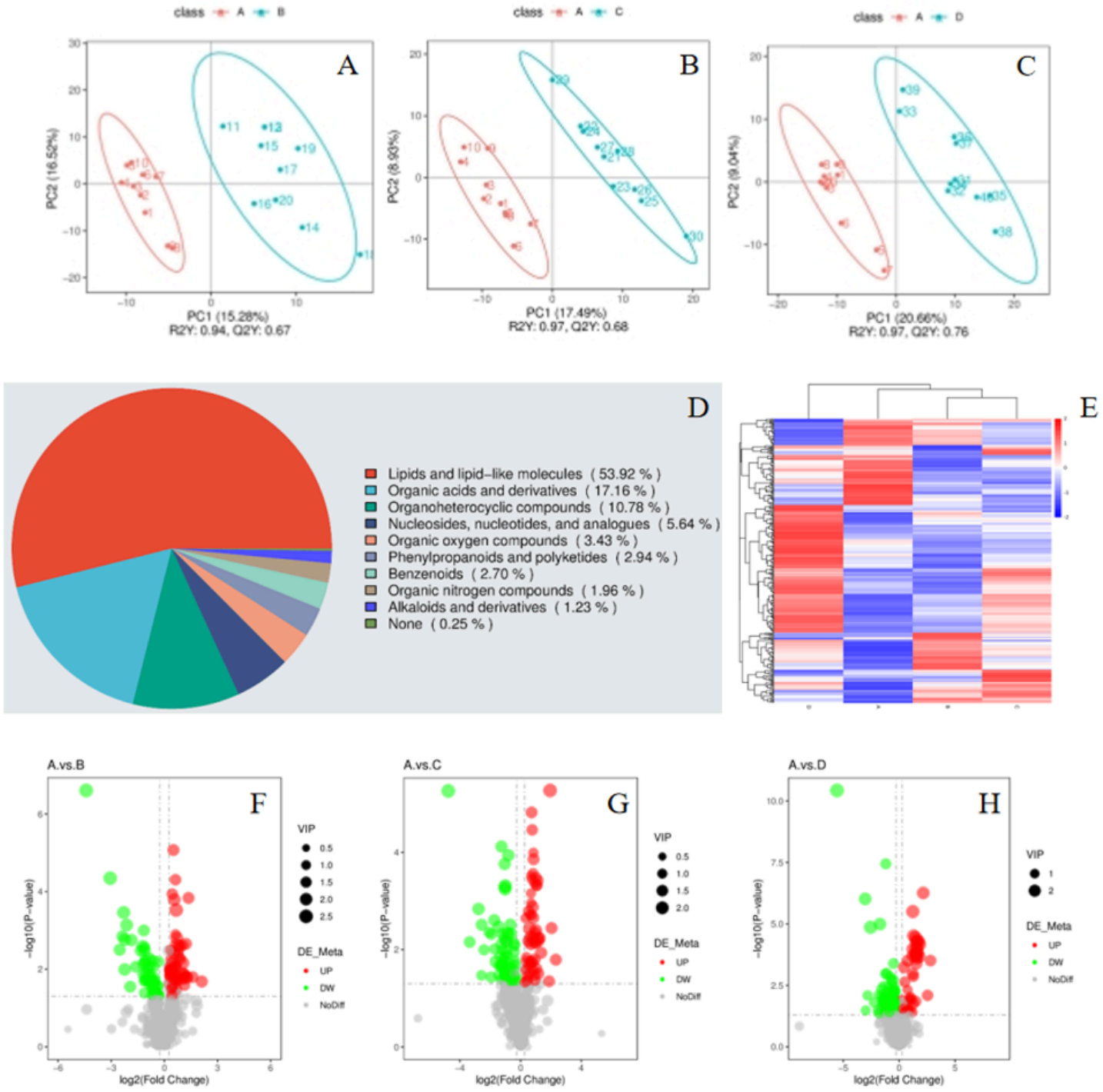


Figure 4

Metabolomic analysis results. (**A**) OPLS-DA Score Plots of Serum Samples between group A and group B. (**B**) OPLS-DA Score Plots of Serum Samples between group A and group C. (**C**) OPLS-DA Score Plots of Serum Samples between group A and group D. (**D**) metabolite classification. (**E**) heatmap of inter-group substances. (**F**) the volcano plots shows the different metabolites between group A and group B. (**G**) the volcano plots shows the different metabolites between group A and group C. (**H**) the volcano plots shows the different metabolites between group A and group D.

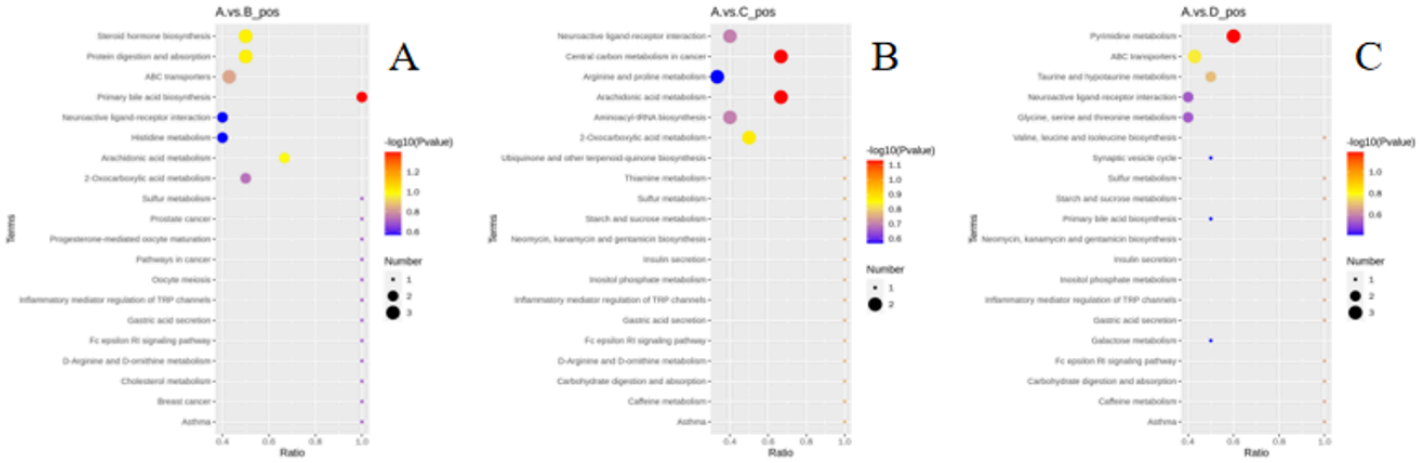


Figure 5

Metabolic pathways of differential serum metabolites. (**A**) group A vs. group B KEGG pathways. (**B**) group A vs. group C KEGG pathways. (**C**) group A vs. group D KEGG pathways.

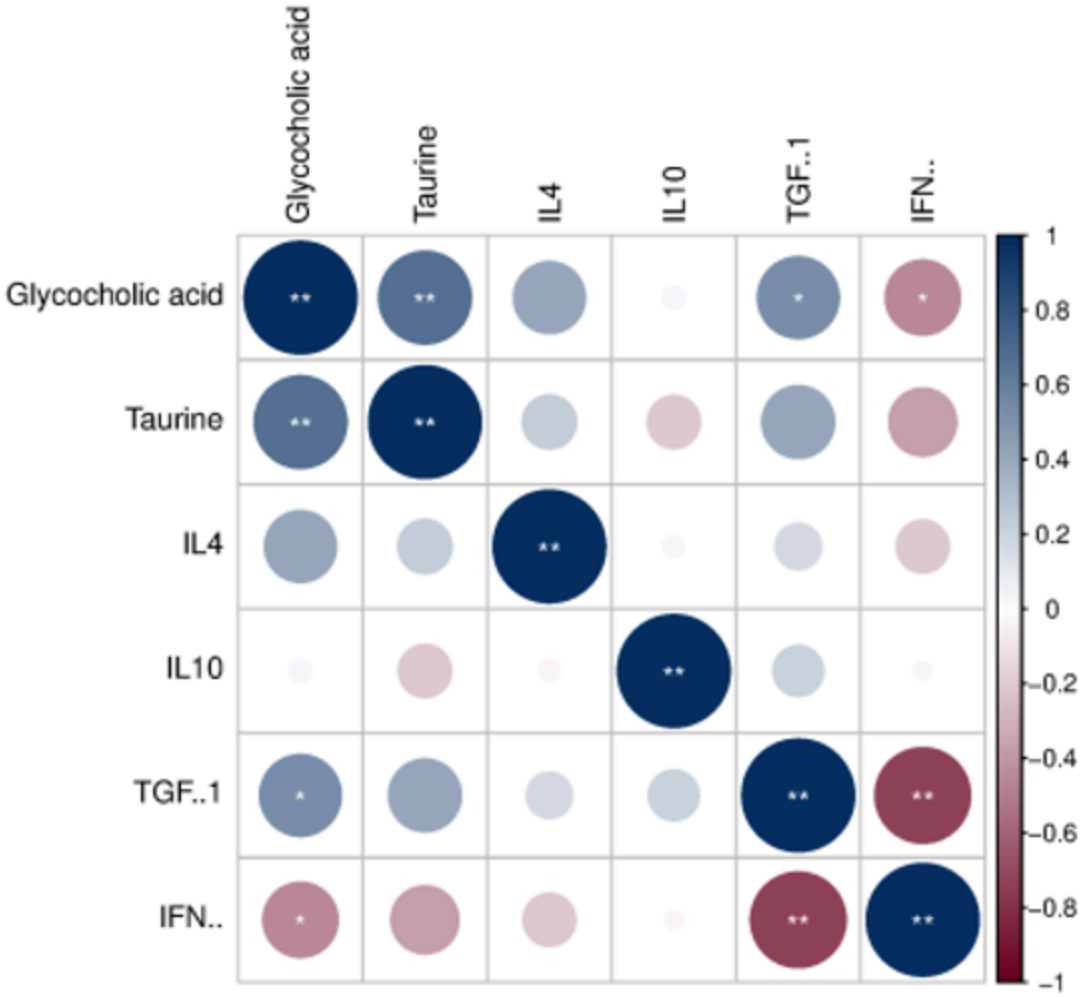


Figure 6

Correlations between differential serum metabolites and inflammatory cytokine levels