Effects of Low Birth Weight on Immune Function in Piglets and Nutritional Effects of Arginine: Postprint

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Abstract

This study aimed to investigate the effects of low birth weight (LBW) on immune function in piglets and the nutritional effects of dietary arginine supplementation. Thirty 4-day-old piglets were selected for the experiment, including 10 normal birth weight (NBW) and 20 LBW piglets. NBW piglets were fed a basal diet (Group), while LBW piglets were fed either the basal diet (Group) or the basal diet supplemented with 1.0% L-arginine (Group), with artificial milk feeding for 21 days. The results showed: 1) Compared with Group , Group piglets had significantly lower final body weight, average daily gain (ADG), and average daily feed intake (ADFI) ($P<0.05$), significantly reduced serum immunoglobulin G content ($P<0.05$), significantly decreased expression of tumor necrosis factor- , interleukin-10, and transforming growth factor- 1 in the spleen ($P<0.05$), significantly reduced expression of Toll-like receptor 2, nuclear factor- B, myeloid differentiation factor 88 (MyD88), and p38 mitogen-activated protein kinase (p38 MAPK) in the spleen ($P<0.05$), significantly decreased expression of MyD88 and p38 MAPK in the thymus ($P<0.05$), and significantly reduced expression of arginase 2 (ARG2) in the thymus ($P<0.05$). 2) Compared with Group , Group piglets showed significantly increased final body weight, ADG, and ADFI ($P<0.05$), a trend toward reduced feed-to-gain ratio ($P=0.07$), significantly elevated spleen index ($P<0.05$), a trend toward increased thymus index ($P=0.07$), significantly higher serum immunoglobulin A (IgA) content ($P<0.05$), a trend toward increased serum immunoglobulin M (IgM) content ($P=0.05$), significantly enhanced expression of interferon- (IFN-) in the spleen ($P<0.05$), and a trend toward increased expression of p38 MAPK in the spleen ($P=0.08$). In conclusion, under the conditions of this experiment, compared with NBW, LBW reduced growth performance, serum IgG content, spleen cytokine gene expression, spleen and thymus immune-related gene expression, and thymus ARG2 expression in piglets; dietary supplementation with 1.0% arginine

improved growth performance, serum IgA and IgM content, spleen index, and spleen IFN- expression in LBW piglets, indicating that dietary arginine supplementation has the effect of improving immune function in LBW piglets, with some indicators reaching the levels of NBW piglets.

Full Text

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Abstract

This study investigated the effects of low birth weight (LBW) on immune function and the nutritional effects of dietary arginine supplementation in piglets. Thirty 4-day-old piglets were selected, including ten normal birth weight (NBW) and twenty LBW piglets. NBW piglets were fed a basal diet (Group), while LBW piglets were fed either the basal diet (Group) or the basal diet supplemented with 1.0% L-arginine (Group) for 21 days using artificial milk feeding. The results showed that: 1) Compared with Group , Group piglets exhibited significantly lower final body weight, average daily gain (ADG), and average daily feed intake (ADFI) ($P<0.05$), significantly reduced serum immunoglobulin G content ($P<0.05$), significantly decreased expression of tumor necrosis factor- , interleukin-10, and transforming growth factor- 1 in the spleen ($P<0.05$), significantly downregulated expression of Toll-like receptor 2, nuclear factor- B, myeloid differentiation factor 88 (MyD88), and p38 mitogen-activated protein kinase (p38 MAPK) in the spleen ($P<0.05$), significantly reduced expression of MyD88 and p38 MAPK in the thymus ($P<0.05$), and significantly decreased arginase 2 (ARG2) expression in the thymus ($P<0.05$). 2) Compared with Group , Group piglets showed significantly higher final body weight, ADG, and ADFI ($P<0.05$), a trend toward improved feed-to-gain ratio ($P=0.07$), significantly increased spleen index ($P<0.05$), a trend toward increased thymus index ($P=0.07$), significantly elevated serum immunoglobulin A (IgA) content ($P<0.05$), a trend toward increased serum immunoglobulin M (IgM) content ($P=0.05$), significantly upregulated interferon- (IFN-) expression in the spleen ($P<0.05$), and a trend toward increased p38 MAPK expression in the spleen ($P=0.08$).

In conclusion, under the experimental conditions, LBW decreased piglet growth performance, serum IgG content, spleen cytokine gene expression, immune-

related gene expression in both spleen and thymus, and thymus ARG2 expression. Dietary supplementation with 1.0% arginine improved growth performance, serum IgA and IgM contents, spleen index, and spleen IFN- expression in LBW piglets, suggesting that arginine supplementation can improve immune function in LBW piglets, with some indices reaching levels comparable to NBW piglets.

Keywords: low birth weight; arginine; organ index; immune; cytokine

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Although advances in genetic breeding and management practices have increased litter size in sows, they have also increased the number of low birth weight piglets, with naturally occurring LBW piglets accounting for 15-20% of litters. Low birth weight significantly elevates pre-weaning mortality rates and the proportion of underdeveloped piglets, increasing both labor and capital costs in pig production. Intrauterine growth retardation (IUGR), defined as impaired embryonic or fetal development during gestation in mammals, represents the primary cause of LBW piglets. Previous studies have demonstrated that compared with normal birth weight piglets, IUGR piglets exhibit significantly reduced proportions of CD4+CD8+ double-positive cells among total T cells in the thymus, diminished peripheral blood lymphocyte responses to lipopolysaccharide (LPS) and concanavalin A (ConA) stimulation, and lower serum concentrations of immunoglobulin A (IgA), immunoglobulin G (IgG), and immunoglobulin M (IgM).

Arginine is an essential amino acid for young mammals that participates in protein synthesis and serves as a precursor for nitric oxide, polyamines, creatine, and agmatine. Circulating concentrations of arginine and its precursors citrulline and ornithine decrease significantly in suckling piglets between 7 and 14 days of age. Research indicates that a 7-day-old suckling piglet requires 2.7 g of arginine daily, while sow milk provides only 1.06 g. Dietary arginine supplementation in milk formula significantly increased plasma arginine concentrations and growth performance in suckling piglets. Under normal conditions, dietary supplementation with 0.2% and 0.8% arginine increased spleen index by 32% and 14%, respectively, in 14-day-old piglets, while 0.6% arginine supplementation increased thymus index by 150%. Under immunosuppressive conditions, arginine supplementation significantly improved spleen and thymus indices, peripheral blood leukocyte counts and lymphocyte proportions, serum IgA content, and IFN- expression. Under immune activation conditions, dietary arginine reduced serum IFN- and interleukin-6 (IL-6) contents and hepatic tumor necrosis factor- (TNF-) content, while inhibiting excessive activation of the Toll-like receptor 4 (TLR4) signaling pathway.

Given that low birth weight affects piglet immune function and arginine plays a crucial role in immune regulation, this study compared immune function between LBW and NBW piglets and evaluated the effects of dietary arginine supplementation on immune function in LBW piglets.

1.1 Experimental Animals and Design

Experimental piglets were selected from 72 newborn "Duroc × Landrace × Large White" piglets born to sows with similar body condition, parity, and farrowing date. Based on previous studies, selection criteria were established: 16 piglets with normal birth weight (1.4-1.6 kg) and 72 LBW piglets (0.8-1.0 kg) were initially identified. At 4 days of age, 30 piglets were selected according to similar body weight and consistent sex ratio, comprising ten NBW piglets and twenty LBW piglets. NBW piglets were fed the basal diet (Group), ten LBW piglets received the basal diet (Group), and the remaining ten LBW piglets received the basal diet supplemented with 1.0% L-arginine (Group). All piglets were fed artificial milk for 21 days.

1.2 Experimental Materials

L-arginine was provided by Ajinomoto (Japan) with 99% purity, and L-alanine was provided by Shanghai Yimengsi Company with 99% purity.

1.3 Experimental Diets

The experimental diet consisted of artificial milk prepared from milk replacer powder mixed with water at a ratio of 1:4 using 40°C warm water. The basal diet was formulated based on previous studies. The arginine-supplemented diet (Group) contained 1.0% L-arginine, with glucose and L-alanine added to achieve isocaloric and isonitrogenous conditions with the basal diet. The composition and nutrient levels of the basal diet are presented in Table 1 .

1.4 Management

The experiment was conducted at the research base of the Institute of Animal Nutrition, Sichuan Agricultural University. All piglets were housed individually in metabolism cages. The facility and cages were thoroughly disinfected before the experiment. Temperature was maintained at 31-32°C during the first week and reduced by 2°C weekly thereafter, with relative humidity controlled at 50-60%. Piglets were fed at 06:00, 09:00, 12:00, 15:00, 18:00, 21:00, and 24:00 daily, with feed provided ad libitum until satiation. Feed intake was recorded to calculate dry matter intake. No antibiotics were administered during the experimental period, and other management practices followed standard operating procedures.

1.5 Sample Collection and Processing

On day 22 of the experiment, piglets were weighed in the morning to record final body weight.

1.5.1 Blood Sample Collection Following weighing on day 22, fasting blood samples were collected from the anterior vena cava into glass tubes. Blood was

centrifuged at 3,500 r/min for 15 minutes to prepare serum, which was collected, labeled, aliquoted, and stored at -20°C for subsequent analysis.

1.5.2 Organ Weighing After blood collection, piglets were anesthetized and slaughtered. The abdominal cavity was opened immediately to isolate the heart, liver, spleen, lungs, kidneys, thymus, and inguinal lymph nodes. Adherent tissues were carefully removed, surface blood was absorbed with filter paper, and organs were weighed using an electronic balance.

1.5.3 Tissue Sample Collection Following organ weighing, spleen and thymus tissues were collected in cryovials, wrapped in aluminum foil, snap-frozen in liquid nitrogen, and stored at -80°C for subsequent analysis.

1.6 Measurements

1.6.1 Growth Performance Average daily gain (ADG) and average daily feed intake (ADFI) were calculated based on initial body weight, dry matter intake, and final body weight. Feed-to-gain ratio (F/G) was also calculated.

1.6.2 Organ Indices Organ indices were calculated using the formula: Organ index (g/kg) = organ weight (g) / body weight (kg).

1.6.3 Serum IgA, IgG, and IgM Contents Serum immunoglobulin concentrations were determined using enzyme-linked immunosorbent assay (ELISA) kits from Shanghai Xinle Biological Technology Co., Ltd., following the manufacturer' s instructions.

1.6.4 Spleen and Thymus Gene Expression Analysis Real-time quantitative PCR was used to measure relative expression levels of cytokine genes [including interleukin-1 (IL-1), interleukin-2 (IL-2), IL-6, interleukin-10 (IL-10), tumor necrosis factor- (TNF-), IFN- , and transforming growth factor- 1 (TGF- 1)], immune-related genes [including Toll-like receptor 2 (TLR2), TLR4, myeloid differentiation factor 88 (MyD88), nuclear factor- B (NF- B), and p38 mitogen-activated protein kinase (p38 MAPK)], and arginine metabolism genes [including inducible nitric oxide synthase (iNOS), endothelial nitric oxide synthase (eNOS), arginase 1 (ARG1), and arginase 2 (ARG2)].

Total RNA was extracted using Trizol Reagent (TaKaRa, Japan) according to the manufacturer' s protocol. RNA quality was assessed using a nucleic acid-protein detector (Beckman DU-800, CA, USA) at 260 nm, with A260/A280 ratios of 1.8-2.0 indicating good RNA purity. cDNA synthesis was performed using a Prime Script™ reagent kit (TaKaRa, Japan), and products were stored at -20°C. Gene sequences were obtained from the National Center for Biotechnology Information (NCBI), and primers were designed using Primer 5 software and synthesized by Shanghai Sangon Biotech Co. Ltd. Primer sequences are listed in Table 2 . Real-time quantitative PCR was performed using an ABI7900HT

Real-Time PCR System (ABI, USA) with SYBR Green I (TaKaRa, Japan) as the fluorescent dye. The reaction mixture (10.0 μL) contained 5.0 μL SYBR Premix Ex Taq™ II (2×), 0.5 μL forward primer, 0.5 μL reverse primer, 3.0 μL double-distilled water, and 1.0 μL cDNA template. After screening three commonly used reference genes [-actin, glyceraldehyde-3-phosphate dehydrogenase (GAPDH), and 18S ribosomal RNA (18S rRNA)], -actin and GAPDH were selected as reference genes for spleen and thymus, respectively. Relative gene expression was calculated using the 2-ΔΔCT method.

1.7 Statistical Analysis

Experimental data were initially organized using Excel 2010 and subsequently analyzed using SPSS 21.0 software with independent samples t-tests for pairwise comparisons. Significance between Group and Group was denoted as P1, between Group and Group as P2, and between Group and Group as P3. Differences were considered significant at $P<0.05$ and trends at $0.05 P<0.10$. Results are expressed as "mean ± standard error."

2.1 Effects of Low Birth Weight on Growth Performance and Nutritional Effects of Arginine

As shown in Table 3 , compared with Group , Groups and exhibited significantly lower final body weight, ADG, ADFI, and F/G ($P<0.05$). Compared with Group , Group showed significantly higher final body weight, ADG, and ADFI ($P<0.05$), with a trend toward improved F/G ($P=0.07$).

2.2 Effects of Low Birth Weight on Organ Indices and Nutritional Effects of Arginine

As shown in Table 4 , compared with Group , Group showed no significant differences in heart, liver, spleen, lung, kidney, thymus, or inguinal lymph node indices ($P>0.10$), while Group exhibited significantly higher thymus and inguinal lymph node indices ($P<0.05$). Compared with Group , Group showed significantly increased spleen index ($P<0.05$) and a trend toward higher thymus index ($P=0.07$).

2.3 Effects of Low Birth Weight on Serum Immunoglobulin Content and Nutritional Effects of Arginine

As shown in Table 5 , compared with Group , Group displayed significantly reduced serum IgG content ($P<0.05$), while Group showed significantly increased serum IgA and IgM contents ($P<0.05$) but significantly decreased IgG content ($P<0.05$). Compared with Group , Group exhibited significantly elevated serum IgA content ($P<0.05$) and a trend toward increased IgM content ($P=0.05$).

2.4 Effects of Low Birth Weight on Cytokine Gene Expression in Spleen and Thymus and Nutritional Effects of Arginine

As shown in Table 6 , compared with Group , Group demonstrated significantly decreased expression of IL-10, TNF- , and TGF- 1 in the spleen (P<0.05), while Group showed significantly increased spleen IFN- expression (P<0.05) and significantly decreased spleen TGF- 1 expression (P<0.05). Group exhibited a trend toward increased thymus IL-2 expression (P=0.08), while Group showed significantly decreased thymus IL-10 expression (P<0.05) and trends toward reduced thymus IL-1 (P=0.08) and IL-6 expression (P=0.08). Compared with Group , Group displayed significantly increased spleen IFN- expression (P<0.05) and trends toward decreased thymus IL-10 (P=0.06) and TGF- 1 expression (P=0.09).

2.5 Effects of Low Birth Weight on Immune-Related Gene Expression in Spleen and Thymus and Nutritional Effects of Arginine

As shown in Table 7 , compared with Group , Group exhibited significantly reduced expression of TLR2, NF- B, p38 MAPK, and MyD88 in the spleen (P<0.05), while Group showed significantly decreased spleen NF- B expression (P<0.05). Group demonstrated significantly lower thymus p38 MAPK and MyD88 expression (P<0.05) with a trend toward reduced thymus NF- B expression (P=0.09), while Group showed significantly decreased thymus expression of TLR4, NF- B, p38 MAPK, and MyD88 (P<0.05). Compared with Group , Group exhibited a trend toward increased spleen p38 MAPK expression (P=0.08) and a trend toward decreased thymus TLR4 expression (P=0.06).

2.6 Effects of Low Birth Weight on Arginine Metabolism Gene Expression in Spleen and Thymus and Nutritional Effects of Arginine

As shown in Table 8 , compared with Group , Group showed a trend toward decreased spleen iNOS expression (P=0.07) and a trend toward increased spleen ARG1 expression (P=0.09), while Group exhibited trends toward decreased spleen iNOS (P=0.08) and ARG2 expression (P=0.07). Group demonstrated significantly reduced thymus ARG2 expression (P<0.05) and a trend toward decreased thymus iNOS expression (P=0.07), while Group showed significantly decreased thymus iNOS, ARG1, and ARG2 expression (P<0.05) with a trend toward reduced thymus eNOS expression (P=0.06). Compared with Group , Group showed no significant differences in iNOS, eNOS, ARG1, or ARG2 expression in either spleen or thymus (P>0.10).

3.1 Effects of Low Birth Weight on Growth Performance and Nutritional Effects of Arginine

Low birth weight impairs piglet growth performance, negatively impacting the swine industry. He et al. reported that IUGR piglets exhibited significantly lower weaning weight at 21 days and ADG compared with normal birth weight

piglets. Our results showed that Group had significantly lower final body weight, ADG, ADFI, and F/G compared with Group , consistent with previous findings and confirming that LBW reduces growth performance. Arginine is an essential amino acid for young mammals; research indicates that sow milk contains insufficient arginine, with each 7-day-old piglet requiring 2.7 g daily while receiving only 1.06 g from milk. Circulating arginine and its precursors decline significantly in suckling piglets aged 7-14 days. Kim et al. demonstrated that arginine supplementation in milk formula enhanced plasma arginine content and growth performance. Wang et al. showed that 0.6% arginine supplementation in IUGR piglets improved growth performance. Our findings that Group had significantly higher final body weight, ADG, and ADFI than Group align with these studies, confirming that arginine promotes growth in LBW piglets. However, Group still had significantly lower final body weight, ADG, and ADFI than Group , indicating that 1.0% arginine supplementation did not fully restore LBW piglet growth performance to NBW levels.

3.2 Effects of Low Birth Weight on Organ Indices and Nutritional Effects of Arginine

Organ indices represent key biological characteristics that reflect functional capacity. Wiyaporn et al. found no significant differences in liver, spleen, or kidney indices between LBW and normal birth weight piglets at birth and 7 days of age. Hu et al. reported similar findings for heart, liver, spleen, and kidney indices in 28-day-old IUGR piglets. Our results showing no significant differences in organ indices between Groups and confirm that LBW does not significantly affect organ development. Tan et al. demonstrated that dietary arginine at 0.2% and 0.8% increased spleen index by 32% and 14%, respectively, while 0.6% arginine increased thymus index by 150% in 14-day-old piglets. Our finding that Group had significantly higher spleen index and a trend toward increased thymus index compared with Group supports these results, indicating that arginine promotes spleen and thymus development in LBW piglets. Notably, Group showed significantly higher thymus and inguinal lymph node indices than Group , suggesting that arginine supplementation elevated thymus development in LBW piglets to NBW levels.

3.3 Effects of Low Birth Weight on Serum Immunoglobulin Content and Nutritional Effects of Arginine

Serum immunoglobulins are primary antibodies mediating humoral immunity, with IgG being the most abundant. Zhong Xiang reported that 7-day-old IUGR piglets had 20.8% lower serum IgG content than normal birth weight piglets. Our results showing significantly reduced serum IgG in Group compared with Group confirm that LBW compromises humoral immune function. Arginine dosage affects immunoglobulin secretion in LBW piglets. Fan Miao found that dietary arginine at 250, 375, and 500 mg/kg increased serum IgA content by 33.33%, 58.10%, and 33.33%, respectively, in 21-day-old piglets, with only the

250 mg/kg dose increasing IgG content by 17.68%. Tan et al. reported that 0.6-0.8% arginine increased serum IgM content by 150-200% at 14 days and by 62-91% at 21 days, while only 0.2% arginine increased IgG content by 12% at 21 days, with higher doses providing no additional benefit. Our finding that Group had significantly higher serum IgA and a trend toward increased IgM compared with Group aligns with these studies, demonstrating that arginine enhances humoral immune function by promoting immunoglobulin secretion, though optimal dosage requires further investigation. Group showed significantly higher serum IgA and IgM but lower IgG than Group , indicating that arginine supplementation restored IgA and IgM levels to NBW standards but not IgG.

3.4 Effects of Low Birth Weight on Cytokine Gene Expression in Spleen and Thymus and Nutritional Effects of Arginine

Cytokines are small proteins synthesized and secreted by immune cells (monocytes, macrophages, T cells, B cells, NK cells, etc.) upon stimulation, representing crucial components of the immune system. TNF- , secreted by mast cells, macrophages, and T cells, recruits neutrophils and macrophages to infection sites, enhancing pathogen clearance. IL-10, produced by Th2 cells, macrophages, and mast cells, inhibits Th1 cell activation. TGF- 1, secreted by T cells and monocytes, interferes with naive T cell differentiation, suppresses macrophage activation and effector T cell proliferation, and reduces pro-inflammatory effects. IL-2, primarily produced by Th1 cells, induces activation of cytotoxic T cells, macrophages, and NK cells. Studies have shown that compared with normal birth weight piglets, newborn IUGR piglets have significantly lower serum IL-10 content and trends toward reduced TNF- , with significantly decreased TNF- and IL-10 expression in the ileum. However, other studies report increased IL-6 expression in the colon and elevated TNF- expression in the proximal small intestine of IUGR piglets. Our results showing significantly decreased spleen IL-10, TNF- , and TGF- 1 expression, with a trend toward increased thymus IL-2 expression in Group , confirm that LBW affects cytokine expression, though effects vary by tissue. The spleen and thymus are rich in T cells, which mediate cellular immunity and regulate cytokine secretion. Lin et al. found that 1-day-old IUGR piglets had significantly lower proportions of CD4+CD8+ double-positive T cells in the thymus. Dong et al. reported reduced blood CD4/CD8 ratios but increased spleen CD8 and thymus CD4 expression in IUGR piglets. Limited research exists on T cells in LBW piglet spleen and thymus, and the relationship between abnormal cytokine secretion and T cell populations requires further investigation. Qu Hongyan demonstrated that arginine supplementation increased serum IFN- content reduced by cyclophosphamide. Han Jie showed that arginine improved cyclophosphamide-induced reductions in leukocyte count, lymphocyte proportion, and IFN- content. IFN- , primarily secreted by Th1 cells, activates macrophages and exhibits antiviral, antitumor, and immunomodulatory activities under strict transcriptional regulation. Our finding that Group had significantly increased spleen IFN- expression and trends toward decreased

thymus IL-10 and TGF- 1 compared with Group supports these results, indicating that arginine improves cytokine expression in LBW piglet spleen. Group showed significantly higher spleen IFN- , lower spleen TGF- 1, reduced thymus IL-10, and trends toward decreased thymus IL-1 and IL-6 compared with Group , demonstrating that arginine supplementation improved spleen IFN- and thymus IL-2 expression in LBW piglets.

3.5 Effects of Low Birth Weight on Immune-Related Gene Expression in Spleen and Thymus and Nutritional Effects of Arginine

Toll-like receptors are pattern recognition receptors that identify pathogen-associated molecular patterns and activate NF- B and MAPK signaling pathways, playing vital roles in immune defense. TLR2 recognizes peptidoglycan, lipopeptides, and lipoproteins, forms heterodimers with other Toll-like receptors, and signals through the MyD88-dependent pathway. Upon ligand binding, the intracellular Toll domain recruits adaptor molecules including MyD88, which interacts with IL-1 receptor-associated kinase (IRAK) and TNF receptor-associated factor 6 (TRAF6) to activate NF- B and induce secretion of immune-related cytokines such as IL-2, TNF- , and IFN- . p38 MAPK, a member of the MAPK family, activates transcription factors including ATF-2, NF-AT, and NF- B when phosphorylated, thereby promoting IFN- secretion. Zhong et al. reported significantly reduced NF- B protein content in the jejunum and ileum of 7-day-old IUGR piglets. Our results showing significantly decreased spleen TLR2, NF- B, MyD88, and p38 MAPK expression, along with reduced thymus p38 MAPK and MyD88 expression and a trend toward lower thymus NF- B expression in Group , confirm that NF- B and p38 MAPK signaling pathways are impaired in LBW piglet spleen and thymus. Arginine can modulate immune signaling pathways to improve piglet immune function. Chen et al. demonstrated that arginine supplementation inhibited excessive activation of the TLR4-MyD88 pathway in spleen, liver, and inguinal lymph nodes during immune challenge. Li et al. showed that arginine inhibited the liver TLR4-NF- B pathway to alleviate damage caused by Escherichia coli. Our finding that Group had a trend toward increased spleen p38 MAPK expression and decreased thymus TLR4 expression compared with Group aligns with these studies, suggesting that arginine modulates immune-related gene expression in LBW piglet spleen and thymus, though whether improved p38 MAPK expression can enhance spleen immune function requires further investigation. Group showed significantly lower spleen NF- B expression and significantly reduced thymus TLR4, NF- B, p38 MAPK, and MyD88 expression compared with Group , indicating that arginine supplementation restored spleen TLR2, MyD88, and p38 MAPK expression to NBW levels.

3.6 Effects of Low Birth Weight on Arginine Metabolism Gene Expression in Spleen and Thymus and Nutritional Effects of Arginine

Nitric oxide is an important endogenous signaling molecule involved in immune regulation. Arginine is the sole precursor for nitric oxide synthesis via nitric oxide synthase (NOS). Two major NOS isoforms exist: eNOS produces physiological nitric oxide levels, while iNOS is induced by bacterial endotoxins, IL-1 , and TNF- to generate large amounts of nitric oxide. Arginase catalyzes arginine conversion to ornithine and urea, representing an alternative arginine utilization pathway that competitively inhibits NOS activity. Two arginase isoforms exist: ARG1 is primarily expressed in the liver, while ARG2 is expressed in various tissues at different levels. NOS and arginase are regulated by Th1 and Th2 cytokines, with Th1 cytokines inducing NOS expression and Th2 cytokines inducing arginase expression. Our results showing trends toward decreased spleen iNOS and increased spleen ARG1 expression, along with reduced thymus iNOS and significantly decreased thymus ARG2 expression in Group , indicate that LBW reduces arginine catalytic utilization in the thymus. Dietary arginine can modulate arginine metabolism in vivo. Huang et al. reported that 0.8% arginine supplementation significantly increased duodenal nitric oxide content in 14-day-old piglets and jejunal NOS activity and plasma ornithine content in 21-day-old piglets, while 0.4% arginine increased plasma ornithine and jejunal spermidine content. Our finding that arginine supplementation did not significantly affect ARG1, ARG2, iNOS, or eNOS expression in spleen or thymus differs from previous studies, possibly due to differences in dosage, duration, or experimental conditions. Group showed trends toward decreased spleen iNOS and ARG2 expression, significantly reduced thymus iNOS, ARG1, and ARG2 expression, and a trend toward lower thymus eNOS expression compared with Group , indicating that arginine supplementation did not improve arginine metabolism gene expression in the thymus of LBW piglets.

Conclusions

1. Compared with NBW piglets, LBW piglets exhibited significantly reduced growth performance, serum IgG content, spleen expression of IL-10, TNF- , TGF- 1, TLR2, NF- B, p38 MAPK, and MyD88, and thymus expression of p38 MAPK, MyD88, and ARG2.
2. Dietary supplementation with 1.0% arginine significantly improved growth performance, serum IgA content, spleen index, and spleen IFN- expression in LBW piglets.
3. Dietary supplementation with 1.0% arginine improved immune function in LBW piglets, with some indices reaching levels comparable to NBW piglets.

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