https://doi.org/10.17221/65/2024-CJAS

Effects of tryptophan supplementation on performance, intestinal morphology and protein abundance of tight junction protein and indoleamine 2, 3-dioxygenase in weaned pigs

Pei Mao[1], Naizhi Hu[1], Mengmeng Gao[1], Lingping Zhao[1], Chengsheng Song[2], Wenfeng Ma[1],*

[1]*College of Animal Science and Technology, Henan University of Science and Technology, Luoyang, China*

[2]*Henan Spring Agriculture and Animal Husbandry Technology, Luoyang, China*

**Corresponding author: a113boy@163.com*

Citation: Mao P., Gao M., Zhao L., Song C., Ma W. (2024): Effects of tryptophan supplementation on performance, intestinal morphology and protein abundance of tight junction protein and indoleamine 2, 3-dioxygenase in weaned pigs. Czech J. Anim. Sci., 69: 493–501.

Abstract: This study determined the effect of tryptophan (Trp) supplementation on the protein abundance of intestinal tight junction and indoleamine 2, 3-dioxygenase (IDO) in weaned pigs. Ninety-six White × Landrace × Duroc piglets (5.31 ± 0.54 kg) were selected in a growth trial and the experiment lasted for 30 days. The basal diet based on corn and soybean contained Trp at 2.4 g/kg. The dietary treatments consisted of a basal diet or a basal diet + 500 mg/kg Trp. On the 31st day, 12 pigs (1 pig per pen) were randomly selected and slaughtered in order to collect samples for subsequent analysis. Dietary supplementation with Trp improved the villus height and protein abundance of Zonula occludens protein 1 (ZO-1) in the duodenum and the jejunum ($P < 0.05$), increased the protein abundance of Claudin-1 in the duodenum ($P < 0.05$) and IDO expression in the ileum ($P < 0.05$), and reduced the urea nitrogen concentration in the serum and the ZO-1 protein abundance of the ileum in weaned pigs ($P < 0.05$).

Keywords: essential amino acid; intestinal protein expression; performance; piglets

Tryptophan (Trp) has been demonstrated to be the second or third most important limiting amino acid for weaned pigs and takes a role in regulating growth, behavior and immune response (Eder 2001; Ettle et al. 2004; Guzik et al. 2006; Ma et al. 2020; Chen et al. 2024). The proven alteration in the main route of Trp catabolism is in the kynurenine pathway, in which the amount of metabolised Trp along the kynurenine pathway accounts for about 95% of the total dietary Trp intake by the participation of the two activated enzymes, which includes Trp-2, 3-dioxygenase (TDO) and

Supported by the the National Natural Science Foundation of China (grant number 31702122) and the National Key Research and Development Program of China (grant number 2018YFD0500603).

https://doi.org/10.17221/65/2024-CJAS

indoleamine 2, 3-dioxygenase (IDO) (Metz et al. 2012; Wirthgen et al. 2013; He et al. 2024; Zhao et al. 2024). It has been proposed that the TDO is only present in the liver, while the IDO is widely expressed in a variety of tissues, and that the simulation of IDO contributes to catalysing the indole epoxidative cleavage of Trp molecules (Okamoto et al. 2007; Wirthgen et al. 2013). The detection of the expression of IDO in the intestine can directly reflect the metabolism of Trp in pigs (Le et al. 2008; Zhao et al. 2024).

Tight junction proteins are also important indicators of intestinal barrier function. These proteins include the proteins of Claudin, Zonula occludens protein 1 (ZO-1) and Occludin (Hu et al. 2013; Wang et al. 2015; Zheng et al. 2021). Several studies have shown that some types of amino acids are preferentially absorbed by the small intestine and play an important role in promoting intestinal morphology and the expression of intestinal tight junction proteins (Wang et al. 2015; Yang and Liao 2019). However, limited studies have been conducted on the effect of Trp on intestinal tight junction proteins in weaned pigs. Therefore, this study evaluated the effects of Trp supplementation on the performance and the protein abundance of the intestinal tight junction and IDO in weaned pigs.

MATERIAL AND METHODS

Animals and experimental design

Ninety-six weaned pigs were randomly divided into two groups, the basal diet (BD) was fed a basal diet, and the experimental group (BD + Trp) was fed a basal diet supplemented with 500 mg/kg Trp (Table 1). The piglets (average weight of 5.31 ± 0.54 kg) were placed in 12 pens with 4 castrated males and 4 females per pen. The experiment lasted for 30 days. On the first 5 days, pigs were allowed a commercial diet before the start of the trial and this was then progressively switched to the experimental diet for the next 5 days. The nutrient concentration of the basal diet was designed according to the nutrition recommendations of the National Research Council (NRC 2012). The basal diet was prepared using corn and soybean meal as the main feed materials. The utilisation efficiency of the crystalline amino acids was considered to reach 100%.

Table 1. Ingredient composition of the basal diet (g/kg, as-fed)

Ingredients	Basal diet
Corn	682.0
Soybean meal	140.0
Fish meal	100.0
Whey powder	30.0
Soybean oil	15.0
Limestone	7.0
Dicalcium phosphate	12.0
Salt	3.0
Vitamin-mineral premix[1]	5.0
L-Lysine HCl	3.2
DL-Methionine	1.2
L-Threonine	1.2
L-Tryptophan[2]	0.4
Calculated values	
DE (Mcal/kg[3])	14.06
Chemical analysis	
Crude protein	188.4
Isoleucine	7.3
Lysine	13.0
Methionine + cysteine	7.7
Threonine	8.5
Tryptophan	2.4
Valine	8.6

[1]Premix provided the following per kg of complete diet for weaned pigs: vitamin A (15 000 IU); vitamin D3 (4 000 IU); vitamin E (50 IU); vitamin K3 (2.5 mg); thiamine,) (1.5 mg); riboflavin (5 mg); niacin (30 mg); pantothenic acid (13.5 mg); pyridoxine (5 mg); folacin (0.7 mg); biotin (45 µg); cobalamin vitamin B12 (30 µg); choline chloride (500 mg); Mn (60 mg); Fe (75 mg); Zn (75 mg); Cu (100 mg); I (0.3 mg); Se (0.3 mg); [2]L-Tryptophan was added at 0.5 g/kg of the diet in place of corn; [3]digestible energy (DE) content of the diets was calculated using energy values for the ingredients obtained from NRC (2012)

Feeding and management

Before the experiment, all the pens and equipments in pig houses were sterilised. Each pen (2.4 × 3.2 m^2) was equipped with a pacifier drinker and a stainless-steel automatic feeder so that the piglets had free access to water and feed. The pig house was kept to the temperature between 26 °C

https://doi.org/10.17221/65/2024-CJAS

with 28 °C, and the humidity to be led between 60% with 70%. At the end of the experiment, the body weight of pigs were recorded to calculate the average daily feed intake (ADFI), average daily gain (ADG) and ADFI/ADG.

Samples collection

After overnight fasting for 12 h on the 30th day, 12 pigs (1 pig per pen) were randomly selected and slaughtered for sample collection. The pigs were sacrificed according to the requirements of the Animal Welfare Committee of Henan University of Science and Technology. Sampling of the intestine and organs were collected for further analysis.

Blood samples were determined primarily by jugular vein puncture (1 pig per pen) and placed in a 5 ml heparin-free vacuum tube (Becton Dickinson Vacutainer, Franklin Lakes, NJ, USA) and then stored on ice for 20 min. After centrifugation at 2 500 × g for 5 min (Biofuge 22R; Heraeus, Hanau, Germany), the serum sample was collected and stored at –20 °C for analysis. The small intestine specimen samples were removed from the mesentery and then placed on ice. Two copies were taken of 2 cm long segments from the middle duodenum, the middle jejunum and the middle ileum (the entire digestive tract without chyme). Additionally, one set of segments was fixed with 10% neutral buffer formalin for histological analysis and the other copy of the intestine was rinsed with phosphate buffered saline (PBS) and placed into three sterile Eppendorf tubes frozen with liquid nitrogen, and immediately stored at –80 °C for subsequent protein abundance measurements.

Chemical determination

The crude protein (CP) content of the diet was determined experimentally using AOAC (2023) (Association of Official Analytical Chemists) methodology. The amino acid (AA) content in the feed, excluding tryptophan, was analysed using an automatic amino acid analyser (LA8080; HITACHI, Tokyo, Japan). Specifically, dietary tryptophan (Trp) was determined through alkaline hydrolysis at 120 °C for a duration of 16 h, utilising High-Performance Liquid Chromatography (E2695; Waters, Mass, USA).

Intestinal morphology analysis

Each segment of the intestine was paraffin-embedded and then cut into a series of 5 μm sections. Six discontinuous sections were selected for hematoxylin and eosin (H&E) staining, and the intestinal morphology was photographed under the microscope (CK-40; Olympus, Tokyo, Japan). Each section of the 6 well-oriented villi (as the distance between the crypt openings and the end of the villi) and its corresponding crypt (from the crypt-villus junction to the base of the crypt) was observed for analysis using an Image Dissector (Lucia Software; v3, Lucia, Kostelec nad Orlicí, Czech Republic). With each pig as the unit, the detection data were finally presented as the average value per pig.

Western blot measurement

Western blotting was detected for the protein abundance of IDO, Claudin-1, Occludin and ZO-1 in intestinal homogenate samples. The intestinal sample frozen in liquid nitrogen was ground to a powder and dissolved in a RIPA buffer of pH 7.4. Next, a protease inhibitors cocktail obtained from Apply Gene (Beijing, China) was added to this. The protein level was measured using the BCA Protein Detection Kit (Pierce, Rockford, IL, USA). A 30 μg of protein content was extracted using a pipette gun and electrophoresed by sodium dodecyl sulfate (SDS) polyacrylamide gels. The protein was transferred to a polyvinylidene fluoride membrane (Millipore, Bedford, MA, USA) and sealed in 5% skimmed milk powder at 4 °C for 1 h and then incubated with diluted primary antibodies against IDO, Claudin-1, ZO-1, and Occludin (Santa Cruz Biotechnology, Santa Cruz, CA, USA). After careful washing, anti-rabbit IgG conjugated with 1 : 7 000 horseradish peroxidase was then incubated with these proteins at room temperature for 1 h. Western band densities were measured by the Western blot luminance reagent (Santa Cruz Biotechnology, Santa Cruz, CA, USA) using Glyceraldehyde 3-phosphate dehydrogenase (GAPDH) in each lane as a loading and

https://doi.org/10.17221/65/2024-CJAS

then quantified by the AlphaImager 2200 (Alpha Innotech, San Leandro, CA, USA).

Serum indicators

Several biochemical indicators in serum were assayed using a CX-4 blood biochemistry analyser (Beckman Coulter, Inc., Fullerton, CA, USA), including glucose (GLU), triglycerides (TG), total protein (TP), cholesterol (CHO), albumin (ALB), aspartate aminotransferase (AST), and alanine aminotransferase (ALT). Serum urea nitrogen (SUN) levels were assayed using a diacetyl oxime colorimetric assay kit (Nanjing Jiancheng Bioengineering Institute, Nanjing, China).

Statistical analyses

The experimental data were analysed using the *t*-test procedure of Statistical Analysis System (SPSS v21.0; SPSS Inc., Chicago, IL, USA). The pen was the experimental unit for the analysis of pig performance, while other data were analysed with a pig as the experimental unit. The results are presented as the least square means and those where $P < 0.05$ was considered statistical significance.

RESULTS

Growth performance

The growth performance of the pigs was presented in Table 2. There was no significant difference in growth performance for weaned pigs that was correlated with dietary supplementation with Trp ($P > 0.05$).

Table 2. Effect of dietary tryptophan (Trp) supplementation on performance of weaned pigs

Items	Treatments		SEM	*P*-value
	BD	BD + Trp		
ADFI (g)	457	454	4.79	0.127
ADG (g)	287	299	10.4	0.833
ADFI/ADG	1.59	1.52	0.058	0.395

ADG = average daily gain; ADFI = average daily feed intake; BD = basal diet; each pen served as an experimental unit and data are means of six pens per treatment

Table 3. Effect of dietary tryptophan (Trp) supplementation on the serum urea nitrogen and metabolite concentrations of weaned pig

Items	Treatments		SEM	*P*-value
	BD	BD+Trp		
SUN (mmol/l)	3.76[a]	2.56[b]	0.267	0.017
ALT (U/l)	67.2	71.5	6.85	0.174
ALB (g/l)	21.1	23.2	0.97	0.156
AST (U/l)	86.1	91.5	8.15	0.262
CHO (mmol/l)	3.14	2.95	0.791	0.462
GLU (mmol/l)	2.77	2.99	0.87	0.819
TP (g/l)	47.5	49.4	3.86	0.154
TG (mmol/l)	120	126	8.04	0.453

[a,b]Means in the same column indicate significant difference between groups ($P < 0.05$); each pen served as an experimental unit and data are means of six pens per treatment

ALT = alanine aminotransferase; AST = aspartate aminotransferase; ALB = albumin; BD = basal diet; CHO = cholesterol; GLU = glucose; SEM = standard error mean; SUN = serum urea nitrogen; TP = total protein; TG = triglyceride

Serum parameters

Serum metabolites and SUN concentration were presented in Table 3. The Trp supplementation decreased the SUN content of pigs ($P < 0.05$). Regarding the other serum metabolites, the results observed in this study were not significant ($P > 0.05$).

Table 4. Effect of dietary tryptophan (Trp) supplementation on intestinal morphology of weaned pigs

Items	Treatments		SEM	*P*-value
	BD	BD + Trp		
Villous height (μm)				
Duodenum	435[b]	478[a]	9.99	0.022
Jejunum	396[b]	464[a]	16.9	0.029
Ileum	334	337	19.8	0.925
Crypt depth (μm)				
Duodenum	381	373	13.7	0.686
Jejunum	316	322	14.1	0.783
Ileum	290	286	20.2	0.887
Villous height: crypt depth				
Duodenum	1.15	1.29	0.072	0.208
Jejunum	1.26	1.45	0.096	0.221
Ileum	1.17	1.21	0.121	0.837

[a,b]Means in the same column indicate significant difference between groups ($P < 0.05$); each pen served as an experimental unit and data are means of six pens per treatment; BD = basal diet; SEM = standard error mean

https://doi.org/10.17221/65/2024-CJAS

Intestinal morphology

The results of the intestinal morphology were shown in Table 4. The Trp supplementation increased the villous height of the duodenum and jejunum ($P < 0.05$). Regarding the other intestinal morphology parameters, the results observed in this study were not significant ($P > 0.05$).

Protein abundance of tight junction

The Western blot analysis data showed the presence of Claudin-1, Occludin and ZO-1 in the small intestine (Figure 1). The inclusion of Trp increased the protein abundance of ZO-1 in the duodenum and the jejunum ($P < 0.05$), and also improved the

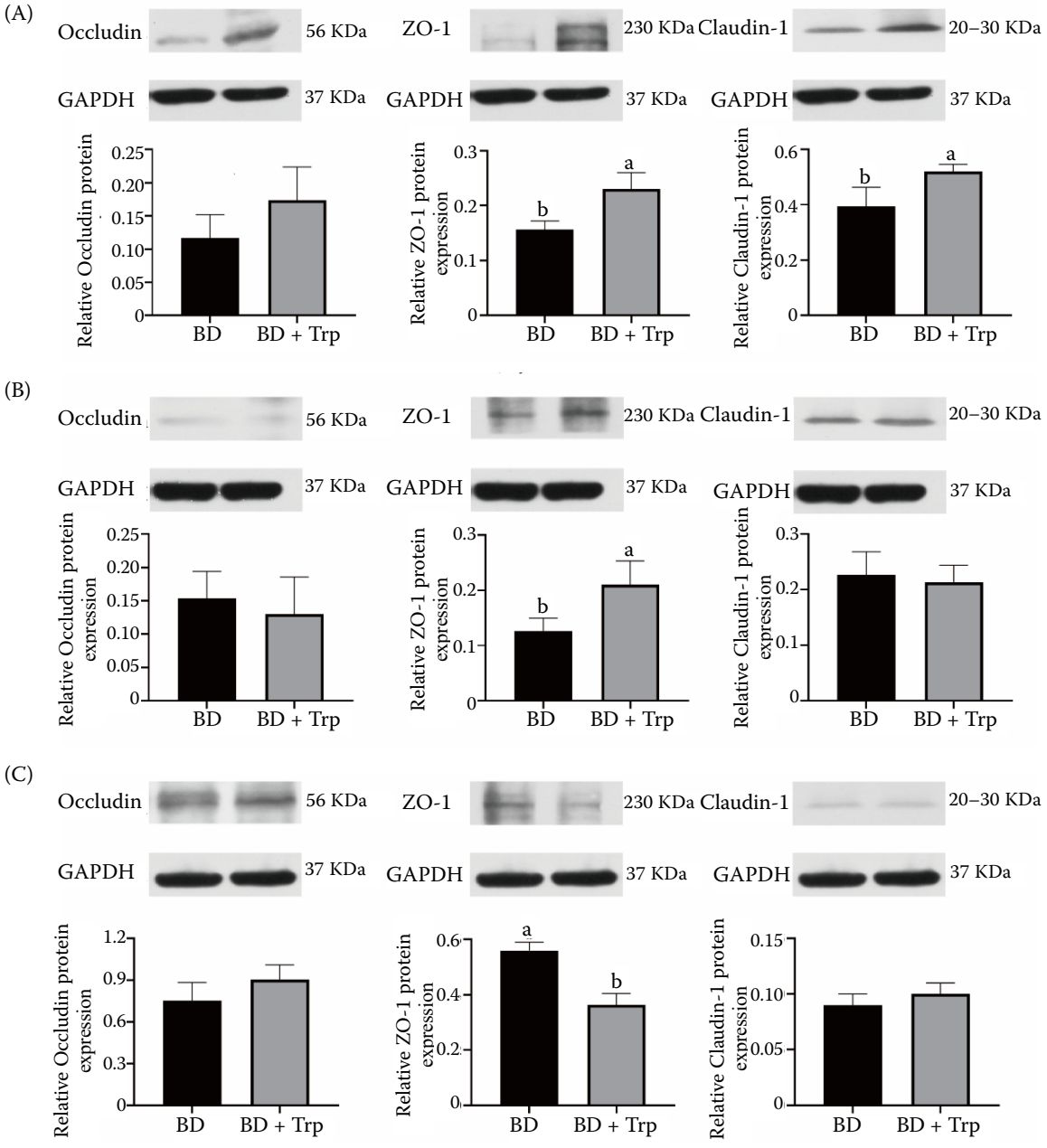

Figure 1. Western blot analysis of the proteins Occludin, ZO-1 and Claudin-1 obtained from a whole homogenised section of small intestine segments obtained from piglets fed a basal diet (BD) or BD + Trp for 30 days after weaned ($n = 6$)
(A) Occludin, ZO-1 and Claudin-1 protein expression level in the duodenum; (B) Occludin, ZO-1 and Claudin-1 protein expression level in the jejunum; (C) Occludin, ZO-1 and Claudin-1 protein expression level in the ileum
[a,b]Means indicate significant difference between groups ($P < 0.05$); GAPDH was used as an internal standard to normalise the signal; GAPDH = Glyceraldehyde 3-phosphate dehydrogenase

https://doi.org/10.17221/65/2024-CJAS

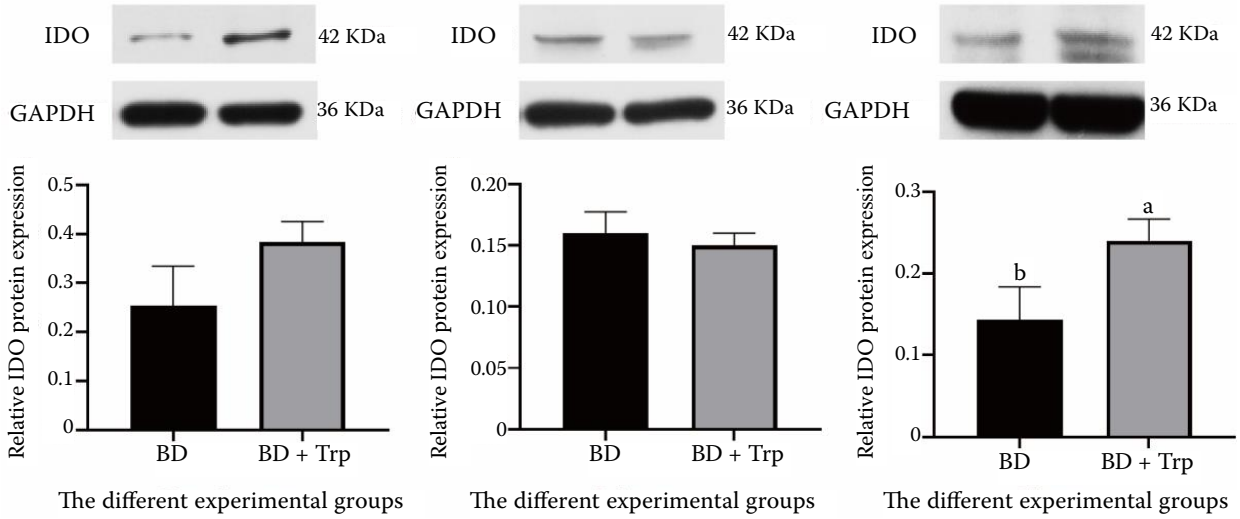

Figure 2. Western blot analysis of the IDO protein obtained from a whole homogenized section of small intestine segments obtained from piglets fed a basal diet (BD) or BD + Trp for 30 days after weaned (n = 6)

[a,b]Means indicate significant difference between groups ($P < 0.05$); GAPDH was used as an internal standard to normalise the signal; GADPH = Glyceraldehyde 3-phosphate dehydrogenase

Claudin-1 protein abundance in the duodenum ($P < 0.05$). However, the protein abundance of ileal ZO-1 was decreased by the dietary Trp supplementation ($P < 0.05$).

Protein abundance of intestinal indoleamine 2,3-dioxygenase

The intestinal protein abundance of IDO in pigs is presented in Figure 2. The protein abundance of IDO in the ileum was increased by the dietary Trp supplementation ($P < 0.05$). Regarding the IDO protein abundance in the duodenum and the jejunum, the results observed in this study were not significant ($P > 0.05$).

DISCUSSION

The Trp is closely involved in regulating the voluntary feed intake, protein synthesis and immune response in animals (Han et al. 1993; Ma et al. 2019; Seo et al. 2023). Studies have shown that Trp is one of the most important limiting amino acids in weaned pigs and also plays an important role in influencing the intestinal development (Liu et al. 2017). The promotion of the villi height and villi height/crypt depth represented an improvement in the intestinal barrier, while the increase in crypt depth reflected a decrease in the maturation rate in small intestinal epithelial cells (Hedemann et al. 2006; Yi et al. 2018). In this study, an improvement in the villi height was obtained by Trp supplementation, which might indicate that the dietary inclusion of Trp can promote the intestinal morphology development in weaned pigs.

The effect of Trp supplementation on serum biochemical metabolites was also evaluated in this study. It was found that the dietary inclusion of Trp only decreased the SUN level in weaned pigs. The SUN concentration was demonstrated to be inversely correlated with the nitrogen utilisation efficiency of animals (Jayaraman et al. 2017; Yoo et al. 2018), and the results of this study might indicate that a moderate addition of Trp further improves the utilisation of nutrients in weaned pigs, which is consistent with the conclusions of the previous studies (Han et al. 1993; Ma et al. 2019).

In addition, the tight junction protein is a key component of the intestinal barrier and works alongside the epithelial cells in regulating the transportation of ions, solutes and water molecules across the intestinal epithelium between adjacent cells (Hu et al. 2013; Wang et al. 2015; Yang et al. 2015). It also play a key role in maintaining a normal permeability and thus preventing pathogens and microorganisms from entering the epithelia (Yang et al. 2015). Tight junctions

https://doi.org/10.17221/65/2024-CJAS

are mainly composed of transmembrane proteins and cytoplasmic proteins, including the Claudin family and Occludin, which are structural proteins constituting a selective barrier (Wang et al. 2016). Additionally, cytoplasmic proteins such as ZO-1 usually assume the role of connecting membrane proteins and the cytoskeleton, and the increase in ZO-1 expression predominantly reflects the decrease in parenteral cell permeability (Zhang et al. 2009). The weaned of piglets often causes damage to the intestinal barrier, resulting in a decline in growth. Therefore, nutritional means can be adopted in order to improve the protein abundance of tight junctions, which can help to restore intestinal health in weaned piglets (Hedemann et al. 2006; Trevisi et al. 2009). In this study, dietary supplementation with Trp increased the abundance of ZO-1 in the duodenum and the jejunum, and decreased the abundance of ZO-1 in the ileum, which indicated that the amount of Trp in this study decreased the permeability of the duodenum and jejunum and increased the ileum permeability.

It has been proposed that altered Trp metabolism through the kynurenine pathway may be an important contributor to the inflammatory response or the simulation of IDO expression (Trevisi et al. 2009). The absorption utilisation efficiency of Trp is also strongly associated with the distribution of IDO enzymes in the intestine (Fujigaki et al. 2001), which can be used as a key modulator to maintain both the homeostasis and the physiological status of pigs (Trevisi et al. 2009). Indoleamine 2,3-dioxygenase is the only rate-limiting enzyme in the intestine for catalysing the Trp catabolism along the kynurenine pathway (Okamoto et al. 2007; Wirthgen et al. 2013). The metabolic status of Trp and the IDO expression are also closely associated with intestinal immunity. In the present study, the dietary supplementation of Trp increased the IDO expression in the ileum, which might reflect the idea that the moderate addition of Trp regulated the immune function of ileum. Although some research has been conducted on the expression of IDO, the role of specificity in IDO activation is not yet completely clear. It is recommended that young pigs are a suitable model animal species for IDO research, in order to provide a basis for further research on immunoregulatory IDO functions for human-based study (Ristagno et al. 2013; Wirthgen et al. 2013). Therefore, the mechanism surrounding the simulation and distribution of IDO needs to be studied further for clarification.

In summary, we found that on the basis of meeting the recommended amount of Trp in dietary nutrition standards, further addition of Trp increased tight junction proteins expression and intestinal morphology parameter associated with no adverse effect on performance, indicating that further appropriate inclusion of Trp could improve the intestinal barrier function of weaned pigs.

CONCLUSION

Under normal physiological conditions, a moderate increase in dietary Trp improved villus height and the protein abundance of ZO-1 in the duodenum and the jejunum, increased the protein abundance of Claudin-1 in the duodenum and IDO expression in the ileum, and reduced the serum urea nitrogen level and the expression of ZO-1 in the ileum of weaned pigs.

Acknowledgement

The authors wish to express their sincere gratitude to all of their colleagues who were involved in this research for their assistance.

Conflict of interest

The authors declare no conflict of interest.

REFERENCES

AOAC – Association of Official Analytic Chemists. Official methods of analysis. 17th ed. Arlington, VA, USA: Association of Official Analytic Chemists; 2003.

Chen C, Hu H, Li Z, Qi M, Qiu Y, Hu Z, Feng F, Tang W, Diao H, Sun W, Tang Z. Dietary tryptophan improves growth and intestinal health by promoting the secretion of intestinal β-defensins against enterotoxigenic Escherichia coli F4 in weaned piglets. J Nutr Biochem. 2024;129:109637.

Eder K, Peganova S, Kluge H. Studies on the tryptophan requirement of piglets. Arch Anim Nutr. 2001 Jan; 55(4):281-97.

https://doi.org/10.17221/65/2024-CJAS

Ettle T, Roth FX. Specific dietary selection for tryptophan by the piglet. J Anim Sci. 2004 Apr;82(4):111521.

Fujigaki S, Saito K, Sekikawa K, Tone S, Takikawa O, Fujii H, Wada H, Noma A, Seishima M. Lipopolysaccharide induction of indoleamine 2,3-dioxygenase is mediated dominantly by an IFN-γ-independent mechanism. Eur J Immunol. 2001 Jul;31(8):2313-8.

Guzik AC, Matthews JO, Kerr BJ, Bidner TD, Southern LL. Dietary tryptophan effects on plasma and salivary cortisol and meat quality in pigs. J Anim Sci. 2006 Aug; 84(8):2251-9.

Han Y, Chung TK, Baker DH. Tryptophan requirement of pigs in the weight category 10 to 20 kilograms. J Anim Sci. 1993 Jan;71(1):139-43.

Hedemann M.S, Jensen BB, Poulsen HD. Influence of dietary zinc and copper on digestive enzyme activity and intestinal morphology in weaned pigs. J Anim Sci. 2006 Dec; 84(12):3310-20.

He T, Li C, Chen Q, Li R, Luo J, Mao J, Yang Z. Combined analysis of lncRNA and mRNA emphasizes the potential role of tryptophan-mediated regulation of muscle development in weaned piglets by lncRNA. J Anim Sci. 2024 Jan 3;102:skae264.

Hu CH, Xiao K, Luan ZS, Song J. Early weaning increases intestinal permeability, alters expression of cytokine and tight junction proteins, and activates mitogen-activated protein kinases in pigs. J Anim Sci. 2013 Mar;91(3): 1094-101.

Jayaraman B, Htoo JK, Nyachoti CM. Effects of different dietary tryptophan: Lysine ratios and sanitary conditions on growth performance, plasma urea nitrogen, serum haptoglobin and ileal histomorphology of weaned pigs. Anim Sci J. 2017 May;88(5):763-71.

Le Floch N, Melchior D, Seve B. Dietary tryptophan helps to preserve tryptophan homeostasis in pigs suffering from lung inflammation. J Anim Sci. 2008 Dec;86(12):3473-9.

Liu W, Mi S, Ruan Z, Li J, Shu X, Yao K, Jiang M, Deng Z. Dietary tryptophan enhanced the expression of tight junction protein ZO-1 in intestine. J Food Sci. 2017 Jan; 82(2):562-7.

Ma W, Mao P, Guo L, Qiao S. Crystalline amino acids supplementation improves the performance and carcass traits in late-finishing gilts fed low-protein diets. Anim Sci J. 2020 Dec;91(1):e13317.

Ma W, Mao P, Fan W, Zhu Y, Guo L. Valine and isoleucine supplementation improve performance and serum biochemical concentrations in growing gilts fed low-protein diets. Can J Anim Sci. 2019 Jul;99(4):921-8.

Metz R, Rust S, DuHadaway JB, Mautino MR, Munn DH, Vahanian NN, Link CJ, Prendergast GC. IDO inhibits a tryptophan sufficiency signal that stimulates mTOR: A novel IDO effector pathway targeted by D-1-methyl-tryptophan. OncoImmunology. 2012 Dec;1(9):1460-8.

NRC. Nutrient Requirements of Swine, 11th ed. National Research Council. National Academy Press; Washington, DC, USA. 2012. 420p.

Okamoto T, Tone S, Kanouchi H, Miyawaki C, Ono S, Minatogawa Y. Transcriptional regulation of indoleamine 2,3-dioxygenase (IDO) by tryptophan and its analogue. Cytotechnology. 2007 Jun;54(2):107-13.

Ristagno G, Fries M, Brunelli L, Fumagalli F, Bagnati R, Russo I, Staszewsky L, Masson S, Li Volti G, Zappala A, Derwall M, Brucken A, Pastorelli R, Latini R. Early kynurenine pathway activation following cardiac arrest in rats, pigs, and humans. Resuscitation. 2013 Nov; 84(11):1604-10.

Seo SK, Kwon B. Immune regulation through tryptophan metabolism. Exp Mol Med. 2023 Jul;55(7):1371-9.

Trevisi P, Melchior D, Mazzoni M, Casini L, De Filippi S, Minieri L, Lalatta-Costerbosa G, Bosi P. A tryptophan-enriched diet improves feed intake and growth performance of susceptible weanling pigs orally challenged with Escherichia coli K88. J Anim Sci. 2009 Jan;87(1):148-56.

Wang B, Wu Z, Ji Y, Sun K, Dai Z, Wu G. L-Glutamine enhances tight junction integrity by activating CaMK kinase 2-AMP-activated protein kinase signaling in intestinal porcine epithelial cells. J Nutr. 2016 Mar; 146(3):501-8.

Wang H, Zhang C, Wu G, Sun Y, Wang B, He B, Dai Z, Wu Z. Glutamine enhances tight junction protein expression and modulates corticotropin-releasing factor signaling in the jejunum of weanling Piglets. J Nutr. 2015 Jan;145(1):25-31.

Wirthgen E, Tuchscherer, M, Otten W, Domanska G, Wollenhaupt K, Tuchscherer A, Kanitz E. Activation of indoleamine 2,3-dioxygenase by LPS in a porcine model. Innate Immun. 2013 Apr;20(1):30-9.

Yang Y, Li W, Sun Y Han F, Hu CAA, Wu Z. Amino acid deprivation disrupts barrier function and induces protective autophagy in intestinal porcine epithelial cells. Amino Acids. 2015 Oct;47(10):2177-84.

Yang Z, Liao S F. Physiological effects of dietary amino acids on gut health and functions of swine. Front Vet Sci. 2019 Jun;6:169.

Yi D, Li B, Hou Y, Wang L, Zhao D, Chen H, Wu T, Zhou Y, Ding B, Wu G. Dietary supplementation with an amino acid blend enhances intestinal function in piglets. Amino Acids. 2018 May;50:1089-100.

Yoo SH, Hong JS, Yoo HB, Han TH, Jeong JH, Kim YY. Influence of various levels of milk by-products in weaner diets on growth performance, blood urea nitrogen, diarrhea incidence, and pork quality of weaning to finishing pigs. Asian-Australas J Anim Sci. 2018 Nov;31(5):696-704.

https://doi.org/10.17221/65/2024-CJAS

Zhang B, Guo Y. Supplemental zinc reduced intestinal permeability by enhancing occludin and zonula occludens protein-1 (ZO-1) expression in weaning piglets. Br J Nutr. 2009 Sep;102(5):687-93.

Zhao X, Pang J, Zhang W, Peng X, Yang Z, Bai G, Xia Y. Tryptophan metabolism and piglet diarrhea: Where we stand and the challenges ahead. Anim Nutr. 2024 Mar; 17:123-33.

Zheng L, Duarte M E, Sevarolli Loftus A, Kim S W. Intestinal health of pigs upon weaning: Challenges and nutritional intervention. Front Vet Sci. 2021 Aug;8:628258.

Received: April 30, 2024
Accepted: November 5, 2024
Publihed online: December 20, 2024