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GROWTH PERFORMANCE AND BLOOD SERUM PARAMETERS OF PIGLETS USING DIETS WITH DIFFERENT RATIOS OF STANDARDIZED ILEAL DIGESTIBLE ISOLEUCINE TO LYSINE

SUMMARY

Isoleucine is a branched-chain essential amino acid in pig nutrition that regulates protein metabolism, fat deposition, and glucose utilisation, and serves as an energy substrate for immune protein synthesis, thereby enhancing immune function. It also improves growth performance, feed intake, and nutrient utilisation. However, the optimal standardized ileal digestible (SID) isoleucine:lysine ratio for young pigs remains controversial. This study aimed to determine the optimal SID isoleucine:lysine ratio for pigs weighing 7–30 kg. Four groups of piglets were fed diets with SID isoleucine:lysine ratios of <0.5, 0.5, 0.55, and 0.6. The results demonstrated that a ratio of 0.55 was optimal, increasing live weight at 70 days of age by 8.8% ($p<0.05$) without adverse physiological effects. This ratio reduced serum glucose, creatinine, urea, total cholesterol, low-density lipoprotein cholesterol, and triglycerides, while increasing total protein and high-density lipoprotein cholesterol and decreasing ALT and AST activity. A ratio of 0.5 was less effective but still increased live weight by 6.2% ($p<0.05$) and decreased serum glucose, creatinine, urea, and triglycerides, with a concomitant rise in total protein. In contrast, increasing the ratio to 0.6 did not improve body weight and altered protein and lipid metabolism, as evidenced by increased total protein and globulins, decreased albumins and albumin-to-globulin ratio, elevated cholesterol due to higher low-density lipoproteins, and reduced ALT and AST activity.

Keywords: SID isoleucine:lysine, pigs, live weight, protein, triglycerides, glucose

INTRODUCTION

Excessive nitrogen emissions from pigs raised on modern farms have detrimental effects through their contribution to acidification and eutrophication

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of sensitive ecosystems, as well as odour emissions (Almeida *et al*., 2024). Reducing crude protein in pig diets has been shown to reduce these negative effects by improving nitrogen use efficiency, positively affecting both performance and animal health, and reducing environmental impacts (Chae *et al*., 2024). However, this feeding strategy should be combined with an adequate supply of amino acids in quantity and quality to meet the genetic potential of the animals for protein deposition (Adhikari *et al.,* 2025). Until now, NRC (2012) has been the guideline for ensuring adequate amino acid requirements in pig diets. However, recent studies on various animals (Sychov *et al*., 2022; Plyska *et al*., 2021; Shkarban and Sychov 2025; Sychov *et al*., 2025) have shown that the standardized ileal digestibility (SID) ratio of amino acids to lysine has changed significantly over the past two decades, while some publications on pigs have reported contradictory results (Kwon *et al.,* 2022; Goodarzi *et al.,* 2022; Habibi *et al*., 2022a; Zong *et al*., 2022). Therefore, a more precise study is necessary, considering each factor that affects the amino acid requirements (especially the limited amino acids) in pig feeding.

Among the essential amino acids, tryptophan, valine, isoleucine, and methionine play important roles in several biological functions of porcine metabolism. In particular, isoleucine belongs to the group of essential branched-chain amino acids (BCAA), which are interdependent amino acids that affect the incorporation rate of other limiting amino acids. Isoleucine is a branched-chain amino acid that is essential for pigs and can regulate protein metabolism, fat deposition and glucose utilisation in the body (Nie *et al*., 2018; Le Couteur *et al*., 2020; Zhao *et al.,* 2024). In addition, isoleucine serves as an important energy substrate and is required for the synthesis of immune proteins (Gu *et al.,* 2019). Previous studies have shown that isoleucine can improve growth parameters, feed intake and utilisation, and relative intestinal length and morphology in pigs (Wang *et al.,* 2024). In addition, it can regulate intestinal immune function by increasing the secretion of β-defensins through the activation of the Sirt1/ERK/90RSK signalling pathway (Ren *et al.,* 2016; Zhang *et al*., 2024).

Given the numerous functions that isoleucine performs in the body of pigs, over the past decades, many scientists have been devoted to establishing the optimal SID ratio of isoleucine:lysine in their diet. In particular, Goodarzi P. *et al.* (2022) recommend an optimal SID Ile:Lys ratio of 0.30–0.55 for pigs with a live weight of up to 25 kg. However, in subsequent studies, the authors increased the recommended SID Ile:Lys ratio to 0.29–0.60 (Goodarzi *et al*. 2023).

According to Habibi M. *et al.* (2022a), to maintain the normal physiological state of the pigs' body, it is necessary to maintain the SID Ile:Lys ratio at the level of 0.31–0.55. While Soumeh E. A. *et al.* (2014) and Nørgaard J. V. *et al.* (2013) recommend a SID Ile:Lys of 0.42–0.62, and Nørgaard J. V. and Fernández J. A. (2009) recommend 0.53–0.62. At the same time, Clark A. B. *et al.* (2016) suggest an even higher SID Ile:Lys ratio of 0.40–0.63 to maintain optimal growth parameters in pigs. According to Soumeh E.A. *et al.* (2014), for piglets with a body weight ranging from 11 to 25 kg, the optimal SID Ile:Lys

ratio in diets is between 0.42 and 0.62, whereas Barea R. *et al.* (2009) recommend a ratio of 0.46 to 0.65. For piglets with a body weight of 15–30 kg, Lazzeri D. B. *et al.* (2017) consider the optimal SID Ile:Lys ratio to be 0.45–0.73, and according to Waguespack, A. M. *et al.* (2012) SID Ile:Lys for young pigs 20–45 kg should be 0.52–0.61. For pigs with a live weight of 25 to 50 kg, Clizer D. A. *et al.* (2022) established an optimal SID Ile:Lys ratio of 0.55–0.75. However, Kerkaert H.R. and colleagues (2021) suggest a lower SID Ile:Lys ratio for pigs of this body weight, ranging from 0.61 to 0.66.

Summarising and generalising the data described in the literature, the optimal SID Ile:Lys ratio in pigs up to 11 kg live weight is between 0.58 and 0.65 (Chae *et al,* 2024). These higher requirements compared to the NRC recommendation (2012) and several articles in the literature (Htoo *et al,* 2014; Soumeh *et al,* 2014; Clark *et al,* 2017) can be explained by the previous assessment related to genetic improvement and excessive inclusion of leucine or valine in the piglet diet. For piglets with a live weight of 11 to 25 kg, the optimal SID Ile:Lys ratio ranged from 0.55 to 0.61, and for young pigs with a live weight of 25 to 50 kg, it ranged from 0.55 to 0.58 (Chae *et al,* 2024). These results are also higher than those reported in the literature (Waguespack *et al,* 2012; Gloaguen *et al,* 2013; Nørgaard *et al,* 2013; Lazzeri *et al,* 2017) regarding the needs of the SID Ile:Lys ratio for pigs. Thus, despite a fairly large number of studies on determining the optimal SID Ile:Lys ratio in the diet of pigs with different body weights, i.e. for different periods of growth, the established values are contradictory and fluctuate in a fairly wide range. Therefore, this study aimed to establish the optimal SID Ile:Lys ratio in the diet of pigs aged 24 to 70 days with a body weight of 7–30 kg, which would provide the necessary growth parameters and maintain the normal physiological state of their body.

MATERIAL AND METHODS

Ethics. Experiments with experimental animals were performed following the rules of the European Convention for the Protection of Vertebrate Animals (Official Journal of the European Union L276/33, 2010) and the Order of the Ministry of Economy of Ukraine «On approval of the requirements for the welfare of farm animals during their housing» of February 18, 2021, were organised. The local Commission approved the experimental protocol on Bioethics of the National University of Life and Environmental Sciences of Ukraine.

Experimental Treatment Groups. From young pigs of the industrial herd, Danish selection (♀Landrace/Large White X ♂Dyurek) aged 24 days, 4 groups of piglets were formed (all animals were either half-brothers or half-sisters), which were kept in 10 m² pens on a slatted floor. Each pen was equipped with a 2.5 m² cover with a heating lamp, which was used in the first week of life after weaning. The experiment was conducted in December 2024 and January 2025.

The housing conditions (microclimate parameters, functioning of automatic drinkers) and the health status of the experimental animals were monitored daily. Before the start of the experiment, the premises were disinfected, and the

experimental pigs were subjected to preventive deworming. The average daily air temperature in the pigsty where the experimental pigs were kept was at 30-23 °C, depending on the period of detention, the relative humidity fluctuated within 70–75%.

The experimental design is presented in Table 1.

Table 1. Experimental design

Groups	Characteristics of feed	SID isoleucine:lysine
1 (control)	Basic feed (BF)	<0.5
Experimental 2	Basic feed (BF)	0.5
Experimental 3	Basic feed (BF)	0.55
Experimental 4	Basic feed (BF)	0.6

Piglets were fed manually using a TR-4 ROTECNA feeding device. 1 feeding device per group (hopper volume – 120 l, intended for piglets 6–40 kg, feeding type – dry, number of animals served by 1 hopper – 30 animals, feeding place width 18.7 cm. Feeding principle: every morning from 9.00 to 10.00 – the rest of the feed from the previous day was taken out, weighed and a new amount of feed was poured in.

The differences between the groups concerned only the composition of the diet, namely the ratio of SID isoleucine to lysine. Thus, in piglets of the Con group, the SID isoleucine:lysine ratio in the diet was unlimited and less than 0.5, in piglets of the Ile: Lys 0.5 group the SID isoleucine:lysine ratio was 0.5, in the Ile: Lys 0.55 group – 0.55, respectively, and in the Ile: Lys 0.6 group – 0.6.

Zootechnical records. Throughout the rearing period, the number of piglets removed from the experiment (due to mortality and culling) was recorded. Also, every seven days, the live weight of pigs was determined on the AXIS BDU300C-0608X scales (Ukraine). Electronic scales AXIS BDU300C-0608X (manufactured in Ukraine) are intended for weighing animals (in particular piglets) and general cargo, with a maximum capacity of 300 kg and a minimum load of 1 kg, a readability and verification scale interval of 50 g, a medium accuracy class, and a tare compensation range up to 100% of the maximum capacity.

Blood sampling. Blood sampling was performed from 8 pigs (♀4 & ♂4) of each group at the end of the experiment. The sampling was performed from the axillary vein, and 1.0–1.5 ml of blood was taken into a K2 EDTA tube. Blood sampling, as well as the selection of randomised individuals, was performed according to generally accepted methods.

Haematological methods. Haematological parameters of pig blood were determined on the IDEXX ProCyte Dx haematological analyser (IDEXX Europe B.V.) at the LLC "Veterinary Diagnostics Centre". The following parameters were determined: erythrocyte, leukocyte, and platelet counts - by impedance change (conductometric method); haemoglobin concentration - by the spectrophotometric method.

Biochemical parameters and enzyme activity of piglet blood serum were determined on an automatic Beckman Coulter biochemical analyser (USA) in the laboratory of LLC "Centre for Veterinary Diagnostics" using the manufacturer's original test systems and commercial diagnostic kits High Technology Inc. (USA). Determination of the main serum metabolites (High Technology Inc., USA). The following reagent sets were used to determine the concentrations of metabolites: glucose – set HT-G242 (method: glucose oxidase, endpoint; λ=500 nm); total protein – HT-T251 (biuret reaction, endpoint; λ=540 nm); calbumin – HT-A203 (reaction with bromocresol green; λ=630 nm); creatinine – HT-C225 (Jaffe method, kinetic; λ=510 nm); urea – HT-U254 (Trinder/uricase method, endpoint; λ=520 nm); inorganic phosphorus – HT-P244 (ammonium molybdate method; λ=340 nm). calcium – HT-C216 (orthocresolphthalein method, endpoint; λ=570 nm); ALT (alanine aminotransferase) – HT-A206 (IFCC method, kinetic; λ=340 nm). The methods were performed according to the kit manufacturer's instructions.

Statistical analysis of biometric data was carried out on a PC using Statistica 10. The significance of differences between groups was assessed using one-way analysis of variance (ANOVA) and the Tukey–Kramer multiple comparison test as a post hoc procedure. The normality of the sample data distribution was tested using the Shapiro-Wilk, Kolmogorov-Smirnov, and Box-Plot tests. If the data distribution was significantly different from normal, the nonparametric Mann-Whitney U-test was used. Differences between groups were considered significant at $p<0.05$.

RESULTS AND DISCUSSION

The experimental young pigs were fed complete mixed feeds, balanced in all nutrients, in which the main nutritional indicators, including the levels of the first essential amino acids (lysine, methionine + cystine, tryptophan, valine) covered the needs of the recommendations of the CVB (2023). Before compiling the diets, the main ingredients (wheat, barley, soybean meal, soybean concentrate and dry aerosol-dried haemoglobin) were analysed for the content of dry matter, crude protein, crude fibre, starch, fat and crude ash. Based on the indicators obtained, the nutritional value of the main ingredients in the feed was calculated according to the formulas and nutritional matrices of the CVB database (2018). The nutritional value of the mixed feed used for feeding piglets is given in Tables 2–4.

Table 2. Nutritional value of the diet for young pigs from 24 to 38 days of age

Indicator	Group of piglets			
	Con	Ile:Lys0.5	Ile:Lys0.55	Ile:Lys0.6
1	2	3	4	5
Dry matter, g.	904.1	904.2	904.2	904.3
Humidity, g.	95.9	95.8	95.8	95.7

Crude protein, g.	179.5	179.9	180.2	180.6
Crude fibre, g.	43.1	43.1	43.1	43.0
Raw ash, g.	47.9	47.9	47.9	47.9
Crude fat, g.	70.1	70.0	70.0	70.0
Starch according to Evers	390.7	390.2	389.8	389.4
Net energy, kcal	2605.1	2605.0	2605.0	2605.0
Net energy, MJ.	10.9	10.9	10.9	10.9
Lysine, g.	13.2	13.2	13.2	13.2
Methionine, g.	5.0	5.0	5.0	5.0
Methionine + Cystine, g.	8.1	8.1	8.1	8.1
Threonine, g.	9.8	9.8	9.8	9.8
Tryptophan, g.	3.0	3.0	3.0	3.0
Valine, g.	9.5	9.5	9.5	9.5
SID Lysine, g.	12.1	12.1	12.1	12.1
SID Methionine, g.	4.7	4.7	4.7	4.7
SID Methionine + Cystine, g.	7.3	7.3	7.3	7.3
SID Threonine, g.	8.5	8.5	8.5	8.5
SID Tryptophan, g.	2.7	2.7	2.7	2.7
SID Valine, g.	8.3	8.3	8.3	8.3
SID Isoleucine, g.	5.3	6.1	6.7	7.3
SID Leucine, g.	12.1	12.1	12.1	12.1
SID Isoleucine/Lysine	0.439	0.5	0.55	0.6
SID Leucine/ Lysine	1.0	1.0	1.0	1.0
Calcium, g.	6.0	6.0	6.0	6.0
Total phosphorus, g.	5.0	5.0	5.0	5.0
Absorbable phosphorus, g.	3.8	3.8	3.8	3.8
Sodium, g.	3.0	3.0	3.0	3.0
Iron, mg.	110.0	110.0	110.0	110.0
Zinc, mg.	150.0	150.0	150.0	150.0
Manganese, mg.	72.0	72.0	72.0	72.0
Copper, mg.	150.0	150.0	150.0	150.0
Iodine, mg.	0.8	0.8	0.8	0.8
Selenium, mg.	0.4	0.4	0.4	0.4
Vit. A, IU	15000	15000	15000	15000
Vit. D3, IU	2500	2500	2500	2500
Vit. E, mg.	200.0	200.0	200.0	200.0
Vit. K3, mg.	2.5	2.5	2.5	2.5
Vit. B1 (Thiamine), mg.	1.875	1.875	1.875	1.875

Vit. B2 (Riboflavin), mg.	6.25	6.25	6.25	6.25
Vit. B3 (Niacin), mg.	25.0	25.0	25.0	25.0
Choline, mg.	300.0	300.0	300.0	300.0
Vit. B5 (Pantothenic acid), mg.	12.5	12.5	12.5	12.5
Vit. B6 (Pyridoxine), mg.	3.75	3.75	3.75	3.75
Vit. H (Biotin), mcg.	125.0	125.0	125.0	125.0
Vit. B9 (Folate), mg.	0.625	0.625	0.625	0.625
Vit. B12 (Cyanocobalamin), mcg.	31.25	31.25	31.25	31.25

Table 3. Nutritional value of the diet for young pigs from 39 to 55 days of age

Indicator	Group of piglets			
	Con	Ile:Lys0.5	Ile:Lys0.55	Ile:Lys0.6
1	2	3	4	5
Dry matter, g.	895.6	895.6	895.7	895.7
Humidity, g.	104.4	104.4	104.3	104.3
Crude protein, g.	174.9	175.3	175.6	176.0
Crude fibre, g.	42.4	42.4	42.4	42.4
Raw ash, g.	46.7	46.7	46.7	46.7
Crude fat, g.	57.4	57.2	57.0	56.7
Starch according to Evers	426.7	426.5	426.3	426.1
Net energy, kcal	2509.0	2509.0	2509.0	2509.0
Net energy, MJ.	10.5	10.5	10.5	10.5
Lysine, g.	12.7	12.7	12.7	12.7
Methionine, g.	4.9	4.9	4.9	4.9
Methionine + Cystine, g.	7.7	7.7	7.7	7.7
Threonine, g.	8.7	8.7	8.7	8.7
Tryptophan, g.	2.8	2.8	2.8	2.8
Valine, g.	9.0	9.0	9.0	9.0
SID Lysine, g.	11.5	11.5	11.5	11.5
SID Methionine, g.	4.5	4.5	4.5	4.5
SID Methionine + Cystine, g.	6.9	6.9	6.9	6.9
SID Threonine, g.	7.5	7.5	7.5	7.5
SID Tryptophan, g.	2.5	2.5	2.5	2.5
SID Valine, g.	7.8	7.8	7.8	7.8
SID Isoleucine, g.	5.2	5.8	6.3	6.9
SID Leucine, g.	11.5	11.5	11.5	11.5
SID Isoleucine/Lysine	0.452	0.5	0.55	0.6
SID Leucine/Lysine	1.0	1.0	1.0	1.0
Calcium, g.	5.5	5.5	5.5	5.5

Total phosphorus, g.	5.0	5.0	5.0	5.0
Absorbable phosphorus, g.	3.5	3.5	3.5	3.5
Sodium, g.	2.5	2.5	2.5	2.5
Iron, mg.	110.0	110.0	110.0	110.0
Zinc, mg.	150.0	150.0	150.0	150.0
Manganese, mg.	72.0	72.0	72.0	72.0
Copper, mg.	150.0	150.0	150.0	150.0
Iodine, mg.	0.8	0.8	0.8	0.8
Selenium, mg.	0.4	0.4	0.4	0.4
Vit. A, IU	15000	15000	15000	15000
Vit. D3, IU	2500	2500	2500	2500
Vit. E, mg.	200.0	200.0	200.0	200.0
Vit. K3, mg.	2.5	2.5	2.5	2.5
Vitamin B1 (Thiamine), mg.	1.875	1.875	1.875	1.875
Vit. B2 (Riboflavin), mg.	6.25	6.25	6.25	6.25
Ex. B3 (Niacin), mg.	25.0	25.0	25.0	25.0
Choline, mg.	400.0	400.0	400.0	400.0
Vit. B5 (Pantothenic acid), mg.	12.5	12.5	12.5	12.5
Vit. B6 (Pyridoxine), mg.	3.75	3.75	3.75	3.75
Vit. H (Biotin), mcg.	125.0	125.0	125.0	125.0
Vit. B9 (Folate), mg.	0.625	0.625	0.625	0.625
Vit. B12 (Cyanocobalamin), mcg.	31.25	31.25	31.25	31.25

Table 4. Nutritional value of the diet for young pigs from 56 to 70 days of age

Indicator	Group of piglets			
	Con	Ile:Lys0.5	Ile:Lys0.55	Ile:Lys0.6
1	2	3	4	5
Dry matter, g.	895.0	895.0	895.0	895.1
Humidity, g.	105.0	105.0	105.0	104.9
Crude protein, g.	173.8	174.0	174.4	174.7
Crude fibre, g.	39.2	39.2	39.1	39.1
Raw ash, g.	51.2	51.2	51.2	51.2
Crude fat, g.	46.9	46.7	46.5	46.3
Starch according to Evers	412.5	412.4	412.2	412.1
Net energy, kcal	2348.0	2348.0	2348.0	2348.0
Net energy, MJ.	10.2	10.2	10.2	10.2
Lysine, g.	12.2	12.2	12.2	12.2
Methionine, g.	4.5	4.5	4.5	4.5

Methionine + Cystine, g.	7.5	7.5	7.5	7.5
Threonine, g.	8.3	8.3	8.3	8.3
Tryptophan, g.	2.7	2.7	2.7	2.7
Valine, g.	8.4	8.4	8.4	8.4
SID Lysine, g.	11.0	11.0	11.0	11.0
SID Methionine, g.	4.2	4.2	4.2	4.2
SID Methionine + Cystine, g.	6.6	6.6	6.6	6.6
SID Threonine, g.	7.2	7.2	7.2	7.2
SID Tryptophan, g.	2.4	2.4	2.4	2.4
SID Valine, g.	7.2	7.2	7.2	7.2
SID Isoleucine, g.	5.2	5.5	6.1	6.6
SID Leucine, g.	11.0	11.0	11.0	11.0
SID Isoleucine/Lysine	**0.473**	**0.5**	**0.55**	**0.6**
SID Leucine/Lysine	**1.0**	**1.0**	**1.0**	**1.0**
Calcium, g.	5.5	5.5	5.5	5.5
Total phosphorus, g.	5.4	5.4	5.4	5.4
Absorbable phosphorus, g.	3.4	3.4	3.4	3.4
Sodium, g.	2.5	2.5	2.5	2.5
Iron, mg.	110.0	110.0	110.0	110.0
Zinc, mg.	150.0	150.0	150.0	150.0
Manganese, mg.	72.0	72.0	72.0	72.0
Copper, mg.	100.0	100.0	100.0	100.0
Iodine, mg.	0.8	0.8	0.8	0.8
Selenium, mg.	0.4	0.4	0.4	0.4
Vit. A, IU	15000	15000	15000	15000
Vit. D3, IU	2500	2500	2500	2500
Vit. E, mg.	200.0	200.0	200.0	200.0
Vit. K3, mg.	2.5	2.5	2.5	2.5
Vitamin B1 (Thiamine), mg.	1.875	1.875	1.875	1.875
Vit. B2 (Riboflavin), mg.	6.25	6.25	6.25	6.25
Ex. B3 (Niacin), mg.	25.0	25.0	25.0	25.0
Choline, mg.	300.0	300.0	300.0	300.0
Vit. B5 (Pantothenic acid), mg.	12.5	12.5	12.5	12.5
Vit. B6 (Pyridoxine), mg.	3.75	3.75	3.75	3.75
Vit. H (Biotin), mcg.	125.0	125.0	125.0	125.0
Vit. B9 (Folate), mg.	0.625	0.625	0.625	0.625
Vit. B12 (Cyanocobalamin), mcg.	31.25	31.25	31.25	31.25

The effect of using different SID isoleucine to lysine ratios in diets for young pigs on their body weight

Isoleucine has traditionally been considered an essential amino acid for pigs, as it cannot be synthesised in their body (Wang *et al.*, 2024). Optimal isoleucine intake positively affects growth performance and the physiological status of pigs (van Milgen *et al.*, 2012). According to the ideal protein concept, the requirement for isoleucine is determined by the SID isoleucine:lysine ratio, with lysine typically being the first limiting amino acid, and isoleucine thus considered the second limiting amino acid when assessing its requirement (Soumeh *et al.*, 2014; van Milgen *et al.*, 2012).

In this study, four different SID isoleucine:lysine ratios, ranging from less than 0.5 to 0.6, were used for young pigs with a live weight of 7–30 kg. At the beginning of the experiment, at 24 days of age, the body weights of young pigs were 7.40–7.43 kg, with no significant difference between the groups (Table 5). Thus, piglets were selected for the control and experimental groups, which were similar in age and body weight.

Table 5. Body weight of piglets depending on the ratio of SID isoleucine to lysine in the diet, kg, M±SEM, n=21/group

Age, days	Group of piglets			
	Con	Ile:Lys0.5	Ile:Lys0.55	Ile:Lys0.6
24	7.43 ± 0.048^{a}	7.42 ± 0.045^{a}	7.40 ± 0.043^{a}	7.42 ± 0.045^{a}
38	11.08 ± 0.149^{a}	11.49 ± 0.138^{b}	11.67 ± 0.193^{bc}	11.13 ± 0.147^{ab}
55	18.42 ± 0.230^{a}	19.35 ± 0.23^{b}	19.79 ± 0.289^{bc}	18.70 ± 0.179^{ab}
70	27.57 ± 0.332^{a}	29.27 ± 0.357^{b}	30.00 ± 0.434^{bc}	28.27 ± 0.262^{ab}

Note: Values are presented as mean ± SEM (n=21 per group). Means within a row with different superscript letters differ significantly (P<0.05, Tukey–Kramer test)

However, during the growing period, the effect of different SID isoleucine:lysine ratios due to the addition of synthetic isoleucine on the body weight of young pigs was revealed. In particular, at 38 days of age, the body weight of piglets in the Ile:Lys0.5 group was 3.7% higher (P<0.05), at 55 days it was 5.0% higher (P<0.05), and at 70 days it was 6.2% higher (P<0.05), compared to the Con group. Young pigs in the Ile:Lys0.55 group were characterized by a higher body weight at 38 days of age by 5.3% (P<0.05), at 55 days of age by 7.4% (P<0.05), and at 70 days of age by 8.8% (P<0.05), compared to the Con group.

At the same time, no statistically significant differences were observed between the Ile:Lys0.55 and Ile:Lys0.5 groups at any stage of the experiment. Furthermore, piglets in the Ile:Lys0.6 group did not differ significantly in body weight from the Con, Ile:Lys0.5, or Ile:Lys0.55 groups throughout the experimental period.

The obtained data are consistent with the meta-analysis published by van Milgen J. *et al*. (2012), in which the authors concluded that the SID Ile:Lys level for maximising pig growth parameters should be 0.50–0.55. In general, studies on

the effect of the dose-dependent SID isoleucine:lysine ratio in diets without the addition of blood products on growth parameters of pigs with a live weight of 7–30 kg are few (Wiltafsky *et al.*, 2009). However, it is known that excessive isoleucine intake can increase BCAA metabolism and thus negatively affect growth parameters in pigs (Soumeh *et al.*, 2014). Thus, according to Nørgaard J. V. *et al.* (2013), pigs fed a diet containing excessive levels of isoleucine are characterised by reduced feed intake and reduced body weight gains. The negative consequences of excessive isoleucine intake may be due to interactions among BCAAs. Wiltafsky M. K. *et al.* (2009) also found that too high a SID isoleucine:lysine ratio can lead to reduced performance in pigs, and suggested that the synthesis of α-keto-β-methylvalerate by excessive isoleucine may activate the branched-chain ketoacid dehydrogenase complex, which can catalyse all irreversible steps of BCAA catabolism, and thus limit leucine metabolism.

Biochemical profile of piglets depending on the ratio of SID isoleucine to lysine in diets

Serum biochemical parameters can reflect the metabolic function in the pig body (Oliveira *et al.*, 2025). This study showed that when using all the studied SID isoleucine:lysine ratios in the diet for young pigs, namely from < 0.5 to 0.6, the clinical biochemistry parameters of their serum were within the physiological norm (Table 5). However, certain differences were still observed within the reference intervals depending on the ratios mentioned above. In particular, a slight increase in the concentration of total protein in the serum of piglets was found in proportion to the increase in the SID isoleucine:lysine ratio in the diet. Thus, in the young pigs of the Ile:Lys0.5 group, the concentration of total protein in the serum was lower by 1.9 % ($p<0.05$), and in the group Ile: Lys 0.55 - by 4.8% ($p<0.05$), while in piglets of the Ile:Lys0.6 group - by 5.1% ($p<0.05$), compared to the Con group.

Table 6. Biochemical parameters of piglet blood serum depending on the ratio of SID isoleucine to lysine in the diet, M±SEM, n=8/group

Indicator	Group of piglets				PI[1]
	Con	Ile:Lys0.5	Ile:Lys0.55	Ile:Lys0.6	
Total protein, g/L	56.7±0.31^a	57.8±0.45^b	59.4±0.55^c	59.6±0.46^c	33–63
Albumin, g/L	31.5±0.87^a	29.5±0.76^a	31.5±0.69^a	29.5±0.95^a	20–39
Glucose, mmol/L	7.5±0.21^a	6.2±0.24^b	5.4±0.25^c	7.7±0.36^a	5.l–8.3
Creatinine, μmol/L	98.8±2.78^a	89.8±2.73^b	88.9±2.32^c	101.9±2.46^a	73–111
Urea, mmol/L	3.8±0.48^a	1.9±0.49^b	1.5±0.42^b	2.6±0.32^c	0.5–3.8
Uric acid, μmol/L	3.6±0.34^a	3.5±0.27^a	3.7±0.22^a	3.2±0.31^a	–
Calcium, mmol/L:					
- general	2.8±0.07^a	2.8±0.06^a	2.9±0.08^a	2.6±0.09^a	2.4–3.3
- ionised	1.3±0.02^a	1.4±0.01^a	1.4±0.01^a	1.3±0.02^a	–
Phosphorus, mmol/L	4.3±0.07^a	4.4±0.03^a	4.3±0.05^a	4.2±0.03^a	2.6–4.0

Note: Values are presented as mean ± SEM (n=21 per group). Means within a row with different superscript letters differ significantly (P<0.05, Tukey–Kramer test). 1 RI - reference intervals according to Egeli A. K. et al. (1998)

Glucose is the primary source of energy and carbon for most eukaryotic cells, as it can be oxidised to provide energy and has a wide range of effects on cell function (Gaster *et al.,* 2000). Amino acids can be involved in the regulation of glucose levels. BCAAs, and especially isoleucine, improve glucose uptake and utilisation. Isoleucine can suppress increased blood glucose concentration (Doi *et al.,* 2003). According to Zhang, S. *et al*. (2016), the intake of isoleucine alone can increase glucose utilisation in pigs. In this study, a decrease in serum glucose concentration was observed in young pigs with an increase in the SID isoleucine:lysine ratio in the diet. Thus, in the young pigs of the Ile:Lys0.5 group, the concentration of glucose in the blood serum was lower by 17.3% ($p<0.05$), and in the young pigs of the Ile:Lys0.55 group - by 2.1 mmol/l or 28.0% ($p<0.05$), compared to the Con group. However, a further increase in the SID isoleucine:lysine ratio in the diet caused an increase in the concentration of glucose in the blood serum of piglets to the level of the Con group. The hypothesis of the mechanism by which isoleucine and leucine regulate the level of glucose in the blood serum may be associated with an increase in glucose uptake by muscles, glucose oxidation in the body and a decrease in gluconeogenesis in the liver of pigs (Doi *et al.,* 2007; Zhang *et al.,* 2016). Glycogen, which is synthesised and stored in the liver and skeletal muscle, plays an important role in the control of blood glucose levels. BCAAs promote glucose uptake by the liver and muscle and enhance glycogen synthesis. Isoleucine lowers plasma glucose levels by increasing muscle glucose uptake by activating the glucose transporter. This involves phosphatidylinositol-3-kinase and protein kinase C, which are insulin-independent (Doi *et al.,* 2005). BCAAs, in addition to mediating muscle glucose uptake and upregulating glycogen synthesis through activation, are also involved in fat and energy metabolism. e above statement is generally consistent with evidence indicating that isoleucine can reduce glycemia by enhancing skeletal muscle glucose uptake and activating glucose transport, including through PI3K/PKC signaling pathways, partially in an insulin-independent manner, thereby potentially supporting glycogen synthesis and storage in muscle. These effects were demonstrated in classic studies by Doi et al., where isoleucine administration in rats reduced plasma glucose levels and increased muscle glucose uptake; subsequent work further clarified the involvement of PI3K-dependent pathways and effects on glucose oxidation and hepatic gluconeogenesis (Doi *et al.,* 2005; 2007).

Also, within the physiological norm, a decrease in the serum concentration of creatinine in young pigs was observed when diets with a SID isoleucine:lysine ratio of 0.5 and 0.55 were used for their feeding. In the piglets of the Ile:Lys0.5 group, the creatinine concentration in the serum was 9.1% lower ($p<0.05$), while in the other group, it was 10.0% lower ($p<0.05$), compared to the Con group. However, further increasing the SID isoleucine:lysine ratio in the diet to 0.6 resulted in an opposite effect, with the creatinine level in the serum of piglets in the Ile:Lys0.6 group rising to levels comparable to those of the Con group. Your observation that feeding young pigs diets with SID isoleucine:lysine ratios of 0.5

and 0.55 lowered serum creatinine within the physiological range, but increasing the ratio to 0.6 restored creatinine to control levels, aligns with emerging evidence that amino acid balance affects nitrogen metabolism and renal biomarkers indirectly rather than directly altering creatinine metabolism. Recent work indicates that functional amino acids such as isoleucine can influence overall nitrogen utilization, protein turnover, and metabolic health in growing pigs, which may in turn modulate serum creatinine concentration as part of broader metabolic shifts rather than a direct renal effect. For example, a 2024 study on dietary functional amino acid supplementation reported improvements in growth and health indicators in pigs, suggesting that balanced dietary amino acids can optimize protein metabolism and reduce metabolic stress markers (e.g., metabolites related to protein catabolism) without necessarily altering renal function directly (França *et al.*, 2024). In another recent analysis of SID branched-chain amino acid (BCAA) requirements, meta-analysis models confirmed that appropriate Ile:Lys ratios support optimal growth performance and efficient amino acid utilization in growing pigs, which likely reflects more efficient muscle protein synthesis and reduced amino acid catabolism — processes that could decrease creatinine release from muscle turnover. Additionally, a 2023 study on optimal Ile:Lys ratios in piglets found similar recommendations around the mid-range of ratios (close to ~0.5–0.55) for performance and physiological balance, supporting your empirical finding that moderate proportions are beneficial (Kerkaert *et al.*, 2021). From a mechanistic standpoint, serum creatinine in pigs tends to reflect lean tissue mass and protein turnover more than acute dietary changes. Thus, the reduction in creatinine at SID Ile:Lys ratios of 0.5 and 0.55 may indicate more efficient amino acid utilization and reduced muscle protein breakdown, whereas the return to control levels at 0.6 suggests that exceeding the optimal balance does not further improve nitrogen metabolism and may instead increase amino acid catabolism back to baseline. This interpretation is consistent with broader literature showing that maintaining amino acid balance near predicted requirements minimizes unnecessary catabolism and supports growth efficiency (França *et al.*, 2024; Sarri *et al*., 2024; Bertolini*т et al*., 2026).

Enriching the pig diet with isoleucine reduces blood urea and affects lipid metabolism (Spring *et al.*, 2020; Habibi *et al*., 2022b). The mechanisms by which isoleucine regulates nitrogen utilisation and lipid metabolism in pigs are still poorly understood. However, in support of this, this study showed that increasing the SID isoleucine:lysine ratio in the diet for young pigs was associated with a decrease in serum urea concentrations within the physiological norm. Urea is a serum by-product of protein catabolism and a key indicator of the efficiency of amino acid utilisation in pigs (Marín-García *et al*., 2022). In young pigs of the Ile:Lys0.5 group, the concentration of urea in the blood serum was lower by 50.0% ($p<0.05$), and in the Ile:Lys0.55 group - by 60.5 % ($p <0.05$), compared to the Con group. However, a further increase in the SID isoleucine:lysine ratio caused, on the contrary, an increase in the serum urea level of piglets of the Ile: Lys0.6 group to the values of the Con group, which indicates a negative effect of

excessive isoleucine consumption on the efficiency of protein utilisation in the body of pigs. This effect can be explained by the fact that pigs fed a balanced diet demonstrate a low level of urea in the blood, which indicates a decrease in protein catabolism, a more efficient use of total nitrogen and, consequently, a decrease in urea synthesis (Waguespack *et al.,* 2012; Toledo *et al.,* 2014). Increased blood urea concentration is associated with decreased protein synthesis and increased protein catabolism, and its level is negatively correlated with amino acids and protein utilisation efficiency (Heo *et al*., 2008). Isoleucine deficiency leads to decreased protein synthesis, which may promote deamination of other amino acids and subsequently to increased blood urea levels (Kwon *et al.,* 2019).

The reduction in serum urea seen at moderate SID isoleucine:lysine ratios followed by a return to control levels at the higher ratio aligns with contemporary evidence that blood urea nitrogen (BUN) reflects the efficiency of nitrogen and amino acid utilization in pigs, rather than acute changes in crude protein alone. Recent studies have confirmed that BUN concentration is a reliable marker of protein degradation and nitrogen utilization, with lower values generally indicating improved matching of dietary amino acid supply to metabolic requirements and reduced excess amino acid catabolism. Such decreases in serum urea are interpreted as evidence of enhanced nitrogen retention and more efficient use of dietary amino acids for protein synthesis, as pigs on diets with better balanced amino acid profiles exhibit reduced hepatic ureagenesis and nitrogen excretion. Furthermore, current pig nutrition research underscores that oversupply or imbalance of indispensable amino acids, including branched-chain amino acids, can diminish the efficiency of nitrogen utilization, often resulting in higher urinary nitrogen outputs and less favorable metabolic profiles. The observed pattern in your data, where moderate optimization of Ile:Lys improved nitrogen metabolism but further increases had no additional benefit, is consistent with meta-analytical findings pointing to evolving and refined standardized ileal digestible amino acid requirements that support nitrogen efficiency only within an optimal range (Wang *et al.,* 2024; Chae *et al.,* 2024).

The activity of serum enzymes in piglets was within the physiological norm (Table 6). However, certain differences were still observed within the reference interval. In particular, in young pigs of the group Ile: Lys 0.55 showed a decrease in ALT activity by 20.5% ($p<0.05$) and AST by 11.7% ($p<0.05$), and in piglets of the group Ile: Lys 0.6 – ALT by 24.2% ($p<0.05$) and AST – by 13.1% ($p<0.05$), compared to the Con group.

Table 7. Activity of serum enzymes in piglets depending on the ratio of SID isoleucine to lysine in the diet, IU/L, M±SEM, n=8/group

Enzyme	Group of piglets				PI[1]
	Con	Ile:Lys0.5	Ile:Lys0.55	Ile:Lys0.6	
ALT	82.5±4.33[a]	73.5±3.23[a]	65.6±3.28[b]	62.5±4.11[b]	15–108
AST	83.0±2.74[a]	81.8±3.01[a]	73.3±3.11[b]	72.1±2.98[b]	8–84

Note: Values are presented as mean ± SEM (n=21 per group). Means within a row with different superscript letters differ significantly (P<0.05, Tukey–Kramer test). 1 RI - reference intervals according to Egeli A. K. et al. (1998)

BCAA, especially isoleucine, is involved in the regulation of amino acid and fatty acid profiles of the pig body (Doudou *et al.,* 2022). This study confirmed that changes in the SID isoleucine:lysine ratio in the diet of young pigs affect the protein profile of their blood serum (Table 7). In particular, no differences were found in the concentration of serum albumins and globulins, as well as in the individual globulin fractions and the albumin-globulin ratio, in the piglets of the Con, Ile:Lys0.5, and Ile:Lys0.55 groups. However, in the piglets of the Ile:Lys0.6 group, a 12.7% decrease in albumins ($p<0.05$), a 13.2% increase in globulins ($p<0.05$), and a 0.45% reduction in the albumin-globulin ratio ($p<0.05$) were observed, compared to the Con group. The increase in globulins in the serum of piglets in the Ile:Lys0.6 group was due to an increase in all fractions, except for β-globulins. Specifically, there was a 2.5% increase in α1-globulins ($p<0.05$), a 6.3% increase in α2-globulins ($p<0.05$), and a 3.2% increase in γ-globulins ($p<0.05$), compared to the Con group.

Branched-chain amino acids, particularly isoleucine, not only influence lipid and nitrogen metabolism but also affect systemic protein profiles in growing pigs, with serum albumin and globulin fractions serving as sensitive indicators of metabolic balance and immune modulation. In the present study, no differences were detected in albumins, total globulins, individual globulin fractions, or the albumin–globulin ratio among piglets fed control or moderate SID Ile:Lys diets, underscoring metabolic stability when amino acids are balanced within recommended ranges. However, at a higher SID Ile:Lys ratio (0.6), a marked decrease in albumin and concomitant increases in total globulins –driven by elevated α1-, α2-, and γ-globulin fractions –suggest a shift in hepatic protein synthesis and possibly mild activation of immune-related protein production. This interpretation is supported by recent research showing that alterations in dietary protein and amino acid supply can significantly affect serum protein fractions in pigs, with both albumin and globulin responding in nonlinear ways to nutritional imbalances (Hlathini *et al.,* 2024). Moreover, contemporary studies on amino acid–balanced diets demonstrate that dietary protein and amino acid composition shape nitrogen metabolism and blood biochemical profiles, including serum proteins, and that deviations from ideal patterns can lead to compensatory responses in immune-related protein fractions.

Table 8. Protein profile of piglet blood serum depending on the ratio of SID isoleucine to lysine in the diet, %, M±SEM, n=8/group

Indicator	Group of piglets			
	Con	Ile:Lys0.5	Ile:Lys0.55	Ile:Lys0.6
Albumins	52.2±1.13^{a}	53.5±1.35^{a}	50.5±1.67^{a}	39.5±1.42^{b}
Globulins	47.3±1.12^{a}	46.5±1.36^{a}	49.5±1.31^{a}	60.5±1.21^{b}
ά1-globulins	2.5±0.27^{a}	3.2±0.29^{a}	2.5±0.22^{a}	5.0±0.26^{b}
ά2-globulins	4.5±0.57^{a}	5.0±0.21^{a}	5.8±0.49^{a}	10.8±0.22^{b}
β-globulins	15.0±0.85^{a}	14.3±0.77^{a}	15.5±0.57^{a}	16.2±0.73^{a}
γ- globulins	25.3±0.56^{a}	24.0±0.97^{a}	25.7±0.73^{a}	28.5±0.67^{b}
Albumin-globulin ratio	1.12±0.048^{a}	1.25±0.196^{a}	1.03±0.074^{a}	0.67±0.061^{b}

Note: Values are presented as mean ± SEM (n = 21 per group). Means within a row with different superscript letters differ significantly (P < 0.05, Tukey–Kramer test)

BCAAs are known to be involved in fatty acid metabolism (Cheng *et al.*, 2010). Insufficient or excessive levels of BCAA in the diet can negatively affect lipid metabolism in pigs (Zhang *et al.*, 2017). Serum triglyceride and total cholesterol levels are related to lipid absorption, and low-density lipoprotein and high-density lipoprotein levels are related to lipid breakdown and transport in pigs (Heng *et al.*, 2020). Therefore, at the final stage of this study, the serum lipid profile of young pigs was analysed depending on the SID isoleucine:lysine ratio in their diet (Table 8). It was found that the content of total cholesterol in the serum of young pigs using all the studied SID isoleucine:lysine ratios in the diet was within the physiological norm. However, certain differences were still observed within the reference intervals. In particular, in young pigs of the Con and Ile:Lys0.5 groups, the concentration of total cholesterol was at the same level. In the piglets of the Ile:Lys0.55 group, the total cholesterol content was 8.7% lower ($p<0.05$) compared to the Con group. However, in the piglets of the Ile:Lys0.6 group, a 10.6% increase in serum cholesterol levels ($p<0.05$) was observed, compared to the Con group.

Table 9. Lipid profile of piglet blood serum depending on the ratio of SID isoleucine to lysine in the diet, mmol/L, M±SEM, n=8/group

Indicator	Group of piglets				PI
	Con	Ile:Lys0.5	Ile:Lys0.55	Ile:Lys0.6	
Total cholesterol	2.65±0.063^{a}	2.68±0.031^{a}	2.42±0.094^{b}	2.93±0.063^{c}	1.6–4.3[1]
Triglycerides	0.58±0.049^{a}	0.37±0.019^{b}	0.30±0.038^{b}	0.46±0.053^{b}	0.2–2.25[1]
High-density lipoproteins	0.88±0.028^{a}	0.95±0.033^{a}	0.99±0.026^{b}	0.93±0.02^{ab}	0.59–2.10[2]
Low-density lipoproteins	1.62±0.059^{a}	1.61±0.019^{a}	1.28±0.062^{b}	1.81±0.042^{c}	0.42–3.72[2]
Very low-density lipoproteins	0.15±0.021^{a}	0.12±0.024^{a}	0.15±0.019^{a}	0.19±0.024^{a}	–

Note: Values are presented as mean ± SEM (n = 21 per group). Means within a row with different superscript letters differ significantly (P < 0.05, Tukey–Kramer test).1 RI - reference intervals according to Egeli A. K. et al. (1998); 2 RI - reference intervals according to Sucheong S. et al. (2012).

High-density lipoproteins, known as "good cholesterol," are a type of lipoprotein that transports excess cholesterol from the body tissues of pigs to the liver for further removal (Kiss *et al*., 2025). Its amount in young pigs of the Con and Ile:Lys0.5 groups was at the same level within the physiological norm. In young pigs of the Ile: Lys 0.55 group, the level of high-density lipoproteins was 12.5% higher ($p<0.05$) compared to the Con group. With a further increase in the SID isoleucine:lysine ratio in the diet of young pigs, no differences in the serum content of high-density lipoproteins were observed in pigs of the Ile: Lys 0.6 group. As for low-density lipoproteins, their concentration in the blood serum of young pigs of the Con and Ile:Lys0.5 groups was at the same level within the physiological norm. While in young pigs of the Ile:Lys0.55 group there was a

decrease in the content of low-density lipoproteins by 21.0% ($p<0.05$) compared to the Con group. Further increase in the SID isoleucine:lysine ratio in the diet of young pigs of the Ile: Lys 0.6 group was accompanied by an increase in the content of low-density lipoproteins in their blood serum by 11.7% ($p<0.05$) compared to the Con group.

Serum triglyceride content is related to lipid absorption (Wang *et al.*, 2024). Isoleucine deficiency is known to dramatically increase fat mobilisation in white adipose tissue and reduce fat quality (Du *et al.*, 2012). Excess isoleucine in the diet of pigs can enhance the synthesis of monounsaturated fatty acids and fat accumulation in skeletal muscle by regulating lipid metabolism (Luo *et al.*, 2019). Therefore, isoleucine may play a potential role in the regulation of lipid metabolism. Oxidative metabolism by isoleucine can reduce lipid accumulation in cells (Solon-Biet *et al.*, 2019). Furthermore, excessive BCAA intake may lead to increased catabolism of all BCAAs, and studies have shown that excessive isoleucine may have an antagonistic effect and negatively affect adipose tissue function and lipid metabolism (Morales *et al.*, 2016; Zhang *et al.*, 2021). This study revealed changes, within the physiological norm, in serum triglyceride concentrations in young pigs depending on the SID isoleucine:lysine ratio in their diet. Thus, in piglets of the Ile:Lys0.5 group, serum triglyceride concentrations were 36.2% lower ($p<0.05$) compared to the Con group. Young pigs of the Ile:Lys0.55 group were characterised by an even lower serum triglyceride concentration, namely 48.3% ($p<0.05$) compared to the Con group. However, with a further increase in the SID isoleucine:lysine ratio in the diet of piglets in the Ile: Lys 0.6 group, a decrease in serum triglyceride concentration was found by only 25.9% ($p<0.05$) compared to the Con group. The increase in triglyceride content is associated with increased serum BCAA levels, insulin resistance in muscles and impaired glucose utilisation, which clearly links BCAA oxidation with glucose and fat metabolism (She *et al.*, 2007). Isoleucine reduces muscle triglyceride levels and increases free fatty acid oxidation in skeletal muscle and liver by promoting the expression of peroxisome proliferator-activated receptor (PPAR-alpha) and uncoupling protein. These results indicate that isoleucine may prevent the development of visceral obesity and high insulin levels by affecting white adipose tissue, liver, and muscle adiposity (Nishimura *et al*., 2010). Inhibition of fat synthesis in adipose tissue has also been reported by many other studies, using animal models or cell lines under conditions of BCAA deficiency (Du *et al.*, 2012; Freudenberg *et al.*, 2013) or excess (Bong *et al.*, 2010; Vianna *et al.*, 2012).

CONCLUSIONS

Based on the obtained findings, for pigs (7–30 kg), the optimal SID Ile:Lys was 0.55, which increased live weight at 70 days by 8.8% ($p<0.05$) without negative physiological effects. At this ratio, serum glucose decreased by 28.0% ($p<0.05$), creatinine by 10.0% ($p<0.05$), urea by 60.5% ($p<0.05$), total cholesterol by 8.7% ($p<0.05$), low-density lipoproteins by 21.0% ($p<0.05$), triglycerides by

48.3% ($p<0.05$), ALT activity by 20.5% ($p<0.05$), and AST by 11.7% ($p<0.05$), whereas total protein increased by 4.8% ($p<0.05$) and high-density lipoproteins by 12.5% ($p<0.05$). The 0.5 ratio was less effective, increasing body weight by 6.2% ($p<0.05$) and reducing glucose by 17.3% ($p<0.05$), creatinine by 9.1% ($p<0.05$), urea by 50.0% ($p<0.05$), and triglycerides by 36.2% ($p<0.05$), while total protein increased by 1.9% ($p<0.05$). Increasing the ratio to 0.6 did not affect growth but led to a 5.1% rise in total protein ($p<0.05$), a 12.7% decrease in albumins ($p<0.05$), a 13.2% increase in globulins ($p<0.05$), and a 10.6% elevation in total cholesterol ($p<0.05$) due to higher low-density lipoproteins, which increased by 11.7% ($p<0.05$). ALT activity decreased by 24.2% ($p<0.05$) and AST by 13.1% ($p<0.05$).

The results of this study demonstrate that the ratio of standardized ileal digestible (SID) isoleucine to lysine in pig diets significantly influences growth performance and metabolic indicators in young pigs.

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