

MINISTRY OF EDUCATION AND
TRAINING

MINISTRY OF AGRICULTURE AND
ENVIRONMENT

**REDUCING THE CRUDE PROTEIN LEVEL BASED ON
AMINO ACID BALANCING IN THE DIET FOR COLOUR
FEATHER BROILER**

MAJOR: NUTRITION AND ANIMAL FEED
CODE: 9.62. 01. 07

DOCTORAL THESIS SUMMARY

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Supervisor:

- 1. Assoc. Prof. Dr. Pham Kim Đang**
- 2 Assoc. Prof. Dr. Tran Thi Bich Ngoc**

Reviewer 1: Assoc. Prof. Dr. Le Dinh Phung

Reviewer 2: Assoc. Prof. Dr. Nguyen Hung Quang

Reviewer 3: Dr. Pham Cong Thieu

**The thesis will be defended before the Thesis Evaluation Council at National
Institute of Animal Science**

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GENERAL INTRODUCTION

1. INTRODUCTION

Vietnam's livestock industry depends on 65-70% of imported feed ingredients. According to the Department of Livestock Production (2024), the amount of imported feed ingredients in 2023 was about 16.8 million tons, equivalent to 6.8 billion USD, excluding ingredients of animal origin. Meanwhile, imported raw materials providing protein for livestock are mainly soybeans meal and other oil cakes were 5.24 million tons (equivalent to 2.63 billion USD), accounting for 31.2% of the total imported feed ingredients and 39.2% of the total import value. The strongly dependence on imported feed ingredients, especially rich-protein ingredients make the livestock industry in general and poultry industry in particular unstable, high feed cost, thereby increasing production costs and reducing profits.

Poultry diet is mixed from many ingredients with different amino acid profiles. Therefore, creating a diet with enough amino acids for poultry requirement and minimize excess amino acids is always a concern. Using a standard ileal digestible (SID) amino acid database with the use of industrial single amino acids was considered an effective solution in reducing crude protein level in the animal diet (Selle et al., 2020). However, in Vietnam, research on the SID amino acid database is still limited in terms of raw material types and the number of samples in the research (Ho Le Quynh Chau, 2014). Currently, Vietnam have not yet had a database of SID amino acid requirements for poultry and normally apply the foreign recommendations. These recommendations were developed for high-yielding or fast-growing chicken breeds, while colored feather broilers dominate the broiler breed structure in Vietnam and was defined as slow-growing broilers (Commission Regulation (EC) No 889/2008). Luong Phuong chicken is one of important chicken breed, often used to create commercial breed for improving growth performance (Desvaux et al., 2008; Nguyen Van Duy et al., 2020). Therefore, the study to determine the database of SID amino acids as a basis for reducing crude protein level in the diet of Luong Phuong broiler chickens is necessary to use protein sources more effectively, reduce feed costs and eliminate odour gas emissions in colored feather broiler production in Vietnam.

2. OBJECTIVES

2.1. General objective

Determine the crude protein reduction level based on amino acid balance in colored feather broiler diet.

2.2. Specific objective

Determine the standardized ileal digestibility ratio of amino acid in common feed ingredients in broiler production in Vietnam and appropriate proportions of standard ileal digestible essential amino acids in broiler diets.

Formulate the diet for commercial colored feather broiler chickens with reducing crude protein level by 2-3% without changing growth performance, productivity and meat quality, reducing odorous gas emissions, reducing feed costs and improving economic efficiency

3. THE NEW CONTRIBUTIONS OF THE THESIS

Providing a database of standard ileal digestibility of amino acids of 15 common feed ingredients in Vietnam poultry production on Luong Phuong chickens, contributing to enrich the database of nutritional value of feed ingredients in Vietnam.

Developing a diet for colored feather broiler chickens with a 2% crude protein reduction for three growth stages based on balancing essential amino acids in the SID form. An experiment to apply the research results to compare a diet with 2% protein reduction to the currently used feed on the farm in two type of chicken breeds including Luong Phuong and Luong Phuong cross Mia chickens. The result showed that, using the low protein diet with the balancing essential amino acid reduced the feed cost per kg of weight gain from 1-84 days by 6.97% and 8.10%, respectively.

4. SCIENTIFIC AND PRACTICAL SIGNIFICANCE

4.1. Scientific significance

The thesis determined the standard ileal digestibility ratio of amino acids in 15 common chicken feed ingredients in Vietnam on Luong Phuong chickens and the appropriate ratio of SID amino acids in the diet for Luong Phuong chickens. The results of the thesis contribute to enriching the database on the nutritional value of feed ingredients and the requirement for SID amino acids for Luong Phuong chickens in Vietnam.

From the database on SID amino acids, the research determined the reduction of 2% crude protein on the basis of balancing standard ileal digestible amino acids on Luong Phuong chicken diet, to reduce feed costs and odour gas emissions. The research results of the thesis were the scientific basis contributing to the development of optimal feed formulation, reducing feed costs and increasing the efficiency of broiler production. Otherwhile, the research results were the references for further research and teaching materials in the field of nutrition and animal feed.

4.2. Practical significance

The database of standard ileal digestibility of amino acids of common feed ingredient in poultry production can be applied in the diet formulation in broiler farms and feedmill.

The database of appropriate ratio of essential amino acid in SID form and the crude protein reduction level in the diet for Luong Phuong chickens can be applied in broiler feed production to save feed costs, reduce odour gas emissions and saving protein source in poultry diet, thereby reducing the pressure on importing protein-rich ingredients.

CHAPTER 1. LITERATURE REVIEW

1.1. Scientific basis of research issue

The research issue of the thesis is the reduction of crude protein levels in broiler diets based on the standardized ileal digestibility ratio of amino acids in feed ingredients and the SID amino acid requirements for broiler. Reducing crude protein content in the diet while balancing amino acids in SID form decreased the use of protein-rich ingredients, improve gut health and protein digestibility, reduce emissions of NH_3 and H_2S gases, minimize the effects of heat stress, and lessen water intake. However, using low-protein diets also face the challenges such as reduced growth performance, impacts on feed palatability, and the high cost of industrial amino acids. Therefore, determining the reasonable level of protein reduction is a key issue of interest.

1.2. Research landscape domestically and internationally

Overview of both Vietnamese and international studies showed that reducing crude protein content in poultry diets is a trend in modern poultry nutrition. Worldwide, research on building databases for evaluating the nutritional value of feed as well as the nutritional requirements for poultry is regularly updated. Based on that, many studies on low-protein diets with the supplementation of both essential and non-essential amino acids have been conducted. However, these studies have mostly been conducted on high-yield broiler breeds, which have different characteristics and growth rates compared to colored feather breeds (slow-growing chickens).

Domestic studies on protein nutrition, although focused on colored feather chickens, but mainly addressed on protein and a few amino acids requirements for that breed. Studies focusing on the ileal digestibility coefficients of amino acids in feed, as well as SID amino acid requirement databases for chickens are still limited. Consequently, studies on reducing crude protein content in broiler diets have not yet been conducted systematically or have not applied the SID amino acid database.

In the context of Vietnam's animal feed industry mainly relied on imported protein sources ingredients, the effective use of protein or the reduction of crude protein in diets without affecting animal productivity is becoming an inevitable trend. Therefore, research to build a database on SID amino acid of feed and SID amino acid requirements for poultry are basis database for formulating low-protein diets in broiler production.

CHAPTER 2. MATERIALS AND METHODS

2.1. RESEARCH SUBJECT

Luong Phuong and Luong Phuong cross Mia broiler.

15 types of feed ingredients were selected to determine the standard ileal digestibility of amino acids divided into two groups including:

- (1) Cereals and cereal by-products: Rice bran extract (India), whole rice bran (Vietnam), wheat bran (India), brown rice (Vietnam), rice bran (Vietnam), broken rice (Vietnam), corn (Vietnam);
- (2) Rich-protein ingredients: Corn DDGS (Europe), corn gluten meal (USA), soybean (Vietnam), soybean meal (Argentina), canola meal (India), fish meal (Vietnam), meat and bone meal (Europe) and blood meal (America).

2.2. TIME AND LOCATION

Time: From 2020 to 2022

Location:

- Department of Animal Nutrition and Feed, National Institute of Animal Science,
- Department of Feed and Livestock Products Analysis, National Institute of Animal Science,
- High Quality Livestock Breeds Centre, Faculty of Animal Science, Vietnam National University of Agriculture.
- Amino Lab, Evonik Company, Singapore
- Center for Environmental Monitoring and Modeling Research, University of Sciences

2.3. EXPERIMENTS

Experiment 1: Determination of standard ileal digestibility of amino acids of some common feed ingredients in poultry production

Experiment 2: Determination of standard ileal digestible lysine levels in the diet of colored feather broilers

Experiment 3: Determination of optimal ratio of standard ileal digestible essential amino acids in the colored feather broiler diet.

Experiment 4: Determination of appropriate crude protein reduction based on the balance of standard ileal digestible essential amino acids in the colored feather broiler diet.

Experiment 5: Testing the low protein diets based on the optimal balance of standard ileal digestible essential amino acids for colored feather broilers

2.4. METHODS

2.4.1. Determination of standard ileal digestibility of amino acids of some common feed ingredients in poultry production

The experiment was conducted on 204 day-old Luong Phuong chickens, the chickens were fed the same diet until the 28 days old (feed ad libitum). On the day 29, the chickens

were weighed, then 192 chickens were selected and divided to 16 treatments, each treatment included 12 chickens raised in 4 cages (2 female cages and 2 male cages), 3 chickens/cage, each cage was a replication. The experiment was conducted on 15 feed ingredients, one treatment to calculate endogenous amino acids (16 treatments x 3 chickens/replication x 4 replications = 192 chickens). After that, the chickens continued to feed commercial diet to adapt to the cages condition.

Feeding and collecting digestive digesta

From the day 46 of age, the chickens step to step change from the commercial feed to experimental feed. From day 50, chick was fed by experimental feed only. Feeding and collection of digestive digesta process was referred the procedure of Kadim and Moughan (1997). Summary as follows:

Chickens were fed the experimental diet and the protein-free diet ad libitum for 5 days. Fasting on the day 6. On the day 7, the digestive digesta were collected: Chick was fed for 1 hour, recorded the feed consumed. After 4 hours of feeding, all experimental chickens (192 chickens) were slaughtered to collect digestive digesta. Chickens were arranged to be fed and slaughtered alternately to ensure that all chickens were slaughtered to collect digesta after 4 hours of feeding. Digestive digesta were collected in the ileum segment determined from Meckel's node to the ileo-caecal valve. Digestive digesta samples from all chickens in one cage were mixed together and stored at -20°C. The total number of digestive samples was 16 treatments x 4 replicates = 64 samples.

Chemical analysis

The samples were analyzed at the Department of Analysis of Feed and Livestock Products, National Institute of Animal Science and at Amino Lab of Evonik Company, Singapore. The basic nutritional composition of the samples was analyzed according to Vietnamese standard; dry matter (TCVN 4326:2001), crude protein (TCVN 4328:2007); total ash (TCVN 4327:2007); Ca (TCVN 1526:2007); total P (TCVN 1525:2001); ash insoluble in acid (TCVN 9474:2012). The amino acid content was determined after hydrolyzing the sample in HCl solution for 24 hours at 110°C, then the amino acids were analyzed by ultra-pressure liquid chromatography (UPLC). The tryptophan content was analyzed according to TCVN 5283:2007.

Statistical analysis

The data was analyzed by descriptive statistics using Excel and Minitab 16 software.

2.4.2. Determination of standard ileal digestible lysine levels in the diet of colored feather broilers.

The experiment was conducted on 750 days-old Luong Phuong chickens and was designed using a one-factor completely randomized design with 5 levels of standard ileal digestible lysine (SID-Lys), each level being a treatment. The SID-Lys levels were set up based on the recommendations of Cobb (2018), Aviagen (2014) and Brazil (Rostagno et al., 2011) for broilers. Accordingly, the standard SID-Lys levels selected were 1.2; 1.05

and 0.95% respectively for the three growth stages, then these levels were designed to increase or decrease by 0.1 and 0.2% respectively.

Table 2.1. Experiment design of determining SID-Lys level in the colour feather broiler

Parameter	TM1	TM2	TM3	TM4	TM5
No of chicks/replicate	30	30	30	30	30
No of replicate	5	5	5	5	5
Total chicks	150	150	150	150	150
Experiment time (day)	84	84	84	84	84
1-28 days old					
Crude protein (%)			20,8*		
ME (Kcal/kg)			3000*		
SID-Lys (%)	1,00	1,10	1,20	1,30	1,40
29-56 days old					
Crude protein (%)			19,0*		
ME (Kcal/kg)			3100*		
SID-Lys (%)	0,85	0,95	1,05	1,15	1,25
57-84 days old					
Crude protein (%)			18,0*		
ME (Kcal/kg)			3150*		
SID-Lys (%)	0,75	0,85	0,95	1,05	1,15

*Note: TM: Treatment *Crude protein and ME level in the diet was determine by averaging of those value in commercial feed for colour feather chickend of Japfa, Dabaco, Guyomarch companies. In the diets, other SID-essential amino acid was balanced based on recommendation of Aviagen (2014).*

Measurements

Total tract nutrient digestibility; nitrogen balance; H₂S and NH₃ gas emission in chicken drop; growth performance and meat productivity and quality.

Chemical analysis

Samples were analyzed at the Department of Analysis of Feed and Livestock Products, National Institute of Animal Science and Amino Lab, Evonik Company, Singapore. The basic nutritional composition of the samples was analyzed according to Vietnamese standard; Dry mater (TCVN 4326:2001), crude protein (TCVN 4328:2007); total ash (TCVN 4327:2007), Ca (TCVN 1526:2007), total P (TCVN 1525:2001). The amino acid content was determined after hydrolyzing the sample in HCl solution for 24 hours at 110°C, then the amino acids were analyzed by ultra-pressure liquid chromatography (UPLC). Tryptophan content was analyzed according to (TCVN 5283:2007).

Statistical analysis

Data were statistically processed using the analysis of variance (ANOVA) method to evaluate the difference among the mean values of the treatments, Tukey-test was used to analyze the difference between two mean values with a confidence level of 95%. Data were statistically processed using minitab 16 software (2012).

A non-linear regression model (broken-line) was used to estimate the SID-lys requirement for maximum ADG and minimum FCR in Luong Phuong broiler diet at different growth stages according to the model of Robbins et al. (2006) using SAS 9.4 software.

2.4.3. Determination of optimal ratio of standard ileal digestible essential amino acids in the colored feather broiler diet.

The experiment was conducted on 750 day-old Luong Phuong chickens and was designed in a one-factor completely random design with 5 treatments, the SID-EAA/Lys ratios including Met + Cys, Thre, Tryp and Agr were 90; 95; 100; 105; 110%, respectively, compared to the recommendations of Aviagen (2014). Each treatment included 150 chickens raised in 5 floor pens as 5 replications. The corresponding protein levels for the 3 stages were 20.8%; 19% and 18%; ME levels were 3000; 3100 and 3150 kcal and SID-lys levels were 1.3; 1.15 and 0.95%.

Table 2.2. Experiment desing of determining optimal ratio of standard ileal digestible essential amino acids in the colored feather broiler diet

Treatments	TM 1	TM2	TM3	TM4	TM5
1-28 days old					
Crude protein (%)			20,8*		
ME (Kcal/kg)			3000*		
SID-Lys (%)	1,30	1,30	1,30	1,30	1,30
SID-Methionine/lysine (%)	36,45	38,48	40,5**	42,53	44,55
SID-Meth+Cys/lysine (%)	67,50	71,25	75**	78,75	82,50
SID-Threonine/lysine (%)	60,30	63,65	67**	70,35	73,70
SID-Tryptophan/lysine (%)	14,04	14,82	15,6**	16,38	17,16
SID-Arginine/lysine (%)	96,30	101,65	107**	112,35	117,7
29-56 days old					
Crude protein (%)			19,0*		
ME (Kcal/kg)			3100*		
SID-Lys (%)	1,15	1,15	1,15	1,15	1,15
SID-Methionine/lysine (%)	37,80	39,9	42**	44,1	46,20
SID-Meth+Cys/lysine (%)	70,65	74,58	78,5**	82,43	86,35
SID-Threonine/lysine (%)	60,30	63,65	67**	70,35	73,70
SID-Tryptophan/lysine (%)	14,04	14,82	15,6**	16,38	17,16
SID-Arginine/lysine (%)	96,30	101,65	107**	112,35	117,7
57-84 days old					
Crude protein (%)			18,0*		
ME (Kcal/kg)			3150*		
SID-Lys (%)	0,95	0,95	0,95	0,95	0,95
SID-Methionine/lysine (%)	37,35	39,43	41,5**	43,58	45,65
SID-Meth+Cys/lysine (%)	70,20	74,10	78**	81,90	85,80
SID-Threonine/lysine (%)	60,30	63,65	67**	70,35	73,70
SID-Tryptophan/lysine (%)	14,04	14,82	15,6**	16,38	17,16
SID-Arginine/lysine (%)	96,30	101,65	107**	112,35	117,70

Measurement, chemical analysis and statistical analysis

Similar as experiment 2

2.4.4. Determination of appropriate crude protein reduction based on the balance of standard ileal digestible essential amino acids in the colored feather broiler diet

The experiment was carried out on 750 day-old Luong Phuong chickens randomly divided to ensure uniformity in body weight and gender into 25 floor pens, each pen had 30 chickens. The experiment was designed using a one-factor completely randomized method with 5 treatments as 5 levels of crude protein in the diet and 5 replications for each treatment.

Table 2.3. Experiment design of determining crude protein reduction level based on balancing amino acid in standard ileum digestible form in colour feather broiler diet.

Treatments	TM 1	TM 2	TM 3	TM 4	TM 5
1-28 days old					
Crude protein (%)	20,8*	19,8	18,8	17,8	16,8
ME (Kcal/kg)			3000*		
SID-Lys (%)			1,3		
SID-Met/Met+Cys/Thre/Tryp/Arg (%)		0,53/0,98/0,87/0,2/1,39			
29-56 days old					
Crude protein (%)	19*	18	17	16	15
ME (Kcal/kg)			3100*		
SID-Lys (%)			1,15		
SID-Met/Met+Cys/Thre/Tryp/Arg (%)		0,48/0,9/0,77/0,18/1,23			
57-84 days old					
Crude protein (%)	18*	17	16	15	14
ME (Kcal/kg)			3150*		
SID-Lys (%)			0,95		
SID-Met/Met+Cys/Thre/Tryp/Arg (%)		0,39/0,74/0,64/0,15/1,02			

Measurement, chemical analysis and statistical analysis

Similar as experiment 2

2.4.5. Development and testing of low protein diets based on the optimal balance of standard ileal digestible essential amino acids for colored feather broilers

Two experimental models were implemented on Luong Phuong chickens and Luong Phuong hybrid Mia chickens. Each experimental model was carried out on 300 day-old chickens and was designed using a completely randomized method with 2 treatments. Each treatment consisted of 150 chickens, raised in 5 separated pens (30 chickens/pen, each pen was a replication). Treatment 2 used the feed currently used by the farm, treatment 1 used a diet with a 2% protein level lower than treatment 2 and supplemented with essential amino acids according to the research results of experiment 2, 3 and 4.

Measurement: Chicken were weighted at 1, 28, 56 and 84 days old; feed intake and, feed conversion ratio (kg feed/kg weight gain); feed cost (VND/kg weight gain).

Statistical analysis: Data was analyzed by analysis of variance (ANOVA) method to evaluate the difference between means using Minitab 16 software (2012).

CHAPTER 3. RESULT AND DISCUSSION

3.1. DETERMINATION OF STANDARD ILEAL DIGESTIBILITY OF AMINO ACIDS OF SOME COMMON FEED INGREDIENTS IN POULTRY PRODUCTION

3.1.1. Endogenous amino acid and standard ileum digestibility amino acid ratio in experimental feed ingredients.

Methionine and cystine were the two amino acids with the lowest values in the endogenous amino acids, accounting for 1.03 and 2.3% of the total endogenous amino acid flow, respectively. The highest amino acid contents in endogenous proteins were glutamic acid, aspartic acid, threonine, serine and proline, ranging from 627.06 to 953.84 mg/kg DM intake.

Cereals and cereal by-products:

Among the four cereals, rice had the lowest SID-AA, ranging from 65.23 to 85.23%, and corn had the highest value with an average SID-AA value of 85.9%. In the raw materials made from paddy rice (paddy rice, broken rice, brown rice), lysine and arginine had high SID-AA, all above 80%, while cystine, methionine and threonine had the lowest SID-AA value from 57.43 to 77.25% except for threonine in brown rice (83.22%). The digestibility of cystine in broken rice and paddy was very low with values of 60.01 and 65.23%, respectively, while these values in brown rice and corn reached 77.25 and 82.27%. Overall, among the four cereal feed ingredient, corn showed superior SID-AA levels compared to ingredients from paddy rice.

In the group of 3 cereal by-products, wheat bran showed superiority in the SID-AA ratio compared to de-oil rice bran and full-fat rice bran with an average SID-AA of 82.12% compared to 49.01 and 73.01%. In wheat bran, glutamic acid and proline were the amino acids with the highest SID. In addition, the SID of sulfur-containing amino acids such as cystine and methionine were also relatively high, reaching over 80%. Histidine and glycine were the amino acids with the lowest SID in wheat bran. As a by-product of rice milling, de-oil rice bran had a much lower SID-AA level than full-fat rice bran, with an average of 73.01% in whole-oil rice bran while less than half of the amino acids of de-oil rice bran were digested in the ileum of broilers (49.01%). The SID of cystine in both types of rice bran was very low, only 35.25 and 62.89%, respectively, while arginine, alanine and glutamic had the highest SID levels.

Protein-rich ingredients

Corn protein-rich by-products showed high digestibility of amino acids with average SID-AA values of 73.28 and 79.77% for DDGS and corn gluten meal, respectively, corn gluten meal showed higher digestibility than DDGS for most of amino acids. Lysine in DDGS showed the lowest digestibility with only 58.89% while the least digestible amino acid in corn gluten meal was histidine (70.84%). In the oilseed and by-product groups, full-fat soybeans showed very high digestibility of all amino acids with average SID-AA of 91.03%, especially SIDs of tryptophan (94.86%) and threonine (94.23%). Soybean meal and canola meal also showed very high digestibility of amino acids with average values of 80.86% and 82.05%, respectively. The SID of cysteine in soybean meal was much lower than that in canola meal (72.57 vs. 84.02%) while the SID of threonine in both ingredients was similar (77.68 and 76%).

Table 3.3. Endogenous amino acid and standard ileum digestibility amino acid ratio of cereals and cereal by-products

Parameter	EAA (mg/kg DM)	Standard ileum digestibility ratio (%)						
		De-oil rice bran	Full-fat rice bran	Paddy rice	Brown rice	Broken rice	Wheat bran	Corn
Protein	12368,6 ± 206,6	55,23 ± 5,40	73,71 ± 1,49	76,24 ± 4,02	80,96 ± 1,75	75,45 ± 2,90	81,96 ± 1,64	85,20 ± 2,13
Methionine	128,25 ± 2,02	52,41 ± 6,68	71,54 ± 2,35	70,35 ± 6,02	76,35 ± 2,63	57,43 ± 8,67	83,57 ± 1,66	86,96 ± 5,47
Cystine	279,5 ± 4,81	35,25 ± 6,30	62,89 ± 1,74	65,23 ± 7,78	77,25 ± 1,93	60,01 ± 7,90	80,26 ± 2,73	82,27 ± 2,04
Methionine+Cystine	407,75 ± 6,74	38,72 ± 11,64	67,13 ± 1,60	67,93 ± 6,83	76,79 ± 2,26	58,57 ± 8,28	81,68 ± 2,24	84,55 ± 3,70
Lysine	409,45 ± 9,19	57,61 ± 5,57	72,71 ± 2,48	84,88 ± 4,49	87,64 ± 1,32	88,82 ± 2,41	80,10 ± 1,95	90,66 ± 1,20
Threonine	830,64 ± 13,87	45,12 ± 9,59	76,60 ± 2,19	71,02 ± 8,18	83,22 ± 1,41	72,5 ± 2,69	81,79 ± 3,07	87,48 ± 3,18
Tryptophan	74,00 ± 4,72*	55,46 ± 5,72	78,38 ± 1,25	78,8 ± 5,49	83,84 ± 0,59	82,56 ± 3,54	84,19 ± 2,4	92,35 ± 2,86
Arginine	404,78 ± 7,26	68,30 ± 4,83	80,74 ± 1,86	85,23 ± 3,09	81,24 ± 1,21	82,92 ± 1,66	81,44 ± 1,50	88,19 ± 1,71
Isoleucine	395,96 ± 6,97	47,72 ± 7,57	73,93 ± 2,66	79,63 ± 5,12	80,78 ± 1,19	76,25 ± 2,04	84,41 ± 1,65	87,68 ± 4,17
Leucine	588,46 ± 11,95	50,38 ± 6,99	73,41 ± 2,22	78,26 ± 4,99	80,08 ± 1,67	77,04 ± 2,34	83,55 ± 1,64	83,55 ± 7,42
Valine	549,02 ± 11,25	50,61 ± 7,24	75,71 ± 2,06	79,42 ± 4,84	81,86 ± 1,07	78,29 ± 1,71	82,55 ± 1,64	86,87 ± 2,95
Histidine	304,12 ± 7,30	47,25 ± 8,32	71,79 ± 1,78	76,78 ± 5,83	80,36 ± 1,54	76,82 ± 2,49	77,98 ± 2,00	81,80 ± 1,91
Phenylalanine	279,24 ± 6,54	53,53 ± 6,23	75,31 ± 2,42	82,18 ± 3,78	80,47 ± 1,67	79,60 ± 2,00	85,05 ± 1,29	86,68 ± 6,31
Glycine	506,12 ± 9,00	41,71 ± 9,60	70,10 ± 1,80	75,63 ± 5,60	80,81 ± 1,63	77,29 ± 2,22	75,46 ± 2,22	84,14 ± 1,20
Serine	692,24 ± 11,21	49,33 ± 8,60	74,41 ± 1,75	72,95 ± 6,85	81,21 ± 1,60	74,14 ± 2,77	81,95 ± 2,38	85,25 ± 2,52
Proline	627,06 ± 9,50	42,76 ± 8,63	70,86 ± 1,54	68,41 ± 6,94	80,68 ± 1,84	72,55 ± 3,11	87,97 ± 1,53	78,92 ± 4,34
Alanine	420,45 ± 7,85	54,61 ± 6,60	76,55 ± 2,17	77,96 ± 4,96	80,86 ± 1,69	78,31 ± 1,97	79,32 ± 1,91	85,94 ± 5,48
Aspartic	915,84 ± 15,3	38,07 ± 10,33	71,23 ± 2,24	78,89 ± 4,93	81,4 ± 1,23	80,29 ± 1,78	78,88 ± 2,27	88,41 ± 1,79
Glutamic	953,84 ± 14,53	53,38 ± 7,3	75,22 ± 1,94	77,72 ± 4,63	78,25 ± 1,84	73,32 ± 3,68	88,00 ± 1,20	84,59 ± 6,27
Average		49,01 ± 7,31	73,01 ± 1,83	75,91 ± 5,55	80,5 ± 1,49	74,36 ± 3,04	82,12 ± 1,79	85,90 ± 3,02

*Note: EAA: Endogenous amino acids; DM: Dry mater; * Reference value from research of Kong and Adeola (2013)*

Table 3.4. Standard ileum digestibility amino acid ratio of protein-rich ingredients

Chỉ tiêu	Standard ileum digestibility ratio (%)							
	Corn DDGS	Corn Gluten Meal	Full-fat soybean	Soybean meal	Canola meal	Fish meal	Meat and bone meal	Blood meal
Protein	75,10±4,29	79,47 ± 1,72	91,32 ± 3,93	80,91 ± 2,46	77,84 ± 1,39	88,59 ± 3,27	68,41 ± 5,35	42,78 ± 4,10
Methionine	82,70±3,32	83,12 ± 5,75	91,95 ± 3,84	84,73 ± 1,66	85,89 ± 0,82	87,82 ± 2,64	65,83 ± 7,10	47,05 ± 7,48
Cystine	70,31±5,58	72,97 ± 3,46	90,28 ± 5,46	72,57 ± 2,64	84,02 ± 1,30	90,41 ± 6,84	48,58 ± 5,24	62,12 ± 6,95
Methionine+Cystine	76,50±4,35	78,78 ± 2,87	91,10 ± 4,66	78,49 ± 2,15	84,80 ± 1,09	88,46 ± 3,57	57,09 ± 6,00	54,70 ± 6,74
Lysine	58,89±6,66	78,55 ± 11,00	90,14 ± 3,29	84,73 ± 1,10	80,14 ± 1,10	91,57 ± 2,89	63,21 ± 7,06	50,65 ± 6,47
Threonine	67,47±5,97	76,39 ± 5,36	94,23 ± 4,63	77,68 ± 2,22	76,00 ± 2,70	94,09 ± 5,32	66,11 ± 6,20	54,72 ± 6,18
Tryptophan	64,69±8,5	93,27 ± 4,35	94,86 ± 2,6	87,55 ± 3,68	84,57 ± 2,76	89,49 ± 6,12	72,78 ± 2,84	52,18 ± 6,75
Arginine	78,49±3,95	82,46 ± 0,89	92,17 ± 3,07	86,12 ± 1,02	88,34 ± 0,69	91,87 ± 2,44	68,22 ± 8,09	50,05 ± 6,62
Isoleucine	75,61±4,36	79,81 ± 3,34	91,26 ± 3,48	81,20 ± 1,81	80,04 ± 1,68	92,80 ± 3,15	69,12 ± 7,07	54,49 ± 11,21
Leucine	85,48±2,83	82,78 ± 8,23	90,96 ± 3,35	79,98 ± 1,89	81,82 ± 1,66	92,74 ± 2,91	67,78 ± 7,13	49,79 ± 6,70
Valine	75,07±4,47	79,35 ± 1,14	92,21 ± 3,74	80,58 ± 1,85	79,40 ± 1,81	93,32 ± 3,10	69,04 ± 6,83	49,84 ± 8,33
Histidine	67,83±5,72	70,84 ± 4,15	90,46 ± 3,80	79,38 ± 1,38	83,81 ± 1,27	87,22 ± 5,28	54,62 ± 7,15	47,07 ± 6,85
Phenylalanine	83,97±2,92	82,89 ± 6,96	91,53 ± 3,11	81,94 ± 1,69	85,47 ± 1,31	92,68 ± 2,41	71,07 ± 7,21	48,73 ± 6,66
Glycine	63,82±6,03	74,19 ± 6,42	89,83 ± 4,09	77,98 ± 1,56	79,18 ± 1,71	85,40 ± 3,35	63,30 ± 8,46	51,31 ± 6,71
Serine	73,26±5,04	80,62 ± 2,17	92,03 ± 3,93	79,77 ± 1,70	77,27 ± 2,16	89,35 ± 4,73	62,88 ± 7,10	53,59 ± 5,46
Proline	76,72±4,23	77,30 ± 3,23	91,16 ± 3,74	79,04 ± 1,79	77,90 ± 1,74	87,40 ± 3,62	60,07 ± 8,36	51,23 ± 6,00
Alanine	82,12±3,29	82,82 ± 6,64	91,04 ± 3,62	80,27 ± 1,58	82,17 ± 1,65	90,05 ± 2,73	65,61 ± 8,04	49,97 ± 6,44
Aspartic	64,53±6,12	78,44 ± 0,66	87,62 ± 3,79	79,79 ± 1,32	78,71 ± 1,35	80,02 ± 5,08	49,08 ± 7,18	48,48 ± 7,06
Glutamic	82,80±3,30	81,63 ± 7,35	89,96 ± 3,28	83,85 ± 1,05	87,65 ± 0,82	90,12 ± 2,96	62,76 ± 7,23	50,41 ± 6,71
Average	73,89±4,50	79,77 ± 1,25	91,03 ± 3,78	80,86 ± 1,56	82,05 ± 1,24	89,74 ± 3,59	63,17 ± 6,64	51,42 ± 6,79

In the group of animal originated protein-rich ingredients, the average SID-AA of the ingredients was: Fish meal: 89.74%; Meat and bone meal: 63.17%; Blood meal: 51.42%. Fish meal showed high and consistent SID of all amino acids with values around 90% except aspartic (80.02%). Cystine and aspartic had the lowest SID in meat and bone meal (48.58 and 49.08%) while methionine and histidine were the least digestible amino acids in blood meal with 47.05 and 47.07%.

3.2. DETERMINATION OF STANDARD ILEAL DIGESTIBLE LYSINE LEVELS IN THE DIET OF COLORED FEATHER BROILERS

3.2.1. Effect of different standard ileal digestible lysine levels in the diet on nutrient total tract digestibility and odour gas emission.

Table 3.5. Effect of different SID-Lys levels in the diet on nutrient total tract digestibility

Treatments	1 (n=5)	2 (n=5)	3 (n=5)	4 (n=5)	5 (n=5)	SEM	P
1-28 days old							
Dry matter (%)	68,89	68,34	68,68	68,70	69,22	0,867	0,966
Crude protein (%)	56,93 ^b	58,71 ^{ab}	59,13 ^{ab}	59,05 ^{ab}	60,29 ^a	0,605	0,020
Organic matter (%)	71,66	71,84	71,66	71,98	72,92	0,838	0,808
29-56 days old							
Dry matter (%)	72,60 ^b	75,01 ^{ab}	75,13 ^{ab}	76,70 ^a	76,98 ^a	0,899	0,026
Crude protein (%)	66,04 ^b	68,71 ^{ab}	69,13 ^{ab}	69,43 ^{ab}	71,15 ^a	0,851	0,012
Organic matter (%)	75,62 ^b	77,99 ^{ab}	78,71 ^{ab}	80,17 ^a	80,31 ^a	0,931	0,017
57-84 days old							
Dry matter (%)	69,41 ^b	71,74 ^{ab}	71,95 ^{ab}	72,72 ^a	72,53 ^a	0,641	0,016
Crude protein (%)	55,34 ^b	56,87 ^{ab}	57,56 ^{ab}	59,67 ^a	60,39 ^a	0,987	0,015
Organic matter (%)	73,48 ^b	75,30 ^{ab}	76,24 ^a	76,77 ^a	76,44 ^a	0,603	0,010

Note: Treatment 3 had SID-Lys 1.2; 1.05 and 0.95 for three growth stages, other treatment will be increase or decrease SID-lys by 0.1 or 0.2 %. Values in the same row with different letters are statistically different (P<0.05)

During the 1-28 days, protein total tract digestibility differed among treatments (P<0.05), treatment 1 with the lowest SID-Lys level had the lowest protein digestibility, but the statistically significant difference only occurred between treatments 1 and 5 (P<0.05). The same trend was observed in the other two stages when treatments 4 and 5 had the highest total tract protein digestibility. The marked improvement in protein digestibility in treatments with high SID-Lys levels resulted in higher dry matter and organic matter digestibility in treatments 4 and 5 than in treatment 1 (P<0.05) and similar to treatments 2 and 3 (P>0.05).

The level of NH₃ and H₂S gas emission in experimental chicken manure is presented in Table 3.7. The results of NH₃ gas emission in chicken manure in the period of 1-28 days did not differ between treatments (P>0.05). In the remaining two stages, the NH₃ gas content tended to decrease when chickens were fed diets with increasing levels of SID-Lys (P<0.05). Treatment 1 with the lowest SID-Lys level gave the highest

emission level in these two stages with 4.83 and 4.49 mg/m³, the lowest in treatment 5 with 4.26 and 3.56 mg/m³, respectively. Meanwhile, changing the SID-Lys content in the diet did not affect H₂S gas emission in experimental chicken manure in all 3 experimental stages.

Table 3.7. Effect of SID-Lys level in the diet to the NH₃ and H₂S emission

Treatments	1 (n=5)	2 (n=5)	3 (n=5)	4 (n=5)	5 (n=5)	SEM	P
1-28 days old							
H ₂ S (mg/m ³)	48,59	44,98	44,33	43,85	42,91	2,013	0,352
NH ₃ (mg/m ³)	5,02	4,84	4,77	4,73	4,46	0,256	0,646
29-56 days old							
H ₂ S (mg/m ³)	42,75	39,12	38,16	37,02	36,56	1,849	0,174
NH ₃ (mg/m ³)	4,83 ^a	4,54 ^{ab}	4,41 ^b	4,36 ^b	4,24 ^b	0,086	0,001
57-84 days old							
H ₂ S (mg/m ³)	37,17	34,86	34,46	33,91	33,60	1,201	0,276
NH ₃ (mg/m ³)	4,49 ^a	3,99 ^{ab}	3,85 ^{ab}	3,79 ^b	3,56 ^b	0,162	0,009

3.2.2. Effect of different standard ileal digestible lysine levels in the diet on growth performance and feed efficiency

Increasing the SID-Lys content in the diet resulted in an increase in ADG ($P < 0.05$) in the first two stages of the experiment. However, the difference in ADG among treatments during the 57-84 days was not statistically significant ($P > 0.05$). Overall, increasing the SID-Lys content in the diet increased the growth rate of the chickens, however, a significant difference was only observed between treatment 1 which had the lowest SID-Lys level compared to treatment 5 (the highest SID-Lys level).

Bảng 3.8. Effect of different SID-Lys level in the diet on average daily weight gain, feed conversion ratio and feed cost.

Treatment	1 (n=5)	2 (n=5)	3 (n=5)	4 (n=5)	5 (n=5)	SEM	P
Average daily weight gain (g/chick/day)							
1-28 days old	11,49 ^{bc}	11,37 ^c	11,59 ^{abc}	12,1 ^{ab}	12,21 ^a	0,170	0,006
29-56 days old	26,72 ^b	27,93 ^{ab}	27,76 ^{ab}	28,34 ^a	28,35 ^a	0,365	0,030
57-84 days old	30,04	30,45	31,45	30,32	30,86	0,540	0,415
1-84 days old	22,75 ^b	23,25 ^{ab}	23,6 ^{ab}	23,58 ^{ab}	23,81 ^a	0,203	0,013
Feed conversion ratio (kg feed/kg weight gain)							
1-28 days old	2,05 ^a	2,09 ^a	1,96 ^{ab}	1,88 ^b	1,91 ^b	0,032	<0,001
29-56 days old	2,61 ^a	2,40 ^b	2,35 ^b	2,32 ^b	2,29 ^b	0,032	<0,001
57-84 days old	3,35	3,39	3,26	3,24	3,18	0,081	0,306
1-84 days old	2,84 ^a	2,78 ^{ab}	2,69 ^{bc}	2,64 ^c	2,60 ^c	0,330	<0,001
Feed cost (đ/kg weight gain)							
1-28 days old	16673 ^{ab}	17114 ^a	16101 ^{abc}	15552 ^c	15981 ^{bc}	260,6	0,004
29-56 days old	21267 ^a	19691 ^b	19372 ^b	19254 ^b	19315 ^b	266,0	<0,001
57-84 days old	26252	26698	25864	25994	25901	641,0	0,819
1-84 days old	21397 ^a	21168 ^{ab}	20446 ^b	20267 ^b	20318 ^b	218,4	0,013

The SID-Lys level in the diet affected feed conversion in the first two stages of the experiment ($P < 0.05$). In the period of 1-28 days, birds fed diets containing 1.3 and 1.4% SID-Lys (treatments 4 and 5) had lower FCR (1.88 and 1.91 kg feed/kg weight gain) than diets 1 and 2 (2.05 and 2.09 kg). A similar trend was observed during the growing period (29-56 days of age). However, there were no statistically significant differences in FCR among diets during the 57-84 days ($P > 0.05$). For the whole experimental period, increasing dietary SID-Lys content reduced FCR ($P < 0.05$), with a reduction of 8.45% in treatment 5 compared to treatment 1 (2.6 vs. 2.84 kg feed/kg weight gain).

The SID-Lys content in the diet significantly affected the feed cost ($P < 0.05$) in the period of 1-28 days; 29-56 days and for the whole experimental period. From 1-84 days old, the lowest feed cost was in treatment 4 with 20,267 VND per kg of weight gain.

Table 3.10. Estimation of optimal SID-Lys level in the diet for different growth stages by broken-line regression

Period (day)	Parameter	Equation	Optimal SID-Lys	P-value	R ²
1 – 28	ADG	$Y = 12,085 - 2,44 (1,347 - \text{SIDLys})$	1,347	0,003	0,436
	FCR	$Y = 1,894 + 0,6404 (1,304 - \text{SIDLys})$	1,304	<0,001	0,513
29 – 56	ADG	$Y = 28,34 - 522 (1,117 - \text{dLys})$	1,117	0,013	0,328
	FCR	$Y = 2,305 + 1,30 (1,076 - \text{SIDLys})$	1,076	<0,001	0,706
57 – 84	ADG	$Y = 31,159 - 11 (0,906 - \text{SIDLys})$	0,906	0,044	0,323
	FCR	$Y = 3,147 + 1,427 (0,907 - \text{SIDLys})$	0,907	0,030	0,307
1 – 84	ADG	$Y = 23,63 - 4,04 (1,10 - \text{SIDLys})$	1,100	0,003	0,323
	FCR	$Y = 2,636 + 1,072 (1,06 - \text{SIDLys})$	1,060	<0,001	0,395

SIDLys: SID-Lys in the diet. Y: ADG or FCR at the different SIDLys level

A broken-line regression model was used to estimate the SID-Lys requirement for optimal ADG and FCR (Table 3.10). The SID-Lys content for maximum ADG was higher than that for minimum FCR. The estimated SID-Lys content for maximum ADG was 1.347; 1.117; 0.907 for the three experimental periods, respectively. Meanwhile, the estimated SID-Lys content for optimal FCR was 1.304; 1.076 and 0.907.

3.2.3. Effect of different standard ileal digestible lysine levels in the diet on meat productivity and meat quality.

Carcass yield (%) was affected by different SID-Lys levels in the diet ($P < 0.05$) with the highest values for chickens fed diets in treatment 4 and 5 (78.83% and 78.67%, respectively). There were significant differences in water holding capacity, pH and toughness among treatments ($P < 0.05$). The pH value of breast meat 15 minutes after slaughter was lowest in the treatment with the highest SID-Lys level. The rate of water

loss after storage was lowest in treatment 4 and highest in treatment 2. Toughness of meat tended to decrease with increasing SID-Lys in the diet ($P < 0.05$).

Table 3.11. Effect of different SID-Lys level in the diet on meat productivity and meat quality

Treatment	1 (n=10)	2 (n=10)	3 (n=10)	4 (n=10)	5 (n=10)	SEM	P
Live weight (g)	2019	2008	2036	2021	2056	21,42	0,596
Carcass weight (g)	1499 ^c	1526 ^{bc}	1542 ^{abc}	1591 ^{ab}	1617 ^a	20,16	0,003
Carcass yield (%)	74,15 ^b	75,83 ^b	75,62 ^b	78,83 ^a	78,67 ^a	0,648	<0,001
Abdominal fat (%)	1,29	1,32	1,65	1,58	1,53	0,190	0,590
Breast meat yield (%)	15,98	16,63	16,78	16,70	15,82	0,403	0,331
Thigh meat yield (%)	20,58	21,03	20,92	20,21	19,93	0,487	0,472
Water lost after 24h (%)	1,86 ^b	1,69 ^b	2,79 ^a	1,45 ^b	2,17 ^{ab}	0,207	0,002
Water lost after processing (%)	15,70 ^{ab}	14,41 ^b	18,86 ^a	15,78 ^{ab}	17,44 ^{ab}	0,855	0,014
pH 15	6,06 ^{ab}	6,18 ^a	6,04 ^{ab}	6,11 ^{ab}	5,86 ^b	0,070	0,046
pH 24	5,84	5,96	5,77	5,81	5,73	0,075	0,287
Toughness (N)	14,17 ^{ab}	16,47 ^a	14,89 ^{ab}	12,08 ^b	12,29 ^b	0,978	0,025

Conclusion: The results of this study showed that although increasing the SID-Lys content in the diet increased growth performance and reduced feed conversion rate of experimental chickens. However, in terms of feed costs, the appropriate SID-Lys levels in the diet for Luong Phuong chickens are 1.30; 1.15 and 0.95% respectively for the 3 growth stages of 1-28; 29-56 and 57-84 days. The corresponding crude protein and metabolizable energy levels were 20.8% and 3000 Kcal/kg; 19.0% and 3100 Kcal/kg; 18.0% and 3150 Kcal/kg. The results of SID-Lys levels in this study are applied for the up coming experiment of the thesis.

3.3. DETERMINATION OF OPTIMAL RATIO OF STANDARD ILEAL DIGESTIBLE ESSENTIAL AMINO ACIDS IN THE COLORED FEATHER BROILER DIET

3.3.1. Effect of different ratio of standard ileal digestible essential amino acid to lysine on nutrient total tract digestibility and odour gas emission

The digestibility of dry matter, protein and organic matter tended to increase during the growing stage, and decrease during the fattening stage. The digestibility of dry matter was similar among treatments in all three experimental stages ($P > 0.05$), ranging from 67.62 to 69.79% in period of 1-28 days; 75.71 to 78.06% in the period 29-56 days and 68.38 to 72.0% in period of 57-84 days. The digestibility of nutrients did not differ among treatments except for the digestibility of protein at the age of 29-56 days with the lowest level in treatment 5 (66.54%).

Table 3.13. Total tract digestibility of nutrient of chicken fed the diet with different SID-EAA/Lys ratio

Treatment	1 (n=5)	2 (n=5)	3 (n=5)	4 (n=5)	5 (n=5)	SEM	P
1-28 days old							
Dry matter (%)	69,58	69,79	69,53	67,62	68,13	1,96	0,902
Crude protein (%)	59,58	59,83	61,78	57,58	58,81	2,58	0,833
Organic matter (%)	73,06	73,18	72,93	70,97	71,53	1,81	0,863
29-56 days old							
Dry matter (%)	78,06	77,72	77,65	77,14	75,71	1,49	0,812
Crude protein (%)	70,44 ^a	71,54 ^a	71,67 ^a	71,39 ^a	66,54 ^b	0,67	0,001
Organic matter (%)	81,45	81,1	80,67	80,35	79,07	1,37	0,773
57-84 days old							
Dry matter (%)	68,38	71,23	68,59	68,82	72,00	1,15	0,152
Crude protein (%)	58,63	57,33	59,45	57,95	56,47	3,16	0,967
Organic matter (%)	71,50	74,54	72,61	72,63	75,74	1,08	0,111

Chickens from 1-28 days had higher gas emissions than those period of 29-56 and 57-84 days. There was no difference in the amount of gas emissions in the experimental chicken feces among the diets with different SID-EAA/lys ratios in all three experimental stages ($P>0.05$).

Table 3.15. The result of odour gas emission in chicken faeces while used the diet with different SID-EAA/lys ratios

Treatment	1 (n=5)	2 (n=5)	3 (n=5)	4 (n=5)	5 (n=5)	SEM	P
1-28 days old							
H ₂ S (mg/m ³)	43,83	43,00	41,72	42,14	42,70	1,219	0,776
NH ₃ (mg/m ³)	4,67	4,63	4,37	4,43	4,58	0,132	0,455
29-56 days old							
H ₂ S (mg/m ³)	38,11	36,70	34,78	35,29	36,62	1,566	0,600
NH ₃ (mg/m ³)	4,47	4,39	4,12	4,29	4,46	0,360	0,952
57-84 days old							
H ₂ S (mg/m ³)	35,88	34,07	32,20	33,26	33,38	1,558	0,561
NH ₃ (mg/m ³)	4,38	4,22	3,94	4,07	4,17	0,160	0,397

Note: Treatment 1-5 had the SID-EAA/Lys at 90, 95, 100, 105, 110% compare with the recommendation of Aviagen (2014).

3.3.2. Effect of different ratio of standard ileal digestible essential amino acid to lysine on growth performance and feed efficiency

Daily weight gain was different among treatments at period of 1-28, 57-84 days old and for the whole experimental period ($P<0.05$). Chickens fed diets with the ratio of SID-EAA/Lys at 100% of the Aviagen (2014) recommendation had the highest average weight gain from 1-84 days old with 23.16g/bird/day. Increasing and decreasing the ratio of SID-EAA/Lys both reduced chickens weight gain, especially at the decrease level of 10% (treatment 1) with the average ADG for the whole period only reaching (22.50g/bird/day) 2.85% lower than treatment 3 (23.16g/bird/day).

Table 3.16. Average daily weight gain, feed conversion ratio and feed cost in chicken fed the diet with different SID-EAA/lys ratio

Treatment	1 (n=5)	2 (n=5)	3 (n=5)	4 (n=5)	5 (n=5)	SEM	P
Average daily weight gain (g/chick/day)							
1-28 days old	12,71 ^a	12,64 ^a	12,97 ^b	12,86 ^{ab}	12,67 ^a	0,079	0,036
29-56 days old	26,42	26,90	27,54	26,91	26,76	0,253	0,070
57-84 days old	28,37 ^b	28,69 ^{ab}	28,97 ^a	28,39 ^{ab}	28,46 ^b	0,148	0,046
1-84 days old	22,50 ^b	22,74 ^b	23,16 ^a	22,72 ^b	22,63 ^b	0,090	0,001
Feed conversion ratio (kg feed/kg weight gain)							
1-28 days old	1,63 ^a	1,633 ^a	1,586 ^b	1,611 ^{ab}	1,633 ^a	0,011	0,046
29-56 days old	2,593 ^a	2,569 ^{ab}	2,514 ^b	2,529 ^{ab}	2,570 ^a	0,027	0,027
57-84 days old	3,324 ^{ab}	3,434 ^{ab}	3,284 ^b	3,468 ^a	3,434 ^{ab}	0,041	0,018
1-84 days old	2,72 ^{ab}	2,76 ^a	2,662 ^b	2,747 ^{ab}	2,757 ^a	0,022	0,028
Feed cost (d/kg weight)							
1-28 days old	18671 ^b	18873 ^b	18414 ^b	18780 ^{ab}	19194 ^a	148,0	0,001
29-56 days old	29614 ^{ab}	29497 ^{ab}	29031 ^b	29424 ^{ab}	30020 ^a	329,00	0,008
57-84 days old	36347 ^b	37876 ^{ab}	36464 ^b	38833 ^a	38741 ^a	452,40	0,001
1-84 days old	28211 ^{bc}	28748 ^{abc}	27969 ^c	29012 ^{ab}	29318 ^a	194,8	<0,0001

Note: Treatment 1-5 had the SID-EAA/Lys at 90, 95, 100, 105, 110% compare with the recommendation of Aviagen (2014). Values in the same row with different letters are statistically different (P<0.05).

Feed conversion ratio had a significant difference between treatments (P<0.05) in all 3 experimental stages. Treatment 3 had the smallest FCR with 2.662 kg feed/kg weight gain and the highest value in treatment 5 with 2.757 kg feed/kg weight gain for the whole experimental time from 1 to 84 days old.

The results showed that treatment 3 had the lowest feed cost (P<0.05) calculated for the whole experimental time which is 27,969 VND/kg weight gain, followed by treatments 1 and 2 with 28,211 and 28,748 VND, and the highest were treatments 4 and 5 with 29,012 and 29,318 VND.

3.3.3. Effect of different ratio of standard ileal digestible essential amino acid to lysine on meat productivity and meat quality

The different level of essential amino acids to lysine did not affect the productivity and meat quality indicators at 84-day-old of Luong Phuong chickens (P>0.05)

Table 3.18. Effect of different SID-EAA/Lys ratio in the diet on the meat productivity and meat quality

Treatment	1 (n=10)	2 (n=10)	3 (n=10)	4 (n=10)	5 (n=10)	SEM	P
Live weight (g)	1874,4	1896,3	1908,3	1891,4	1886,5	89,58	0,996
Carcass weight (g)	1456,0	1486,1	1500,6	1487,4	1475,4	71,15	0,987
Carcass yield (%)	77,65	78,41	78,67	78,6	78,23	0,527	0,630
Abdominal fat (%)	1,41	1,40	1,42	1,43	1,43	0,269	0,993
Breast meat yield (%)	20,61	20,67	20,8	20,86	20,61	0,333	0,973
Thigh meat yield (%)	15,91	16,11	16,33	16,23	15,93	0,401	0,900

Treatment	1 (n=10)	2 (n=10)	3 (n=10)	4 (n=10)	5 (n=10)	SEM	P
Water lost after 24h (%)	1,81	1,94	1,81	1,83	1,75	0,116	0,827
Water lost after processing (%)	18,32	18,57	18,43	18,5	18,35	1,053	0,991
Toughness (N)	14,35	14,74	14,51	14,75	14,60	0,691	0,961
Ph 15	6,16	6,18	6,19	6,20	6,19	0,071	0,995
Ph 24 h	5,84	5,91	5,85	5,86	5,87	0,041	0,763

Summary: The results showed that the appropriate SID-EAA/Lys level for Luong Phuong chickens was 100% compared to the recommendation of Avigen (2014), with the SID-lysine level at the period of 1-28, 29-56 and 57-84 days old being 1.30; 1.15 and 0.95%, respectively. The results of SID-EAA/Lys level in this study were used for the following experiment.

3.4. DETERMINATION OF APPROPRIATE CRUDE PROTEIN REDUCTION BASED ON THE BALANCE OF STANDARD ILEAL DIGESTIBLE ESSENTIAL AMINO ACIDS IN THE COLORED FEATHER BROILER DIET

3.4.1. Effect of crude protein reduction in the diet based on balance of standard ileal digestible essential amino acids on nutrient digestibility and odour gas emission

The total tract protein digestibility tended to increase as crude protein content decreased ($P < 0.05$), but the increase was only significant when dietary protein level decreased by 4%. Protein digestibility was highest in treatment 5 with 62.23; 72.48 and 61.44% corresponding to 3 growth stages, while protein digestibility in treatment 1 (highest crude protein level) reached 58.62; 68.21 and 57.65% respectively.

Table 3.20. Effect of crude protein reduction in the diet based on balance of standard ileal digestible essential amino acids on nutrient total tract digestibility

Treatment	1 (n=5)	2 (n=5)	3 (n=5)	4 (n=5)	5 (n=5)	SEM	P
1-28 days old							
Dry matter (%)	70,78	70,23	70,16	70,74	70,06	0,870	0,959
Crude protein (%)	58,62 ^b	59,07 ^{ab}	60,52 ^{ab}	60,52 ^{ab}	62,23 ^a	0,777	0,038
Organic matter (%)	74,05	74,00	74,62	75,30	75,27	0,787	0,641
29-56 days old							
Dry matter (%)	74,11	74,64	74,84	74,82	75,03	0,706	0,906
Crude protein (%)	68,21 ^b	69,39 ^{ab}	70,71 ^{ab}	71,65 ^{ab}	72,48 ^a	0,878	0,025
Organic matter (%)	79,22	79,05	79,07	79,11	80,2	0,464	0,387
57-84 days old							
Dry matter (%)	73,24	73,01	72,69	72,63	72,9	1,214	0,997
Crude protein (%)	57,65 ^b	58,62 ^{ab}	60,72 ^{ab}	60,79 ^{ab}	61,44 ^a	0,760	0,012
Organic matter (%)	76,69	76,17	76,34	76,63	76,44	1,028	0,993

Note: Treatment 1 had crude protein level for three experimental periods as follow: 20,8; 19,5 và 18%, the other treatment had the protein reduction from 1-4% at all three growth stages. Values in the same row with different letters are statistically different ($P < 0.05$).

Reducing crude protein level with the addition of some essential amino acids tended to significantly reduce H₂S and NH₃ emissions at the 1-28 and 29-56 days old period. However, at the 57-84 days old stage, only NH₃ emission levels showed differences among treatments (P<0.05). At 1 and 2% protein reduction, the amount of H₂S emitted was not different to the control treatment, the difference was only significant when the protein level was reduced by 3-4% (P<0.05). Meanwhile, NH₃ gas content decreased significantly as soon as the protein level was reduced by 1%. At 2% protein reduction, compared to the control (treatment 3), H₂S and NH₃ emissions decreased by 8.3% and 23.18% respectively for period of 1-28 days old; 9.4 and 27.92% (29-56 days old); 2.19 and 28.57% (57-84 days old).

Table 3.22. Effect of crude protein reduction in the diet based on balance of standard ileal digestible essential amino acids on H₂S and NH₃ emission

Treatment	1 (n=5)	2 (n=5)	3 (n=5)	4 (n=5)	5 (n=5)	SEM	P
1-28 days old							
H ₂ S (mg/m ³)	42,94 ^a	41,29 ^a	39,37 ^{ab}	36,65 ^b	35,21 ^b	1,275	0,020
NH ₃ (mg/m ³)	4,53 ^a	3,92 ^b	3,48 ^{bc}	3,37 ^c	3,13 ^c	0,517	0,011
29-56 days old							
H ₂ S (mg/m ³)	35,14 ^a	34,22 ^{ab}	31,83 ^{ab}	30,80 ^{bc}	30,14 ^c	0,952	0,005
NH ₃ (mg/m ³)	4,37 ^a	3,74 ^b	3,15 ^c	2,93 ^c	2,89 ^c	0,097	<0,001
57-84 days old							
H ₂ S (mg/m ³)	31,07	30,84	30,39	29,94	28,59	0,709	0,147
NH ₃ (mg/m ³)	3,99 ^a	3,42 ^b	2,85 ^c	2,76 ^c	2,64 ^c	0,130	<0,001

3.4.2. Effect of crude protein reduction in the diet based on balance of standard ileal digestible essential amino acids on growth performance and feed efficiency

In the whole experimental period (1-84 days old), the chicks increased their average weight from 22.72 to 26.87g/bird/day (table 3.23) and increased when the crude protein level increased. The period of 1-28 days of age showed more sensitivity to changes in protein level, when reducing 2% crude protein (treatment 3), the average daily weight had a significant difference compared to treatment 1 (P<0.05). However, in the period of 29-56 and 57-84 days of age and the whole experimental period, it can be seen that reducing 1 and 2% crude protein in treatments 2 and 3 did not show a statistically significant difference compared to treatment 1 (P>0.05).

Reducing crude protein level on the basis of SID-EAA balance increased FCR of the chickens (P<0.05). In the period of 1-28 days old, reducing 1% protein in the diet did not affect FCR (1.46 and 1.47 kg feed/kg weight gain) (P>0.05), however, when the reduction of 2; 3; 4%, FCR increased significantly to 1.53; 1.59 and 1.60 respectively in treatments 3, 4, 5 (P<0.05). In the period of 29-56 days of age, there was no difference in FCR in treatment 1 compared to treatments 2 and 3 which had a protein reduction of 1 and

2%. Overall, for the whole experimental period, treatment 1 had the lowest FCR with 2.77 kg feed/kg weight gain, increasing gradually in treatments 2, 3, 4 and highest in treatment 5 with 3.11 kgTĂ/kg of weight gain. There was no statistically significant difference between treatment 1 and treatments 2 and 3 ($P>0.05$).

Table 3.23. Effect of crude protein reduction in the diet based on balance of standard ileal digestible essential amino acids on growth performance and feed efficiency

Nghiem thức	1 (n=5)	2 (n=5)	3 (n=5)	4 (n=5)	5 (n=5)	SEM	P
Average daily weight gain (g/chick/day)							
GĐ 1-28 nt	19,28 ^a	19,03 ^{ab}	18,29 ^{bc}	17,75 ^{cd}	17,48 ^d	0,177	<0,001
GĐ 29-56 nt	35,02 ^a	34,21 ^{ab}	33,82 ^{ab}	30,9 ^{bc}	29,15 ^c	0,865	<0,001
GĐ 57-84 nt	26,32	25,40	25,61	25,39	21,53	1,230	0,087
GĐ 1-84 nt	26,87 ^a	26,21 ^{ab}	25,91 ^{ab}	24,68 ^b	22,72 ^c	0,461	<0,001
Feed conversion ratio (kg feed/kg weight gain)							
GĐ 1-28 nt	1,46 ^c	1,47 ^c	1,53 ^b	1,59 ^{ab}	1,60 ^a	0,014	<0,001
GĐ 29-56 nt	2,58 ^b	2,63 ^b	2,62 ^b	2,84 ^a	2,90 ^a	0,041	<0,001
GĐ 57-84 nt	4,00	4,08	4,06	4,14	4,65	0,160	0,056
GĐ 1-84 nt	2,77 ^c	2,81 ^c	2,84 ^c	2,97 ^b	3,11 ^a	0,028	<0,001
Feed cost (đ/kg weight)							
GĐ 1-28 nt	16642 ^b	16294 ^b	16749 ^{ab}	17097 ^a	16987 ^a	162,9	<0,001
GĐ 29-56 nt	29353 ^{ab}	29298 ^{ab}	28674 ^b	30594 ^a	30843 ^a	449,5	0,012
GĐ 57-84 nt	44250	44060	43205	43429	48181	1684,9	0,252
GĐ 1-84 nt	30082 ^{ab}	29884 ^b	29543 ^b	30373 ^{ab}	32003 ^a	486,2	0,017

3.4.3. Effect of crude protein reduction in the diet based on balance of standard ileal digestible essential amino acids on meat productivity and meat quality

Meat yield indicators including carcass yield, thigh meat and abdominal fat rate did not differ among treatments ($P>0.05$). Only breast meat rate tended to decrease gradually when crude protein level decreased. When the protein level was reduced from 1-3%, it did not affect the breast meat rate of chickens, however, when the reduction reached 4% (treatment 5), the breast meat ratio decreased significantly, corresponding to a decrease of 8.86% compared to treatment 1. Meat quality indicators including water loss and pH 15 minutes after slaughter did not differ among treatments. Notably, when the crude protein level was reduced by 4%, the pH reduction from slaughter to 24 hours was slowed down, as shown by the pH₂₄ index in treatment 5 having the highest value ($P<0.05$), different from treatments 1,2,3, while pH₁₅ had no difference among treatments. Tenderness of breast meat was affected by crude protein level in the diet ($P<0.05$).

Table 3.25. Effect of crude protein reduction in the diet based on balance of standard ileal digestible essential amino acids on meat productivity and meat quality

Treatment	1 (n=10)	2 (n=10)	3 (n=10)	4 (n=10)	5 (n=10)	SEM	P
Live weight (g)	2256 ^{ab}	2270 ^a	2134 ^{ab}	2066 ^{ab}	1876 ^b	95,32	0,038
Carcass weight (g)	1761,2	1760,6	1657,9	1612,1	1458,7	77,55	0,052
Carcass yield (%)	78,0	77,6	77,7	77,9	77,8	0,904	0,997
Thigh meat yield (%)	15,8 ^{ab}	16,4 ^a	16,4 ^a	15,5 ^{ab}	14,4 ^b	0,373	0,002
Breast meat yield (%)	20,8	20,1	19,8	19,9	20,5	0,513	0,571
Abdominal fat (%)	1,38	1,58	1,95	2,07	2,16	0,412	0,624
Water lost after 24h (%)	2,48	2,41	3,26	2,63	2,32	0,355	0,388
Water lost after processing (%)	17,77	16,66	18,86	16,69	18,93	0,846	0,197
Toughness (N)	16,27 ^b	16,38 ^b	20,42 ^{ab}	21,15 ^{ab}	25,58 ^a	1,319	0,001
Ph 15	6,11	6,13	6,11	6,20	6,17	0,082	0,909
Ph 24 h	5,76 ^b	5,77 ^b	5,78 ^b	5,84 ^{ab}	5,90 ^a	0,027	0,009

Summary: Reducing the crude protein level of the diet by 2% based on the SID-EAA balance with SID-Lys content of 1.30; 1.15 and 0.95% respectively for the three growth stages and the content of other essential amino acids were balanced according to the recommendations of Aviagen (2014) did not affect the body weight, daily weight gain and FCR of Luong Phuong broiler chickens. Reducing at 2% crude protein in the diet had the lowest feed cost of 29,543 VND/kg weight gain, 539 VND/kg lower than the control treatment, equivalent to 1.79%. At this reduction level, the emission of NH₃ and H₂S at the growth stages decreased compared to the control treatment from 23.18 to 28.57% and 2.19 to 9.4%, respectively. Reducing at 4% crude protein reduced the breast meat yield, increased pH_{24h} and increased the toughness of the breast meat

3.5. DEVELOPMENT AND TESTING OF LOW PROTEIN DIETS BASED ON THE OPTIMAL BALANCE OF STANDARD ILEAL DIGESTIBLE ESSENTIAL AMINO ACIDS FOR COLORED BROILERS

Table 3.27. Growth performance and feed efficiency in Luong Phuong trial

Treatment	1 (n=5)	2(n=5)	SEM	P
Body weight (g)				
1 day old	32,83	32,97		
28 days old	488,2	531,7	5,805	0,002
56 days old	1276,4	1370,4	24,52	0,035
84 days old	2027,0	2127,4	29,13	0,051
Average daily weight gain (g/chick/day)				
1-28 days old	16,26	17,81	0,208	0,002
29-56 days old	28,15	29,95	0,898	0,206
57-84 days old	26,81	27,04	0,982	0,875

1-84 days old	23,74	24,93	0,347	0,051
Feed conversion ratio (kg feed/kg weight gain)				
1-28 days old	1,80	1,63	0,029	0,005
29-56 days old	2,72	2,66	0,042	0,304
57-84 days old	4,02	3,98	0,112	0,812
1-84 days old	3,00	2,89	0,038	0,075
Feed cost (VND/kg weight gain)				
1-28 days old	22594	23463	381,5	0,164
29-56 days old	33471	35494	550,9	0,035
57-84 days old	46927 ^b	52482 ^a	1412,9	0,031
1-84 days old	36010 ^b	38708 ^a	474,0	0,007

Table 3.28. Growth performance and feed efficiency in Luong Phuong cross Mia trial

Treatment	1 (n=5)	2 (n=5)	SEM	P
Body weight (g)				
1 day old	31,80	31,75		
28 days old	396,3	426,7	5,18	0,006
56 days old	980,0	1079,6	32,74	0,075
84 days old	1634,0	1738,8	31,65	0,057
Average daily weight gain (g/chick/day)				
1-28 days old	13,01 ^b	14,10 ^a	0,186	0,006
29-56 days old	20,85	23,32	1,134	0,174
57-84 days old	23,36	23,54	1,179	0,917
1-84 days old	19,07	20,32	0,374	0,057
Feed conversion ratio (kg feed/kg weight gain)				
1-28 days old	1,94	1,75	0,041	0,016
29-56 days old	3,37	3,18	0,145	0,404
57-84 days old	3,97	4,13	0,124	0,385
1-84 days old	3,29	3,19	0,051	0,249
Feed cost (VND/kg weight gain)				
1-28 days old	24343	25149	562,7	0,367
29-56 days old	41290	42038	1860,1	0,785
57-84 days old	46320	54554	1631,3	0,012
1-84 days old	39469	42658	649,9	0,013

The results obtained when applying low crude protein level with balancing of some essential SID-EAA in the broiler diets are as follows:

- The growth performance was stable and there was no significant difference between the experimental and control group, specifically:

+ Luong Phuong chickens had body weight at 84 days old, average weight gain and FCR from 1-84 days old respectively of 2027.0 g/chick; 23.74g/chick/day; 3.0 kg feed/kg weight gain; reduced 6.97% in feed cost.

+ Luong Phuong cross Mia chickens had body weight at 84 days old, average weight gain and FCR from 1-84 days old respectively of 1634.06 g/chick; 19.07g/chick/day; 3.29 kg feed/kg weight gain; reduced feed cost by 8.1% per kg of weight gain.

CHAPTER 4. CONCLUSIONS AND RECOMMENDATIONS

4.1. Conclusions

Researches of the thesis were determined the standard ileal digestibility ratio of amino acids of 15 common feed ingredients in poultry production in Vietnam on Luong Phuong chickens including: (1) Group of cereals and cereal by-products: De-oil rice bran, fullfat rice bran, wheat bran, brown rice, broken rice, paddy rice, corn. (2) Group of protein-rich ingredients: Corn DDGS, corn gluten meal, soybeans, soybean meal, canola meal, fish meal, meat and bone meal and blood meal.

Determining the appropriate SID-Lys level in the diet of Luong Phuong chickens corresponding to three growth stages (1-28 days old; 29-56 days old; 57-84 days old) are: 1.3%; 1.15% and 0.95%.

The ratio of essential amino acids in the standard ileal digestible form (methionine, methionine + cystine; threonine; tryptophan and arginine compared to lysine) was determined:

- 1-28 days old: 40.5; 75; 67; 15.6 and 107% corresponding to the level in the diet of 0.53; 0.98; 0.87; 0.20 and 1.39%.

- 29-56 days old: 42; 78.5; 67; 15.6 and 107% corresponding to the level in the diet of 0.48; 0.9; 0.77; 0.18 and 1.23%.

- 57-84 days old: 41.5; 78; 67; 15.6 and 107% corresponding to the level in the diet of 0.39; 0.74; 0.64; 0.15 and 1.02%.

Determining the appropriate crude protein reduction level based on the balance of essential amino acids in the standard ileal digestible form as above in the diet of Luong Phuong chickens is 2% with the dietary protein level is 18.8%; 17% and 16% corresponding to the three growth stages of 1-28 days old; 29-56 days old and 57-84 days old. At this reduction level, the feed cost/kg of weight gain calculated for the period of 1-84 days old decreased by 539 VND, equivalent to 1.792%; the amount of NH₃ and H₂S gas emissions at the end of the growth stages decreased by 23.18 - 28.57% and 2.19 - 9.4% compared to the control experiment.

Apply the research results of 2% crude protein reduction with the ratio of essential amino acids in the standard ileal digestible form as above in the diet to compare with the feed currently used in farm on Luong Phuong and Luong Phuong crossbred Mia chicken. The results showed that the growth performance and feed efficiency of chickens in the experimental treatment were equivalent to the control treatment, however, the feed cost per kg of weight gain calculated for the period of 1-84 days of age decreased compared to the control treatment by 6.97% for Luong Phuong chickens and 8.10% for Luong Phuong cross Mia chicken.

4.2. Recommendations

Using the database of standard ileal digestibility ratio of amino acid in common feed ingredients and appropriate ratios of standard ileal digestible essential amino acids in the diet to formulate diets for colored feather broilers.

Applying a crude protein reduction of 2% corresponding to dietary protein content of 18.8%; 17% and 16% for three growth stages of 1-28; 29-56; 57-84 days old in formulating diets for colored feather broilers at broiler farm and feedmill.