Livestock Science 181 (2015) 114–120

Contents lists available at ScienceDirect

Livestock Science

journal homepage: www.elsevier.com/locate/livsci

Simultaneous determination of standardized ileal digestible tryptophan and lysine for barrows from 15 to 30 kg live weight

T.J. Pasquetti [a,*], P.C. Pozza [b], I. Moreira [c], T.C. Santos [b], L.M. Diaz-Huepa [b], L.D. Castilha [b], D. Perondi [d], P.L.O. Carvalho [e], S.W. Kim [f]

[a] Departamento de Zootecnia, Universidade Estadual de Mato Grosso do Sul, Rodovia Aquidauana, Aquidauana, MS 79200-000, Brazil
[b] Departamento de Zootecnia, Universidade Estadual de Maringá, Avenida Colombo, 5790, Jd. Universitário, Maringá, PR, 87020-900, Brazil
[c] Departamento de Zootecnia, Universidade Tecnológica Federal do Paraná, Estrada para Boa Esperança, Dois Vizinhos, PR 85660-000, Brazil
[d] Departamento de Zootecnia, Universidade Estadual Paulista, Via de Acesso Prof. Paulo Donato Castellane, Jaboticabal, SP 14884-900, Brazil
[e] Departamento de Zootecnia, Universidade Estadual do Oeste do Paraná, Rua Pernambuco, 1777, Marechal Cândido Rondon, PR 85960-000, Brazil
[f] Department of Animal Science, North Carolina State University, Raleigh, NC 27695, United States

ARTICLE INFO

Article history:
Received 27 November 2014
Received in revised form
19 September 2015
Accepted 22 September 2015

Keywords:
Lysine
Pigs
Response surface
Requirements
Standardized ileal amino acid digestibility
Tryptophan

ABSTRACT

The digestible amino acid (AA) determination of ingredients, or experimental diets, are essential in obtaining the real levels of AA that are being studied, as well as of other indispensable AA, because the lack of a single indispensable AA may limit pig growth performance. Two experiments were conducted to determine the optimum levels and the ideal ratio of standardized ileal digestible (SID) Trp and Lys for barrows from 15 to 30 kg of live weight. The first experiment (Exp. 1) was conducted to determine the SID AA content of the basal diet used in the growth performance study (Exp. 2). Ten crossbred barrows (21.5 ± 1.6 kg live weight) were used in Exp. 1. Pigs were fed a basal diet and a protein free diet to determine the SID of Trp, Lys, and all other indispensable AA of the basal diet. In Exp. 2, 64 barrows (15.0 ± 1.6 kg) were allotted to 16 diets in a 4×4 factorial arrangement of treatments (1.55, 1.85, 2.15, and 2.45 g SID Trp and 9.72, 11.12, 12.52, and 13.92 g SID Lys/kg) with 4 replications per treatment. In both Exp. 1 and 2, each pig was considered as an experimental unit. Growth performance, Longissimus muscle (LM), backfat thickness, and plasma urea nitrogen (PUN) were evaluated. Average daily feed intake and average daily gain increased linearly ($P < 0.05$) with increasing levels of SID Trp. There was a quadratic effect ($P=0.04$) of dietary SID Trp and Lys levels on the gain:feed, and the optimal levels were estimated to be 2.1 (0.65 g/Mcal ME) and 11.98 g/kg (3.71 g/Mcal ME), respectively, resulting in a SID Trp:Lys of 17.5:100. A quadratic effect of SID Trp ($P=0.021$) and Lys ($P=0.026$) levels on LM was observed, and the optimum levels were 2.08 and 11.36 g/kg diet, respectively, with a Trp:Lys of 18.3:100. The amount of PUN decreased linearly ($P=0.004$) as SID Lys increased. For pigs used in our study (between 15 and 30 kg live weight), the SID Trp and SID Lys requirements are 2.10 and 11.98 g/kg, respectively. The corresponding Trp:Lys are 17.5:100.

1. Introduction

Most of the previous research evaluating amino acid (AA) requirements for pigs were designed to determine the requirement of 1 AA at a time and did not consider metabolic interactions (antagonism and synergism) that may occur. Furthermore, few researchers have approached the simultaneous study of digestible Trp and Lys levels for pigs.

Tryptophan is the fourth limiting AA in practical diets, based on cereals, for pigs in growing phase (Jansman et al., 2007), while Lys is the first. Because of its constancy in body protein and its metabolic destination, especially to lean tissue deposition, Lys is considered the most important nutrient for meat deposition in the bodies of growing pigs. Trptophan, however, also plays an essential role in protein synthesis, and as proposed by Jansman et al. (2007), its effect extends to feed intake regulation, being the precursor of serotonin (5-HT), which plays a central role in this process.

It is important to know the ratio between Lys and other AA before proceeding with feed formulation. The Trp:Lys reported by Baker and Chung (1992) was 18:100 for pigs from 10 to 20 kg of body weight. Recently Naatjes et al. (2014), using broken-line, curvelinear plateau and exponential models, for pigs from 15 to 35 kg of live weight, observed a better feed:gain for a diet based on wheat when the Trp:Lys was 17.0:100, 18.5:100, and 19.7:100,

* Corresponding author. Fax: +55 67 3904 2907.
E-mail address: pasquettizoo@yahoo.com.br (T.J. Pasquetti).

http://dx.doi.org/10.1016/j.livsci.2015.09.021
1871-1413/

respectively. For a diet based on corn the better ratios were 19.9:100, 20.9:100, and 20.9:100, respectively.

Differences in digestible Trp:Lys are observed in many studies, especially because of mathematical models and response used (Naatjes et al., 2014), as well as environmental factors (Le Floc'h and Seve, 2007), sex and genetic potential of animals. Furthermore, most of the studies did not determine the ileal digestibility of AA contained in the ingredients or in the diet with the lowest level of digestible Trp and Lys.

The digestible AA determination of ingredients, or experimental diets, are essential in obtaining the real levels of AA that are being studied, as well as of other essential AA, as the lack of a single essential AA may limit pig performance. The current study was conducted to determine simultaneously the requirement for standardized ileal digestible (SID) Trp and Lys and the ratio between the 2 AA for barrows between 15 and 30 kg live weight.

2. Material and methods

Two experiments were carried out in the Fazenda Experimental de Iguatemi at Universidade Estadual de Maringá/UEM and in the Nutrition Lab of the Animal Science Department. The experiments were approved by the UEM Animal Care and Use Committee (CEUA, Protocol no. 050/2011).

2.1. Standardized ileal digestibility of amino acids of the basal diet (Exp. 1)

2.1.1. Animals, experimental design and diets

This study was conducted to determine the digestible Trp and Lys contents of the basal diet and to establish whether other indispensable AA were limiting or not. Ten crossbred barrows (Landrace × Large White and Pietrain), averaging 21.5 ± 1.57 kg of live weight, were individually allotted to metabolism cages, and distributed to a randomized block design. Treatments consisted of a basal diet, with the lowest levels of digestible Trp (1.5 g/kg diet) and Lys (9.0 g/kg diet) and a protein free diet (PFD) used to determine the endogenous losses of AA (Table 1).

The basal diet was formulated based on corn, soybean meal, soybean oil, AA, vitamins, minerals and additives, to meet the requirements proposed by Rostagno et al. (2011) for barrows from 15 to 30 kg of body weight, except for the crude protein, Trp, and Lys levels. In the PFD, rice hulls was used as fiber source. Chromic oxide (Cr_2O_3) was used in the diets to determine the indigestibility factor. Diets were provided twice a day (at 0800 h and 1500 h) and water was provided ad libitum. Feed was moistened and provided in amounts calculated based on metabolic size (BW0.75).

2.1.2. Surgery procedure and digesta collection

Animals were submitted to an adaptation period of 5 d to the experimental diets and cages. Sampling of ileal digesta was performed as proposed by Lazzeri (2011), which was adapted from Apolônio et al. (2002). Before proceeding with surgery, tranquilizers and an anesthetic were injected into the pigs. After depilation and disinfection of the surgical site in the abdominal cavity, laparotomy was preceded through a ventral incision in the cephalocaudal direction, of approximately 10 cm. The segment of distal ileum was exteriorized, and the digesta flow between the ileocecal valve and the end of the ileocecal fold was closed with a hemostatic forceps. A segment of approximately 20 cm of the ileum has been cut and removed, washed with distilled water and dried with a paper towel. The ileal digesta from each treatment (basal diet and PFD) was collected and stored in plastic containers, and then the samples were lyophilized and composed by treatment for subsequent analysis. After collection procedure, pigs were euthanized.

Table 1
Composition of the basal diet and protein-free diet.[a]

Item	Basal diet	Protein-free diet
Ingredient (g/kg)		
Corn	742.4	–
Soybean meal (45.2% CP)	136.8	–
Corn gluten meal (60% CP)	60.0	–
Dicalcium phosphate	15.9	20.6
Limestone	8.2	7.0
Soybean oil	1.2	–
Vitamin-trace mineral premix[b]	5.0	5.0
Sodium bicarbonate	4.6	–
Sand	1.5	–
Salt	1.4	5.0
Glutamic acid	7.7	–
L-Lys≅HCl (78.4%)	4.9	–
L-Thr (98.0%)	1.9	–
DL-Met (99.0%)	1.2	–
L-Trp (98%)	0.2	–
L-Val	1.2	–
L-Ile	0.5	–
Antioxidant[c]	0.1	0.1
Cromic oxide	5.0	5.0
Starch	–	727.0
Rice hulls	–	30.0
Sugar	–	200.0
Antibiotic[d]	0.05	0.00
Calculated composition (g/kg unless indicated otherwise)		
ME (kcal/kg)	3,230	3,341
CP	165.2	1.2
Ca	7.7	7.7
Available P	3.8	3.8
Na	2.0	2.0
SID Lys	9.00	–
SID Thr	6.89	–
SID Met + Cys	6.13	–
SID Trp	1.50	–
SID Val	7.55	–
SID Ile	6.01	–
SID Leu	16.13	–
SID Arg	7.76	–

[a] SID = standardized ileal digestible, ME = metabolizable energy; and CP = crude protein.
[b] Provided per kilogram of diet: vitamin A,10,000 IU; vitamin D3, 2000 IU; vitamin E, 40 IU; vitamin K3, 3.12 mg; vitamin B1, 1.57 mg; vitamin B2, 4.0 mg; vitamin B6, 2.18 mg; vitamin B12, 23 μg; calcium pantothenate, 15.2 mg; niacin, 30.38 mg; folic acid, 0.3 mg; biotin, 0.12 mg; choline, 360 mg; Zn, 125 mg; Fe, 81 mg; Cu, 12 mg; Mn, 40.3 mg; I, 1.12 mg; Se, 0.32 mg; Co, 0.2 mg; and antioxidant, 5 mg.
[c] Antioxidant = butylated hydroxytoluene.
[d] Antibiotic: Leucomag (Ceva Animal Health, Paulínia, Brazil).

2.1.3. Chemical analysis

Dry matter and chromic oxide were measured in the basal diet and PFD, as previously described by the AOAC (2012) and Kimura and Miller (1957), respectively. The AA composition of diets, in the digesta and rice hulls, was analyzed using ninhydrin for post-column derivatization and norleucine as the internal standard. Performic acid was used to oxidize the AA, which was neutralized with sodium metabisulfite (Llames and Fontaine, 1994; Commission Directive, 1998), and then hydrolyzed (AOAC, 2012). Amino acid quantification was performed as proposed by Rosenfelder et al. (2015). Tryptophan was determined by fluorescence detection using high performance liquid chromatography (Commission Directive, 2000).

2.2. Standardized ileal digestible Trp and Lys levels for crossbred barrows from 15 to 30 kg live weight (Exp. 2)

2.2.1. Animals, experimental design and diets

Sixty-four individually housed crossbred barrows (Tybor × Topigs 20; 15.00 ± 1.63 kg) were allotted to 16 treatments in