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Poultry

Metabolizable energy of raw materials

A comparison of values obtained either from chick assays or prediction equations for poultry



Key Information

- Energy is the main contributor to feed cost, and affects directly poultry performance.
- Nutritionists usually obtain metabolizable energy values from feedstuff composition tables.
- Table values cannot be adjusted to variation in nutrient composition of the feedstuff. Therefore prediction equations have been developed for the individual feedstuffs.

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Standardized ileal digestible tryptophan to lysine ratios

to optimize performance of starting, growing and finishing pigs, and factors affecting the optimum tryptophan ratio

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Revised amino acid recommendations

by Evonik

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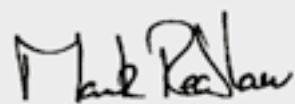
Research Highlights: Page 31 – 34

Editorial

Dear Reader,

Energy is a critical component of any feed, Adhemar Oliveira describes a number of approaches for managing and measuring energy content of feed ingredients based upon this he proposes a practical approach of how to implement this knowledge in daily practice. The role of tryptophan in pig diets is also discussed in an article by John Htoo. We are also pleased to bring you a new updated version of our amino acid recommendations for a whole range of poultry species as well as swine. These new recommendations are based upon our latest trial results supplemented by available data from the international literature.

Happy reading.



Dr. Mark Redshaw

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- Prediction equations need to be used cautiously as the predicted number can considerably differ from the energy table values.
- A method is proposed how to use equations for adjustment of the energy values found in tables. This allows for a more sustainable use of the energy in the feedstuffs.

Broilers use dietary energy for various purposes

Broilers obtain energy from feedstuffs by digesting and absorbing nutrients such as glucose from sugar or starch, fatty acids, or amino acids from proteins. Nutrients are then transported to various tissues by the blood stream. Once inside the cells, nutrients may be stored as energy containing substance such as protein, fat and glycogen or may be oxidized as a readily available source of energy for metabolic processes. This available energy can be used for countless physiological and biochemical processes.

Increasing the energy level in broiler diets may improve weight gain and feed conversion ratios (Figure 1). However, excessive energy intake may linearly increase fat deposition rate (FDR), whereas protein deposition rate (PDR) may achieve a maximum (Figure 2). This PDR maximum may be due to amino acid deficiency or imbalance, or because the animal has achieved its maximum genetic potential although recent work has established that modern broilers continue to respond to increasing levels of balanced protein (Lemme *et al.*, 2009).

Oliveira Neto (1999) measured PDR as a difference between the percentage of broiler body protein at 22 and at 42 days of age. The same procedure was applied for FDR. Fat deposition rate increased by 9.2% when dietary metabolizable energy (ME) levels were raised from 3150 to 3300 kcal/kg whereas it increased only 7.2% when metabolizable energy increased from 3000 to 3150 kcal/kg. In contrast, PDR

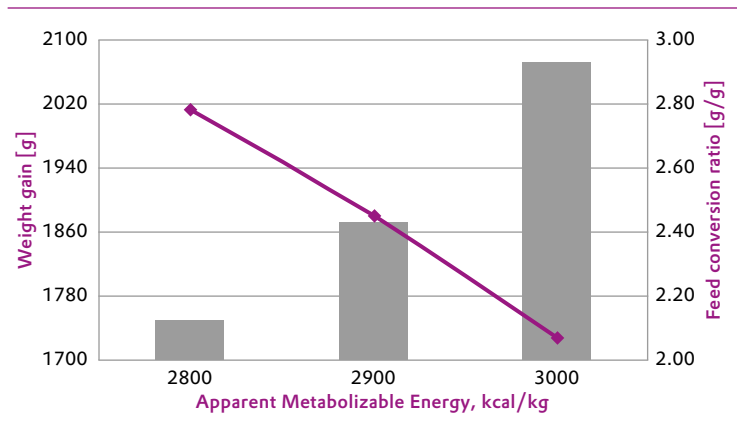


was maximized at 3108 kcal ME/kg with no further improvement with higher dietary energy levels. Thus, dietary energy supply above 3108 kcal ME/kg could only be used for fat deposition.

These observations demonstrate the effect of dietary energy on body composition. Dietary energy concentration above the requirement for PDR potential is used for fat deposition mainly as abdominal fat or subcutaneous fat. Excessive body fat deposition is not desired because it finally increases production costs due to inefficient use of feed and impaired carcass quality.

Once the dietary metabolizable energy content for optimal performance is known, nutritionists need to formulate the diets accordingly. In this context, due to competition within the global poultry industry it is very important to evaluate and consider the correct apparent metabolizable energy (AME) value of feeds because any excess may lead to a loss of profitability. Therefore, nutrient analysis of raw materials is highly important and feed formulation must be adjusted accordingly. However, metabolizable energy levels of raw materials cannot be evaluated by routine laboratory analysis; instead biological trials are required. This limits the information available on energy content, although it remains the main cost factor in poultry feeds. Alternative ways to assess ingredient energy levels such as prediction equations have therefore been established.

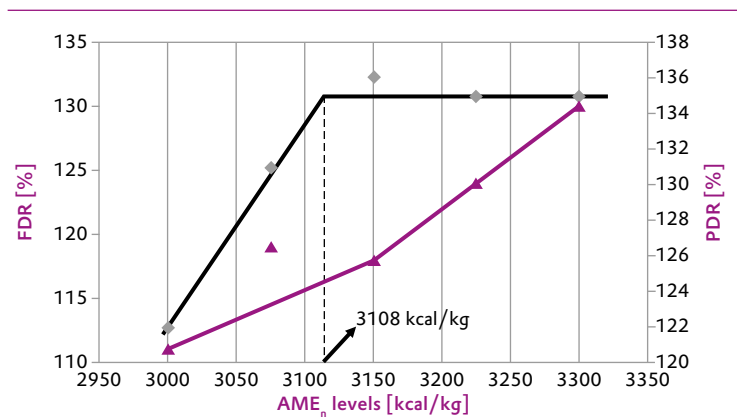
Figure 1



■ Weight gain [g]
◆ Feed conversion ratio [g/g]

Effect of AME_n on the performance of 29 to 56-day-old broilers (adapted from Bertechini, 1987)

Figure 2



▲ FDR [%] = fat deposition rate
◆ PDR [%] = protein deposition rate

Effect of feed energy levels on fat and protein content broilers at 42 days in relation to that measured at 22 days of age (100%) (Oliveira Neto *et al.*, 1999)

The aim of this article is to discuss opportunities and limitations of the use of prediction equations developed to estimate metabolizable energy values in poultry feed raw materials. Energy values presented in feedstuff composition tables currently used in the feed industry are assessed. As these values are obtained by biological assays, the determined ME contents are greatly influenced by the methodology applied. This might be a major explanation for the differences in energy value among ingredient composition tables. The precision of several prediction equations used to estimate ME values in feedstuffs is evaluated, and energy values obtained by these equations are compared with feedstuff composition tables.

Energy of feed ingredients can be assessed in various ways

Energy is available to the animals through the oxidation of organic compounds including proteins, fats, and carbohydrates. Finally, energy appears as energy rich metabolites such as adenosine tri-phosphate (ATP) together with by products such as CO₂, H₂O, and heat. Moreover, energy potential differs between nutrients (Table 1).

Table 1

Starch	Glucose	Protein	Fat
3.7 kcal/kg GE	4.2 kcal/kg GE	5.6 kcal/kg GE	9.4 kcal/kg GE

Gross energy (GE) of different nutrients used by poultry. Brody (1994) and NRC (1998).

Dietary energy of ingredients or compound feed can be expressed as gross energy, digestible energy, metabolizable energy, and net energy (Figure 3).

Gross energy (GE) is the energy released as heat by the complete burning of the organic matter. GE in feedstuffs or feeds is measured by using bomb calorimetry. This assay can actually be performed in a laboratory but due to different digestibility or utilization of energy (see below) of the individual raw materials this assay is not of relevance for practical poultry feeding.

Digestible energy (DE) of the feedstuff is GE minus energy excreted with feces, i. e., it is the feedstuff energy absorbed by the animal after digestion. In birds, it is difficult to separate feces from urine, therefore DE is not applicable in poultry.

Apparent metabolizable energy (AME) is defined as DE minus the energy excreted with urine and with gases such as methane. The production of gases by monogastric animals is negligible and is thus not taken into consideration. The AME is usually used for raw material and compound feed assessment as well as for setting specifications. However, metabolizable energy systems can be distinguished into apparent metabolizable energy corrected for nitrogen balance (AME_n) and true metabolizable energy (TME).

The most commonly used system for poultry feedstuffs is AME_n. The correction for nitrogen balance was first proposed by Hill and Anderson (1958), who assumed that the ingested nitrogen that was not retained would appear in the excreta mainly in the form of uric acid which is excreted by the kidney. Gross energy for complete uric acid oxidation is about 8.22 kcal per gram of nitrogen (N) retained. The authors proposed a correction of 8.22 kcal/g N in order to adjust AME values to a 0-balance nitrogen retention. Despite the criticism that only 60 – 80 % of the excreted nitrogen can be assigned to uric acid (NRC, 1994), AME_n is still widely used.

According to Sibbald (1982), the nitrogen balance concept is required in order to correct the effect of the age of the birds used for measuring the energy content of feedstuffs. In young birds, the dietary protein retained as body tissue (growth) will not be catabolized, and therefore this protein (nitrogen) would not contribute to the energy content in feces and urine. On the other hand, adult birds which are often used for ME determination have higher catabolism because amino acids are only used for maintenance and thus a high proportion of the ingested protein is degraded to uric acid. This relationship was nicely demonstrated by Rodrigues (2000, Table 2). The formulas below aid understanding of nitrogen balance and how it influences ME values.

$$AME = (GE \text{ intake} - GE \text{ excretion}) / DM \text{ intake}$$

$$AME_n = [(GE \text{ intake} - GE \text{ excretion}) / DM \text{ intake}] - 8.22 * (N \text{ intake} - N \text{ excretion})$$

where:

- GE = gross energy
- AME = apparent metabolizable energy
- AME_n = apparent metabolizable energy corrected for nitrogen balance
- DM = dry matter
- N = nitrogen

Table 2

Feedstuff	Young birds			Roosters		
	AME	AME _n	Difference	AME	AME _n	Difference
Corn	3,749	3,699	50	3,444	3,736	- 292
Gluten meal	4,314	4,108	206	3,772	3,982	- 210
Soybean meal	2,508	2,337	171	2,187	2,459	- 272
RFFSB	3,550	3,400	150	3,503	3,736	- 233
Micronized soybeans	4,260	4,104	156	4,003	4,180	- 177
Average	3,439	3,280	159	3,231	3,458	- 227

AME and AME_n (kcal/kg DM) values as determined by metabolism assay with young and adult birds, using total excreta collection.

RFFSB – roasted full fat soybeans
Adapted from Rodrigues (2000)

True metabolizable energy (TME) is defined as the AME corrected for the endogenous energy losses in the feces and urine. Endogenous energy losses result from the excretion of energy present in digestive fluids and mucus, and in slaughtered cells of the intestinal mucosa. These losses originate from animal metabolism and must not be related to the ingested feedstuff. TME may also be corrected for nitrogen balance, revealing TME_n.

CVB (2004) further developed the AME_n formulation and adjusted them to broiler and layer requirements.

Net energy (NE) is defined as metabolizable energy minus heat increment associated with the metabolic utilization of ME and with the energy cost of feed intake and digestion. In order to calculate NE, either the heat increment provoked by the ingredient or the NE/ME ratio, which is specific for each feedstuff, needs to be known. This ratio represents the efficiency of the utilization of ME for energy retention. The net energy system is currently widely used in swine nutrition; however, in poultry research has not yet substantiated benefits over ME systems.

As listed in Figure 3 various methods are used to describe the nutritional value in terms of energy, when talking about broiler nutrition this is mainly ME based systems. It needs to be mentioned that one important rule should be considered in feed formulation: Recommendation and feed evaluation should use the same system otherwise requirement figures and nutritional figures are not synchronized.

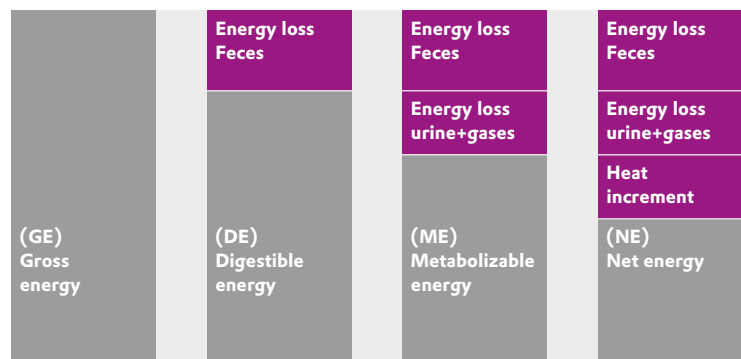


Figure 3
Definition of different energy systems in animals by considering various ways of energy excretion



Factors that influence energy values

The current feedstuff composition tables (NRC, 1994; Rostagno *et al.*, 2005; INRA, 2004; CVB, 2004) show higher variation of AME_n values for animal products compared to those for vegetable ingredients (Table 3). When the energy values of animal byproduct meals provided in the tables are compared, differences are substantial:

- Meat and bone meal (51 %) – 527 kcal/kg NRC (1994) vs. INRA (2004)
- Feather meal + 389 kcal/kg NRC (1994) vs. INRA (2004)
- Offal meal + 309 kcal/kg AME_n Rostagno *et al.* (2005) vs. NRC (1994)

Although lower, variation of AME_n in vegetable ingredients is also important, considering their high level of inclusion in poultry feeds. Taking corn as an example, 250 kcal/kg of difference is observed between the energy level recommended by Rostagno *et al.* (2005) and by INRA (2004). This may cause a 150 kcal/kg difference in AME_n value of feeds when corn is included at 60 % – just because of the difference in the energy value between those composition tables.

These discrepancies may be partially explained by the influence of several factors (Moughan *et al.*, 2000):

- the methodology applied in the experiments used to determine feedstuff energy values;
- the physical and chemical composition of the feedstuffs;
- the presence of anti-nutritional factors;
- the age of the birds used in these experiments;
- the inclusion level of the tested feedstuffs in the test feed;
- the particle size of the tested feedstuff.

Although it is recognized that feedstuff energy values are influenced by several factors, these are not taken into consideration in feed formulation software. Calcium, phosphorus, crude protein, and amino acid levels are frequently corrected in the matrices according to laboratory analyses, but energy values remain in most of the cases unchanged, as their determination depend on biological assays. Companies commonly do not have the physical structure or dedicated people to perform biological trials, limiting the knowledge on the energy levels of the feedstuffs used in their feed mills.

Considering these limitations, prediction equations were developed in order to account for the chemical composition of the feedstuffs (March and Biely, 1973; Sibbald and Price, 1975; Villamide and San Juan, 1998; Vieites, 1999; Rodrigues *et al.*, 2000; Nascimento *et al.*, 2002). The common aim being to improve the assessment of the nutritional value of the ingredients with respect to their energy content.

Prediction equations to determine ME in feedstuffs used in poultry feeds

In order to estimate energy value as a function of the chemical composition of feedstuffs, simple or multiple regression equations are applied. In these equations, many nutrients, such as protein, ether extract (fat), crude fiber, and ash are positively or negatively correlated with the metabolizable energy value of a determined feedstuff. These regression equations are called prediction equations. Prediction equations can be obtained using the stepwise backward elimination procedure. This method eliminates independent variables that do not have significant effects on metabolizable energy value. The choice of the best prediction equations is based on coefficient of determination (R²) values.

Table 3

Ingredients	Rostagno <i>et al.</i> (2005)	NRC (1994)	INRA (2004)	CVB (2004)
Corn	3,381	3,350	3,131	3,210
Corn gluten 60 %	3,696	3,720	3,561	3,179
Sorghum	3,192	3,288	3,227	3,143
Wheat	3,046	3,120	2,892	2,866
Soybean oil	8,790	8,370	9,011	8,598
Soybean meal 45 %	2,256	2,230	–	1,829
Soybean meal 48 %	2,302	2,440	2,223	–
Meat bone meal 45 %	2,445	–	–	2,257
Meat bone meal 51 %	2,638	2,150	2,677	–
Feather m. 79–83 %	2,734	2,360	2,749	2,720
Offal meal 57 %	3,259	2,950	–	–

Apparent Metabolizable Energy corrected for nitrogen balance (AME_n, in kcal/kg)
Feedstuff energy values for poultry

The prediction equation used in many recommendations has been published by Janssen (1989). As example, a general formula that can be used for many plant ingredients is presented below

$$\text{AME}_n \text{ (kcal/kg)} = 4.31 * \text{dCP} + 9.29 * \text{dF} + 4.14 * \text{dNFE}$$

where:

AME_n = apparent metabolizable energy corrected for nitrogen balance (kcal/kg)

dCP = digestible crude protein (g/kg)

dF = digestible fat (g/kg)

dNFE = digestible nitrogen free extract (g/kg)

However, digestibility of crude protein, fat and nitrogen free extract needs to be known in order to correctly determine the AME_n. But, digestibility of the different nutrients is not necessarily known. Therefore, equations were developed using those nutrients which can be routinely analyzed in the feed mill laboratory including ether extract, crude fiber, crude protein, ash, calcium, and phosphorus. Equations including NDF (neutral detergent fiber), ADF (Acid detergent fiber), lignin, starch, and sugar are also available but assays are less widely

established in the laboratories. The following prediction equation

$$\text{AME}_n = 5167.2 - 8.62 * \text{CP} - 131.97 * \text{CF} - 183.43 * \text{Ash} - 14.71 * \text{Starch (Rodrigues, 2000)},$$

can be used as an example, because all nutrients (CP = crude protein; CF = crude fiber) were provided as crude nutrients and digestibility of the nutrients was not considered.

Prediction equations have been developed for:

- complete Feeds;
- groups of feedstuffs;
- individual feedstuffs.

In general, prediction equations for individual feedstuffs are superior to those established for groups of feedstuffs or for feeds (Larbier and Leclercq, 1994). However, good prediction equations for individual feedstuffs are highly dependent on the number of samples used. The wider the range of chemical composition in the feedstuff samples used to develop the equation, the higher the precision of the estimated metabolizable energy values.



Prediction equations in literature

Some studies to develop prediction equations were carried out using a pool of different feedstuff (corn, corn gluten, wheat), because it increases the variation of chemical composition values and then it can improve the R^2 value. The main problem of most prediction equations is that when applied to a raw material sample that is not representative of the reference pool used to generate the equation, the accuracy suffers. The prediction equation may give much higher or lower energy values and may be further away from the true value than feed composition tables. This obviously limits the utilization of prediction equations. Table 4 shows differences between AME_n values of different corn samples published by Rostagno *et al.* (2005) and Rodrigues (2000) as determined in biological assay and also shows values estimated by using various prediction equations developed by Rostagno *et al.* (2005), Rodrigues (2000) and Janssen (1989). For instance, prediction equation 1 provides a very precise metabolizable energy estimation of corn 1 (3881 vs. 3879). The explanation is that equation 1 was developed by Rostagno *et al.* (2005) based on the composition of corn 1 from the table of the same author used to generate this equation. On the other hand, when equation 1 is used to estimate the energy values of corn 2, 3, and 4, differences of 143, 334, and 287 kcal/kg, respectively, are observed between estimated and observed values. These corn samples were not part of

the pool used by Rostagno (2005) to create his equations. Actually corn 2, 3 and 4 were used in Rodrigues (2000) study where the author measured AME using biological assays and after that developed prediction equations to estimate his corn energy values. When equation 1 (from Rostagno *et al.*, 2005) was used to estimate energy values of other corn samples (2, 3 and 4) from Rodrigues (2000) it was not able to estimate reasonable AME values if we compare estimate and observed energy values (Table 4).

The differences between observed energy values obtained by biological assay for those corns (corn 1 = 3,881; corn 2 = 3,699; corn 3 = 3,529; corn 4 = 3,647 kcal/kg) can be explained by factors in the methodology used such as the younger age of the birds used in the assay of Rodrigues (2000) compared to Rostagno (2005).

Interestingly the average estimated energy value of the three equations used in this exercise still resulted in substantially different estimated energy values.

Another interesting thing that can be observed in Table 4 is that equation 2 predicted systematically lower values than Equation 1 which is the logical consequence of using different factors for the same nutrients. In contrast Equation 3 produced considerably higher AME_n values. A reason might be that the starch analysis included in Equation 3 has a higher analytical error compared to crude protein and fat analysis.

Table 4

Ingredients	AME_n Table	AME_n estimated by prediction equations		
	Biological assay	Equation 1	Equation 2	Equation 3
Corn 1	3,881	3,879	3,787	4,208
Corn 2	3,699	3,842	3,747	4,169
Corn 3	3,529	3,863	3,769	4,240
Corn 4	3,647	3,934	3,842	4,253
Average (kcal/kg)	3,689	3,880	3,786	4,218
SD (kcal/kg)	146	39	41	37
CV (%)	4	1	1	1

AME_n Table – apparent metabolizable energy corrected for nitrogen balance, as determined by biological assays.

AME_n value of Corn 1 was obtained from the feedstuff composition table of Rostagno *et al.* (2005).

AME_n values of Corn 2, 3, and 4 were taken from the study of Rodrigues (2000).

Equation 1 – $AME_n = 39.78*CP + 69.68*Fat + 35.40*NFE$ (Rostagno *et al.*, 2005).

Equation 2 – $AME_n = 36.21*CP + 85.44*Fat + 37.26*NFE$ (Janssen, 1989).

Equation 3 – $AME_n = 4887.3 - 5.42*CP - 32.74*NDF - 127.52*Ash - 8.15*Starch$ (Rodrigues, 2000).

SD – standard deviation; CV (%) – coefficient of variation; CP = crude protein; NFE = nitrogen free extract.

Corn AME_n values observed in feedstuff composition tables and AME_n values estimated by prediction equations (on dry matter basis)

The differences in the calculated AME_n values for the four individual corn samples are smaller than the differences found in the bioassay. These small differences result in a low standard deviation for the predicted values, indicating that the chemical composition analysis of the feedstuffs was not able to correctly predict metabolizable energy. Moreover, other factors obviously impact estimating feedstuff energy values. For instance, composition of the starch

component (amilopectin to amilose ratio) and corn's physical characteristics influence its energy values (Barbarino, 2001).

Many prediction equations have been published for a wide range of raw materials. Tables 5 and 6 present some prediction equations published in literature for vegetable and animal feedstuffs respectively, all parameters are entered in the equations in g/kg.

Table 5

Feedstuff	Equation	DM basis	Author
Millet	AME _n = 36.20*CP + 69.68*Fat + 38.09*NFE	100	Janssen (1989)
	AME _n = 39.78*CP + 69.68*Fat + 35.40*NFE	100	Rostagno <i>et al.</i> (2005)
Corn	AME _n = 4887.3 – 5.42*CP – 32.74*NDF – 127.52*Ash – 8.15*Starch	100	Rodrigues (4) (2000)
	AME _n = 5167.2 – 8.62*CP – 131.97*CFiber – 183.43*Ash – 14.71*Starch	100	Rodrigues (5) (2000)
	AME _n = 36.21*CP + 85.44*Fat + 37.26*NFE	100	Janssen (1989)
	AME _n = 37.05*CP + 85.47*Fat + 38.21*NFE	100	Rostagno <i>et al.</i> (2005)
DDGS	TME _n = 2957.1 + 43.8*Fat – 79.1*CFiber	86	Batal and Dale (1) (2006)
	TME _n = 2582.3 + 36.7*Fat – 72.4*CFiber + 14.6*CP	86	Batal and Dale (2) (2006)
	TME _n = 2732.7 + 36.4*Fat – 76.3*CFiber + 14.5*CP – 26.2*Ash	86	Batal and Dale (3) (2006)
Corn germ	AME _n = 21.12*CP + 87.23*Fat + 32.29*NFE	100	Janssen (1989)
Corn gluten 60 CP	AME _n = 40.95*CP + 88.26*Fat + 33.12*NFE	100	Janssen (1989)
	AME _n = 40.08*CP + 88.26*Fat + 40.57*NFE	100	Rostagno <i>et al.</i> (2005)
Sorghum	AME _n = 31.03*CP + 77.11*Fat + 37.69*NFE	100	Janssen (1989)
	AME _n = 4412 – 90.43*ADF	100	Moir and Connor (1977)
	AME _n = 3152 – 357.79*Tanic acid	100	Gous <i>et al.</i> (1982)
Wheat, w. midds, w. germ	AME _n = 34.92*CP + 63.10*Fat + 36.42*NFE	100	Janssen (1989)
	AME _n = 4754.02 – 48.38*CP – 45.32*NDF	100	Nunes (1) (2000)
	AME _n = 4536.71 – 29.55*CP – 89.17*CFiber + 40.30*Fat – 231*Ash	100	Nunes (2) (2000)
	AME _n = 4222.41 + 67.10*Fat – 473.46*Ash	100	Nunes (3) (2000)
	AME _n = 3994.87 – 48.82*NDF (R ² = 0.91)	100	Nunes (4) (2000)
Triticale	AME _n = 34.49*CP + 62.16*Fat + 35.61*NFE		Janssen (1989)
	AME _n = 37.32*CP + 62.24*Fat + 35.31*NFE	100	Rostagno <i>et al.</i> (2005)
Canola meal	AME _n = 32.76*CP + 83.52*Fat + 13.25*NFE	100	Janssen (1989)
	AME _n = 31.46*CP + 69.60*Fat + 12.75*NFE	100	Rostagno <i>et al.</i> (2005)
SBM, FFSB, RFFSB, MSB, JSSBE	AME _n = 1822.76 – 99.32*CFiber + 60.50*Fat + 286.73*Ash – 52.26*Starch	100	Rodrigues (1) (2000)
	AME _n = 2822.19 – 90.13*CFiber + 49.96*Fat	100	Rodrigues (2) (2000)
	AME _n = – 822.33 + 69.54*CP – 45.26*ADF + 9.81*Fat	100	Rodrigues (3) (2000)
SBM 45CP	AME _n = 37.50*CP + 46.39*Fat + 14.9*NFE	100	Janssen (1989)
	AME _n = 39.61*CP + 46.45*Fat + 12.63*NFE	100	Rostagno <i>et al.</i> (2005)
RFFSB	AME _n = 2769 – 59.10*CFiber + 62.10*Fat	100	Janssen (1989)
	AME _n = 37.50*CP + 79.34*Fat + 19.46*NFE	100	Rostagno <i>et al.</i> (2005)
S. Extrusada	AME _n = 38.79*CP + 87.33*Fat + 18.22*NFE	100	Rostagno <i>et al.</i> (2005)

DDGS = Distillers Dried Grains with solubles. Corn gluten – with 60% of crude protein. Corn germ with 20% of ether extract; SBM = soybean meal, FFSB = full-fat soybeans; JSSBE = Jet Sploder soybeans; RFFSB = roasted full fat soybeans; MSB = micronized soybeans; CFiber = crude fiber; NFE = nitrogen free extract; NDF = neutral detergent fiber; ADF = acid detergent fiber; CP = crude protein; DM = dry matter.

Prediction equations for individual plant feedstuffs used in poultry feeds

Table 6

Ingredient	Equations	DM basis	Author
MBM 38CP	$AME_n = 33.95 \cdot DM - 45.79 \cdot Ash + 60.02 \cdot Fat$	As is	Janssen (1989)
	$AME_n = 4.31 \cdot dCP + 9.29 \cdot dFat$	100	Rostagno <i>et al.</i> (2005)
	$AME_n = -2021.65 + 56.08 \cdot CP + 66.49 \cdot Fat$	100	Vieites (1999)
Fish meal	$AME_n = 35.89 \cdot DM - 34.10 \cdot Ash + 42.11 \cdot Fat$ (from 60 to 67% of CP)	As is	Janssen (1989)
Feather meal	$AME_n = 2928.39 + 75.5209 \cdot Ash - 676.968 \cdot Ca + 600.986 \cdot AGD$	100	Nascimento <i>et al.</i> (2002)
	$AME_n = 3553.27 + 124.254 \cdot Ash - 307.156 \cdot P$	100	Nascimento <i>et al.</i> (2002)
	$AME_n = 3041.64 + 7.67521 \cdot Fat - 469.885 \cdot Ca + 544.717 \cdot AGD$	100	Nascimento <i>et al.</i> (2002)
Offal meal	$TME_n = 2904 + 65.1 \cdot Fat - 54.1 \cdot Ash$	92	Dale <i>et al.</i> (1993)
	$TME_n = 1728 + 77.9 \cdot Fat - 40.7 \cdot Ash + 6.0 \cdot CP$	92	Dale <i>et al.</i> (1993)
	$AME_n = 4592.56 - 45.6345 \cdot Ash - 135.306 \cdot Ca + 273.728 \cdot P - 844.303 \cdot AGD$	100	Nascimento <i>et al.</i> (2002)
	$AME_n = 4723.02 - 60.5854 \cdot Ash - 1040.3 \cdot AGD + 10.1511 \cdot PEP$	100	Nascimento <i>et al.</i> (2002)
	$AME_n = 7669.37 - 55.154 \cdot CP - 78.2412 \cdot Ash - 264.726 \cdot Ca + 471.567 \cdot P$	100	Nascimento <i>et al.</i> (2002)

MBM = meat and bone meal; CP = crude protein, AGD = average geometric diameter; Ca = calcium; P = phosphorus; dFat = digestible fat; dCP = digestible crude protein; DM = dry matter; PEP = digestibility in pepsin 0.002%

Prediction equations for individual feedstuffs of animal origin used in poultry feeds

Practical application of prediction equations

As shown above prediction equations for AME_n and TME_n estimation are available. The question is now how to apply them properly in daily business. A suggestion is given below explaining how prediction equations can be used in order to adjust feedstuff AME_n and TME_n values in the least cost formulation process (Table 7). The example demonstrates how table values can be adjusted by means of the prediction equations. For this six steps must be taken using AME_n values as an example:

- 1 Both the chemical composition and the AME_n values of the feedstuff to be analyzed are obtained from the composition tables of feedstuffs which are used as reference for the least cost formulation. As example we took, roasted full fat soybeans (RFFSB) in the table of Rostagno *et al.* (2005); AME_n value is 3,281 kcal/kg.
- 2 From the available AME_n prediction equations, such as those shown in Table 5, one is chosen. In our example we use the one of Janssen (1989). AME_n is calculated using the chemical composition data of RFFSB as given by the composition table. The calculated AME_n value was 3,512 kcal/kg.

- 3 A new sample of the RFFSB batch of interest need to be analyzed for the nutrients which are needed for the energy equation i. e. crude fiber and fat.
- 4 The prediction equation is applied to estimate the energy value of this particular RFFSB batch. In the example, the calculated AME_n content was 3,390 kcal/kg.
- 5 The difference between the AME_n value calculated from the table chemical composition and from the analyzed chemical composition is derived. In this example, the difference is -122 kcal/kg (3,512 - 3,390).
- 6 Finally, this difference of -122 kcal is taken in order to adjust the AME_n value of RFFSB referenced in tables. In the example the adjusted AME_n value was 3,159 kcal/kg (3,281 - 122). This last step permits that different RFFSB batches can be corrected according to its analyzed chemicals variation, but use a table value as standard.

This example demonstrated that the estimated AME_n value of RFFSB using table proximate values is higher than the AME_n value from the composition table. This confirms the disagreement of numbers determined directly and

indirectly by equations discussed earlier in this paper (Table 4). It needs to be mentioned that this adjustment can only account for differences in nutrient composition of the ingredients compared to table values. Thus, the 231 kcal/kg (3,512 estimated – 3,281 observed) or a 7.0% difference once more emphasizes that AME_n values estimated by equations need to be used with caution. However, as demonstrated in the example shown in Table 7 it can be concluded these equation values can be very useful for adjustment of the nutritional matrices in least cost formulation.

Validation of prediction equations with biological assays

Statistical methods can be used to verify whether the applied prediction equations provide a reliable estimation of feedstuff energy values. For instance, the Pearson correlation analysis determines if the energy values in the composition tables are correlated with those estimated by prediction equations. This method allows determining the precision of prediction equations in estimating AME_n in feedstuffs based on their chemical composition.

Firstly it is necessary to evaluate if correlations are significant between the biological assay and prediction equations and for this generally is used “T test (P<0.05)”. After that the correlation between variables should be tested, for example using the Pearson correlation which was used in this study.

Correlation can be explained as a number between –1 and +1 that measures the degree of association among two variables. At the present case variables are: 1) the observed energy value in poultry assay and, 2) that estimated by prediction equations.

When the correlation has a positive value, there is a positive association between the biological assay and the predicted values (example: high observed energy values tend to be associated with high estimate energy values).

On the other hand, when a negative correlation is observed it means that variables have a inverse association (when one has high energy values the other one has low energy values).

Correlations that have values above ± 0.70 indicates strong associations between the variables.

In this review it was used many prediction equations from the literature (Tables 5 and 6) were tested using Pearson’s correlation (shown in Table 8). Equations that have both a significant (P<0.05) and a correlation ± 0.70 are highlighted and would be acceptable.

Table 7

Nutrient	RFFSB Table of Rostagno et al. (2005)	RFFSB New sample/analyzed nutrients*	Difference
Crude protein	37.0	36.0	- 1.0
Fat	17.86	16.86	- 1.0
Crude fiber	6.20	7.20	+ 1.0
AME _n – feedstuff table	3,281	-	
AME _n – estimated by equations ¹	3,512 *	3,390 *	122
New adjusted AME _n	-	3,159	

AME_n – apparent metabolizable energy corrected for nitrogen balance (kcal/kg).

RFFSB – roasted full fat soybeans.

* New sample (used in one feed mill) – reduction of crude protein and ether extract in 1%, and increase of crude fiber in 1%.

¹ Prediction equation of Janssen (1989) – AME_n = 2,769 – 59.10*Crude Fiber + 62.10*Fat.

Example of the practical use of prediction equations to correct nutritional matrices of raw materials used in broiler feeds

Although Pearson’s correlation indicates that some prediction equations can be used in practice, caution should be taken, once differences between observed and estimated AME_n values are high sometimes.

Conclusions

Dietary energy levels directly influence broiler performance and production costs. Therefore, the real values of energy present in raw materials and the one used by nutritionists to formulate feed for broilers should be as similar as possible. This avoids rations with differing AME_n levels, on one hand energy excess levels raise feed cost and on the other hand low energy levels impairs animal performance.

Energy values provided in feedstuff composition tables, obtained by biological assays, should still be considered as the reference for poultry feed formulation. It is recommended to use these table values as starting values and correct them with regression equations.

In spite of the currently available regression equations not being accurate enough to estimate feedstuff energy directly, they are valuable tools to correct feedstuff energy values from bioassays reported in ingredient composition tables. These corrected energy values are then ideal for use in least cost formulation.

Table 8

Biological assay	Equations	Observations, n	Correlation	Significance T test
Corn and byproducts				
Rodrigues (2000)	Rodrigues (2) (2000)	14	+0.68	0.004
	Rodrigues (3) (2000)	14	+0.59	0.014
	Rostagno <i>et al.</i> (2005)	14	+0.54	0.023
	Rodrigues (1) (2000)	14	+0.47	0.046
	Janssen (1989)	14	+0.35	0.111
	Rodrigues (4) (2000)	14	+0.25	0.196
Soybeans and byproducts				
Rodrigues (2000)	Rodrigues 2 (2000)	19	+0.98	0.001
	Rodrigues 3 (2000)	19	+0.98	0.001
	Rostagno <i>et al.</i> (2005)	19	+0.97	0.001
	Janssen (1989)	19	+0.94	0.001
	Rodrigues 1 (2000)	19	+0.93	0.001
Wheat and byproducts				
Nunes (2000)	Rostagno <i>et al.</i> (2005)	11	+0.97	0.001
	Nunes 1 (2000)	11	+0.97	0.001
	Nunes 3 (2000)	11	+0.95	0.001
	Nunes 2 (2000)	11	+0.93	0.001
	Janssen (1989)	11	+0.87	0.001
Meat and bone meal (35 to 51 % CP)				
Vieites (1999)	Janssen (1989)	11	+0.85	0.001
	Vieites 3 (1999)	11	+0.63	0.018
	Rostagno <i>et al.</i> (2005)	11	+0.62	0.021
	Vieites 2 (1999)	11	-0.22	0.257
	Vieites 1 (1999)	11	-0.24	0.240
Offal meal				
Nascimento (2002)	Nascimento 3 (2002)	9	+0.82	0.003
	Dale <i>et al.</i> 2 (1993)	11	+0.68	0.025
	Janssen (1989)	11	+0.43	0.091
	Rostagno <i>et al.</i> (2005)	11	+0.42	0.100
Feather meal				
Nascimento (2002)	Nascimento 1 (2002)	6	+0.99	0.001
	Nascimento 2 (2002)	6	+0.98	0.001
	Nascimento 3 (2002)	6	+0.97	0.001
	Janssen (1989)	11	+0.38	0.176
	Rostagno <i>et al.</i> (2005)	11	+0.30	0.238

Pearson's correlation between mean AME_n values obtained by biological assays and mean AME_n values estimated by prediction equations



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Standardized ileal digestible tryptophan to lysine ratios

to optimize performance of starting, growing and finishing pigs, and factors affecting the optimum tryptophan ratio



Key information

- In the literature, there are considerable variations in the tryptophan requirements and optimum dietary tryptophan to lysine ratios for pigs, which may be influenced by many factors such as statistical models used, dietary large neutral amino acid levels, health status and genotypes.
- The optimum tryptophan to lysine ratios between corn-soybean meal based and barley-wheat-corn-peas based diets are similar when diets are formulated on standardized ileal digestible (SID) basis.
- Data evaluation of published tryptophan dose-response studies by exponential regression revealed that the optimum SID tryptophan to lysine ratio to optimize feed intake and body weight gain is 22% for starting pigs (7–25 kg body weight; BW) and 20% for growing pigs (20–50 kg BW). The SID tryptophan to lysine ratio to optimize body weight gain and feed conversion is 19% for finishing pigs (85–125 kg BW).
- The economic optimum SID tryptophan to lysine ratio for maximum net income is calculated to be 22% for starting pigs (7–25 kg BW) and 19% for finishing pigs (85–125 kg BW) which are in line with the physiological optimum estimates.
- A higher tryptophan requirement in starting pigs may be attributed to an increased need of tryptophan for optimizing feed intake (serotonin synthesis), immune function and stress control.

Introduction

Weaning is a stressful time for pigs and it is often associated with gut disorders (e.g. diarrhea), low feed intake and poor growth. In spite of strict bio-security controls, today's commercial swine operations still face serious sub-clinical disease challenges. Health status of pigs can impact feed intake, production efficiency, nutrient excretion into the environment, and profitability. Voluntary feed intake of the pig determines nutrient and energy intake levels in a diet. Thus, it is very important for optimum growth performance especially in weaned pigs for which sufficient feed intake is challenging.

Tryptophan (Trp) is an essential dietary amino acid (AA) which is required for body protein synthesis and maintenance, as well as it is involved in various metabolic pathways such as the control of immune response and synthesis of serotonin which plays a key role in the regulation feed intake and stress response (Henry *et al.*, 1992). Furthermore, it has been shown that Trp influences the animal's behavior. Short-term dietary supplementations of L-Tryptophan above requirement levels were found to reduce aggression behaviors in weaned (Martinez-Trejo *et al.*, 2009) and in growing pigs (Li *et al.*, 2006). Tryptophan is the first-limiting AA in some feed ingredients such as corn, meat and bone meal and fish meal. In corn-soybean meal (SBM) diets, Trp is usually considered as the third-limiting AA (Cromwell, 2004), and in European swine diets it is the fourth-limiting AA (Le Floc'h and Seve, 2007). With the growing ethanol production in the United States (U.S.), corn-dried distiller's grains with soluble (DDGS), a by-product of the ethanol industry, have become available for use as a livestock feed. A significant portion of these DDGS have been used in swine feeds in recent years. Tryptophan is the third-limiting AA in DDGS (Shurson *et al.*, 2008), and Trp becomes increasingly limiting with increases in the dietary inclusion level of DDGS. In pigs, failures to provide adequate Trp supply typically will result in decreased voluntary feed intake, followed by impaired performance. Thus, an adequate dietary supply of Trp is crucial for optimum feed intake and performance.

With increasing pressures to reduce nitrogen (N) excretion into the environment and drastic changes in price of ingredients, it has become

increasingly more important to know reliable requirement estimates for essential AA. Ideally pigs should be provided with a balanced diet which exactly meets all nutrient requirements for most efficient production. Over- and under-supply of AA can be best managed by applying the ideal protein concept (IPC) which provides a perfect profile of essential and non-essential AA in the diet without any excess or deficiency. The use of commercially available supplemental AA such as L-Lysine sources, L-Threonine, L-Tryptophan, and DL-Methionine in the diets makes it easier to meet pigs' AA needs as close as possible while reducing the dietary crude protein (CP) level and N excretion. For an effective application of this concept, it is imperative to know the ideal Trp:Lys ratio in pig diets.

To date, there are considerable variations in Trp requirements of pigs and optimum dietary Trp:Lys ratios among published data. These variations may be attributed to differences in experimental methodology, diet compositions and other factors. The objectives of this article is not to describe metabolic roles of Trp in pig nutrition (which was addressed in a previous AMINONews® article, July 2009) but rather to briefly review Trp requirement and optimum dietary Trp:Lys ratio estimates for pigs of various body weight (BW), and discuss some factors that may contribute to the differences in Trp requirements and ratios. The second part focuses on estimating the optimum standardized ileal digestible (SID) Trp:Lys ratios in diets for starting, growing, and finishing pigs by analysis of compiled data from the literature as well as from Evonik's recent collaborative research on Trp.

Review of Tryptophan requirements of pigs

While a moderate amount of research has been conducted to evaluate the Trp requirement of starting and growing pigs, research on the Trp requirement of finishing pigs is limited. Tryptophan research in pigs started about six decades ago when Beeson *et al.* (1949) studied the effects of a Trp deficiency in growing pigs, and Shelton (1951) first reported the Trp requirement of weaned pigs. The Trp requirement estimates of 19 studies and the NRC (1998) values, covering pigs of various BW are summarized in Table 1.

Table 1

BW, kg	CP, %	SID Lys, %	SID Trp, %	SID Trp, g/d	Diet type	Breeds ²	Sex ³	AB ⁴	Statistical method	Reference
5–10	23.7	1.19	0.22	1.10	Corn-SBM⁵	n. a.⁶	M	n. a.	Growth model	NRC, 1998
5–7	n. a.	1.35	0.21	0.63	Corn-peas	LW/LR x DR	M	+	Broken-line	Guzik <i>et al.</i> , 2002
5–10	20.0	(1.30)	(0.23)	(0.73)	Cornstrach-peas-CGM ⁷	LW	M	–	Linear	Seve <i>et al.</i> , 1991
6–10	n. a.	1.19	0.20	1.06	Corn-peas	LW/LR x DR	M	+	Broken-line	Guzik <i>et al.</i> , 2002
6–16	22.4	(1.39)	0.15	0.72	Corn-CGM-fishmeal	LW/LR x DR	M	+	Broken-line	Burgoon <i>et al.</i> , 1992
6–22	n. a.	(1.10)	(0.16)	1.23	Corn-SFM ⁸	LW x LR	M	+	Linear	Borg <i>et al.</i> , 1987
10–20	20.9	1.01	0.18	1.90	Corn-SBM	n. a.	M	n. a.	Growth model	NRC, 1998
10–16	n. a.	1.01	0.18	1.39	Corn-peas	LW/LR x DR	M	+	Broken-line	Guzik <i>et al.</i> , 2002
11–22	n. a.	n. a.	0.14	1.36	Corn-whey-cornstarch	LW x LR	M	+	Broken-line	Han <i>et al.</i> , 1993
11–37	18.1	(1.15)	(0.23)	n. a.	Corn-CGM	LW x LR	M	–	Linear	Schutte <i>et al.</i> , 1989
20–50	18.0	0.83	0.15	2.80	Corn-SBM	n. a.	M	n. a.	Growth model	NRC, 1998
18–35	12.0	(0.84)	(0.17)	(3.28)	Corn-SBM	LW x Hampshire	M	+	Broken-line	Russell <i>et al.</i> , 1983
20–35	13.7	(0.89)	0.13	1.60	Corn starch-herring meal	n. a.	M	–	Broken-line	Henry <i>et al.</i> , 1986
20–40	16.2	(1.09)	0.18 ⁹	2.55 ⁹	Barley-corn-Cassava	GY x NL	M	–	Linear	Schutte <i>et al.</i> , 1995
22–50	15.6	(0.92)	0.10	2.28	Corn-CGM-fishmeal	LW/LR x DR	M	+	Broken-line	Burgoon <i>et al.</i>, 1992
25–50	13.3	0.87	0.20	3.39	Corn-barley-peas	LR/LW x Pietrain	G	–	Exponential ¹⁰	Eder <i>et al.</i> , 2003
30	n. a.	0.87	0.17 ¹¹	n. a.	Corn-peas-SBM	LW/LR x DR	B	–	Broken-line	Guzik <i>et al.</i> , 2005 a
50	n. a.	0.70	0.14 ¹¹	n. a.	Corn-peas-SBM	LW/LR x DR	B	–	Broken-line	Guzik <i>et al.</i> , 2005 a
50–80	15.5	0.66	0.12	3.10	Corn-SBM	n. a.	M	n. a.	Growth model	NRC, 1998
50–80	13.1	0.80	> 0.17	3.71	Corn-barley-peas	LR/LW x Pietrain	G	–	Exponential	Eder <i>et al.</i> , 2003
55–97	12.3	(0.73)	0.06	2.86	Corn-barley-peas	LW/LR x DR	M	+	Broken-line	Burgoon <i>et al.</i> , 1992
70	n. a.	0.61	0.10	3.30	Corn-peas-SBM	LW/LR x DR	B	–	Broken-line	Guzik <i>et al.</i> , 2005 a
80–120	13.2	0.52	0.10	2.90	Corn-SBM	n. a.	M	n. a.	Growth model	NRC, 1998
80–115	10.7	0.56	0.12	3.77	Corn-barley-peas	LR/LW x Pietrain	G	–	Exponential	Eder <i>et al.</i> , 2003

BW = body weight
 CP = crude protein
 SID = standardized ileal digestibility

¹ Values in parenthesis are on a total basis.
² LW = Large white; LR = Landrace; DR = Duroc; GY = Great Yorkshire.
³ M = Mixed-sex; B = Barrows; G = Gilts.
⁴ AB = antibiotics (+/-: with/without).
⁵ SBM = Soybean meal.
⁶ n. a. = Not available.

⁷ CGM = Corn gluten meal.
⁸ SFM = sunflower meal.
⁹ Values are on AID basis.
¹⁰ Estimated at 95% of asymptotic response.
¹¹ Plasma urea nitrogen was used as response parameter, otherwise growth performance criteria were used in other studies.

Review of Trp requirement estimates for pigs of various body weight categories¹

In general, older studies (e.g. Borg *et al.* 1987; Burgoon *et al.* 1992) reported Trp requirements that are below the NRC (1998) recommendations, while Guzik *et al.* (2002) more recently reported that the SID Trp requirement was 0.21, 0.20 and 0.18% for pigs of 5 – 7, 5 – 10 and 6 – 10 kg BW, respectively which are close to or slightly below the NRC (1998) value of 0.22% (5 – 10 kg BW). On the other hand, higher than NRC (1998) Trp requirement estimates were reported mainly from experiments conducted in Europe. For example, Eder *et al.* (2003) estimated the SID Trp requirement to be 0.20% for growing pigs (20 – 50 kg BW), 0.17% for growing-finishing pigs (50 – 80 kg BW), and 0.12% for finishing pigs (80 – 115 kg BW), respectively which exceed the NRC (1998) recommendations of 0.15, 0.12 and 0.10% SID Trp for pigs of the three corresponding BW categories (Table 1).

From the data summary in Table 1, it is clear that there were differences in dietary CP level, genotypes and use of in-feed antibiotics among the experiments. All experiments used corn as the main dietary component but other ingredients varied among studies. A majority of the researchers used older genetics, mixed-sex pigs and applied the broken-line regression to estimate the Trp requirement. Interestingly, the trials that obtained higher Trp requirement estimates (i. e. Eder *et al.*, 2003) used gilts of Pietrain cross which is known to have a higher lean gain and applied exponential regression. Additionally, older studies reported the Trp estimates on a total basis while more recent studies reported on SID basis. Overall, the published Trp requirement data vary considerably. Therefore, it is difficult to conclude for a commonly agreeable Trp estimate for a given BW category of pigs.

Review of optimum Trp to Lys ratios in pig diets

The first proposal for IPC referred directly to the ratio of essential AA in the diet without any access or deficiency. As research progressed in this area, Fuller *et al.* (1989), Chung and Baker (1992), and Cole and van Lunen (1994) further developed the IPC. The optimum Trp:Lys ratios given by these authors ranged from 18 to 19% in diets for growing pigs. Based on the requirement estimates for Lys and Trp, the average value of NRC (1998) for optimum Trp:Lys ratio in grower diets is 18%. A recent literature review suggested an optimum Trp:Lys ratio of 17% for grower diets (Susenbeth, 2006).

In the IPC, the concentrations for each of other essential amino acids (EAA) are expressed as a percentage of Lys, which is set at 100%. A clear advantage of applying IPC is that the requirements of other EAA can be estimated as long as the requirement of Lys is known. Additionally, formulating diets according to IPC allows for the most efficient and economical use of dietary protein while minimizing N excretion to the environment. The IPC was first introduced almost 30 years ago (ARC, 1981). Due to differences in the availability of the individual AA among ingredients, the IPC should be based on digestible AA. Generally, the SID is considered to be the most correct measure for availability, and it is suggested that SID values should be used in feed formulation (Stein *et al.*, 2007). Therefore, for obtaining maximum accuracy in balancing the dietary AA, the IPC should be based on the SID of the individual AA (Boisen, 2003).

Both the requirement and ratio of individual AA can be determined by “dose response” studies wherein the performance data are usually used as response criteria in *ad libitum* feeding condition. However, it is important to mention that the experimental designs for AA requirement and for AA ratio are different. In an AA requirement trial, the AA under investigation must be first-limiting while all EAA (including Lys) have to be supplied at or above the requirement in the diets to ensure that these AA will not limit the performance. In an AA ratio trial, the AA under investigation and Lys (or the reference AA) must be first- and second-limiting, respectively while all other EAA need to be supplied at or preferably slightly above requirement to avoid underestimation of the test AA and Lys ratio (Boisen, 2003).



Table 2

BW, kg	Dietary CP, %	SID Trp:Lys, %	Diet type	Breeds ¹	Sex ²	AB ³	Statistical method	Reference
7–16	20.9	> 19.5	Wheat-barley-peas	PIC	M	–	Linear	Guzik <i>et al.</i> , 2005
7–17	19.3	> 20.3	Wheat-corn-SBM	LW x LR	B	–	Linear	Pluske and Mullan, 2000
9–24	18.3	23.1	Corn-SBM ⁴	n. a. ⁵	M	–	Exponential	Jansman and van Diepen, 2007
9–24	18.3	21.4	Wheat-barley-peas	n. a.	M	–	Exponential	Jansman and van Diepen, 2007
11–26	18.5	> 23.0	Wheat-barley-corn	LW/LR	M	–	Linear	Lynch <i>et al.</i> , 2000
89–123	9.3	14.5–17.0	Corn-SBM	EB x Newsham	B	–	Broken-line Quadratic	Kendall <i>et al.</i> , 2007
10–20	20.9	18.0	Corn-SBM	n. a.	M	n. a.	Growth model	NRC, 1998
20–50	18.0	18.0	Corn-SBM	n. a.	M	n. a.	Growth model	NRC, 1998
80–120	13.2	19.0	Corn-SBM	n. a.	M	n. a.	Growth model	NRC, 1998

BW = body weight

CP = crude protein

SID = standardized ileal digestibility

¹ LW = Large white; LR = Landrace; EB = Monsanto Choice Genetics.

² M = Mixed-sex; B = Barrows; G = Gilts.

³ AB = Antibiotics (+/-: with/without).

⁴ SBM = Soybean meal.

⁵ n. a. = Not available.

Review of optimum Trp:Lys ratios in different pig diets

Research related to Trp has received more attention around year 2000 partly due to increases in availability of economically priced supplemental AA coupled with the application of reduced protein diets to minimize N excretion. For the present review, the SID Trp:Lys estimates of 6 studies and the NRC (1998) ratio values are given in Table 2. Kendall *et al.* (2007) reported the SID Trp:Lys ratio of 14.5 to 17.0% for 89 – 123 kg finishing pigs which is lower than the NRC (1998) value of 19%. In contrast, all other Trp:Lys ratios for starting and growing pigs were higher than the NRC (1998) values despite differences in experimental setup (e.g., ingredients, genetics, statistical models). Therefore, there is a need to re-evaluate the optimum Trp:Lys ratio in pig diets.

Factors influencing the Trp requirement and optimum Trp:Lys ratios in swine diets

Literature data clearly indicates that both Trp requirements and optimum dietary Trp:Lys ratios in swine diets vary greatly. Many factors may have attributed to these variations existing among the published literature, and some, if not all, main factors, such as statistical models, dietary CP and large neutral amino acids (LNAA) levels, health status and use of in-feed antibiotics, genotypes and sex, digestibility of Trp in feed ingredients used, and the accuracy of AA analyses should be considered in estimating the optimum dietary Trp supply.

Statistical models

As reported in Tables 1 and 2, there is considerable variation in the Trp requirement and optimal Trp:Lys ratio. The differences in these published requirements and ratios can in part be attributed to the different statistical models that were used in the studies. Requirements of AA are defined for groups or populations. There is no universally accepted definition of requirement because the AA needs of individual animals in a population typically vary, and different opinions exist to which percentage of population should be taking into account as requirement (Baker, 1986).

Linear broken-line regression is largely used probably because it describes an objective break point of the two lines as the requirement. However, the broken-line model assumes that the dose response of a nutrient is linear until the requirement is met and above which no significant change in response can be expected (Robbins *et al.*, 2006). In reality, a population of animals exhibit a smooth nonlinear response to a specific nutrient (Curnow, 1973; Morris, 1983; Baker, 1986; Schutte and Pack, 1995), therefore, the broken-line model may then be biologically inadequate, and underestimate the requirement (Robbins *et al.*, 2006). Some used the quadratic broken-line model or combination of quadratic and broken-line models to estimate AA requirement for curvilinear data sets (Kerr *et al.*, 2004; Robbins *et al.*, 2006).



Ideally a model fit to response data should meet both mathematical and biological considerations (Mercer, 1992). In this regard, the exponential regression model is more suitable than broken-line because it best describes the growth responses to limiting nutrients which are generally curvilinear (Curnow, 1973; Schutte and Pack, 1995). In the exponential regression analysis an optimum is generally estimated by arbitrarily setting a point at 90 or 95% of the maximum curvilinear response (Baker, 1986). Generally, the broken-line regression analysis predicts the lower requirement values than those determined by the exponential regression.

Dietary CP and large neutral amino acids levels

In addition to its need for growth, Trp is needed for serotonin synthesis which plays a role in feed intake regulation (Henry *et al.*, 1992). Studies have demonstrated that insufficient dietary Trp will result in reduced feed intake and growth performance in pigs (e.g. Henry *et al.*, 1992; Eder *et al.*, 2003). The dietary level of CP or rather LNAA (i.e. Leu, Ile and Val, Phe, Tyr) can affect the optimum Trp:Lys ratio due to potential imbalances between Trp and the LNAA. Jansman *et al.* (2002) demonstrated that feed intake and subsequent BW gain of starting pigs were maximized at the apparent ileal digestible (AID) Trp:Lys ratio of 23% when fed a 17% CP diet (1.0% AID Lys) with a high Trp:LNAA ratio (0.07), whereas for 20% CP diet (1.0% AID Lys) with a low Trp:LNAA ratio (0.04), feed intake and BW gain were lower and did not further increase above the Trp:Lys ratio of 19%. This was due to a lower concentration of Trp relative to LNAA in the 20% CP diet, and due to subsequent reduced formation of serotonin and consequently the feed intake because Trp shares and competes with the LNAA for transport through the blood-brain-barrier into the brain. The CP levels of diets used in Trp requirement and ratio studies varied considerably (Table 1 and 2), and hence, the dietary CP contents or Trp:LNAA ratios may have affected the Trp estimates.

Health status of pigs and the use of in-feed antibiotics

One of the differences among the Trp requirement studies (Table 1) was the use of antibiotics (AB) in the diets. Trials conducted in the US mostly included AB in the diets whereas those carried out in Europe did not use AB. Pig diets have been fortified with AB to improve growth performance and health status during the past six decades. Becker *et al.* (1955) first demonstrated that the Trp requirement of starting pigs was higher when fed diets without AB compared with AB-fortified diets. It has been shown that the withdrawal of AB increased the EAA requirements and the optimum dietary SID EAA (Thr:Lys) ratio for maximum growth performance in growing-finishing pigs (Bikker *et al.*, 2003, 2007).

It is well accepted that the use of in-feed AB is an effective means to improve the health status of pigs, especially in young pigs. The use of AB for growth promotion in livestock diets has been banned in the European Union since January 2006 but not in other countries including the U.S. The withdrawal of AB may allow higher microbial growth in the digestive tract, reduce ileal digestibility of amino acids (Dierick *et al.*, 1986), increase incidence of *E. coli* diarrhea (Mateos *et al.*, 2000), and negatively affect the health (immune) status and performance of the pigs because intestinal bacteria can modulate health and nutrition of the pigs. Bacteria in the upper gastrointestinal tract compete with the host (pig) for readily available AA, but at the same time the intestinal microbial synthesis of AA may also be utilized by the pig to meet its AA requirement (Torrallardona *et al.*, 2003).

Tryptophan also plays a role in proper function of the immune system by its catabolism through the kynurenine pathway. Tryptophan that is not utilized for protein synthesis is primarily (> 95%) metabolized via the kynurenine pathway which is induced by the interferon gamma during infection and tissue inflammation (Botting, 1995). In pigs suffering from inflammation, a decline in plasma Trp concentration was observed (Melchior *et al.*, 2004) indicating an increased use of Trp for immune functions during sub-clinical disease conditions at the expense of growth performance. Overall, it is possible that the requirement of Trp or dietary Trp:Lys ratio is higher when pigs fed diets without AB compared with AB-fortified diets. This may also be true for pigs kept under sub-clinical disease, poor sanitation or stressful conditions (e.g. period after weaning or during early lactation).

Genotypes and sex

Pig genotypes differ in their genetic potentials to deposit lean and fat in the body. The rate and composition of BW gain during the growing-finishing period can affect the AA requirement. For example, crosses of Pietrain, a popular breed in Europe, are leaner but have a lower feed intake capacity (Van Oeckel *et al.*, 1997), and are generally more sensible to stressful conditions compared to other pig breeds. The dietary AA (Trp) requirements of modern genotypes with a greater capacity for body growth and protein accretion are higher than that of the older genotypes (Friesen *et al.*, 1994; Kendall *et al.*, 2008). Gilts and barrows also differ in their pattern of lean and fat deposition. Because gilts usually have a higher lean deposition rate (Schinckel and de Lange, 1996), and generally consume less feed than barrows (Ekstrom, 1991), the dietary AA (Trp) requirement of gilts, expressed as a percentage of the diet, is higher than that of barrows (Warnants *et al.*, 2008). Hence, differences in genotype and sex of pigs among different experiments may also affect the Trp requirements.

Total content and digestibility of Trp in common feedstuffs

Differences in the ingredients used among experiments also may influence the Trp estimates. As shown in Figure 1, there is a great deal of variation in total and SID contents of Trp in common feedstuffs used in swine diets. This means that if the requirement and ratio of Trp estimates are determined on total basis, and if ingredients used are different among experiments, it is likely that the results will not be the same. If dietary content of AA are balanced on SID basis and at the same requirement level, differences in ingredients used should not impact on the performance response as demonstrated by Jansman and van Diepen (2007).

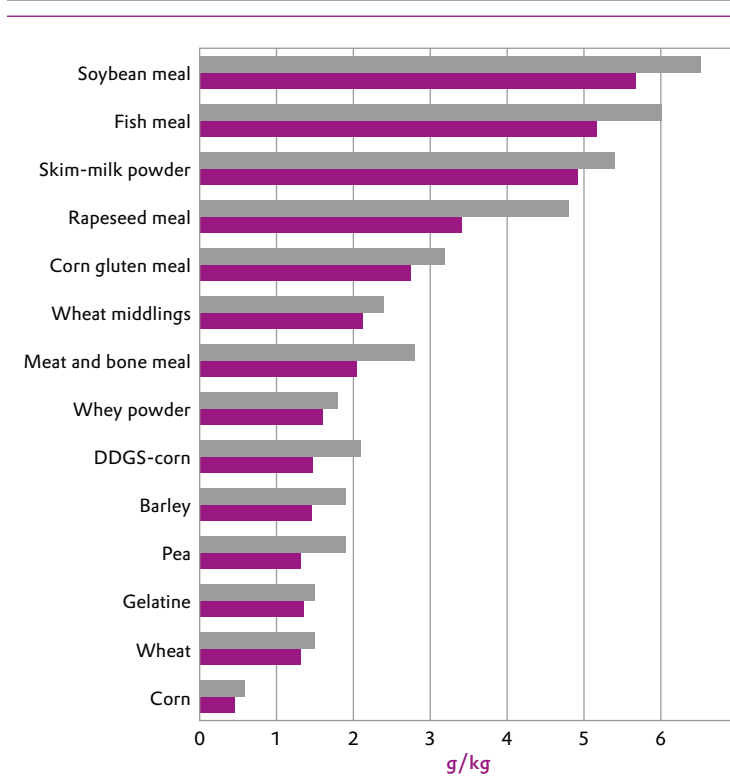
Accuracy of diet mixing and amino acid analyses

Proper mixing of diets depends on the skill of personnel and the capacity of the mixer. It is not uncommon that the analyzed dietary Trp results obtained from laboratories are somewhat different from the calculated values. In addition, AA analyzed values also usually vary among different laboratories (Fontaine and Eudaimon, 2000; Cromwell *et al.*, 2003), especially for Trp (Sato *et al.*, 1984) because unlike other AA, Trp analysis requires an additional step of alkaline hydrolysis. Therefore, the differences in feed mixing and AA analysis may partly contribute to the variations observed among the published literature.

Types of response criteria

Most of the researchers used growth performance data such as average daily feed intake (ADFI), average daily gain (ADG) and feed conversion ratio (FCR) as response criteria while some researchers used plasma urea nitrogen (PUN) to estimate the Trp requirements (Table 1). As a limiting AA increases towards its optimal value, more protein is synthesized which will lead to increased N retention, improved animal performance and decreased PUN. As such, it is generally assumed that the measurement of PUN provides an indirect measurement of changes in protein synthesis. However, it seems that the acceptance of using PUN varies among scientists. For some, PUN is a valid and useful parameter to estimate AA requirement (e.g. Coma *et al.*, 1995; Guzik *et al.*, 2002), however, some found it difficult to fit the PUN data sets for regression analysis to derive optimum AA level (e.g., Parr *et al.*, 2003, 2004). Generally, fitting PUN data leads to a lower AA requirement or optimum ratio (e.g., Guzik *et al.*, 2002; Kerr *et al.*, 2004). Therefore, the response variables used to estimate will likely affect the Trp requirement.

Figure 1



■ Total Trp ■ SID Trp

Total and SID of Trp in common feedstuffs used in swine diets (AMINODat® 3.0)

Evaluation of Trp dose-response data for estimating optimum dietary SID Trp:Lys ratios

The optimum Trp:Lys ratio estimates among different experiments vary considerably. In such instances, the evaluation (meta-analysis) of available published literature is worth doing to yield more conclusive results (Sauvant *et al.*, 2008). Because experiments that evaluated the optimum Trp ratio in pig diets are limited, dose response data of suitable Trp requirement studies were included in the data pool for estimating optimum SID Trp:Lys ratios provided that the dietary Lys was not over-supplied (compared with Lys recommendations by Evonik, 2009) in these studies.

The ideal ratios of Trp, Thr and sulfur AA to Lys for maintenance are higher than for protein deposition (NRC, 1998); therefore, it is reasonable to think that the ideal Trp:Lys

ratio changes over the pig life. Thus, the available literature data were divided for starting, growing and finishing pigs for the present evaluations. Because individual trials differed in dietary CP, genotypes, sex and environmental conditions, the performance data such as ADFI, ADG and FCR of all trials were pooled after converting them to a relative scale (% of maximum response) within each study for a better fit. The response in performance to a limiting nutrient can best be described by exponential regression analysis (Schutte and Pack, 1995). Therefore, the exponential regression (SAS Inst., Inc., Cary, NC) which also seemed to fit better to dose responses (Figures 2 – 4), was used to estimate the optimum SID Trp:Lys ratios at 95% of the asymptotic response.

Table 3

BW, kg	SID Trp:Lys range, %	Dietary CP, %	SID Lys, %	Diet type	Breed ¹	Sex ²	AB ³	Reference
7-16	14.5 to 19.5	20.9	1.10	Wheat-barley-peas	PIC	M	-	Guzik <i>et al.</i> , 2005 a
7-16	14.5 to 19.5	20.9	1.10	Wheat-barley-peas	PIC	M	-	Guzik <i>et al.</i> , 2005 b
7-16	14.5 to 19.5	20.9	1.10	Wheat-barley-peas	PIC	M	-	Guzik <i>et al.</i> , 2005 c
7-17	16.0 to 21.0	19.3	1.12	Wheat-corn-SBM ⁴	LW x LR	B	-	Pluske and Mullan, 2000
10-16	12.9 to 22.8	n. a. ⁵	1.01	Corn-peas	LW/LR x DR	M	+	Guzik <i>et al.</i> , 2002
9-24	14.0 to 23.0	18.3	1.03	Corn-SBM	n. a.	M	-	Jansman and van Diepen, 2007 a
9-24	14.0 to 23.0	18.3	1.03	Wheat-barley-peas	n. a.	M	-	Jansman and van Diepen, 2007 b
11-26	14.0 to 23.0	18.5	1.12	Wheat-barley-corn	LW/LR cross	M	-	Lynch <i>et al.</i> , 2000
10-26	16.0 to 24.0	18.0	1.04	Barley-corn-cassava	LW x LR	M	-	Schutte <i>et al.</i> , 1989

BW = body weight

CP = crude protein

SID = standardized ileal digestibility

¹ LW = Large white; LR = Landrace; DR = Duroc.

² M = Mixed-sex; B = Barrows; G = Gilts.

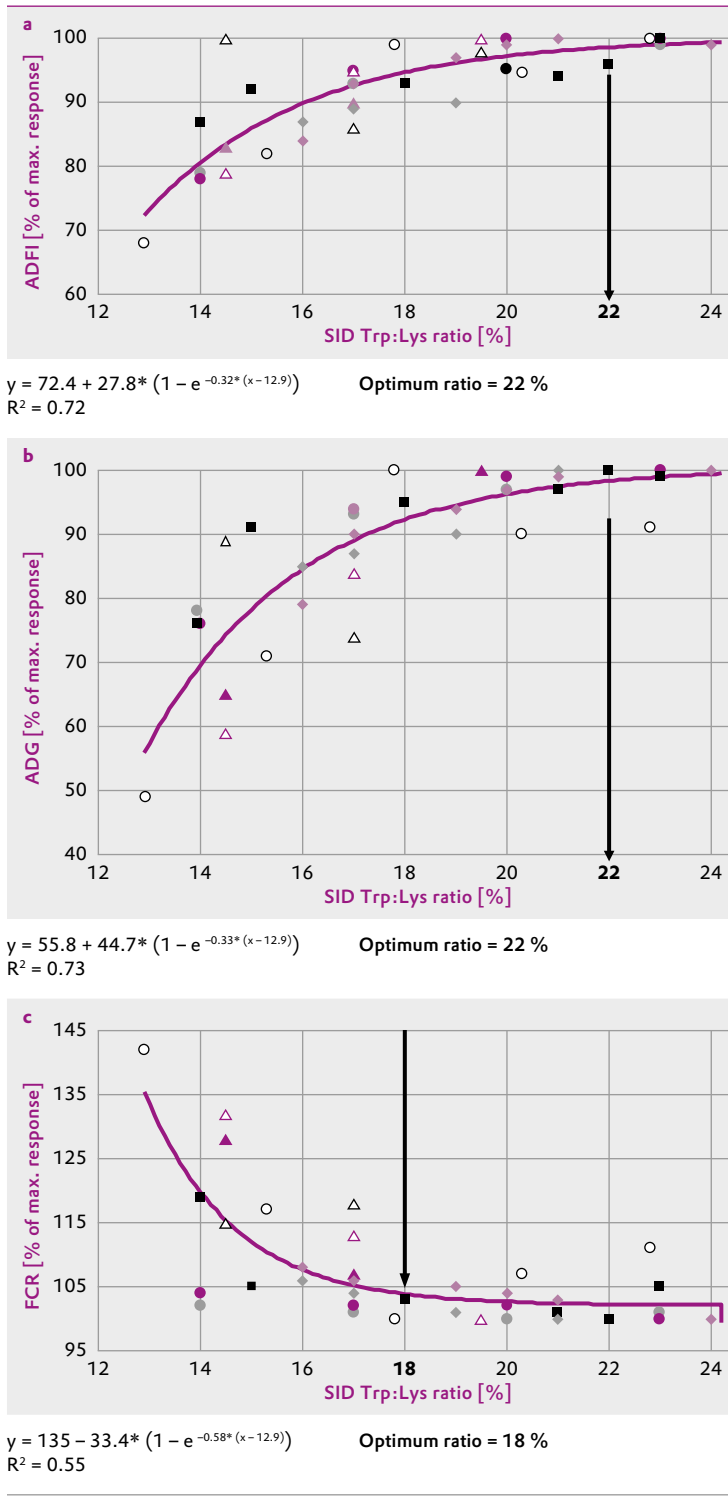
³ AB = Antibiotics (+/-: with/without).

⁴ SBM = Soybean meal.

⁵ n. a. = Not available.

Summary of the experiments used in the data analysis for the starting pigs

Figure 2



- Guzik *et al.*, 2002
- Jansman and van Diepen, 2007a
- Jansman and van Diepen, 2007b
- △ Guzik *et al.*, 2005a
- △ Guzik *et al.*, 2005b
- ▲ Guzik *et al.*, 2005c
- Lynch *et al.*, 2000
- ◆ Pluske and Mullan, 2000
- ◆ Schutte *et al.*, 1989

Relative response in average daily feed intake (ADFI), average daily gain (ADG) and feed conversion ratio (FCR) of starting pigs (7–25 kg BW) to graded dietary SID Trp : Lys ratios

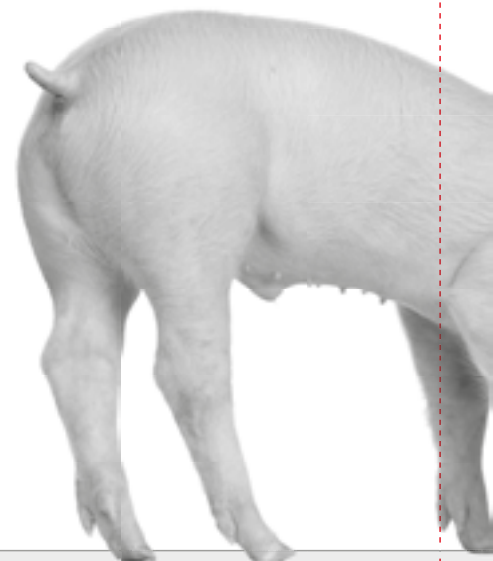
Evaluation of Trp dose-response data for starting pigs

A total of nine data sets from six dose-response studies were used to estimate the SID Trp : Lys ratio in diets for starting pigs (7 – 26 kg BW). All trials met the requirements for an AA ratio design wherein dietary Lys was marginally limiting for the given BW ranges (Table 3). All experiments were conducted within a similar range of dietary SID Trp : Lys ratios which ranged from 12.9 to 24.0%. The ingredients used and genotypes vary considerably between the studies. Details of the experimental data are given in Table 3.

Fitted exponential plots of ADFI, ADG and FCR as functions of optimum SID Trp : Lys ratio in starting pig diets are illustrated in Figure 2. The optimum SID Trp : Lys ratio that maximized both ADFI and ADG determined by exponential regression analysis, was 22%. The SID Trp : Lys ratio that minimized FCR was calculated to be 18%.

Evaluation of Trp dose-response data for growing pigs

Evaluation of the Trp : Lys ratio in grower diets is basically nonexistent. Therefore, Evonik has recently conducted some collaborative research trials with two Universities to determine the optimal Trp : Lys ratio for growing pigs. The published Trp : Lys ratios derived from experiments which used European type of diets seem to be higher than those derived from corn-SBM based US type diets (Table 2). Therefore, both types of diets were used in these experiments. All ingredients were analyzed for total AA content and diets formulation was based on the SID basis using the SID AA of Evonik (2009).

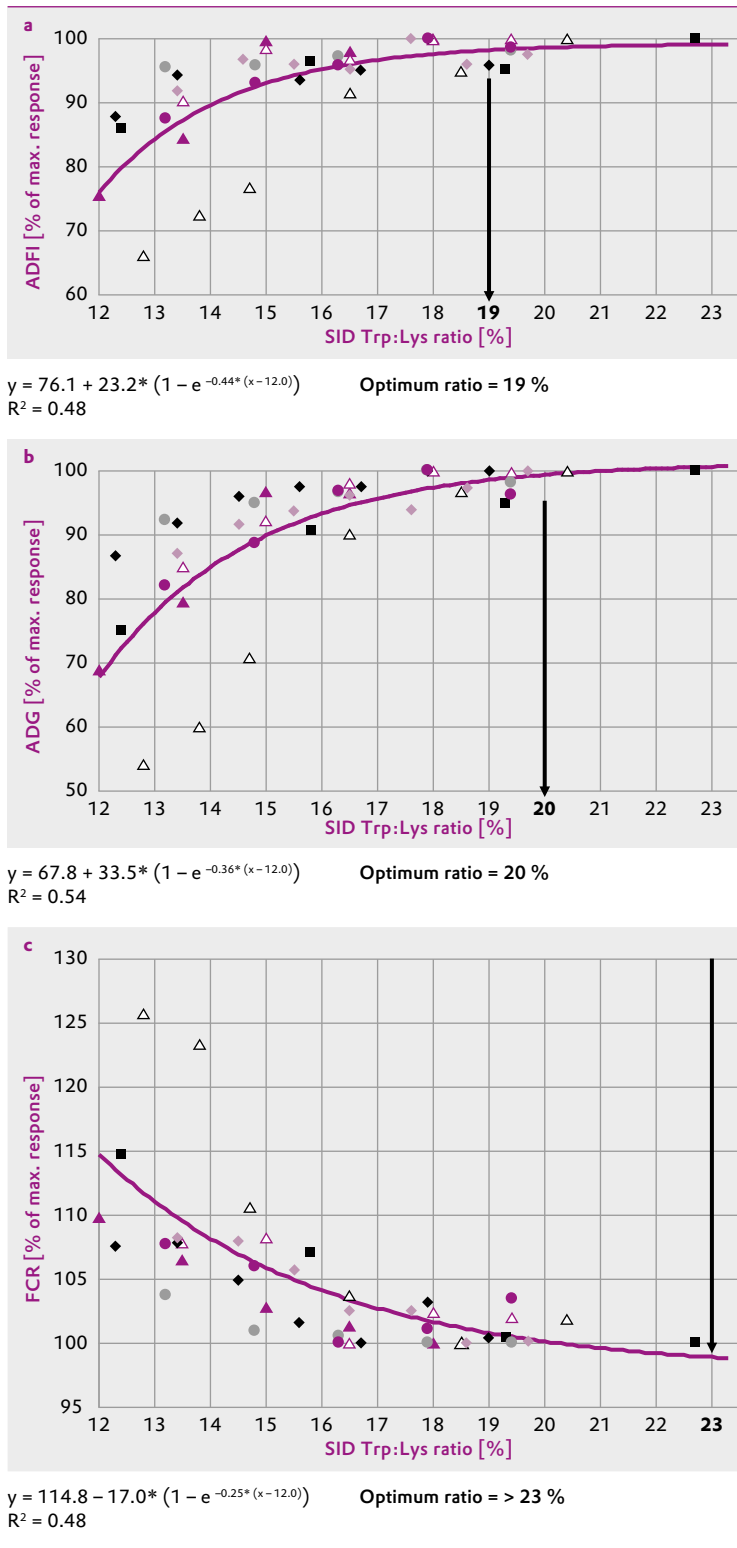


Three experiments (25 – 40 kg BW) were conducted by the research team of Prof. Lindemann at the University of Kentucky, USA (Lindemann, 2007). Experiment 1 and 3 used corn-SBM based diets while barley-wheat-peas based complex diets were used in Exp. 2. The dietary Lys was set at the same sub-optimal level in all three experiments. The dietary EAA level were supplied at requirement level in Exp. 3 whereas in Exp. 1 and 2, the dietary EAA were balanced according to ideal ratio relative to the Lys content which was marginally limiting (Table 4). Although the ADG of pigs fed corn-SBM based diets were lower at low SID Trp:Lys ratios, generally the maximum ADG and shape of response curves were similar to their counterparts that received the complex diets (Figure 3).

Two other experiments (15 – 35 kg BW) were conducted by the research team of Prof. Susenbeth at the University of Kiel, Germany (Naatjes and Susenbeth, 2009). Experiment 1 used wheat-barley-SBM complex diets whereas corn-SBM diets were used in Exp. 2. The dietary Lys was set at the same marginally limiting level in both experiments and other EAA levels met requirement level (Table 4). The growth responses were similar irrespective of diet composition (Figure 3). Dose response data sets from Schutte *et al.* (1995) and Eder *et al.* (2003) were added to the data analysis. The dietary Trp:Lys ratios in the study of Schutte *et al.* (1995) were on AID basis, however, these values should be similar to the ratios based on SID basis (Van Cauwenberghe and Relandeau, 2000). Details of the experimental data are given in Table 4.

Fitted exponential plots of ADFI, ADG and FCR as functions of optimum SID Trp:Lys ratios in growing pig diets are illustrated in Figure 3. The optimum SID Trp:Lys ratio that maximized ADFI determined by exponential regression analysis was 19%. Exponential regression estimated an optimum SID Trp:Lys ratio of 20% to optimize the ADG. The SID Trp:Lys ratio that minimized FCR was calculated to be higher than 23%.

Figure 3



- ▲ Lindemann, 2007a
- △ Lindemann, 2007b
- △ Lindemann, 2007c
- ◆ Naatjes and Susenbeth, 2009a
- ◆ Naatjes and Susenbeth, 2009b
- Eder *et al.*, 2003
- Schutte *et al.*, 1995a
- Schutte *et al.*, 1995b

Relative response in average daily feed intake (ADFI), average daily gain (ADG) and feed conversion ratio (FCR) of growing pigs (20–50 kg BW) to graded dietary SID Trp:Lys ratios

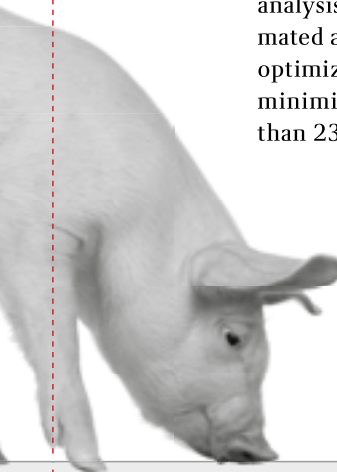


Table 4

BW, kg	SID Trp:Lys range, %	Dietary CP, %	SID Lys, %	Diet type	Breed ¹	Sex ²	AB ³	Reference
15–35	12.7 to 19.4	16.6	1.05	Wheat-barley-SBM	Pietrain x dbNaima	M	–	Naatjes and Susenbeth, 2009 a
15–35	13.8 to 20.1	19.5	1.05	Corn-SBM ⁴	Pietrain x dbNaima	M	–	Naatjes and Susenbeth, 2009 b
25–40	12.0 to 18.1	13.1	0.66	Corn-SBM	LW/LR x DR	M	+	Lindemann, 2007 a
25–40	13.1 to 18.1	12.8	0.66	Barley-wheat-peas	LW/LR x DR	M	+	Lindemann, 2007 b
25–40	13.1 to 20.9	14.01	0.66	Corn-SBM	LW/LR x DR	M	+	Lindemann, 2007 c
20–40	13.6 to 19.8 ⁵	16.2	0.97	Barley-corn-cassava	GY x LR	M	–	Schutte <i>et al.</i> , 1995 a
20–40	13.6 to 19.8 ⁵	16.2	0.97	Barley-corn-cassava	GY x LR	M	–	Schutte <i>et al.</i> , 1995 b
25–50	12.8 to 23.1	13.3	0.87	Corn-barley-peas	LR/LW x Pietrain	G	–	Eder <i>et al.</i> , 2003

BW = body weight

CP = crude protein

SID = standardized ileal digestibility

¹ LW = Large white; LR = Landrace; DR = Duroc; GY = Great Yorkshire.

² M = Mixed-sex; B = Barrows; G = Gilts.

³ AB = Antibiotics (+/-: with/without).

⁴ SBM = Soybean meal.

⁵ Values are on AID basis.

Summary of the experiments used in the data analysis for the growing pigs

Evaluation of Trp dose-response data for finishing pigs

A total of 5 data sets from 3 published papers were used to estimate the SID Trp:Lys ratio in diets for finishing pigs (80 – 125 kg BW). Kendall *et al.* (2007) conducted all 3 trials according to the AA ratio design wherein dietary Lys was marginally limiting. Requirement dose response data sets of Eder *et al.* (2003) and Guzik *et al.*

(2005) were added to the data pool because the dietary Lys levels and BW ranges were similar to Kendall *et al.* (2007). The SID Trp:Lys ratios ranged from 10.9 to 23.5% among the experiments. Ingredients and genotypes and use of AB vary between the studies. Details of the experimental data are given in Table 5.

Table 5

BW, kg	SID Trp:Lys range, %	Dietary CP, %	SID Lys, %	Diet type	Breed ¹	Sex ²	AB ³	Reference
89–114	10.9 to 21.8	8.7	0.55	Corn-SBM ⁴	EB x Newsham	B	–	Kendall <i>et al.</i> , 2007 a
91–123	13.0 to 23.5	9.3	0.55	Corn-SBM	PIC	B	+	Kendall <i>et al.</i> , 2007b
99–123	13.0 to 21.0	10.0	0.55	Corn-SBM	PIC	B	–	Kendall <i>et al.</i> , 2007c
80–115	12.3 to 22.9	10.7	0.56	Corn-barley-peas	LR/LW x Pietrain	G	–	Eder <i>et al.</i> , 2003
80–105	11.5 to 23.1	10.7	0.52	Corn-feather meal	LW/LR x DR	B	–	Guzik <i>et al.</i> , 2005d

BW = body weight

CP = crude protein

SID = standardized ileal digestibility

¹ LW = Large white; LR = Landrace; DR = Duroc; EB = Monsanto Choice Genetics.

² B = Barrows; G = Gilts.

³ AB = Antibiotics (+/-: with/without).

⁴ SBM = Soybean meal.

Summary of the experiments used in the data analysis for the finishing pigs

The ADFI responses of finishing pigs were inappropriate for regression analysis. Fitted exponential plots of ADG and FCR as functions of optimum SID Trp:Lys ratio in finishing pig diets are illustrated in Figure 4. The optimum SID Trp:Lys ratio that maximized ADG determined by exponential regression analysis was 18%. The SID Trp:Lys ratio that minimized FCR was calculated to be 20%.

Estimation of economic optimum Trp:Lys ratios

The ultimate goal of pig producers is to achieve maximum profitability. Thus, it is worthwhile to determine the optimum dietary Trp:Lys ratio that maximizes economic return based on the actual production situation. This economic optimum can be calculated by combining the growth performance responses (analyzed by exponential regression) with key economic figures such as prices of ingredients, L-Tryptophan and pork meat. For the current examples, the relative performance data of starting and finishing pigs were calculated back to absolute values by setting the maximum absolute value at 100%. The maximum performance levels were set at 600g ADG and 1.55 FCR for starting pigs and 970g ADG and 3.30 FCR for finishing pigs, respectively.

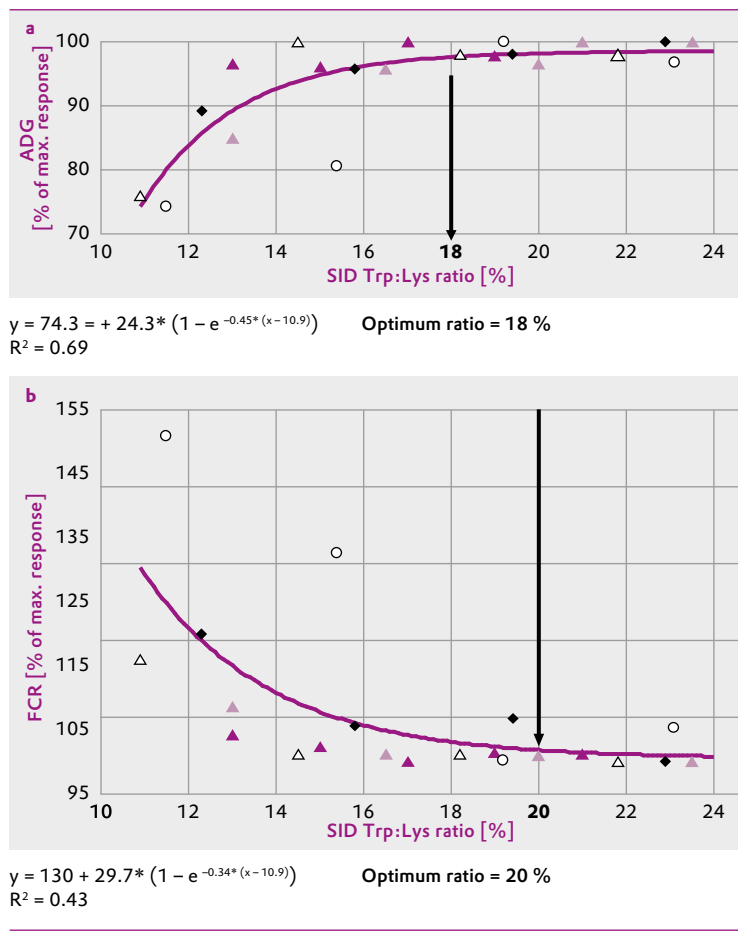
As an economic parameter, gross margin/pig/day were calculated based on two price scenarios of L-Tryptophan (30 or 33 US\$/kg). A low and high prices of basal feed without L-Tryptophan were set at 0.30 and 0.33 US\$/kg for starter diet, and 0.24 and 0.30 US\$/kg for finisher diet, respectively. The price of pork meat was set at 1.3 US\$/kg BW gain. The performance responses of starting and finishing pigs used in the calculation are given in Figure 2 and 4, respectively. The economic parameters were calculated as follows:

$$\text{Cost per kg feed} = \text{basal feed cost} + [(\text{cost/unit L-Tryptophan} - \text{cost/unit basal feed}) * \text{supplemented L-Tryptophan units}]$$

$$\text{Cost per kg BW gain} = \text{FCR} * \text{cost per kg feed}$$

$$\text{Gross margin per pig per day} = (\text{pork price} - \text{cost per kg BW gain}) * \text{ADG}$$

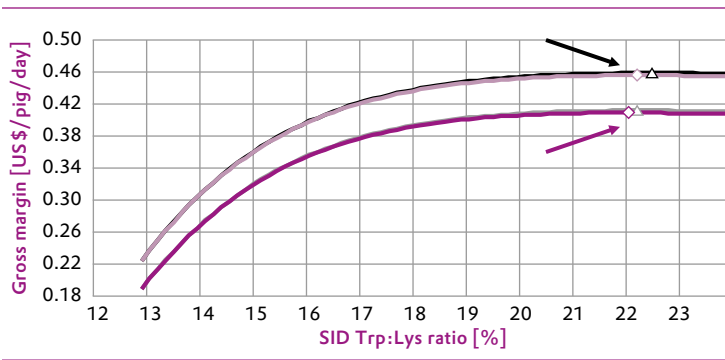
Figure 4



- △ Kendall et al., 2007a
- ◆ Eder et al., 2003
- ▲ Kendall et al., 2007b
- Guzki et al., 2005d
- ▲ Kendall et al., 2007c

Relative response in average daily gain (ADG) and feed conversion ratio (FCR) of finishing pigs (80–125 kg BW) to graded dietary SID Trp:Lys ratios

Figure 5



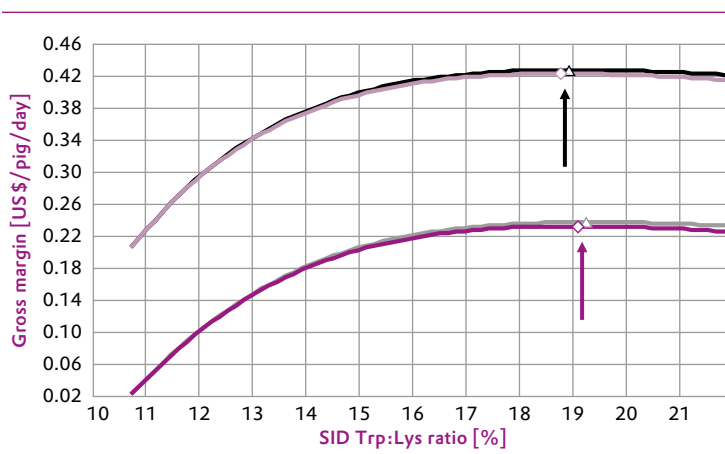
- ▲ Optimum SID Trp:Lys = 22.5% (price of L-Tryptophan: 30 US\$/kg)
- ◇ Optimum SID Trp:Lys = 22.2% (price of L-Tryptophan: 33 US\$/kg)
- ▲ Cost of basal diet without L-Tryptophan: 0.35 US\$/kg
- ◇ Optimum SID Trp:Lys = 22.2% (price of L-Tryptophan: 30 US\$/kg)
- ◇ Optimum SID Trp:Lys = 22.1% (price of L-Tryptophan: 33 US\$/kg)
- ◇ Cost of basal diet without L-Tryptophan: 0.35 US\$/kg

Effect of varying L-Tryptophan and feed prices on economic optimum SID Trp : Lys ratio in diets for starting pigs (7–25 kg BW) based on gross margin

The effects of varying L-Tryptophan and feed prices on economic return of starting and finishing pigs are illustrated in Figure 5 and 6, respectively. Based on the gross margin response curve and a basal feed price of 0.30 US\$/kg, the economic optimum SID Trp:Lys ratios were 22.5 and 22.2% for L-Tryptophan prices of 30 and 33 US\$/kg, respectively for starting pigs. When the cost of basal feed price was at 0.35 US\$/kg, the economic optimum SID Trp:Lys ratios changed to 22.2 and 22.1% based on L-Tryptophan prices of 30 and 33 US\$/kg, respectively (Figure 5).

For finishing pigs, the economic optimum SID Trp:Lys ratio were 19.0 and 18.9% based on L-Tryptophan prices of 30 and 33 US\$/kg, respectively based on the gross margin response curve and a basal feed price of 0.24 US\$/kg. When the cost of basal feed price was increased to 0.30 US\$/kg, the economic optimum SID Trp:Lys ratios shifted to 19.3 and 19.2% based on L-Tryptophan prices of 30 and 33 US\$/kg, respectively (Figure 6).

Figure 6



- ▲ Optimum SID Trp:Lys = 19.0% (price of L-Tryptophan: 30 US\$/kg)
- ◇ Optimum SID Trp:Lys = 18.9% (price of L-Tryptophan: 33 US\$/kg)
- ▲ Cost of basal diet without L-Tryptophan: 0.24 US\$/kg
- ◇ Optimum SID Trp:Lys = 19.3% (price of L-Tryptophan: 30 US\$/kg)
- ◇ Optimum SID Trp:Lys = 19.2% (price of L-Tryptophan: 33 US\$/kg)
- ◇ Cost of basal diet without L-Tryptophan: 0.30 US\$/kg

Effect of varying L-Tryptophan and feed prices on economic optimum SID Trp : Lys ratio in diets for finishing pigs (80–125 kg BW) based on gross margin

An increase in feed price slightly elevated the economic optimum SID Trp:Lys ratio while price changes for supplemented L-Tryptophan (10% increase) had only little effect on the economic optimum Trp:Lys ratio in both starting and finishing diets. Overall, the economic optimum SID Trp:Lys ratio to maximize the gross margin was at least 22% in starter diets which agrees well with the optimum SID Trp:Lys ratio for maximum ADFI and ADG which was also estimated at 22%. The optimum SID Trp:Lys ratio in finisher diets to maximize gross margin was determined to be 19% which is an intermediate value compared with the optimum Trp ratios to maximize the ADG (18%) and minimize the FCR (20%).



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Revised amino acid recommendations

by Evonik

An updated set of Evonik's nutritional recommendations has been published at the "www.aminoacidsandmore.com" for poultry species including broilers, laying hens, turkeys as well as Pekin ducks and swine. Compared to the previous version some changes are introduced with respect to the presentation of the recommendation.

- The amino acid recommendations are given on the basis of standardized ileal digestible (SID) and not on total amino acids. An exception is Pekin ducks due to a lack of data. It should be noted that SID amino acids for laying hens and turkeys refer to digestibility coefficients determined in broilers (Lemme *et al.*, 2004) as specific information on amino acid digestibility of raw materials in laying hens and turkeys is scarce.
- The set of amino acids comprise lysine, methionine, methionine+cystine, threonine, tryptophan, arginine, isoleucine, leucine, valine and histidine for each species. For broilers, laying hens and swine also figures for SID phenylalanine+tyrosine are given. For Pekin ducks data on isoleucine and leucine is insufficient and not included.
- Recommendations for dietary protein levels have been omitted. Diets should be formulated on basis of SID amino acids considering the whole set of essential amino acids which finally will set the protein level.

For broilers recommendations are given for males and females separately because requirements of females differ from those of males (Table 1). There are of course other factors such as phase length, dietary energy level or pellet quality influencing the optimal dietary amino acid levels. The impact of the mentioned factors can be simulated with QuickChick which is a software providing levels of SID amino acids for broilers in a more flexible way (Lemme, 2006). The amino acid recommendation are based on the concept of ideal protein and, compared to the previous recommendations, the SID methionine to SID lysine ratio has been adjusted so that the methionine to cystine ratio is kept constant across all feeding phases (55:45).

Recommendations for laying hens have recently completely been revised (Lemme, 2009 a). For this revision a meta-analysis on

methionine dose-response studies published during the last 20 years has been performed in order to update the recommendation for the optimal daily SID methionine intake. Accordingly, the optimal intake increased by 25 mg/d to 415 mg SID methionine per day. In a second step available studies on ideal protein in laying hens have been compiled and the ideal amino acid profile for laying hens has been revised. Apart from the fact that now more amino acid are included in the recommendation for laying hens major difference to the previous set of recommendations is an increase of the SID threonine to SID lysine ratio from 62% to 70% although literature would suggest an even higher ratio. Combination of both the updated optimal SID methionine intake and the updated ideal amino acid profiles allowed for giving recommendation for 11 essential amino acids as given in Table 2 + 3.

Also the amino acid recommendation for turkeys has recently been revised (Lemme, 2009b). Whereas the previous amino acid recommendations just gave numbers for turkeys according to phase feeding program with 4-week periods throughout whole production cycle, the revised recommendations as shown in Table 4 distinguish between male and female turkey on the one hand and between heavy and medium heavy turkey production on the other hand. In contrast to the broiler recommendations adjustment to gender is not carried out by adjustment of amino acid levels but by adjusting the phase feeding program. While phases 1 to 4 (until day 91) are similar for both sexes phase 5 is shortened by two weeks and phase 6 feed is thus introduced earlier (day 106 female, day 120 male). In some markets turkeys are grown for 20 or 22 weeks (heavy turkeys) while in other markets birds are slaughtered earlier (medium heavy). However, research suggested that reduction of the dietary amino acid level during phase 1 and 2 (until day 35) improves overall production as this strategy reduces late mortality. However, as late mortality is not such an issue in medium heavy turkey production and compensation of the growth depression which is a consequence of reduced amino acid supply can not be ensured, this amino acid reduction in phases 1 and 2 is not recommended for medium heavy turkeys.

Table 1

Days	Metabol. Energy (MJ/kg)	(kcal/kg)	Lys	Met	Met+ Cys	Thr	Trp	Arg	Ile	Leu	Val	His	Phe+ Tyr
Male Broilers – Recommendations for Standardised Ileal Digestible Amino Acids (% of diet)													
1–12*	12.70	3030	1.27	0.50	0.92	0.80	0.20	1.30	0.86	1.36	1.00	0.42	1.47
13–22	12.90	3080	1.09	0.44	0.81	0.70	0.18	1.13	0.75	1.17	0.87	0.36	1.26
23–35	13.00	3100	1.00	0.42	0.76	0.65	0.16	1.05	0.71	1.07	0.80	0.33	1.16
36–48	13.20	3150	0.95	0.40	0.74	0.63	0.16	1.01	0.68	1.02	0.77	0.31	1.10
>49	13.40	3200	0.89	0.39	0.70	0.60	0.15	0.96	0.65	0.96	0.73	0.29	1.03
Female Broilers – Recommendations for Standardised Ileal Digestible Amino Acids (% of diet)													
1–12*	12.70	3030	1.25	0.50	0.91	0.79	0.20	1.28	0.85	1.34	0.99	0.41	1.45
13–22	12.90	3080	1.04	0.42	0.77	0.67	0.17	1.08	0.72	1.11	0.83	0.34	1.21
23–35	13.00	3100	0.93	0.39	0.70	0.61	0.15	0.98	0.66	1.00	0.75	0.31	1.08
36–48	13.20	3150	0.83	0.35	0.64	0.55	0.14	0.88	0.59	0.88	0.67	0.27	0.96
>49	13.40	3200	0.73	0.31	0.57	0.49	0.12	0.78	0.53	0.78	0.59	0.24	0.84

* equals a cumulated feed intake about 350 – 400g/broiler.

Recommendations for Broilers

Table 2

	Lys	Met	Met+ Cys	Thr	Trp	Arg	Ile	Leu	Val	His	Phe+ Tyr
Laying Hens – Recommendations for Standardised Ileal Digestible Amino Acids (daily amino acid intake/mg)											
Amino acid intake (mg/day)	831	415	756	582	174	864	665	997	731	249	997

Recommendations for Laying Hens

Table 3

Feed intake (g/day)	Metabol. Energy (MJ/kg)	(kcal/kg)	Lys	Met	Met+ Cys	Thr	Trp	Arg	Ile	Leu	Val	His	Phe+ Tyr
Laying Hens – Recommendations for Standardised Ileal Digestible Amino Acids (% of diet)													
80	11.82	2820	1.04	0.52	0.95	0.73	0.22	1.08	0.83	1.25	0.91	0.31	1.25
85	11.82	2820	0.98	0.49	0.89	0.68	0.21	1.02	0.78	1.17	0.86	0.29	1.17
90	11.82	2820	0.92	0.46	0.84	0.65	0.19	0.96	0.74	1.11	0.81	0.28	1.11
95	11.82	2820	0.87	0.44	0.80	0.61	0.18	0.91	0.70	1.05	0.77	0.26	1.05
100	11.82	2820	0.83	0.42	0.76	0.58	0.17	0.86	0.66	1.00	0.73	0.25	1.00
105	11.82	2820	0.79	0.40	0.72	0.55	0.17	0.82	0.63	0.95	0.70	0.24	0.95
110	11.82	2820	0.76	0.38	0.69	0.53	0.16	0.79	0.60	0.91	0.66	0.23	0.91
115	11.82	2820	0.72	0.36	0.66	0.51	0.15	0.75	0.58	0.87	0.64	0.22	0.87
120	11.82	2820	0.69	0.35	0.63	0.48	0.15	0.72	0.55	0.83	0.61	0.21	0.83

Recommendations for Laying Hens

Table 4

Sex	Weeks	Metabol. Energy (MJ/kg)	(kcal/kg)	Lys	Met	Met + Cys	Thr	Trp	Arg	Ile	Leu	Val	His
Heavy Turkeys – Recommendations for Standardised Ileal Digestible Amino Acids (% of diet)													
male	1–2	11.50	2740	1.55	0.58	0.98	0.87	0.25	1.63	0.95	1.70	1.05	0.54
female	1–2												
male	3–5	11.70	2790	1.41	0.53	0.90	0.79	0.23	1.48	0.86	1.55	0.95	0.49
female	3–5												
male	6–9	12.10	2890	1.31	0.50	0.84	0.74	0.21	1.37	0.80	1.44	0.88	0.46
female	6–9												
male	10–13	12.50	2980	1.14	0.44	0.74	0.65	0.19	1.20	0.70	1.25	0.77	0.40
female	10–13												
male	14–17	12.80	3050	1.01	0.40	0.67	0.58	0.17	1.06	0.62	1.11	0.68	0.35
female	14–15												
male	18–22	13.20	3150	0.91	0.36	0.61	0.53	0.15	0.96	0.56	1.00	0.61	0.32
female	16–20												
Medium Heavy Turkeys – Recommendations for Standardised Ileal Digestible Amino Acids (% of diet)													
male	1–2	11.50	2740	1.63	0.61	1.03	0.91	0.27	1.72	1.00	1.79	1.10	0.57
female	1–2												
male	3–5	11.70	2790	1.49	0.56	0.95	0.83	0.24	1.56	0.91	1.63	1.00	0.52
female	3–5												
male	6–9	12.10	2890	1.31	0.50	0.84	0.74	0.21	1.37	0.80	1.44	0.88	0.46
female	6–9												
male	10–13	12.50	2980	1.14	0.44	0.74	0.65	0.19	1.20	0.70	1.25	0.77	0.40
female	10–13												
male	14–17	12.80	3050	1.01	0.40	0.67	0.58	0.17	1.06	0.62	1.11	0.68	0.35
female	14–15												

Recommendations for Turkeys

Table 5

Days	Metabol. Energy (MJ/kg)	(kcal/kg)	Lys	Met	Met + Cys	Thr	Trp	Arg	His	Val
Pekin Ducks – Recommendations for Total Amino Acids (% of diet)										
1–21	12.20	2940	1.16	0.42	0.76	0.84	0.21	0.94	0.42	0.77
22–49	12.60	3000	0.90	0.42	0.77	0.66	0.20	0.76	0.32	0.59

Recommendations for Peking Ducks

Amino acid recommendation for Pekin ducks (Table 5) are unchanged but just extended by including methionine (previously only methionine+cystine), histidine and valine. No results on basis of SID amino acids were available but, however, some more details can be obtained by an AMINONews® contribution by Dr. Hou (2007).

The amino acid recommendations for swine (Table 6) have been updated by reviewing the optimum amino acid ratio data from both Evonik trial results and available published literature. Some changes have been made on tryptophan recommendations for piglets and growing-finishing pigs, and methionine + cystine, threonine and tryptophan recommendations for sows.

Table 6

Body weight (kg)	Net Energy** (MJ/kg)	(kcal/kg)	Lys	Met	Met+ Cys	Thr	Trp	Arg	Ile	Leu	Val	His	Phe+ Tyr	Lys (g/MJ NE)	Lys (g/Mcal NE)
Growing Swine – Recommendations for Standardised Ileal Digestible Amino Acids (% of diet)*															
<10	10.70	2560	1.35	0.45	0.81	0.85	0.30	0.57	0.81	1.35	0.92	0.43	1.28	1.26	5.30
10–20	10.40	2480	1.18	0.39	0.71	0.74	0.26	0.50	0.71	1.18	0.80	0.38	1.12	1.13	4.80
20–30	10.20	2440	0.98	0.34	0.61	0.64	0.20	0.39	0.59	0.98	0.67	0.31	0.94	0.96	4.00
30–40	10.00	2390	0.90	0.31	0.56	0.59	0.18	0.36	0.54	0.90	0.61	0.29	0.86	0.90	3.80
40–70	9.80	2340	0.81	0.28	0.51	0.54	0.16	0.29	0.49	0.81	0.55	0.26	0.77	0.83	3.50
70–105	9.60	2290	0.71	0.25	0.46	0.50	0.14	0.23	0.43	0.71	0.48	0.23	0.68	0.74	3.10
Reproductive Swine – Recommendations for Standardised Ileal Digestible Amino Acids (% of diet)*															
Sows Gestation	8.90	2130	0.59	0.21	0.39	0.41	0.13	0.53	0.35	0.57	0.40	0.21	0.59	0.66	2.80
Sows Lactating	9.80	2340	0.85	0.28	0.51	0.58	0.19	0.48	0.50	0.97	0.66	0.34	0.96	0.87	3.60

* high lean growth potential
 ** to convert data use: ME=NE/0.74
 DE=NE/0.71

Recommendations for Swine

The current Evonik recommendations for isoleucine and valine take the possible interactions of isoleucine with the other branched-chain amino acids into account, which can occur when the dietary levels of leucine and valine are higher than those recommended by the ideal protein ratios.

The current tryptophan recommendations (SID basis) for pigs with body weight ranges of <10 kg, 10 – 19 kg, 20 – 30 kg, 30 – 40 kg, 40 – 70 kg, 70 – 105 kg are 0.30, 0.26, 0.20, 0.18, 0.16 and 0.14% (a slight increase from 0.24, 0.21, 0.19, 0.17, 0.15 and 0.13% from the last publication), respectively (Htoo, 2009). The recommended dietary levels of methionine, methionine + cystine, threonine and tryptophan for gestating sows are 0.21, 0.39, 0.41 and 0.13% (a slight increase from 0.19, 0.34, 0.37 and 0.11% from the last publication), respectively. The current methionine, methionine + cystine and tryptophan recommendations for lactating sows are 0.28, 0.51 and 0.19% (a slight change from 0.29, 0.54 and 0.16% from the last publication), respectively. The recommendations for lysine and net energy levels remain unchanged.



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Ducks

Dynamics of yolk sac content absorption and intestine development in ducklings fed mixtures with increasing dietary methionine level

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The experiment was carried out with ducklings fed-control feed mixture (Met level 0.3%) and mixtures supplemented with dl-methionine at the dose of 0.03 (II); 0.07 (III) 0.12 (IV) and 0.18% (V). In 5 h post-hatch (12 birds) and on days 3, 4, 6 always 18 birds were taken out from each treatment and killed, then the yolk sac (YS) and intestine were removed. The weight and chemical composition of YS residues (in them also amino acids) were analysed.

Moreover on days 1, 3, 4 and 6, the amino acid pattern of YS residual protein was elaborated as well as length and weight of intestine were recorded. Increased doses of supplemental Met improved body weight gain after 4 days of duckling life. All determined parameters were not affected by treatment. Development of intestines, yolk sac weight and yolk sac composition (DM, fat, protein, amino acid composition) changed with age; however, without any clear effect from dietary methionine level. No differences between sex and analysed parameters were stated.

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Broilers

Effect of inclusion of cellulose in the diet on the inevitable endogenous amino acid losses in the ileum of broiler chicken

H. Kluth and M. Rodehutsord

The objective of this study was to determine the inevitable endogenous amino acid (AA) loss at the terminal ileum of broilers that were fed diets with 2 different fiber levels using a regression approach. The design of the study was a randomized complete block employing a factorial arrangement of treatments with 3 CP levels (50, 90, and 130 g/kg) and 2 fiber levels. The fiber level was adjusted by inclusion of cellulose at the expense of cornstarch. The AA pattern of the CP was the same in all diets. Titanium dioxide was used as indigestible marker. Six cages of 8 birds were allocated to each diet. The experimental diets were offered for *ad libitum* consumption for 3 d, starting on 21 d of age. Digesta were sampled on a cage basis from the distal two-thirds of the intestine section between Meckel's diverticulum and 2 cm anterior to the

ileo-ceca-colonic junction. Inevitable endogenous CP and AA losses were determined by extrapolating the linear regressions between intake and prececal flow toward zero intakes. The inevitable losses of CP and AA, expressed in relation to DM intake, were significantly increased by increased cellulose inclusion in the diet. Amino acids with the greatest loss were Glu, Asp, and Thr, whereas Met was the AA with the lowest loss. The ranking of the concentrations of AA of inevitable CP loss was very similar between the 2 fiber levels. This ranking also was similar in comparison to published values for the endogenous AA losses in broilers. It was concluded that the fiber level in the diet can affect the amount of AA inevitably lost at the terminal ileum and that all AA are affected to a similar extent. The results suggest that there is no effect of enhanced fiber level in the diet on AA composition of prececal endogenous CP loss in broilers. These findings can be considered in modeling the AA requirements of broilers.

Poultry Science; 2009; 88: 1199–1205





Sulfur amino acid deficiency upregulates intestinal methionine cycle activity and suppresses epithelial growth in neonatal pigs

C. Bauchart-Thevret, B. Stoll, S. Chacko and D. G. Burrin

We recently showed that the developing gut is a significant site of methionine transmethylation to homocysteine and transsulfuration to cysteine. We hypothesized that sulfur amino acid (SAA) deficiency would preferentially reduce mucosal growth and antioxidant function in neonatal pigs. Neonatal pigs were enterally fed a control or an SAA-free diet for 7 days, and then whole body methionine and cysteine kinetics were measured using an intravenous infusion of [$1-^{13}\text{C}$;methyl- $^2\text{H}_3$]methionine and [^{15}N]cysteine. Body weight gain and plasma methionine, cysteine, homocysteine, and taurine and total erythrocyte glutathione concentrations were markedly decreased (-46% to -85%) in SAA-free compared with control pigs. Whole body methionine and cysteine fluxes were reduced, yet methionine utilization for protein synthesis and methionine remethylation were relatively preserved at

the expense of methionine transsulfuration, in response to SAA deficiency. Intestinal tissue concentrations of methionine and cysteine were markedly reduced and hepatic levels were maintained in SAA-free compared with control pigs. SAA deficiency increased the activity of methionine metabolic enzymes, i.e., methionine adenosyltransferase, methionine synthase, and cystathionine-synthase, and S-adenosylmethionine concentration in the jejunum, whereas methionine synthase activity increased and S-adenosylmethionine level decreased in the liver. Small intestine weight and protein and DNA mass were lower, whereas liver weight and DNA mass were unchanged, in SAA-free compared with control pigs. Dietary SAA deficiency induced small intestinal villus atrophy, lower goblet cell numbers, and Ki-67-positive proliferative crypt cells in association with lower tissue glutathione, especially in the jejunum. We conclude that SAA deficiency upregulates intestinal methionine cycle activity and suppresses epithelial growth in neonatal pigs.

American Journal of Physiology – Endocrinology and Metabolism; 2009; 296: E1239–E1250

Estimation of the optimum ratio of standardized ileal digestible isoleucine to lysine for eight- to twenty-five-kilogram pigs in diets containing spray-dried blood cells or corn gluten feed as a protein source

M. K. Wiltafsky, J. Bartelt, C. Relandeau and F. X. Roth

Two growth assays and 1 nitrogen (N) balance trial were conducted to determine the standardized ileal digestible (SID) Ile:Lys ratio in 8- to 25-kg pigs using either spray dried blood cells or corn gluten feed as a protein source. In Exp. 1, 48 individually penned pigs (initial BW = 7.7 kg) were used in a 6-point SID Ile titration study (analyzed SID Ile of 0.36, 0.43, 0.50, 0.57, 0.64, and 0.72%) by addition of graded levels of L-Ile. The basal diet contained 1.00% SID Lys, 18.4% CP, and 13.6 MJ ME/kg. Diets were based on wheat, barley, corn, and 7.5% spray-dried blood cells as a protein source. Dietary SID Leu and Val levels were 1.61 and 1.02%, respectively. For the 35-d period, ADG, ADFI, and G:F increased linearly ($P < 0.01$) and quadratically ($P < 0.04$) with increasing SID Ile:Lys. Estimates of optimal SID Ile:Lys ratios were 59% for ADG and ADFI. In Exp. 2, 24 N balances were conducted using the Exp. 1 diets (12 pigs; individually penned; average BW = 11.5 kg). Pigs were fed 3 times daily with an amount equal to 1.0 MJ ME/kg BW 0.75. Prep-

aration and collection periods (7 d each) were repeated after rearranging the animals to treatments. Increasing the dietary SID Ile:Lys ratio increased N retention linearly ($P < 0.01$), and N utilization linearly ($P < 0.01$) and quadratically ($P < 0.01$). An optimal SID Ile:Lys ratio of 54% was estimated for N retention. In Exp. 3, 48 individually-penned pigs (initial BW = 8.0 kg) were fed grain-based diets in a 6-point SID Ile titration (analyzed SID Ile of 0.35, 0.41, 0.49, 0.56, 0.62, and 0.69%). Dietary SID Ile was elevated by graded addition of L-Ile. The basal diet contained 0.97% SID Lys, 16.8% CP, and 13.6 MJ ME/kg. In contrast to Exp. 1 and 2, spray-dried blood cells were excluded and corn gluten feed was used as a protein source. Dietary SID Leu and Val were set to 1.05 and 0.66%. For the 42-d period, ADG, ADFI, and G:F increased linearly ($P < 0.01$) and quadratically ($P < 0.01$) with increasing SID Ile:Lys. Estimated optimal SID Ile:Lys ratios were 54, 54, and 49 for ADG, ADFI, and G:F, respectively. These experiments suggest that the optimal SID Ile:Lys ratio depends on diet composition. In Exp. 1, AA imbalances because of high Leu contents may have led to increased Ile nutritional needs. For ADG and ADFI, an optimum SID Ile:Lys ratio of 54% was estimated for 8- to 25-kg pigs in diets without Leu excess.

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Ideal amino acid balance for sows during gestation and lactation

S. W. Kim, W. L. Hurley, G. Wu and F. Ji

Improving efficiency of protein utilization is important for pregnant sows under restricted feed allowance and for lactating sows with limited feed intake. Sows have limited ability to support the growth of fetuses and mammary glands during late gestation and to support mammary growth and milk production, especially during first lactation period. A series of studies was conducted to characterize requirements and ideal ratios of AA for 1) fetal growth, 2) mammary gland growth of gestating sows, 3) maternal tissue gain of gestating sows, 4) mammary gland growth of lactating sows, and 5) maternal tissue gain of lactating sows. A total of 97 pregnant sows and their fetuses and a total of 174 lactating sows and their nursing piglets were used for these studies to collect fetal tissues, mammary tissues, and maternal tissues for AA analysis. Requirements and ideal ratios of AA for

sows changed dynamically depending on stages of pregnancy. Suggested daily requirements for true ileal digestible Lys were 5.57 and 8.78 g, and relative ideal ratios for Lys:Thr:Val:Leu (on basis of AA weight) were 100:79:65:88 and 100:71:66:95 for d 0 to 60 and d 60 to 114 of gestation, respectively. Requirements and ideal ratios of AA for lactating sows changed dynamically depending on potential amounts of protein mobilization from maternal tissues, which are related to voluntary feed intake and milk production. Suggested ideal ratios for Lys:Thr:Val:Leu were 100:59:77:115 and 100:69:78:123 if BW losses of sows during 21 d of lactation are 0 and 33 to 45 kg, respectively. To optimize efficiency of dietary protein utilization by sows, the dietary AA content and ratios can be adjusted by stages of pregnancy (i. e., phase feeding) and by expected feed intakes or parities of sows during lactation (i. e., parity-split feeding) considering the dynamic changes in the requirements and ideal ratios of AA.

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