Feed Formulation in Broiler Chickens Based on Standardized Ileal Amino Acid Digestibility

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Summary

Analysis of ileal contents rather than excreta is a more reliable method for assessing amino acid digestibility in poultry. Debate will continue among nutritionists about the relative merits of apparent and true digestible amino acid systems. However, there is no doubt that all digestible amino systems are superior to the use of total amino acids in feed formulations. Present methods of evaluating amino acid availability have specific applications and shortcomings. Standardized ileal digestibility is discussed as the concept of choice and some areas for future research are highlighted. Performance data in growing broiler chicks demonstrate the advantages of feed formulation based on standardized ileal digestibility of amino acids in feed ingredients.

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Introduction

It is known that a proportion of dietary amino acids is excreted undigested and that individual raw materials differ widely in this respect. Thus, the higher the inclusion levels of raw materials with low amino acid digestibility in diets formulated on the basis of total amino acids, the less reliable will be the prediction of performance (Esteve-Garcia *et al.*, 1993; Fernandez *et al.*, 1995; Pertillä *et al.*, 2001a). In this situation, costly safety margins are usually applied to avoid potential reductions in performance.

Knowledge of digestibility coefficients (DC) for individual amino acids in raw materials and the requirement of digestible amino acids for a defined production target (such as maximising growth, breast meat yield and/or profitability, or minimising feed conversion ratio and/or feed costs per kg gain or breast meat) therefore enables formulation of diets closer to the requirements of the animals. Diets based on digestible amino acids may encourage the use of alternative protein sources, because such formulations will improve the precision of least cost diets and reduce nitrogen output from poultry operations. Finally, diet formulations on a digestible amino acid basis may also offer economic benefits (Rostagno *et al.*, 1995).

A large volume of published data on the amino acid digestibility of raw materials for poultry is available, but there is considerable confusion in the terminology used due to differences in the methodology employed for determining the DC (Ravindran and Bryden, 1999). The different methodological approaches of assessing amino acid digestibility are briefly reviewed, with emphasis on ileal digestibility. The need for correcting digestibility estimates for endogenous amino acid losses will be highlighted, followed by a discussion on the measurement of inevitable losses. Finally, the concept of 'standardizing' ileal digestibility values by correcting for basal endogenous amino acid recoveries will be introduced, along with a table of "Standardized Ileal Amino Acid Digestibility for Broilers" for a range of raw materials.

What is Digestibility?

Digestibility can be defined as the fraction of a certain nutrient ingested with the diet that is absorbed by the bird, i.e. not excreted in the faeces. Digestibility can therefore be calculated by measuring dietary amino acid input (AA_{diet}) and excreta amino acid output ($AA_{excreta}$) as follows:

$$\text{Digestibility (\%)} = ((AA_{diet} - AA_{excreta})/AA_{diet}) \times 100$$

The assay of digestibility has become the most favoured technique for estimating amino acid availability. Digestibility assays may be separated into two main categories: excreta and ileal digestibility. Excreta digestibility involves the collection of excreta from intact or caecectomized birds. For measurement of ileal digestibility, the digesta are collected from the distal part of the ileum. The latter method is technically more complex, but it eliminates some confounding factors.

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Limitations of Excreta Digestibility Determined by Precision Feeding Assay

Determination of excreta digestibility was the most commonly used method during the early days of digestibility research. In particular, large volume of published data on excreta amino acid digestibility was generated using the precision feeding assay developed by Sibbald (1979) that gained wide acceptance during the 80's and 90's in North America and Europe. In this assay, adult cockerels are fasted for 24-48 hrs and force-fed a defined amount of the feedstuff under test by placing it directly into the crop. The excreta are then collected for a sufficiently long period on the assumption that all undigested components have been excreted. A major advantage of this assay is that many raw materials can be tested in a relatively short time with few birds because the cockerels can be used several times. Excreta digestibility measurements from the precision feeding assay, however, suffer from several drawbacks.

- First, the excreta contain not only amino acids from the faeces but also those excreted with the urine. It would therefore be more accurate to refer to this measurement as 'metabolizability' rather than digestibility. Although some sources point out that renal AA excretion is negligible (O'Dell *et al.,* 1960; Bragg *et al.,* 1969; Terpstra, 1978), there is some evidence suggesting that this is not always the case (McNab, 1995).
- Second, the excreta-based measurements ignore the effects of hindgut micro-organisms on protein digestion or protein utilization and the contribution of microbial proteins to the AA profile and concentrations in the faeces. This source of error can be largely overcome by caecectomizing the cockerels (Parsons, 1986).
- The precision feeding assay is also criticised to impair animal welfare because force-feeding and fasting of birds do not represent "normal" feeding behaviour.
- The force-fed feed consists entirely of the test ingredient and this may exert a significant influence on the digestive processes.
- Stimulation or rather non-stimulation of the secretion of certain digestive enzymes and the deficiency situation induced by the fasting are other relevant concerns.
- Perhaps the major concern for the broiler industry is the application of digestibility estimates generated in adult cockerels, which are physiologically mature, to growing birds. Published data on the effects of age on AA digestibility are limited and contradictory (ten Doesschate *et al.*, 1993; Perttilä *et al.*, 2001a; Huang *et al.*, 2000), but the results clearly suggest that the apparent ileal AA digestibilities of feeds for adult cockerels and growing poultry are different.

Ileal Digestibility as an Alternative in Growing Poultry

The criticisms against the use of the precision feeding assay as listed above can be overcome by the alternative methodology of estimating ileal or precaecal digestibility (Ravindran and Bryden, 1999). In this method, digesta are recovered from the distal part of the ileum and analysed. As a result, urine AA as a source of error and the modifying effects of hindgut fermentation are eliminated. Somewhat inconsistent differences between excreta and ileal DC have been reported in literature (ten Doesschate *et al.*, 1993; Karakas *et al.*, 2001; Perttilä *et al.*, 2001b; Ravindran *et al.*, 1999) making a systematic investigation of raw materials necessary.

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The ileal digesta can be removed either through an intestinal cannula or by the slaughter method. With the former method, the bird can be used for several tests, but obtaining sufficient amounts of digesta is laborious and the insertion of the fistula requires considerable surgical skill. Moreover, this methodology can be employed only in mature birds. With the slaughter method, the animals are killed humanely, the small intestine is immediately surgically exposed and the ileum separated. Ileum is defined as that portion of the small intestine extending from Meckel's diverticulum to a point a few cm proximal to the ileo-cecal junction. The digesta are then gently recovered from the lower half of the ileum by flushing with distilled water. Digesta from several birds often have to be pooled to obtain sufficient quantities for analysis. At least four replicates per ingredient should be run, which ultimately involves the use of considerable numbers of animals. In order to quantify feed intake indigestible markers which do not affect nutrient digestibility and which have a high recovery rate (of almost 100 %) have to be added to the test diet. The calculation of the DC then includes the marker concentrations in the diet (I_{diet}), and excreta ($I_{excreta}$):

$$DC\ (\%) = 100 - ((I_{diet} \times AA_{excreta}) / (I_{excreta} \times AA_{diet}) \times 100).$$

The use of markers eliminates the need to measure feed intake and the feed can be offered *ad libitum*. With *ad libitum* feeding - depending on the raw material to be tested - a suitable test diet has to be formulated because the test substance on its own could lead to considerable partial deficiency symptoms, imbalances or feed refusal. On the other hand, it stimulates 'normal' digestive processes through diet composition. This has a direct effect on endogenous excretions, which consist of digestive enzymes, mucoproteins (mucin) and desquamated cells, which will be discussed later. The raw material under test usually serves as the sole amino acid source. The diet is fortified with minerals and vitamins. Energy is added in form of carbohydrates (purified starch, dextrose) and fats. Calculation of the DC assumes that the AA digestibility of the diet is representative of that of the raw material. This assumption, however, may create a small error because the diet itself stimulates some endogenous AA excretions. This error increases with a decreasing AA content in the raw material and hence in the diet.

The Need to Correct for Endogenous AA Losses

Another relevant debate is about the fact that not all amino acids found in excreta or digesta originate from the diet, but that some are of endogenous origin. This raises two fundamental questions: whether or not to correct for endogenous AA contribution and, if yes, how those losses should be determined and what estimates should be used in the correction. The excreta and ileal assays described above for estimating digestibility determine 'apparent' values and do not account for endogenous AA secretions, which can have a variable effect on the calculated DC. As shown in Figure 1, this effect is most pronounced when protein/ amino acid intake is low.

The relative significance of endogenous AA losses vs. AA levels in the excreta or digesta decreases as the dietary amino acid intake increases. Hence, DCs of raw materials with low AA levels, such as cereals and grain legumes, are most affected. The impact of this effect can be demonstrated by data obtained from Table 2 (standardized figures) and their respective apparent DCs. The apparent DC values were transformed into values of 'standardised' DC by correction

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for 'basal' endogenous AA recoveries. The difference between apparent DC (89 %) and the standardised DC (90 %) is about 1 percentage point for lysine in soybean meal (47 % CP). In the case of wheat (13 % CP), however, the difference amounts to 7 percentage points (79 vs. 86 %).

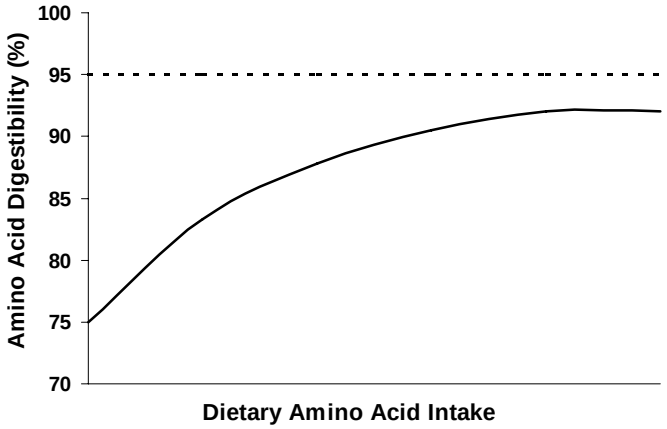

Figure 1. Relationship Between Dietary AA Intake and Apparent (continuous line) or Standardised (dotted line).

In addition to being influenced by dietary AA intakes, apparent digestibility values of individual ingredients may also not be additive when combined in diet formulations. In particular, there may be associative effects when high levels of poorly digestible ingredients are used. The limitations of apparent ileal digestibility values can be overcome by standardising these estimates through corrections for basal endogenous losses. The challenge, however, is to agree on what constitutes basal endogenous losses and how these should be determined.

Measurement of Endogenous AA Losses

Endogenous AA losses at the ileal level can be divided into a basal (or non-specific) and a specific fraction. The basal losses are related to the dry matter intake and are independent of the raw material or diet composition (Butts *et al.*, 1993). In contrast, the specific losses are influenced by the inherent characteristics of the raw material, such as the presence of anti-nutritional factors that may stimulate endogenous secretions. The methodological approaches to measure endogenous AA losses in poultry have been reviewed by Ravindran and Bryden (1999). In theory, data from classic methods of measuring endogenous losses (e.g. protein-free diet, feeding of highly digestible proteins such as casein, wheat gluten or the regression method) may be considered representative of 'basal' losses occurring irrespective of raw material or dietary composition. 'Raw material-specific' losses are not measured by the classic methods.

A method where specific losses are automatically included in the DC determination is the 'regression method', which involves feeding diets containing increasing levels of the test substance. In the regression of (apparent) AA absorption (in g) in relation to AA intake (in g), the slope of the regression line corresponds to a digestibility including specific losses. By definition the slope of the line is determined both by the amount of non-digested AA and specific

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losses, but not by basal losses. The latter can be estimated by extrapolation of the regression line to an AA intake of zero. This extrapolation is subject to large estimation errors, however, especially in case of a wide gap between the lowest data point and a (theoretical) zero intake (Ravindran and Bryden, 1999). A much greater drawback of this method is the technical complexity involved in estimating just one DC, especially when it comes to determining ileal DC. This is probably why the regression method has not yet gained wide acceptance, although it has been employed to determine ileal DC in some raw materials (Short *et. al.*, 1999, Rodehutscord *et al.*, 2004).

Data from several other methods to determine endogenous AA losses are available. The methods include the feeding of protein-free diets, feeding diets with highly digestible casein or feeding wheat gluten, and fasting the birds. The fasting method, where the animals are fasted for extended periods and excretions are then assumed to be of endogenous origin is not appropriate for two reasons. Firstly, this method creates an abnormal physiological state in the animal, and secondly, losses cannot be set in relation to dry matter intake. Dry matter intake is recognised to be a major determinant of basal endogenous losses (Butts *et al.*, 1993).

The protein-free diet method is based on the assumption that the test diet enables normal digestive processes to take place and that the excreted AA are completely of endogenous origin. However, the method is criticised because secretion of proteolytic enzymes is not stimulated (Ravindran and Bryden, 1999). This inaccuracy led to the incorporation of highly digestible protein sources, such as wheat gluten or enzymatically hydrolyzed or guanidinated casein, into the test diets. It is assumed that these proteins are 100 % digestible and that excreted AA therefore represents the basal losses. The studies of Cremers *et al.* (2002), however, indicate the existence of a dose-response relationship. These findings may suggest either that the test substances are not 100% digestible, or that the basal losses are affected by the dietary protein content, or the proteins (or peptides) themselves give rise to specific losses of varying amounts. In the guanidinated casein method, lysine in the casein is converted to homoarginine. By analysing lysine and homoarginine levels in the digesta samples, the endogenous lysine losses are calculated. Conclusions are then drawn based on the behaviour of lysine and homoarginine in relation to other AA, which is a drawback of this method (Ravindran and Bryden, 1999). The feeding of enzyme hydrolyzed casein (EHC) should at least remove the uncertainty concerning the 100 % digestibility. As a result of enzyme pre-treatment, this casein product consists entirely of free AA or very small peptides (< 5,000 Da). Endogenous protein is assumed to be much larger (> 10,000 Da) and the separation of < 5000 Da fraction provides a measure of endogenous losses. Some studies have shown, however, that endogenous excretions may contain certain amounts of small-structured protein molecules and this may entail an error of estimation (Ravindran and Bryden, 1999).

As can be_seen in Table 1, there are marked differences between the various methods to determine endogenous losses both with regard to excretions and in the ranking of different AA. It is noteworthy that the within-method variation is also relatively large, which may be attributed to differences in experimental methodology. Variables such as the choice of marker, the age of the birds, the feeding regime, etc. can all influence the estimation of endogenous losses. Although each method suffers from some limitations and published data on endogenous losses at the ileal

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level in growing poultry are limited, averaged data from repeated experiments using the EHC-methods are considered as the best measure of basal losses since most of the drawbacks described for the other methods are overcome by this method. As shown in Table 1, the lowest ileal endogenous losses were reported for methionine and the highest for threonine, with losses for the remaining amino acids falling between these two extremes. This ranking is consistent with data reported with the other methods used to determine endogenous losses (Cremers, 2002). The relatively high threonine losses are also consistent with findings in swine (Jansman *et al.*, 2002). It is assumed that mucosal secretions, such as mucin contribute to the high threonine losses since the threonine content of mucin is relatively high (about 15%).

Compilation of Standardized Ileal Digestibility of Major Raw Materials for Broilers

The majority of apparent DC data used in this compilation is based on assays, analyses and calculations carried out by the University of Sydney and Degussa AG. Where available and deemed acceptable, literature data have also been included in the table. Standardized digestibility estimates for a total of 17 raw materials have thus been compiled (Table 2). In cases where the AA digestibility of the diet was assumed to be the AA digestibility of the raw material, the apparent DCs were first pre-corrected. This was done on the assumption that the non-protein carrier fraction of the test diet will account for a proportion of endogenous losses. Literature data where digestibility was determined by the difference method do not require this correction and were included directly. Weighted means were then calculated, with the number of observations per method serving as the basis for weighting. In the next step, apparent DC (aDC) was standardised for interactions between the AA content of raw materials (from AminoDat 2.0®) and endogenous losses (Table 1). The calculation of standardized values (sCD) was as follows:

sDC (%) = aDC (%) + ((basal endogenous AA losses, as g/kg DM intake) / (AA content of the raw material, as g/kg DM) x 100))

The sDC are independent of the method by which the aDC were originally estimated. Moreover, these values are additive when used in practical feed formulations. The levels of standardized ileal digestible AA in raw materials (Table 2) can be calculated using the sDC and the gross AA content.

Standardization Particularly Affects Amino Acid Digestibility Coefficients of Cereal Grains

In Table 2, the standardized ileal AA digestibility of several ingredients important in broiler nutrition are shown. The number of experiments or observations vary between raw materials, with digestibility coefficients ranging between 46 % and 95 %. In case of animal by-products, the digestibility coefficients for meat and bone meal are somewhat lower than those found for fish meal but higher than those found for feather meal. However, it is well established that the AA digestibility of animal by-products are dependent on processing conditions (primarily processing temperature) and raw material variability (causing differences in ash content).

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As expected, the strongest effect of the standardization was observed for grains. The differences between standardized and apparent ileal digestible AA ranged between 0 and 17 percentage points for grains while it ranged only between 0 and 7 percentage points for plant protein sources and animal by-products. Among AA threonine digestibility was most affected by the standardization, which is likely to be related to the high content of threonine in endogenous proteins (Table 1). For example, the standardized ileal threonine digestibility of corn, wheat, soybean and meat and bone meal was 17, 14, 3 and 3 percentage points higher, respectively, than the apparent digestibility estimates. Corresponding increments for methionine were only 4, 3, 1 and 1 percentage points, respectively.

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Table 1. Basal Endogenous CP and AA Losses Determined at the End of the Small Intestine by Different Experimental Techniques (means ± SD; mg/kg dry matter intake).

Method	Regression analysis x ± SD	N-free diet x ± SD	Guanidinated casein x ± SD	Wheat gluten x ± SD	EHC* x ± SD
No of observations**	3	7	3	2	5
Crude Protein	16367 ***	6277 ±1369.5	16060 ±878.4	11370 ±3010.0	9234 ±1255.5
Methionine	307 ***	143 ±41.6		136 ±24.1	79 ±14.2
Cystine	408 ***	209 ±16.1		280 ±65.1	169 ±26.8
Met + Cys	715 ***	350 ±49.2		411 ±98.4	257 ±38.6
Lysine	764 ±520.3	293 ±86.0	580 ±190.8	444 ±94.5	255 ±48.3
Threonine	912 ±249.0	494 ±148.0	1207 ±439.2	867 ±170.3	571 ±78.1
Tryptophan		109 ***		176 ±43.5	82 ±6.0
Arginine	715 ±221.1	329 ±100.8	507 ±192.2	384 ±93.1	216 ±51.4
Isoleucine	652 ±404.7	335 ±87.3	1160 ±382.2	477 ±118.4	390 ±144.5
Leucine	1281 ±278.6	475 ±47.7	853 ±306.2	554 ±136.6	381 ±86.5
Valine	817 ±58.9	420 ±54.3	1160 ±208.8	583 ±213.1	449 ±119.5
Histidine	349 ±34.7	193 ±166.0	255 ±77.8	201 ±53.8	209 ±86.8
Phenylalanine	677 ±146.7	289 ±63.3	305 ±289.9	407 ±111.9	237 ±80.8

* Enzymatically hydrolyzed casein

** No of observations might be lower for single amino acids

*** n = 1 for these parameters

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Table 2. Standardized Ileal Crude Protein and Amino Acid Digestibility Coefficients of Feedstuffs for Broilers (%).

	Obs.*	CP	Lys	Met	Cys	M+C	Thr	Trp	Arg	Ile	Leu	Val	His	Phe
Grains														
Barley	3	90	88	88	88	89	85	69	85	90	86	85	86	83
Corn	6	90	92	94	87	90	85	81	93	95	94	92	95	94
Sorghum	5	86	90	89	79	84	83	87	88	90	88	87	84	89
Rice pollard	3	68	76	71	65	68	66	50	78	66	66	68	80	65
Triticale	3	87	85	90	87	88	87	86	83	91	88	87	90	86
Wheat	11	88	86	91	90	91	87	86	85	94	90	90	90	90
Wheat middlings	3	78	80	83	74	78	73	79	80	82	80	77	80	78
Plant protein sources														
Corn gluten meal	1	86	76	88	78	83	79	66	86	86	91	85	86	88
Cotton seed meal	4	78	65	72	74	73	68	80	88	71	73	74	81	81
Lupines	5	86	87	89	83	85	83	82	91	85	85	84	89	85
Peas / Beans	8/1	76	85	73	65	68	78	66	87	77	76	72	82	77
Rapeseed meal	68	76	80	84	77	80	73	80	87	79	82	79	85	83
Soybean meal	37	90	90	91	82	86	85	89	93	89	89	88	92	89
Sunflower meal	3	84	87	92	80	87	82	87	93	89	88	87	88	90
Animal by-products														
Feather meal	1	57	57	61	49	51	53	46	68	73	66	67	60	68
Fish meal	4	80	86	86	71	82	80	78	82	85	85	83	78	82
Meat and bone meal	30	65	69	72	49	62	62	55	77	69	71	70	71	70

* Number of observations for standardized ileal digestibility coefficients

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Table 3. Recommended Standardized Ileal Digestible Amino Acid Levels for Broilers Based on Optimum Dietary Lysine Content and the Ideal Protein Concept.

Phase	Starter I		Starter II		Grower		Finisher I		Finisher II	
Period, days	1-5		6-14		15-35		35-45		>45	
	stand. ileal		stand. Ileal		stand. Ileal		stand. ileal		stand. ileal	
dig. lysine, g/MJ ME	1.03		0.98		0.85		0.75		0.70	
Energy, MJ ME/kg	12.6		12.8		13.0		13.2		13.4	
Ileal dig. Lysine, % of diet	1.30		1.25		1.11		0.99		0.94	
Ileal dig. Protein, % of diet*	22.0		20.0		18.0		17.0		16.5	
	% of diet	Lys = 100	% of diet	Lys = 100	% of diet	Lys = 100	% of diet	Lys = 100	% of diet	Lys = 100
Met	0.59	*45*	0.56	*45*	0.49	*44*	0.43	*43*	0.40	*43*
Met+Cys	0.94	*72*	0.91	*73*	0.83	*75*	0.76	*77*	0.74	*79*
Thr	0.82	*63*	0.80	*64*	0.72	*65*	0.65	*66*	0.63	*67*
Trp	0.21	*16*	0.20	*16*	0.18	*16*	0.17	*17*	0.16	*17*
Arg	1.33	*102*	1.29	*103*	1.17	*105*	1.06	*107*	1.01	*107*
Val	1.03	*79*	0.99	*79*	0.89	*80*	0.80	*81*	0.76	*81*
Ile	0.88	*68*	0.85	*68*	0.78	*70*	0.71	*72*	0.69	*73*
Leu	1.39	*107*	1.34	*107*	1.19	*107*	1.06	*107*	1.01	*107*

* If all essential amino acids are considered as minimum constraint in linear feed programming, ileal digestible protein contents will be around or higher as given in the table. However, these levels should be seen only as guidelines. Crude protein levels are then approximately two percentage points higher than ileal digestible protein.

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Standardized Ileal Digestible Amino Acids in Combination with the Ideal Protein Concept

The required complement to the effective use of standardized ileal digestible AA in feed formulations is a recommendation for optimum dietary AA concentrations for broilers. Feed formulations are based on amino acids rather than on protein as it is recognised that birds have a requirement for AA and not for protein *per se*. The recommendations presented in Table 3 are based on the Ideal Protein Concept using lysine as the reference amino acid. The basic premise of this concept is to ensure optimum utilization of all essential AA since in an Ideal Protein, all AA are in balance and no AA is in relative excess. The concept further assumes that, whilst absolute requirement for AA may vary between various practical situations, the ratios between these AA remain fairly stable. Therefore, optimum amino acid levels for various production stages have to be determined only for lysine and optimum levels for the rest of the essential AA are then obtained simply by multiplying the lysine level with the respective optimum ratios (Lemme, 2003a). Not only the ratios between AA but also the ratio between AA and dietary energy should be considered in feed formulations. Therefore, standardised ileal digestible lysine (and crude protein) values shown in Table 3 are presented in relation to metabolizable energy.

Application of Standardized Ileal Digestibility in Broiler Feed Formulation

The aim of the present study conducted at the Federal University of Viçosa, Brazil, was to evaluate whether the application of numbers from the "Standardized Ileal Amino Acid Digestibility Table" above results in improvements in performance prediction compared with feed formulation based on total amino acids.

A total of 1584 male ROSS 308 birds were distributed to 72 floor pens with 22 birds each. From day 1 to 14 all birds received a commercial starter diet. At day 15 the 72 pens were equally assigned to 9 dietary treatments resulting in 8 replications per treatment. The 9 experimental mash diets and water were offered for free consumption from day 15 to 35. Average body weight of the broilers was 376 g. The experimental design comprised three factors:

- diets were formulated either on total or on standardized ileal digestible (SID) amino acids;
- either corn or sorghum was used as grain;
- two inclusion levels of cottonseed meal (CSM) were established (Table 4).

Diet compositions are given in Table 5. Diets were formulated to be iso-energetic and iso-nitrogenous. Amino acid levels were chosen to marginally limit performance in order not to mask the effects of formulating the diets either on total or digestible amino acids by oversupply of essential amino acids. Diets of treatments II, III, VI, and VII were formulated to contain the same total Lys, Met+Cys, and Thr content as diet I whereas diets of treatments IV, V, VIII, and IX were formulated to contain the same SID Lys, Met+Cys, and Thr content as diet I. Protein and total amino acid contents were confirmed by analyses except for Met and Met+Cys which were systematically 0.05%-points lower than expected since diet formulations had been adjusted to analysed amino acid contents of the raw materials prior to feed production. Body weights and

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feed consumption were measured from day 15 to 35. Subsequently weight gain and feed conversion was calculated. Data were analysed by ANOVA and differences between treatments with $p<0.05$ were considered significant (LSD).

Table 4. Experimental Design.

Treatment	Grain	Diets formulated on Amino acids	Inclusion of cottonseed meal, %
I	Corn	Total / Digestible	-
II	Corn	Total*	6
III	Corn	Total*	12
IV	Corn	standardized ileal digestible*	6
V	Corn	standardized ileal digestible*	12
VI	Sorghum	Total*	6
VII	Sorghum	Total*	12
VIII	Sorghum	standardized ileal digestible*	6
IX	Sorghum	standardized ileal digestible*	12

***** total or SID Lys, Met+Cys, and Thr levels were kept identical to the control (treatment I)

Table 5. Experimental Diets.

Treatment	I	II	III	IV	V	VI	VII	VIII	IX
Ingredients, %									
Corn	58.9	57.3	55.6	57.3	55.7				
Sorghum						57.1	55.5	57.2	55.6
Soybean meal	34.9	29.9	25.0	29.8	24.8	28.4	23.5	28.3	23.3
Cottonseed meal		6.0	12.0	6.0	12.0	6.0	12.0	6.0	12.0
Soybean oil	2.44	3.00	3.56	2.97	3.51	4.56	5.08	4.53	5.03
DL-Met	0.19	0.18	0.17	0.19	0.20	0.20	0.19	0.22	0.23
L-Lys-HCl		0.05	0.11	0.08	0.17	0.13	0.18	0.16	0.24
L-Thr		0.02	0.04	0.03	0.06	0.04	0.05	0.05	0.08
Vitamins & Minerals	3.57	3.55	3.52	3.63	3.56	3.57	3.50	3.54	3.52
Energy and nutrients, %									
Energy, kcal ME/kg	3000	3000	3000	3000	3000	3000	3000	3000	3000
Energy, MJ ME/kg	12.55	12.55	12.55	12.55	12.55	12.55	12.55	12.55	12.55
Protein*	20.4	20.4	20.4	20.4	20.4	20.4	20.4	20.4	20.4
Total Lys*	**1.10**	**1.10**	**1.10**	1.12	1.14	**1.10**	**1.10**	1.12	1.14
Total Met+Cys*	**0.83**	**0.83**	**0.83**	0.84	0.85	**0.83**	**0.83**	0.85	0.86
Total Thr*	**0.78**	**0.78**	**0.78**	0.79	0.80	**0.78**	**0.78**	0.79	0.80
SID Lys**	**0.99**	0.97	0.95	**0.99**	**0.99**	0.97	0.95	**0.99**	**0.99**
SID Met+Cys**	**0.75**	0.73	0.72	**0.75**	**0.75**	0.73	0.72	**0.75**	**0.75**
SID Thr**	**0.66**	0.65	0.64	**0.66**	**0.66**	0.65	0.64	**0.66**	**0.66**

***** calculated values, confirmed by analysis

** SID = standardized ileal digestible

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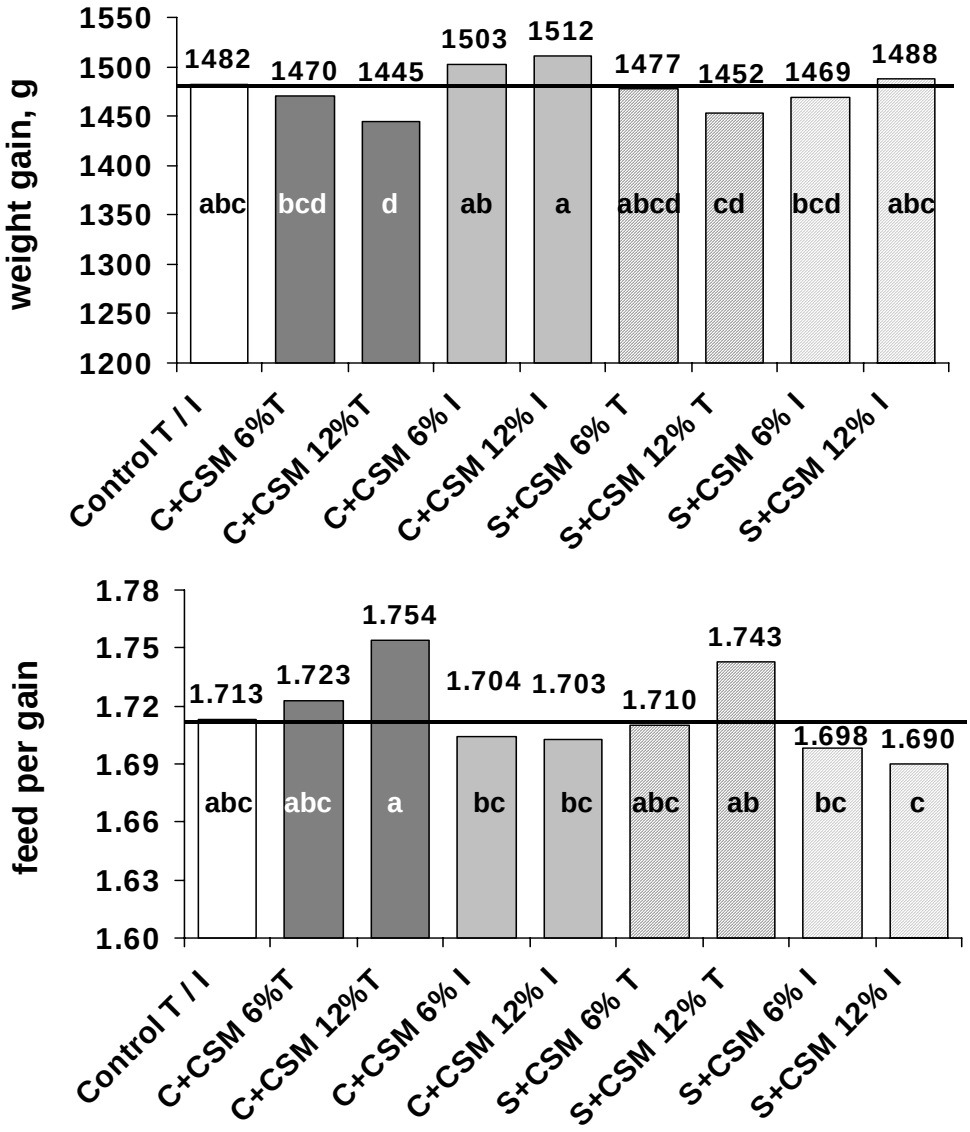

Figure 2. Weight Gain (top) and Feed Conversion (bottom) of 15-35 Day-old Male Ross 308 Broilers Fed Diets Based Either on Corn (C) or Sorghum (S), Containing Either 6 % Or 12 % Cottonseed Meal (CSM), and Formulated either on Total (T) or Standardized Ileal Digestible (I) Amino Acids.

As shown in Figure 2, weight gain was reduced in treatments formulated on total amino acids compared with the corn-soybean meal control. This adverse effect increased with increasing CSM inclusion level, whilst performance could be maintained when diets were formulated on SID amino acids. This finding leads to the conclusion that the higher the inclusion of critical (low digestible) raw materials, the more important is to formulate on digestible amino acid basis in order to avoid loosing animal performance. Moreover, this study shows that using standardized ileal digestibility coefficients results in an improved performance consistency in broiler production. Feed conversion data showed similar effects: feed conversion was impaired

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with increasing CSM level when formulated on total amino acid basis, while feed conversion could be maintained when diets were optimised on SID amino acid basis.

The outcome of this experiment can be seen as a validation of the standardized ileal digestibility figures given above – at least for the raw materials used in this experiment. Knowledge about amino acid digestibility in raw materials enables the nutritionist to more accurately use critical ingredients and to reduce safety margins in formulations.
Although not all essential amino acids are commercially available, it might be useful to consider or at least to monitor them in feed formulation since broiler experiments have consistently shown that the full benefits of the application of the Ideal Protein Concept can only be achieved when all essential AA are in balance (Lemme, 2003b). This is of particular importance especially during the early starter and starter phases.

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