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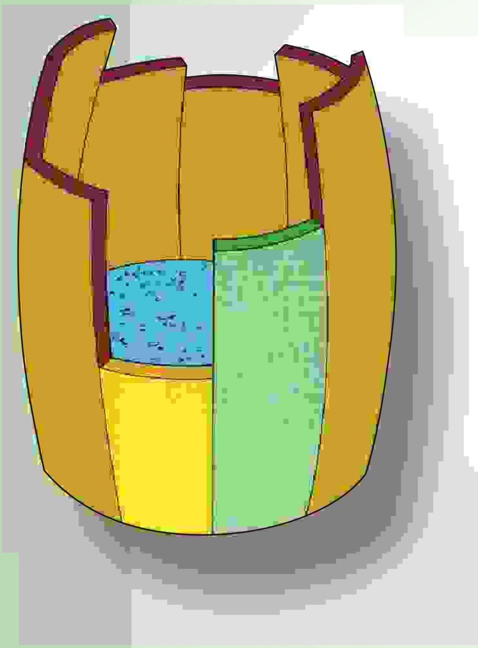
AWT

Amino acids in animal nutrition



Lys
Met
Thr
Trp
Ile
Val
Arg
Leu

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Amino acids in animal nutrition

Responsible for the content

Dr. J. Häffner, Aventis Animal Nutrition

Dr. D. Kahrs, Lohmann Animal Health

Dr. J. Limper, Degussa-Hüls AG

J. de Mol, Novus

Dr. M. Peisker, ADM

Dr. P. Williams, Anitox

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Editor:
Arbeitsgemeinschaft für Wirkstoffe
in der Tierernährung e.V. (AWT)
Contact: Dr. E. Süphke
Roonstr. 5
D-53175 Bonn
Tel. + 49 228/ 35 24 00
Fax + 49 228/ 36 13 97

AWT Trade Association

AWT is a German trade association with an international presence representing the professional, scientific technical and trade interests of leading manufacturers and processors of additives used in animal nutrition.

Scope and objective of the association

- Safeguard member interests and act as their representative in dealing nationally with authorities, government departments, legislative bodies, professional organisations and otherinstitutions
- Representation of German interests in additives internationally.
- Co-operate to harmonise applications for the approval of feed additives
- Brief and advise members on all professional matters, particularly with respect to current legislation.
- Provide information for the general public on the use, safety and quality of feed additives in animal nutrition.

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Spithal 4 · D-29468 Bergen/Dumme
Telephone ++49 (0) 58 45 - 98 81 - 0 · Telefax ++49 (0) 58 45 - 988 111
E-Mail: mail@agrimedia.com · Internet: www.agrimedia.com

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1. Introduction

The requirement for greater efficiency in animal production has led to a trend for highly specialised units where optimum yields have been achieved using animals of high genetic merit. Advances in husbandry techniques have also been made in response to the needs for rearing these high genetic merit livestock. Increases in egg yield per hen (Table 1) which have been gained over the last decade is a further example of the improvements which have been achieved from a combination of good breeding and good husbandry techniques. The nutrition of livestock has played a critical and essential role in these developments and is a component which needs to be continually updated as new scientific information becomes available.

Year	Number of eggs/ hen/ year
1950	120
1960	157
1970	215
1980	242
1990	266
1995	284

Table 1
Improvements in yield of eggs from laying hens in the Federal Republic of Germany

Protein was recognised very early on as an »organic body building substance« of strategic importance to an organism. It was with the development of feed nutrient analysis early in the nineteenth century, that the first qualitative assessment of production animal feeds was made possible. Yet, despite these developments, in practice, formulations continued to be dominated by the concept that formulations were considered »more in terms of components« rather than a holistic approach of regarding the ration as a whole. Even though it is well accepted that amino acids, as building blocks of protein, play an essential role in the nutritional composition of a feedstuff, it has only been the economic incentive which has resulted in their use in feed formulation. However there has been a gradual evolution in the thinking and more emphasis is now being given to thinking in terms of total nutrient supply.

However, even to the present day great weight is still attached to the crude protein content of a feed, as a measure of its protein value. In addition, shortcomings in analytical techniques have been a factor contributing to the concept of minimum levels of dietary crude protein, which often are in excess of requirement for various types of mixed feed The myth is perpetuated by the incorporation of these values into feedstuff legislation.

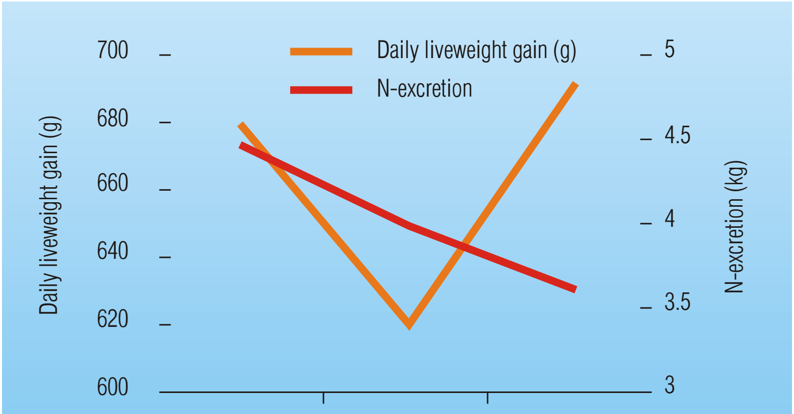
Only as a result of relatively recent advances in analytical techniques has it been possible to demonstrate that an evaluation of protein to the point of the effectively utilised essential amino acids is required to optimise feed formulation and that this yields both practical and financial benefits. It is only then that the crude protein concept looses its overrated status which it has held since the start of nutrient evaluation.

The world population is presently increasing by approximately 220,000 people per day. With this in mind it is not difficult to appreciate that the requirement for protein to feed the world population rises correspondingly. Thus as agricultural production becomes evermore concentrated in specific regions it becomes even more important to use all natural resources as wisely and sparingly as possible. The concept of sustainable agriculture is not solely related to animal production but also incorporates aspects of animal feeding and the use of feeding regimes adjusted to demand.

Amino acids are feed additives which are an integral part of these feeding regimes. In some instances they may act as replacements for naturally produced proteins, helping to save and spare protein and reduce nitrogen excretion. In the future, amino acids will become even more important in order to ensure that animal production does not waste precious feed resources required by the human population, that animal production systems are environmentally compatible and that the optimum use is made of the potential of breed improvement.

Amino acids and mixed feeds

Compound feeds used either as the sole component of a ration or as supplements to other feed materials, represent the major part of the total agricultural economic outlay and remains one of the most important concepts in agriculture. The past decade has seen major increases in compound feed production coupled with a trend towards larger livestock units and higher performance levels. This has increased the importance attached to the utilisation of new findings in the areas of physiology of nutrition and in the design of new compound feed formulas. These developments place continuously higher demands on the formulator to produce feeds capable of meeting the requirements of companies and the end user for high perfor-



Group	1	2	3
Crude protein*	18/16%	16/14%	16/14%
Lysine supplementation	–	–	0.20%

* Starter/ Finishing diet

Figure 1
The influence of amino acid supply on daily liveweight gain and N-excretion

mance, cost-effective and environmentally friendly feeds. The following diagram shows the result of a very simple experiment with only one small supplement of an amino acid to a pig fattening feed (figure 1).

Lowering the protein content of the feed by 2% from 18 to 16% in the starter and 16 to 14% in the finishing diet, resulted in markedly poorer growth compared with the controls. However, after the addition of lysine to the low protein diets the level of performance was raised to that of the controls. Moreover, the pigs given the low protein diets with supplemental lysine excreted significantly less nitrogen compared with the controls.

For many years industrial production of synthetic amino acids has provided the opportunity to achieve improved animal performance coupled with a saving in protein use and above all at a lower cost for the production of feed. In addition to these benefits, both animal producers and consumers have become increasingly more aware of the additional positive effects such as improved animal health and a reduced nitrogen load on the environment.

2.1 The nutrient protein

2.1.1 Significance and composition

Protein (from the Greek: proteios = the first or the most important) is the most important and quantitatively major component of all organisms and as such is a prerequisite of all life. In feed for animals feed proteins cannot be replaced by any other nutrient. Protein containing compounds are found in every cell and account for the major proportion of the protoplasm. They assist in the nutrition of all animal cells and thus in the maintenance, growth and reproduction of the whole organism. However they can only fulfil this role in association with other nutrients which provide energy, with vitamins, elements both in bulk and trace amounts and water.

The major elements in protein are carbon (C), oxygen (O) and hydrogen (H) the same as in fats and carbohydrates. In addition proteins contain nitrogen (N), frequently sulphur (S) and sometimes phosphorous (P).

The level of these elements in protein is relatively constant (%).

C:	51.0 - 55.0
H:	6.5 - 7.3
O:	21.5 - 23.5
N:	15.5 - 18.0
S:	0.5 - 2.0
P:	0 - 1.5

Proteins are high molecular weight, modular compounds. The modular nature is part of the chemical structure since each protein is made up of approximately 20 different amino acids. This unique structure which consists of amino acids in particular sequences gives to each protein a high degree of specificity with respect to the function which each individual protein performs. For example, enzymes are proteins with catalytic function whilst immune bodies display a defence function. Muscle protein enables animals to undertake physical work and bone, skin and connective tissues have a supportive and protective function. However in animal production, the primary target is for the animal to produce muscle protein which is the major essential component of meat.

The series of amino acids within the protein molecule is genetically predetermined and referred to as the amino acid sequence (figure 2). The amino acids are linked by peptide bonds between the

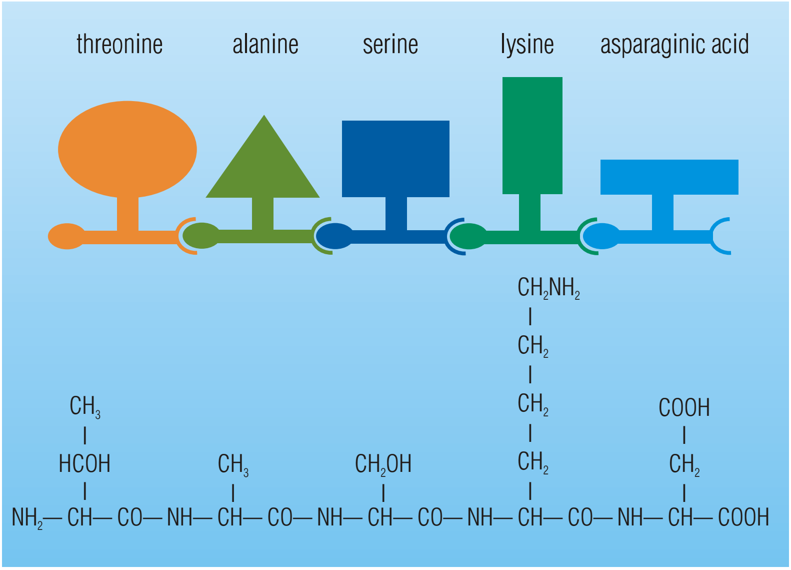


Figure 2
Amino acids as building blocks of proteins

carboxyl group of the one and the -amino group of the other amino acid. A peptide chain can comprise up to 500 amino acids and since there are only approximately 20 different amino acids, several will be repeated in the peptide chain.

2.1.2 Classification

Proteins can be classified into three groups according to their structure and solubility:

Sclero-proteins; which have a fibrous structure and do not dissolve in water. They are comprised of a long linear chain as a supportive and paraplastic substance .Typical examples are collagen (connective tissue, cartilaginous substance) and keratin (skin, hair, wool, feathers).

Sphero-proteins; which are essentially firmly intertwined and soluble in water or dilute saline. This group comprises, albumins, globulins, histones, prolamines and glutelines.

Proteides; which are made up of proteins but also contain a non-protein prosthetic group which tends to be firmly bound to the protein. Depending on the type of prosthetic group proteides are:

- Metalloproteins (e.g. haemoglobin)
- Phosphoproteins (e.g. casein)
- Lipoproteins (e.g. serum lipoproteins)
- Nucleoproteins (e.g. nucleic acid + protein)
- Glycoproteins (e.g. seromucoids)
- Chromoproteins (e.g. myoglobin)

The type of binding between the protein portion and the prosthetic group differs according to the type of prosthetic group and each different type of proteide has a different function in the organism.

Apart from the proteins already mentioned, compounds containing nitrogen but with a non-protein nature are also found in animal tissues. These compounds include alkaloids, amides (asparagine, glutamine, urea), betaine, choline and purines. They are grouped together as non-protein nitrogen (NPN) compounds. By definition, amino acids outside the protein unit are also classified as NPN compounds.

From the point of view of the fundamentals of nutrition, amino acids are equivalent to proteins. It is primarily for this reason that the main emphasis in the nutrition of monogastric animals has shifted from a focus on protein as a whole to a focus on individual amino acids.

2.1.3 Determination

The nitrogen content of different proteins is relatively constant and varies only slightly from an average value of 16%. The level of protein in a feedstuff is usually measured by determining the nitrogen content according to the conventional Kjeldahl method. The protein level of the feed can then be calculated by multiplying the analysed N-level by a factor of 6.25. Since NPN compounds are also included in the N-determination, the protein level calculated in this way is correctly referred to as crude protein. Other procedures can be employed to differentiate true and crude proteins (for example precipitation reactions).
The Near Infra Red Reflectance Spectroscopy (NIRS) technique can be used for fast evaluation of the crude protein level of feeds. It is paramount for this method, that a sufficiently large number (circa 80) of chemically verified analytical values are available for instrument calibration. Furthermore that the population of samples used for calibration is representative in terms of type and composition of the

type of feedstuffs to be tested.
The amount of protein in a feed provides a certain amount of information concerning its nutritional value. However, it must be used with care since it contains the NPN portion which cannot be utilised by monogastric animals apart from the amino acids which as explained earlier are classed as NPN and furthermore, protein level and protein quality are not correlated with each other (see chapter 2.1.6 Protein quality).

2.1.4 Digestion and apsorption

Amino acids chemically bound in proteins must be separated from the parent protein unit, before they can pass from the lumen of the gut across the intestinal wall (absorption) into the blood. This separation occurs in the lumen of the gut with the help of proteolytic digestive enzymes (proteases). The activity of the proteolytic enzymes is aided by the secretion of dilute hydrochloric acid in the stomach. Presence of the acid, acidifies the ingested feed in the stomach which results in denaturation of the protein. The process starts with denaturation of the protein and continues with the cleavage into individual amino acids or as pairs of amino acids (dipeptides), tripeptides and up to five to six amino acid units in length (oligopeptides).
The break-down of the peptide chains is carried out by endopeptidases (pepsin, trypsin, chymotrypsin) which cleave at the centre of a chain and exopeptidases which cleave from the terminal ends. The amino acids and oligopeptides are absorbed by mucosal cells which line the surface of the intestine and finally enter the bloodstream as free amino acids. Specific transport systems are responsible for the absorption of amino acids. The absorbed amino acids are transported via the portal vein into the liver, which is the principal organ for the metabolism of amino acids.

2.1.5 Metabolism and protein synthesis

The metabolism of protein is made up of two opposing processes which run in parallel. The accretion of proteins (synthesis) and the break down of protein (degradation / proteolysis) occur at one and the same time. Synthesis predominates in young growing animals and the protein is built into muscle whereas in mature animals a balance is reached between synthesis and degradation with no increase in the mass of the muscle but

with continuous turnover.
Since the amino acid sequence of a protein is genetically predetermined, all the required amino acids must be present at the same time (synthesis synchronous). The organism is able to compensate for a deficiency of non-essential amino acids within certain limits through autosynthesis. However, protein synthesis comes to a halt if one of the essential amino acids is lacking.

Amino acids which are not synthesised into protein or that are released from protein during degradation must be broken down and excreted since the body has no mechanism to store them. The carbon skeletons of amino acids are metabolised to supply energy and the liberated ammonia which is derived from the nitrogenous component must be "detoxified" and removed from the body. This is achieved via the synthesis in mammals of urea (in poultry uric acid) which is a process with a very high energy requirement. During periods of severe energy deficiency, protein may be metabolised to supply energy for the upkeep of vital processes. However, compared to the metabolism of fats and carbohydrates, efficiency of the process is very low.
From the above it can be seen that:

- Protein and energy metabolism cannot be considered as unconnected. In feed formulation this is taken into account by considering the ratio of the limiting amino acids with respect to the metabolisable or net energy content of the feed and
- the matching of amino acids provided for metabolism with the actual requirement for metabolism must be as precise as possible in terms of both quantity and composition (see chapter 2.2.4 Ideal protein).

Apart from muscle growth only limited amounts of protein can be stored. Some storage occurs in the liver. Otherwise, the degradation of protein is relatively rapid and is expressed by the half life. For example, digestive enzymes which have a half life of only a few days are particularly affected and are thus highly susceptible to changing metabolic conditions with respect to amino acid supply. Hence a transient deficiency in amino acids for the synthesis of the enzyme proteins can show up as a loss in performance.

Great importance is therefore attached to the concept of the continuous supply of free amino acids from the feed into the animal's metabolism (amino acid flux). This needs to be taken into account, when adding free amino acids to mixtures of feed. The free amino acids can be rapidly absorbed and thus are available sooner at the site where synthesis is taking place and out of phase with the remainder of the protein derived amino acids.

2.1.6 The quality of protein

The quality of a protein can be determined by its potential to cover the physiological requirements in terms of amino acids for maintenance and performance (growth, reproduction, production of milk and eggs). The quality of protein required is different dependent on the animal species, age, genotype and sex as well as on the performance level. It follows therefore that there are two important factors with respect to protein quality, a) the amino acid profile which is the ratio of essential amino acids in the protein and b) their availability (see 2.3). Ingredients that contain excellent quality proteins are dried skimmed milk powder and complete egg protein. They have the essential amino acids in the correct proportions and they are all highly available. Some plant proteins lack certain essential amino acids and thus are unable to closely match the requirement of the animal in terms of their amino acid composition.

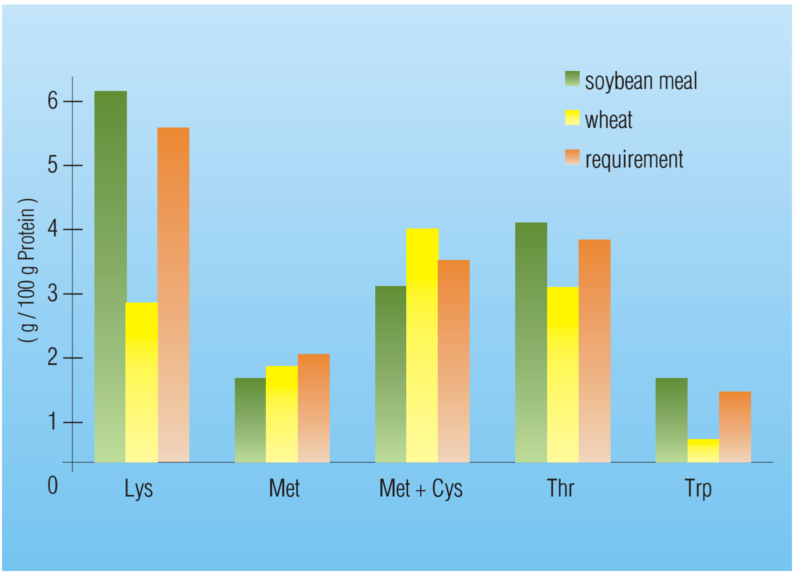


Figure 3
Amino acid composition of protein in soybean meal and wheat compared to the requirements of fattening pigs of 30 - 50 kg

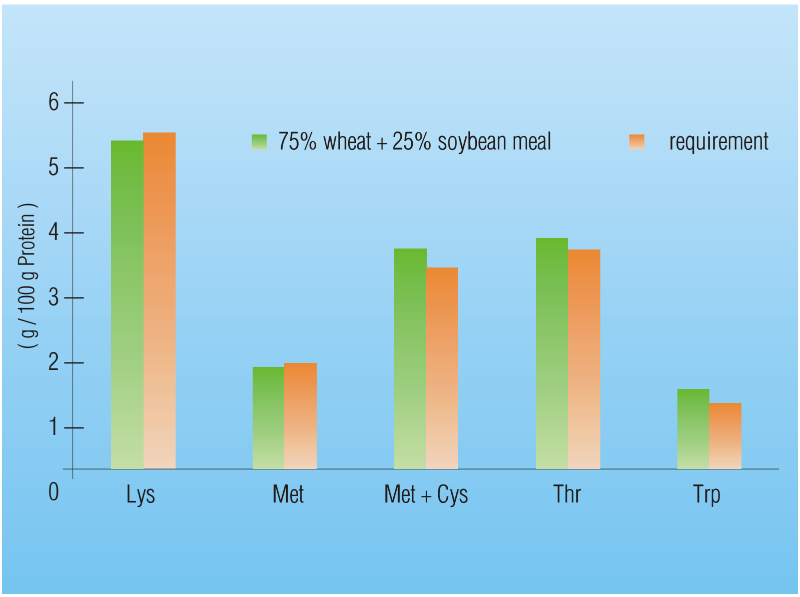
Figure 3 demonstrates that soybean meal alone, with the exception of amino acids rich in sulphur (methionine, cystine), can supply all essential amino acids necessary to satisfy the requirements of a fattening pig (30 - 50 kg liveweight). However wheat has an amino acid profile which first meets the requirements for amino acids rich in sulphur.

By combining the two protein sources in the appropriate proportions it is possible to achieve the requirements in terms of limiting amino acids (figure 4). The protein quality of this mix is greater than that of either of the two individual ingredients.

The quality of the protein in a feed ingredient or compound feed can best be determined in trials with animals. In the simplest method the growth per unit of protein consumed is measured (protein efficiency ratio - PER). Interactions between protein and fat gain and their effect on the body composition are not taken into consideration.

Net Protein Utilisation (NPU) defines the ratio of the retained protein to ingested protein. The Biological Value (BV) relates the amount of protein retained to the

Figure 4
Amino acid content of a mixture of soybean meal and wheat protein relative to the amino acid requirements of fattening pigs (30 - 50 kg live weight



amount of feed protein absorbed. These indexes of protein quality are normally measured by the determination of N-balance, which is a measure of the total nitrogen (N) excreted in faeces and urine relative to the N consumed. In addition to the specific requirement relative to the production of the animal, the other key factor is the amount of feed protein consumed.

There is no universal protein quality standard, which could be employed without taking into account the species of animal and efficiency of protein synthesis. A protein quality must always refer to a specific situation with respect to protein use. In practise, standards of requirement for amino acids are usually given for age and performance levels for all of the major livestock species. With the aid of these standards the quality of a feed protein can be estimated as shown in the example given above.

For a particular feed ingredient or mixture of ingredients supplying protein, the amino acid, which is in shortest supply compared with the requirement of the animal, is defined as the first-limiting amino acid. The first limiting amino acid limits the value of protein (chemical score). In the previous example the amino acid lysine is the first limiting amino acid for the protein in wheat protein and methionine or the sum of sulphur containing amino acids for soybean protein.

Unfortunately, the total content of amino acids in a feed protein, which are determined by chemical analysis, is not fully available for metabolism. A measure of the total amino acid content of a feed protein in comparison with a set of requirements can therefore only give a first approximation with respect to the quality

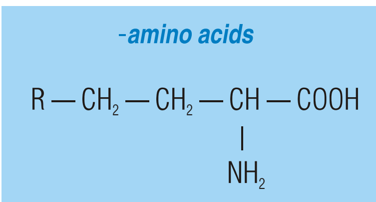
2.2 Amino acids

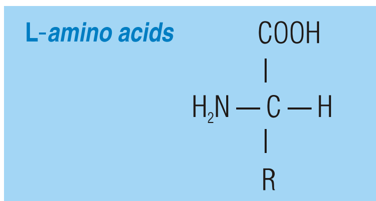
2.2.1 Chemcal structure

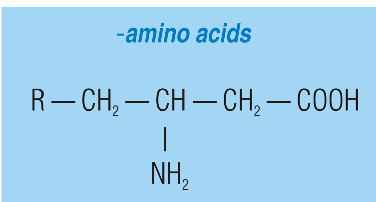
Amino acids are characterised by the two characteristic functional groups in the molecule, as indicated by their nomenclature, by the amino group NH_2 and the carboxyl group COOH.

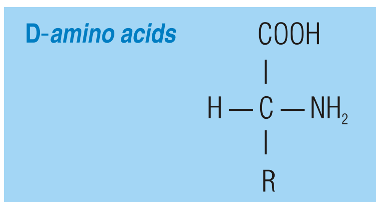
The so-called -amino acids occur exclusively as structural protein units in which the amino group is bound to the -position of the carboxylic acid group (carboxyl group). There are other amino acids in which the amino group is bound in the -position of the carboxylic acid group as shown in the following figure:

Glycine is the simplest representative of an -amino acid. In all other amino acids which are found in protein, an aliphatic or aromatic substituent (=R) is attached to the -carbon atom, in addition to the amino group. This substituent may possess other functional groups. In the general formula shown below, the -carbon atom is surrounded by four different substituents all asymmetrically substituted. There are two additional different types of amino acids, the so-called optically active compounds. Optically active compounds differ with respect to the spatial arrangement of the four substituents on the -carbon atom. This gives rise to the two forms termed the L-form and D-form, which are also known as optically active isomers.
L- and D-amino acids exist as mirror images of each other in the same way as the right hand is to the left.
Apart from differences in the physiological efficacy in animal organisms, the chemical









and physical characteristics of optical isomers are the same except for one property. They differ in the optical rotation of the polarised light, hence the term optical isomers.

Amino acids found in proteins belong to the L-series. If an animal is supplied with amino acids in both the D- and L-forms (that is a 50:50 mixture of L- and D-amino acids, also called " a racemic mixture") then the D-form has to be converted into the L-amino acid before it can participate in metabolism. This can be achieved by deamination to the -keto form and subsequent amination into L-amino acid. This conversion process is dependent on the animal species and takes place for individual amino acids with varying efficiency. In the case of methionine (and tryptophan in the pig) this conversion is so effective that from a nutritional point of view, separation of the D and L- isomers is not necessary ***(see chapter 4, Amino acid supplementation).***

2.2.2 Classification of amino acids

To date 20 different amino acids have been identified following direct hydrolysis of common nutritional feed proteins. Based on chemical principles amino

Neutral amino acids	Acidic amino acids	Basic amino acids
Alanine	Asparaginic acid	Arginine
Asparagine	Glutamic acid	Histidine
Cysteine/Cystine[1]		Lysine
Glutamine		
Glycine		
Hydroxyproline		
Isoleucine		
Leucine		
Methionine		
Phenylalanine		
Proline		
Serine		
Threonine		
Tryptophan		
Tyrosine		
Valine		

Table 2 *Classification of amino acids according to their chemical characteristics*

[1] Tow molecules of cysteine produce one molecule of cystine.

acids can be divided into three main categories, the neutral, acidic and basic amino acids.

This classification is based on the different types of substituents (R) which are present on the -carbon atom in addition to the amino group. Acidic amino acids possess a second carboxyl group in the substituent R position, whilst the basic amino acids have an additional basic group.

2.2.3 Essential amino acids

The animal itself is capable of synthesising about half of the above named amino acids. These amino acids are termed non-essential amino acids. However, about 10 amino acids (depending on the species) cannot be synthesised by the animal and a source must be supplied in the feed. For this reason they are termed the essential amino acids. The essential amino acids are listed in table 3.

The classification of amino acids into essential and non-essential should not be taken to imply that non-essential amino acids are not required for the synthesis of body protein. The terminology non-essential simply indicates that the animal is able to synthesise these non-essential amino acids or convert them from one into another. To undertake such amino acid inter conversions the animal requires sources of carbohydrates and suitable nitrogen compounds.

Table 3 *Essential dietary amino acids for poultry, pigs and humans*

Amino acid	Poultry	Pig	Humans
Arginine [1, 2]	±	±	±
Histidine [1]	+	+	±
Isoleucine	+	+	+
Leucine	+	+	+
Lysine	+	+	+
Methionine	+	+	+
Phenylalanine	+	+	+
Threonine	+	+	+
Tryptophan	+	+	+
Valine	+	+	+

[1] Arginine and histidine are non-essential in humans. However, they become essential under conditions of increased requirement as in rapid growth during early life, in stress and trauma).

[2] Arginine is essential in young chicks.

	Plants	Animals
Biosynthesis	All amino acids (including those essential amino acids required by animals).	A limited number of amino acids (only non-essential amino acids).
Conclusion	Plants require only a supply of simple nitrogenous compounds (via inoganic fertiliser).	The animal must receive a dietary supply of those amino acids which it cannot synthesise itself[1] (essential amino acids).

[1] Exception: The microbial protein synthesis in the rumen of ruminants contributes to the supply of essential amino acids..

In contrast to animals, plants are able to synthesise all amino acids from simple nitrogen and carbon containing compounds (see above).
Certain essential amino acids will be discussed in more detail due to their particular structure and the significant role that they play in metabolism:

Sulphur containing amino acids

The two sulphur containing amino acids methionine and cysteine each contain a sulphur atom and they are present in animal and plant proteins in varying proportions. Methionine is an essential amino acid whereas cysteine is non-essential. But, depending on the species of animal cysteine may be responsible for up to 50% of the dietary methionine requirement. Recent scientific findings show that this proportion is clearly under 50% in high performance animals. In addition to its essential role as a protein building block and precursor of cysteine, methionine is also involved in a number of other biosynthetic pathways. Methionine is involved in the biosynthesis of compounds such as choline, creatine and adrenaline via the release of a methyl group and the formation of S-adenosyl-methionine in the methyl donor pathway. Thus methionine indirectly plays an important role in fat and hormone metabolism, in the conduction of stimuli in the nervous system and in hepatic metabolism. In the organism, cysteine is produced from methionine via S-adenosyl-methionine-cystathionine. Cysteine is subsequently further metabolised into taurine or via a number of intermediate stages transformed into sulphate. It is preferable however that the animal's requirements for sulphate are met by inorganic S-compounds.

Basic and aromatic acids

Lysine is one of the key essential amino acids. Arginine is essential in young chicks but can be replaced with citrulline. Phenylalanine and tyrosine, which are both aromatic amino acids, are also interchangeable. The total requirement for phenylalanine and tyrosine can be met by phenylalanine alone but the converse does not occur for tyrosine. Tyrosine can provide only approximately 50% of the requirement for phenylalanine.

2.2.4 Limiting amino acids and ideal protein

Protein synthesis is an indispensable part of the growth process of an animal. For protein to be correctly synthesised the required essential and non-essential amino acids must be present at the site of synthesis according to the requirements of the genetic code. If an amino acid is required to extend the protein chain but is not present at the exact location of synthetic activity then protein synthesis is halted. Nevertheless, if the amino acid in question is non-essential the body is capable of obtaining a supply via autosynthesis from an alternative amino acid. However, if it is an essential amino acid that is absent which cannot be synthesised in the body, then this amino acid »limits« protein synthesis.

Hence a limiting amino acid must be present in the diet in sufficient quantity to meet the total requirements of the animal. The first amino acid to limit protein synthesis is termed the first limiting amino acid, once this has been supplied the next to limit synthesis becomes the second limiting amino acid and so on. In conventional diets offered to poultry it is usually the sulphur containing amino acids methionine and cysteine which are first limiting whereas for pigs offered conventional diets lysine is the first limiting amino acid. A sufficient quantity of the limiting amino acids in the diet to meet requirements also governs whether the other amino acids are efficiently utilised for protein synthesis.

This principle can be illustrated by the »Liebig barrel« where the level of fill in the barrel represents the capacity for protein synthesis of the animal (figure 5). The capacity of the barrel is »limited« by the shortest stave (the first limiting amino acid). However, if the shortest stave is lengthened (dietary supplementation with the first limiting amino acid) then the capacity increases to the level of the »second-limiting« stave.

The most important factor for the efficient utilisation of feed protein in order to achieve a required level of performance is the balance of the essential amino acids in the diet compared with the physiological requirements for target performance. Cole and Fuller (1988) showed that for pigs, differences in the requirements for essential amino acids between sexes and in different breeds were mainly of a quantitative nature. The relative amounts of essential amino acids needed for the synthesis of 1 g of protein were the same. This led to the concept that for the growth of pigs there was one ratio of amino acids that was »ideal«. Since in pigs, lysine tended to be the first limiting amino acid it was decided that lysine would be taken as a reference value and the remaining essential amino acids would be referred to lysine, taking lysine as 1. For pigs, this ratio of essential amino acids compared to lysine is designated as ›the ideal protein‹ or the ›ideal amino acid profile‹ for pigs. Since poultry have different requirements for some essential amino acids, namely the sulphur containing amino acids for feather growth, they have a different ratio of essential amino acids relative to lysine and hence have a different »ideal amino acid profile« to pigs. Recent work with pigs has shown that the requirement for essential amino acids (sulphur containing amino acids, threoni-

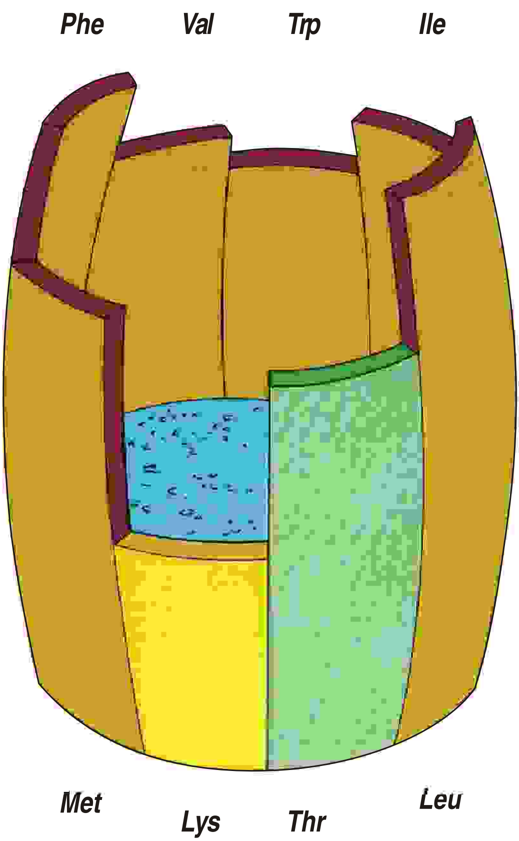


Figure 5
The »Liebig barrel«: Limitation in protein synthesis due to the lack of an essential amino acid.

ne and tryptophan) for the basic maintenance of the animal compared with the requirement for growth increases with increased live weight hence the ideal amino acid profile changes with increasing live weight.

The »ideal amino acid profile« or since protein provides amino acids, the »ideal

Table 4
Ideal digestible amino acid ratios for pigs

Amino acid	***(in % of lysine)***		
	5-20 kg	***20-50 kg***	***50-100 kg***
Lysine	100	100	100
Threonine	65	67	70
Tryptophan	17	18	19
Methionine	30	30	30
Cystine	30	32	35
Met + Cys	60	62	64
Isoleucine	60	60	60
Valine	68	68	68
Leucine	100	100	100
Phenylalanine + Tyrosine	95	95	95
Arginine	42	30	18
Histidine	32	32	32

(Baker, 1997)

Table 5
Ideal ratios of digestible amino acids for broilers

Amino acid	***(in % of lysine)***	
	0-21 Tage	***21-42 Tage***
Lysine	100	100
Met + Cys	72	75
Methionine	36	37
Cystine	36	38
Arginine	105	105
Valine	77	77
Threonine	67	73
Tryptophan	16	17
Isoleucine	67	67
Histidine	31	31
Phenylalanine + Tyrosine	105	105
Leucine	111	111

(Baker & Han, 1997; Baker, 1997)

protein« requirement, differs from one animal species to another and within an animal species differs depending on age and level of production. This is because the requirements for specific essential amino acids and hence the ratio, differs between species and differs according to the requirement in the body whether it be for maintenance, growth or reproduction. The concept of ›ideal protein‹ aids considerably the task of the feed formulator. If the dietary requirements of an animal in terms of lysine are known and the ideal amino acid profile for that animal and stage of production is known, then the requirement for all the other amino acids can be estimated.

The most recent data concerning ideal amino acid ratios are based on amino acids digested in the ileum in order to take into consideration the influence of amino acid losses during digestion and absorption (tables 4 and 5).

2.3 Amino acid availability

2.3.1 Fundamental considerations

Proteins molecules cannot be absorbed intact through the walls of the intestine, the molecules are too large. Thus for an amino acid contained in a feed protein to reach the site of protein synthesis, it must be released from its parent protein. Release from the parent protein occurs during digestion in the gut after which either the individual amino acids or as di and tri peptides are absorbed through the intestinal wall (see chapter 2.1.4 on Digestion and Absorption). Hence factors which have an influence on digestion and absorption in the animal also play a decisive role in the availability of amino acids.

In addition amino acids may undergo major changes during feed processing such that although they still reach the site of protein synthesis, i.e. they are digested and absorbed, they can no longer be used as a structural entity for protein synthesis. Thus, the availability of a feed amino acid is represented by the proportion of that present in the diet which is available without restriction to participate in all metabolic processes for which the amino acid is required.

The classification into digestible amino acids, which is an assessment used to estimate the value of feed proteins is based on the assumption that digested and absorbed amino acids are biologically available.

Digestibility, which is a measure of the proportion of the feed, absorbed and not

Table 6
Effect of replacing 50% soybean meal-protein by meat and bone meal in a diet for pigs growing from 20-45 kg live weight

	soybean meal	50% replacement of soybean meal-protein		
Group	1	2	3	4
AA-supplement	-	-	+ Trp	+ Trp + Lys
Crude protein (%)	15,2	15,2	15,2	15,3
Trp (%)	0,12	0,09	0,12	0,12
ileal digestible Trp (%)	***0,09***	0,06	***0,09***	***0,09***
Lys (%)	0,70	0,66	0,66	0,74
ileal digestible Lys (%)	***0,60***	0,53	0,53	***0,60***
Gain (g/d)	690[1]	590[2]	610[2]	710[1]
kg feed/kg gain	2,42[1]	2,63[2]	2,63[2]	2,49[1]

[1,2] Values with different superscript in the same row differ significantly (p 0,05)

excreted, can be measured at different points of passage along the digestive tract. When digestibility is measured at the end of the small intestines then this is referred to as the pre-caecal or ileal digestibility. This method essentially excludes the effect of microbial activity in the large intestines where the bacteria can either synthesise or degrade amino acids. The bacterial inter-conversion of amino acids results in erroneous estimates of the quantity absorbed. Determination of ileal digestible amino acids results in a more precise estimate of the amounts required for specific levels of performance and aids in diet formulation compared with using total tract digestibility. An example of the benefit to be gained is shown in Table 6.

Replacement of 50% of the crude protein from soybean meal, contained in a diet based on corn and soybean meal (group 1) with meat and bone meal (groups 2 to 4) results in a significant reduction of the daily live-weight gain and poorer feed conversion ratio in group 2. Dietary supplementation with tryptophan alone (group 3) to the level of control group 1 did not significantly improve performance. In group 4, in addition to tryptophan lysine was supplied to achieve the same level of ileal digestible lysine. The level of performance was restored to that of the controls. This example clearly demonstrates that any supplementary amino acid should be supplied according to requirements based on the ileal digestible amino acids. If estimates had been based on the gross lysine content of the diet, the actual requirement of lysinewith respect to that in Group 4 would have been underrestimated since the digestibility of lysine in meat and bone meal is lower than that in soybean meal.

Efforts are made to minimise the influence of amino acids of endogenous origin on the measurement of ileal digestibility by using a number of correction factors. Account must be taken of the amount and composition of the endogenous secretions that are dependent on a number of factors. Endogenous amino acid secretion is influenced by, level of dry matter intake, the amino acid profile of the feed protein, the level of non-starch polysaccharides in the feed, the intestinal kinetics, the microbial

Table 7
True ileal digestibility coefficients of amino acids measured in pigs [1] and in poultry [2] of a range of raw materials and protein supplements

	Lysine		Methionine		M + C		Threonine		Tryptophan	
	pig	poultry	pig	poultry	pig	poultry	pig	poultry	pig	poultry
Wheat	80	83	89	89	89	87	84	82	90	93
Barley	79	80	85	85	85	84	81	84	78	
Rye	73	75	80	77	81	75	73	78	66	56
Corn	77	82	89	93	88	88	83	85	87	90
Triticale	85	84	90	89	90	85	82	85		
Fieldbeans	89	90	79	84	80	82	83	87	75	79
Peas	83	87	78	82	73	78	76	83	75	82
Soy meal 44	88	87	90	89	87	84	84	83	82	84
Linseed meal	78	80	86	91	85	82	69	82	74	89
Sunflower seed meal	81	86	89	94	86	88	83	86	79	91
Peanut meal	87	77	88	87	87	82	91	85	76	
Cotton seed meal	64	60	75	78	72		68	67	65	66
Fish meal	93	85	92	90	90	90	92	84	79	69
Meat and bone meal	83	78	85	84	78	71	83	76	78	71
Skimmed milk powder	96		95		90		90		85	

[1] At the end section of the small intestines

[2] Evaluated in caecectomised cockerels

colonisation of the intestines plus other factors. The quantitative determination and estimation of the magnitude of each of the individual effects is therefore scarcely possible in animal experimentation. Frequently the terms digestibility and availability are interchanged. However, this can result in incorrect estimates of requirements since even digested and absorbed amino acids are not always completely available for protein synthesis.

Taking all into consideration, the principle unfolds of physiologically active amino acids (=utilisable). These can be estimated for limiting amino acids from N-balance trials or measurements of weight gain. Such measurements take into account the availability of amino acids at the metabolic level a factor that is not considered by measuring digestibility alone. Only at the metabolic level can the term availability be truly justified for a feed amino acid.

2.3.2 Influential factors

As has already been shown, digestibility is one of the most important factors that influence the availability of amino acids in feed. Differences in the digestibility of individual amino acids are an inherent characteristic of different raw materials. The true ileal digestibility coefficients of different amino acids in a range of raw materials and protein supplements are shown in table 7. The values have been drawn from a number of different sources and therefore represent a wide range of experimental techniques.

With respect to the nutritional evaluation of feedstuffs the information in table 7 represents a major advance compared with the use of gross amino acid content. If the evaluation of digestibility is carried out at the end section of the small intestine (terminal ileum) the effect of microbiological conversion which occurs in the large intestine is essentially eliminated. Furthermore, if the endogenous secretion of amino acids is estimated, then the true ileal digestibility may be calculated. Diet formulation based on ileal digestible amino acids gives a more accurate estimate of the amino acids available to the animal and is of greatest value with poor digestibility feedstuffs.

Technical processing is used to a great extent in feed production and proteins and meal are treated with steam and heat. For proteins damaged by heat during processing, the measurement of ileal digestible amino acids gives a better estimation of the amino acids available to the animal compared with gross content or total digestibility. However the thermal damage which may occur to an amino acid during processing is not accurately measured by the ileal digestibility technique since although the amino acid can be absorbed it cannot partake in metabolic reactions due to structural damage. This loss in ability to participate in metabolism can only be estimated by measuring the physiologically active amino acids.

Examples of different processing techniques and resulting reactions and effects that can restrict the availability of certain amino acids are shown in table 8. The degree of heat used in the process is particularly important. Individual feeds are subjected to a number of different thermal treatments such as the toasting of soy beans and soybean meal, rapeseed products, peas and field beans; autoclaving of animal meals and feather meal; pasteurising of fish meal or drying of corn gluten and wet cereal. Complete mixtures of feed materials for technical, nutritional, physiological and hygienic reasons are subjected to intense heat treatment (during pelleting up to 80°C; expanding up to 110°C; extruding up to 130°C).

In complete mixtures of feed raw materials components such as reducing sugars are frequently present which under the action of heat, favour the formation of Maillard reaction products with lysine. At present the effect of these processing procedures

Verfahren	***Reaction***	***Affected amino acids***
Heating (Drying, toasting)	Mailard Racemisation Degradation Cross linking	reactionLysine
Protein extraction	Protein-polyhenol reaction	Lysine, methionine, cystine tryptophan
Alkaline treatment	Racemisation Degradation Cross linking	Lysine, methionine, cystine phenyl alanine, histidine threonine
Storage (peroxide formation)	Oxidations products + Amino acids	Methionine, cystine, tryptophan, lysine

Table 8
Effects of processing procedures and possible damage to amino acids

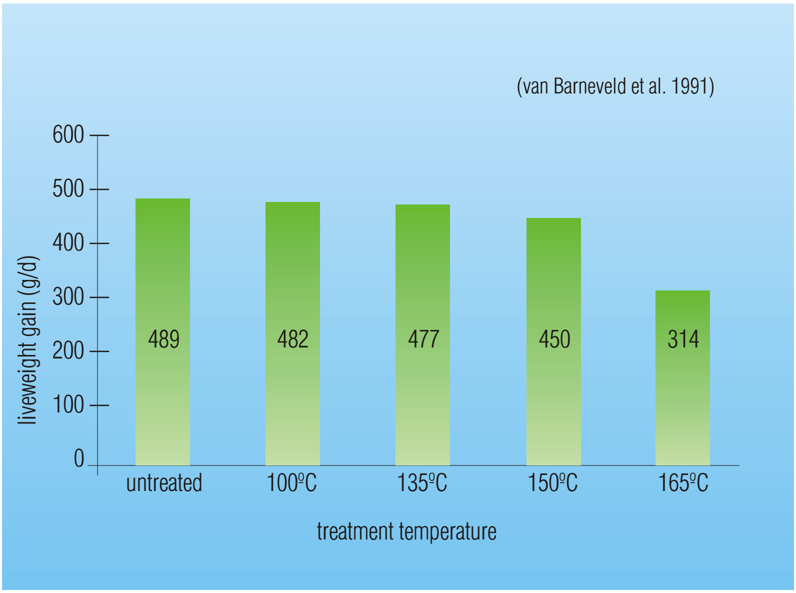
on amino acid availability has not been extensively investigated for individual proteins and mixes, particularly with a view to precisely predicting the effects. However from the point of view of the feedstuff evaluation and formulation, the use of ileal digestible amino acids represents a real progress compared to using the gross amino acid content and is a concept which should be more widely adopted by feed formulators. By accounting for the different losses in the process of digestion/absorption, for example of lysine in cereals, a more accurate estimate of the requirement for supplementation can be made.

2.3.3 Limitations of the evaluation systems

As shown in Table 7 the ileal digestibility of amino acids can be estimated for various raw materials. Apart from diet formulation based on the total amino acid content, research is in progress to determine requirements of key digestible amino acids. Work is also in progress to estimate the availability of amino acids at the physiological level but further research is required.

The availability of amino acids in cereal and protein that have not been damaged by heat can be estimated by digestibility alone. For proteins which have undergone thermal treatment in vitro availability

Figure 6
Daily liveweight gain of pigs (g/day) given rations formulated with 0.36 g ileal digestible lysine/MJ digestible energy and heat treated peas



tests are available (Carpenter test), but presently they are not widely adopted in the feedstuff industry.

The value of using ileal digestible amino acids was demonstrated in trials with fattening pigs (figure 6). Although all diets were formulated with the same amount of digestible lysine, the efficiency of growth decreased with increasing intensity of the heat treatment.
It has proven difficult to accuratel y estimate the degradation rate or catabolism of the essential amino acids in metabolic processes. In the future it will be even more important to be able to accurately measure amino acid catabolism in order to precisely estimate the requirement of utilisable amino acids, particularly for the limiting amino acids.

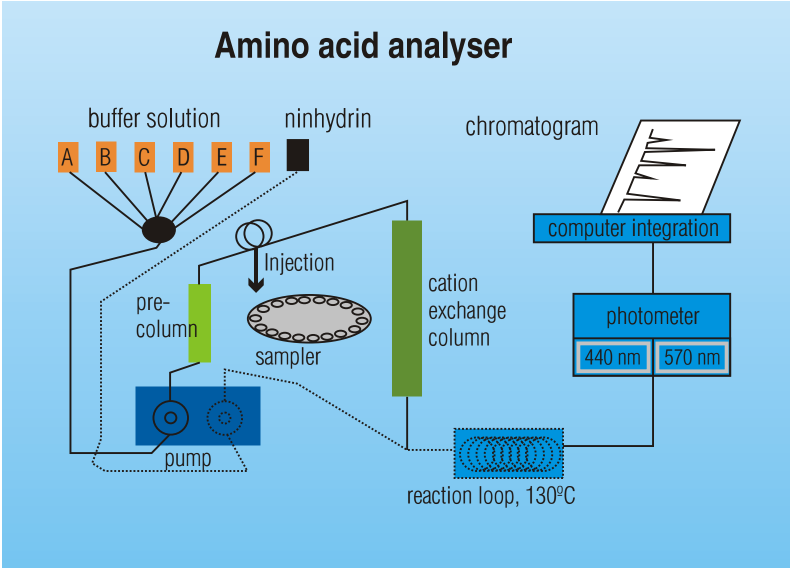
Future research must be aimed at developing adequate systems for the evaluation of amino acid availability (utilisable amino acid), which are able to provide reliable predictions of availability even under the conditions of modern feed processing procedures. The limitations of the present system that considers only digestion and absorption will not be sufficient to satisfy future requirements.

2.4 Analysis

One of the most important prerequisites for the formulation of mixed feeds according to an animal's dietary requirements is precise knowledge of the quantitative amino acid composition of feed ingredients. The determination of the amino acid levels in feedstuff must be considered from two points of view, with reference to methodology and technology.
In order to determine the concentration of amino acids in feed, the protein is first broken down into the individual amino acids by hydrolysis with dilute hydrochloric acid. The quantity of amino acids present in the hydrolysate is determined by ion exchange chromatography using an amino acid analyser or HPLC (high pressure liquid chromatography)(see figure 7).

The solution obtained from the hydrolysis of the protein that is to be measured in the amino acid analyser is loaded onto a separation column packed with a cation exchange resin. The column is rinsed with a selection of buffers at different temperatures. The process separates the amino acids on the column and they can be separately eluted. The individual amino acids are then reacted with the colour reagent ninhydrin in the analyser

Figure 7
Diagram of an amino acid analyser assembly



and finally converted to a specific bluish violet or yellow colour in a reaction loop at 130C. The intensity of the colour (pe-ak) is measured in a photometer, the peak area is evaluated by a computer and the chromatogram printed. The peak position corresponds to a specific amino acid and the area of the peak to the quantity of each amino acid present. This selective detection technique enables a precise quantitative estimation of the amino acid composition of feed protein free of inter-ference from external factors.

For the sulphur containing amino acids methionine and cystine an oxidation step is first required in order to protect them from a partial degradation during hydro-lysis. The oxidation of sulphur containing amino acids is carried out using perfor-mic acid that results in the formation of methionine sulphonium from methionine and two molecules of cysteic acid from cysteine.

Following alkaline hydrolysis tryptophan is measured separately since it is destroyed during acidic hydrolysis with hydrochloric acid. Several detection methods are available.

The accuracy of a laboratory determination of amino acids should be reproducible within a range of variation of 3 - 4%. In

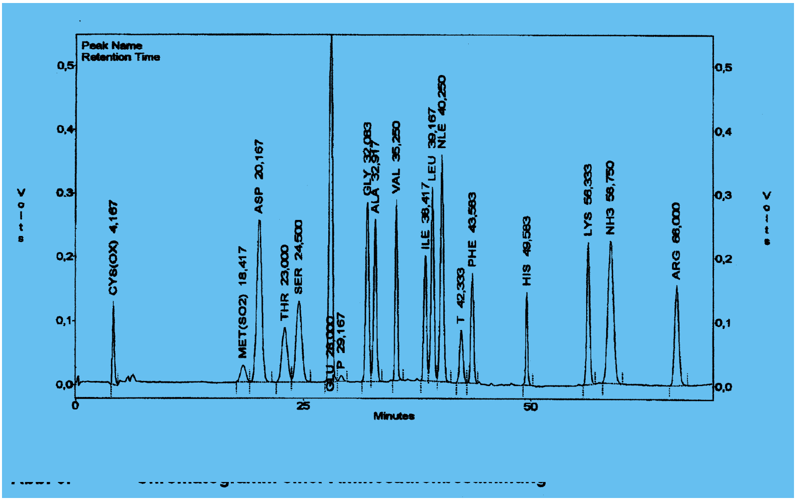


Figure 8
Chromatogram of an amino acid analysis

the past, amino acid analysis has been standardised for feedstuffs both within the European Union (EU) and also in the USA.

The proportion of supplementary amino acids such as DL-methionine, L-lysine HCl, threonine and tryptophan in a feed, can be determined relatively simply by extracting a mixed feed sample with dilute hydrochloric acid at room temperature and analysis of the extract by ion exchange chromatography in the amino acid analyser or via HPLC. The methionine hydroxy analogues (see chapter 4.5) are extracted from the feedstuff with an aqueous extraction agent and determination by HPLC.
Near Infra Red Reflectance Spectroscopy (NIRS) has been used for many years in the feed industry for the rapid measurement of moisture content, crude protein, fat and fibre content as well as for other feed components. Recently NIRS has been developed for the determination of amino acid concentrations in feed ingredients. The advantages of NIRS are that results are provided within minutes and the technique does not require any additional preparation or reagents apart from the grinding of the sample.

The NIRS method is based on the construction of a comprehensive calibration data set produced from samples that are analysed and their nutritive content evaluated by means of reference methods. NIRS calibrations have been constructed for the amino acid composition of several different types of raw materials of both plant and animal origin (for example soy, wheat, meat and bone meal). In addition, the concentration of ileal digestible amino acids in raw materials can also be estimated by NIRS. However, the analysis by NIRS of mixed feeds for the determination of the concentration of supplemental amino acids has not been met with the same success. Nevertheless the technique is of great value for optimising the utilisation of raw material according to their amino acid content.

2.5 Amino acid concentrations in feedstuffs

Different raw materials are characterised by their amino acid composition. In addition within a specific type of raw material but between different samples, differences in amino acid composition may occur as a result of cultivation and geographic influences. Therefore when mean tabular values of amino acid composition are used, consideration must be given to the fact that the actual levels of amino acids in the material may differ considerably from the value given in the tables.

Furthermore, when referring to the different sets of tables for the formulation of mixed feeds, particular attention must be paid to the manner in which the animal's requirements are expressed and how the tabular values are presented (gross, apparent, true digestible amino acids). Additional attention must be paid to the units:

- content present in the raw material (in % or in g/kg)
- content present in the dry matter of the raw material (in % or in g/kg)
- content in % of the crude protein

The values presented in Tables 9 11 are related to a defined dry matter content. Alterations in dry matter content need to be considered when formulating a diet. From a practical point of view, stating the crude protein content is important. If the crude protein content of the sample deviates from the tabular value, then the amino acid content shifts in the same direction. There is an increasing tendency to calculate the amino acid composition of a diet using ileal digestible amino acid levels to ensure the amino acid composition of the diet meets the requirement of the animal. Thus in the following tables for a selection of the raw materials, the true amino acids digestible in the ileum are also presented.

Table 9
Amino acid content in feedstuffs in %

	TS	RP	Lys	Thr	Met	M+C	Try
Field bean	88	25,0	1,57	0,90	0,19	0,50	0,22
Dried brewers yeast	88	50,5	3,43	2,42	0,81	1,34	0,57
CCM	55	5,7	0,15	0,20	0,11	0,23	0,04
Field peas	88	20,0	1,46	0,78	0,21	0,53	0,19
Barley	88	10,5	0,38	0,36	0,18	0,42	0,12
Oats	88	12,6	0,53	0,44	0,22	0,58	0,14
Coconut meal, extr.	88	18,5	0,47	0,57	0,28	0,58	0,14
Linseed mealextr.	89	34,0	1,19	1,23	0,60	1,19	0,50
Alfalfa	88	17,0	0,74	0,70	0,25	0,43	0,24
Corn88	8,5	0,25	0,31	0,18	0,37	0,06	
Corn germ meal	88	11,2	0,47	0,44	0,20	0,43	0,10
Corn gluten meal	88	60,5	1,02	2,08	1,43	2,52	0,31
Corn gluten feed	88	19,0	0,58	0,68	0,32	0,72	0,11
Malt sprouts	92	26,0	1,20	0,87	0,35	0,66	0,20
Rapeseed meal, extr.	88	34,8	1,95	1,53	0,71	1,59	0,45
Rye	88	9,6	0,39	0,34	0,17	0,42	0,09
Soybean meal, extr. 44 %	88	44,0	2,75	1,76	0,64	1,31	0,57
Soybean meal, extr. 48 %	88	47,6	2,98	1,89	0,69	1,40	0,61
Sunflower meal	90	36,2	1,29	1,35	0,84	1,48	0,43
Tapioca	88	3,3	0,12	0,11	0,04	0,09	0,04
Sugar beet plup	88	9,4	0,39	0,31	0,11	0,21	0,07
Whear	88	12,7	0,34	0,37	0,20	0,48	0,15
Wheat bran	88	15,7	0,65	0,53	0,25	0,57	0,25
Wheat midds	88	15,9	0,57	0,51	0,26	0,58	0,20
Blood meal	91	88,8	7,69	3,85	1,03	2,17	1,42
Feather meal	91	83,5	2,12	3,98	0,58	4,91	0,56
Fish meal, 55 %	91	56,3	4,10	2,31	1,53	2,08	0,53
Fish meal, 65 %	91	64,8	4,81	2,64	1,77	2,43	0,66
Meat meal	88	54,7	2,98	2,01	0,80	1,47	0,43
Meat and bone meal, 50 %	91	49,1	2,51	1,59	0,68	1,18	0,28
Meat and bone meal, 55 %	91	53,0	2,82	1,79	0,78	1,33	0,35
Meat meal, 50 %	91	48,8	2,44	1,63	0,68	1,24	0,30
Meat meal, 55 %	91	53,6	2,70	1,88	0,75	1,46	0,35
Poultry by-product meal	91	57,7	3,32	2,18	1,11	1,76	0,48
Skimmed milk powder	93	35,8	2,76	1,58	0,89	1,17	0,49
Whey powder	93	11,8	0,87	0,70	0,16	0,40	0,17
Whey powder, partially extr.	93	23,9	1,80	1,33	0,34	0,81	0,37

Table 10
True ileal digestible amino acid content of feedstuff in % (measured in pigs)

	DM	*CP*	*Lys*	*Thr*	*Met*	*M+C*	*Try*
Field bean	88	28,5	1,59	0,88	0,18	0,38	0,19
Field peas	88	20,0	1,12	0,58	0,16	0,39	0,13
Barley	88	10,0	0,29	0,27	0,14	0,32	0,09
Oats	88	10,5	0,32	0,23	0,14	0,37	0,10
Alfalfa	88	17,0	0,36	0,39	0,15	0,17	0,12
Corn	88	9,0	0,20	0,27	0,17	0,34	0,06
Corn germ meal	88	11,2	0,27	0,29	0,16	0,31	0,07
Corn gluten meal	88	62,0	0,90	1,88	1,39	2,32	0,27
Corn gluten feed	88	20,0	0,45	0,53	0,31	0,55	0,10
Rapeseed, extr.	88	35,5	1,51	1,16	0,66	1,38	0,34
Rye	88	9,6	0,27	0,24	0,12	0,31	0,06
Soybean meal, extr. 44 %	88	44,0	2,40	1,44	0,54	1,06	0,48
Soybean meal, extr. 48 %	88	47,6	2,76	1,70	0,62	1,24	0,59
Sunflower meal	90	34,0	0,94	1,00	0,64	1,08	0,35
Tapioca	88	2,5	0,05	0,06	0,03	0,05	0,01
Wheat	88	11,5	0,26	0,28	0,16	0,42	0,12
Wheat bran	88	15,7	0,47	0,36	0,18	0,43	0,15
Blood meal	91	85,0	7,37	3,41	0,85	1,50	0,95
Feather meal	91	83,5	1,00	2,91	0,33	2,82	0,26
Fish meal, 60 %	91	59,0	4,20	2,20	1,40	1,90	0,45
Fish meal, 65 %	91	64,8	4,70	2,50	1,70	2,20	0,61
Meat and bone meal, 50 %	91	49,1	2,01	1,34	0,49	0,90	0,16
Meat and bone meal, 55 %	91	53,0	2,35	1,53	0,54	1,06	0,17
Meat meal, 50 %	91	48,8	1,95	1,26	0,57	0,81	0,22
Meat meal, 55 %	91	53,6	2,15	1,46	0,63	0,95	0,25
Poultry by-product meal	91	57,7	2,69	1,55	0,87	1,30	0,35
Skimmed milk powder	93	35,0	2,76	1,32	0,78	1,02	0,42

Table 11
True digestible amino acid content in feedstuff in % (measured in poultry)

	DM	CP	Lys	Thr	Met	M+C
Field bean	88	25,0	1,43	0,79	0,15	0,39
Field peas	88	20,0	1,38	0,66	0,17	0,39
Barley	88	10,5	0,30	0,27	0,14	0,34
Oats	88	12,6	0,47	0,37	0,19	0,48
Âlfalfa	88	17,0	0,44	0,48	0,18	0,25
Corn	88	8,5	0,23	0,27	0,17	0,33
Corn gluten meal	88	60,5	0,92	1,93	1,39	2,34
Corn gluten feed	88	19,0	0,42	0,52	0,27	0,53
Rapeseed meal, extr.	88	34,8	1,56	1,22	0,63	1,29
Rye	88	9,6	0,31	0,27	0,13	0,34
Soybean meal, extr. 44 %	88	44,0	2,48	1,57	0,59	1,16
Soybean meal, extr. 48 %	88	47,6	2,68	1,68	0,63	1,24
Sun flower meal	90	36,2	1,04	1,09	0,72	1,20
Tapioca	88	3,3	0,09	0,08	0,03	0,07
Wheat	88	12,7	0,31	0,32	0,18	0,44
Wheat bran	88	15,7	0,47	0,38	0,19	0,42
Wheat midds	88	15,9	0,47	0,38	0,20	0,47
Blood meal91	88,8	6,85	3,39	0,94	1,80	
Feather meal	91	83,5	1,29	2,71	0,44	2,75
Fish meal, 56 %	91	56,3	3,65	2,08	1,38	1,81
Fish meal, 65 %	91	64,8	4,46	2,46	1,66	2,11
Meat and bone meal, 42 %	91	42,9	1,59	0,95	0,45	0,63
Meat and bone meal, 48 %	91	48,1	1,84	1,16	0,53	0,82
Meat meal, 47 %	91	47,1	1,86	1,23	0,55	0,81
Meat meal, 54 %	91	53,7	2,13	1,50	0,64	1,02
Poultry by-product meal	91	57,7	2,66	1,68	0,92	1,32
Poultry by-product, rich in feathers	91	56,7	1,46	1,91	0,47	1,90

3.1 Broiler fattening

Many diverse factors are considered when estimating nutrient requirements. Researchers and National councils in different countries have stipulated different optimum levels of energy contents for broiler finishing diets. Furthermore, the composition of feeds used in the growing stages and their period of application are influenced by availability of raw materials in different countries. Finally the weight when the bird is slaughtered is influenced by variation in consumer demand.

Ration formulation must also account for the changing voluntary intake of the bird as intake is reduced in response to the high energy concentration of poultry finishing diets (expressed as megajoule N-adjusted metabolisable energy [abbreviated: MJ ME N-adj.]). The amino acid concentration of the diet must be increased in order that the absolute intake of amino acids does not fall. The recommended amino acid intake throughout the growth to slaughter of broilers, which is generally divided into three phases, is given in (Table 12). For the reasons given above, the required quantity of amino acids (g) per unit energy (MJ ME) together with the given energy level, in addition to the level in the diet (%) are presented.

Only the proportion of amino acids that can be digested and absorbed from feed protein is made available for metabolism, the remainder is eliminated as part of the faeces. The digestibility of amino acids can differ between different raw materials even though the ingredients may have the same amino acid content. Thus ingredients may differ markedly in their "value" as an amino acid supplier to the organism. In order to account for these differences in amino acid digestibility there has been a tendency for recommendations of amino acid requirements to be made on the basis of digestible amino acids which increases the precision of formulation of the amino acid requirements of the bird.

3.2 Laying hens (incl. rearing)

At peak lay under commercial conditions, the most advanced hybrid birds produce eggs with an average laying efficiency of 95%. However in such flocks the performance of individual hens varies widely.

Therefore sub-optimal nutrient supply will primarily penalise the performance of

Table 12
Recommendations for the digestible amino acid content of complete diets for broilers

					Total amino acid % of diet					Digestible amino acids % of diet			
Feed type	Period (Lebens-woche)	Energy level (Men) (MJ/kg)	(kcal/kg)	Crude protein (%)	Lys (%)	Met (%)	Met +Cys (%)	Thr (%)	Trp (%)	Lys (%)	Met (%)	Met +Cys (%)	Thr (%)
Starter diet	1. - 3.	13,2	3150	21,0	1,24	0,56	0,96	0,77	0,22	1,09	0,52	0,84	0,65
Grower diet	4. - 7.	13,4	3200	20,0	1,12	0,52	0,92	0,70	0,20	0,99	0,48	0,81	0,59
Finisher dietl	ab 7.	13,6	3250	18,0	0,98	0,43	0,82	0,65	0,18	0,86	0,40	0,72	0,55
Starter diet	1. - 3.				0,94	0,43	0,73	0,58	0,16	0,83	0,40	0,64	0,49
Grower diet	4. - 7.	Amino acids (g/MJ ME)			0,84	0,39	0,69	0,52	0,14	0,74	0,36	0,61	0,44
Finisher dietl	ab 7.				0,72	0,32	0,60	0,48	0,13	0,63	0,29	0,53	0,40

Basis: Diet with 88% dry matter; feeding ad libitum; N-adjusted metabolisable energy

the best individual birds. Thus feed formulations are designed to contain all essential ingredients and predominantly amino acids to meet the requirements of birds with the highest performance level during peak lay over weeks 21 to 42 (Table 13).

Table 13
Recommendations for the digestible amino acid content of complete diets for laying hens

					Total amino acid % of diet					Digestible amino acids % of diet			
Feed type	Period (week of-life)	Energy level (MJ ME /kg)	Feeds intake (g) Day	Crude protein (%)	Lys (%)	Met (%)	Met +Cys (%)	Thr (%)	Trp (%)	Lys (%)	Met (%)	Met +Cys (%)	Thr (%)
Chick rearing diet	1. - 6.	13,2	7-40	18,5	0,86	0,39	0,76	0,59	0,16	0,76	0,36	0,67	0,50
Pullets diet I	7. - 12.	13,4	40-60	15,0	0,70	0,33	0,65	0,48	0,13	0,62	0,30	0,57	0,40
Pullets diet II	13. - 20.	13,6	60-80	13,0	0,60	0,30	0,57	0,41	0,12	0,53	0,28	0,50	0,34
Laying diet													
for light hens	ab 21.	12,1	105	16,0	0,84	0,40	0,74	0,55	0,15	0,74	0,37	0,65	0,46
for heavy hens	ab 21.	11,9	115	15,0	0,77	0,37	0,61	0,50	0,14	0,68	0,34	0,60	0,42
Amino acid requirement in mg/hen/day from week 21					880	420	780	575	160	770	390	690	480

Basis: Diet with 88% matter; ad/libitum feeding; N-adjusted metabolisable energy

Table 14
Recommendations for the digestible amino acid content of complete diets for turkeys

Period	Week	Feed intake (g/day)		Turkey weight at the end of the Period (kg)		Energy-level of the feed	Crude protein content of the feed	Total amino acids % of diet					Total amino acids % of diet			
		Female	Male	female	Malel.	MJ ME/kg)	(%)	Lys (%)	Met (%)	Met+ Cys (%)	Thr (%)	Trp (%)	Lys (%)	Met (%)	Met+ Cys (%)	Thr (%)
1	1 - 2	21	33	0,27	0,30	11,7	29,0	1,85	0,65	1,11	1,10	0,32	1,58	0,60	1,01	0,89
2	3 - 5	73	128	1,16	1,31	11,9	27,0	1,75	0,60	1,07	1,02	0,28	1,41	0,56	0,93	0,80
3	6 - 9	164	188	3,36	4,02	12,2	24,0	1,50	0,58	1,00	0,92	0,26	1,23	0,52	0,84	0,71
4	10 - 13	277	360	6,10	7,92	12,8	21,0	1,30	0,52	0,89	0,85	0,22	1,01	0,44	0,71	0,58
5	14 - 17	307	490	8,38	11,92	13,1	18,0	1,10	0,46	0,78	0,68	0,18	0,88	0,40	0,63	0,51
6	18 - 22	-	590	-	15,82	13,5	15,0	0,96	0,40	0,68	0,61	0,16	0,75	0,36	0,55	0,44
Rearing						11,5	15,0	0,65	0,32	0,60	0,50	0,14	0,57	0,29	0,53	0,24
Breeding						11,7	16,5	0,75	0,37	0,68	0,58	0,15	0,66	0,34	0,60	0,49

3.3 Breeding and fattening of turkeys

Turkeys have higher protein requirements than broilers. However, there is poor agreement between different sets of recommendations with respect to the energy levels of turkey finishing diets, the differences mainly apply to the later finishing period.

The nutritional requirements of turkeys during the fattening period differ according to the sex of the bird and whether a heavy or light type is finished. Hens are finished over a shorter period compared with cocks and the final finishing weight differs (Table 14). The high growth rate of turkeys demands an appropriately high supply of amino acids.

3.4 Water fowl

There are few literature references with respect to the amino acid requirements of ducks and geese but they are of particular relevance to Asia and Eastern European countries where large numbers of these birds are found. A selection of the amino acid requirements of waterfowl is presented in Table 15.

Animal species and category	Week	Metabolisable energy (Kcal/kg)	Crude protein content (%)	Lys (%)	Met (%)	Met+Cys (%)	Thr (%)
Starter ducks	1 - 3	2900	20	1,1	0,48	0,83	0,62
Finish. ducks	3 - 8	2950	16	0,94	0,38	0,66	0,53
Rear. ducks	f. 8.	2900	14	0,72	0,28	0,51	0,42
Breed. geese		2400	14	0,71	0,27	0,49	0,41
Starter geese	1 - 5	2900	20	1,10	0,40	0,70	0,62
Finish. geese	f. 5.	2950	15	0,82	0,38	0,66	0,52
Breed. geese		2900	14	0,71	0,32	0,55	0,45

Table 15
Recommendations for the digestible amino acid content of complete diets for ducks and geese

3.5 Pig production and fattening

During the growth of pigs from birth to slaughter weight, the daily energy and nutrient requirement as well as the daily feed intake, changes considerably. In the growing pig the protein and amino acid requirement is dependent on the target level of performance. Thus the nutrient content of diets for piglets and fattening pigs must be adjusted according to:

- Target daily live-weight gain
- Target energy level of the diet
- Daily feed intake or the degree of dietary restriction

Target nutrient composition for different growth phases of fattening pigs are presented in Table 16. The recommendations for protein and energy supply are based on a mean daily live-weight gain of 750 g.

In practice, the different types of diet are applied over time periods of different length and for different weight ranges according to the genotype of pig. In general the protein and amino acid content of the diet declines during the course of the fattening period in response to the increased feed intake of the fattening pig and the reduction in the rate of protein deposition and increase in fat deposition. In figure 9, lysine requirements have been expressed

Table 16
Protein and Energy requirements of fattening pigs over individual weight ranges

Weight range	Duration (days)	Daily feed intake in the period	Daily weight	Feed conversion gain	ME (MJ) hprotein rate	Daily requirement of crude protein	Lys
		(kg)	(g)			(g)	(g)
bis 5	21,0	approx. . 0,2	240	-	sow milk plus diet supplement		
5 - 15	30,0	approx. . 0,5	333	1,50	sow milk pus diet supplement		
15 - 25	20,0	1,10	500	2,20	12 - 13	200	13,8
25 - 35	15,6	1,54	645	2,39	18 - 19	260	16,2
35 - 45	13,7	1,90	728	2,60	22 - 23	300	18,4
45 - 55	12,8	2,20	782	2,81	25 - 26	330	20,5
55 - 65	12,3	2,45	813	3,02	29 - 30	360	21,3
65 - 75	12,2	2,66	824	3,23	32 - 34	380	22,1
75 - 85	12,2	2,84	819	3,46	34 - 36	370	22,2
85 - 95	12,5	2,99	799	3,74	36 - 38	360	21,8
95 - 105	13,0	3,11	768	4,06	38 - 40	340	20,2
Ø 5 - 105				2,89			

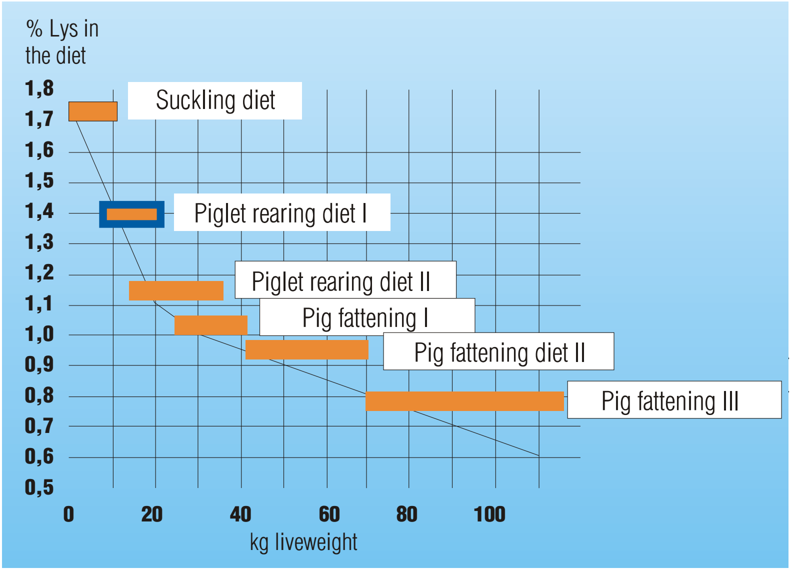


Figure 9
Lysine requirement of growing pigs in % of the diet compared to the content in mixed feed

as a curve. The lysine requirement is expressed as percent of the diet and decreases with increasing live weight of the pig. The smaller drop between 50 and 70 kg live weight corresponds to the period of maximum muscle growth. The lysine content of a complete diet must be adjusted to take account of all these factors in such a way that the minimum daily requirements are met without excessive nutrient waste.

In the past, a single diet has often been fed to pigs during the entire fattening period. However such a concept does not correspond to the changing nutrient requirements of the pig and is no longer appropriate with the desire to optimise nutrient use and reduce nitrogen excretion. In addition when protein prices are high this feeding strategy results in a waste of protein and is therefore not economically viable. The alternative to a single diet strategy is phase feeding, where the nutrient supply is adjusted according to the nutrient requirements of the pig during different growth phases. By adjusting the nitrogen supply according to the protein demands of the pig, the protein is utilised more efficiently and a reduction of 20% in N-excretion can be achieved. If in addition the crude protein content of the diet is further reduced by accurately matching the amino acid requirements of the pig and feeding essentially an ideal protein a further reduction of 20% in N-excretion can be achieved.

With low protein diets ideally matched to the amino acid requirements of the pig, less energy is required for the degradation of excessive dietary protein thus the energy intake of the animal needs to be adjusted and reduced to maintain carcass quality. Since in the metabolisable energy system, diets formulated with low levels of protein tend to be undervalued from the point of energy, it is advantageous to formulate diets on the basis of net energy. Recommendations for the levels of amino acids in complete diets for pigs are shown in Table 18. The values presented are good indications for use in general practise however adjustments should be made according to the genotype of the pig, the potential for protein deposition and environmental factors.

The table shows that at the start of the fattening period higher levels of dietary energy are recommended compared with later stages as the pig approaches slaughter weight. This is because the young pig needs to compensate for its limited appetite with a high energy density in the diet to achieve the levels of energy intake to meet the potential for muscle growth. Conversely with conventional diet formulations, the appetite of the pig during the

later fattening periods is no longer the limiting factor to achieve sufficient nutrient intake for muscle gain

Table 17
Dietary recommendations for the amino acid requirements of pigs given complete diets.

Basis:
- Complete diet with 88% DM
- The fattening of barrows and gilts is not separated
- Pigs with capacity for high lean tissue gain
- Dietary energy concentration may vary with feed restrictions and targeted daily live weight gain

						Dietary recommendation of the total amino acid requirements of pigs					Dietary recommendation for the ileal digestible amino acid requirements of pigs				
	Weight from - to) kg	Age or pregnancy period (from - to in weeks)	Daily increases (g)	MJ ME/ kg	Crude protein (%)	Lys (%)	Met (%)	Met+ Cys (%)	Thr (%)	Trp (%)	Lys (%)	Met (%)	Met+ Cys (%)	Thr (%)	Trp (%)
Pregnant sows	–	1. - 12.	–	12,0	12,5	0,70	0,23	0,42	0,42	0,14	0,57	0,19	0,34	,034	0,11
Lactating sows	–	–	–	13,2	16,5	1,00	0,35	0,62	0,60	0,17	0,74	0,30	0,50	0,47	0,13
Suckling diet	3,5 - 8	2. - 5.	200	13,5	20,0	1,75	0,53	1,05	1,14	0,32	1,49	0,45	0,89	0,97	0,27
Piglet rear-ing diet I	7 - 20	4. - 10.	400	13,3	17,5	1,40	0,42	0,84	0,91	0,25	1,19	0,36	0,71	0,77	0,21
Piglet rear-ing II II	15 - 35	8. - 12.	550	13,0	16,5	1,15	0,38	0,71	0,77	0,22	0,98	0,32	0,61	0,65	0,19
Pig fatten-ing diet I	25- 40	11. - 14.	600	13,2	16,0	1,05	0,35	0,65	0,70	0,20	0,89	0,29	0,55	0,60	0,17
Pig fatten-ing diet II	40 - 70	15. - 20.	800	13,0	14,5	0,90	0,31	0,62	0,67	0,19	0,81	0,27	0,52	0,57	0,16
Pig fatten-ing diet III	70 - 105	21. - 25.	750	12,8	12,5	0,80	0,26	0,52	0,56	0,16	0,68	0,22	0,44	0,48	0,14

3.6 Calf rearing and fattening

Target growth rates for calves are:

Week 1 - 8	700 / 800 g/day
Week 1 - 12	750 - 900 g/day

The calves highest consumption of milk replacer occurs during weeks 4 to 7 when it is limited to a consumption of 6-8 litres per day at a level of 100 - 125 g milk replacer / litre.

From three weeks of age in addition to milk replacer, the calf is offered a calf starter or calf rearing diet plus good quality hay in order to stimulate good rumen development and function. This is coupled with a marked reduction in the level of crude protein and essential amino acids in the total diet. Such changes are made possible by the development of a viable microbial population that supplies microbial protein and contributes an increasing proportion of the total protein requirements.

The gradual transition from a pure milk or milk replacer based diet to one more suited to adult ruminants allows the use of milk substitute feeds of various quality and price. The sooner weaning is achieved from the milk substitute feed (early weaning), the sooner the requirements for high quality protein decline. In the first weeks of life a milk substitute rich in casein is advantageous however from five weeks of age onwards a milk substitute feed in which the amino acids are predominantly derived from whey and soy protein can be adopted. Milk substitute feeds containing high levels of non-milk proteins and a low proportion of skimmed milk need to be enriched with methionine and lysine.

During weeks 1-8 of life the average daily weight gain of growing calves is approximately 1200 g and in weeks 9 - 16 approximately 1400 g. These high rates of gain can be achieved by continuously increasing the amounts of milk offered to the calves and raising the concentration of the milk powder in the milk. Alternatively during the second phase the concentration of protein in the milk replacer and the amino acid content is often reduced.

3.7 Dairy cows

In dairy cows, diets are predominantly formulated on the basis of crude protein or protein utilised in the duodenum, in contrast to diets for pigs and poultry that are based on the amino acid content and often ileal digestible amino acids. This

Stage of growth	Type of feed	Weeks of age	Crude protein (%)	Lys (%)	Met (%)	Met+ Cys (%)	Thr (%)
Rearing	Skimmed milk powder	1.-12.	21	1,70	0,55	0,77	0,95
	casein+ whey protein	1.-12.	20	1,55	0,50	0,75	0,86
	whey protein +soy	5.-12.	20	1,45	0,48	0,72	0,78
Finishing	Starter milk	1.-8. 40-100 kg	23	1,80	0,60	0,80	1,00
	finishing milk	9.-16. 100-180 kg	19	1,50	0,50	0,70	0,83

Table 18
Amino acid recommendations for calves (milk replacer feed)

situation has arisen due to the difficulties in estimating the supply of protein and amino acids to the intestine of the dairy cow when they possess a functional rumen. In ruminants supply from the microbial population must be estimated in addition to the supply from dietary protein that is not degraded by the ruminal bacteria (the non-degradable protein fraction). Recent research has focussed on defining the amino acid composition of the microbial fraction, the quantity of microbial protein produced in the rumen and the amino acid requirements of dairy cows. Such data has allowed the development of computer programs to predict the amino acid requirements of dairy cows and has resulted in several countries such as France and the USA adopting formulation systems for dairy cows based on digestible amino acids. Thus the amino acid requirements for milk production can be precisely estimated and diets for dairy cows are regularly supplemented with the first limiting amino acids.

Limiting amino acids

Several recnt publications have identified either methionine or lysine as being the first limiting or co-limiting amino acids in the dairy cow given a range of standard diets. The basis for this is as follows:

- In terms of amino acid composition microbial protein is well suited to the qualitative amino acid requirements of dairy cows. However, compared with microbial protein, most dietary proteins have lower levels of

- methionine and lysine relative to the total content of essential amino acids. Lysine and cystine often have lower intestinal digestibility than other amino acids in rumen undegraded protein (UDP).

- The contribution of lysine to the total amino acids in UDP is often lower than in the same feeds before exposure to ruminal fermentation. However, this view is still subject to discussion in the literature. Lysine and methionine are the first/ co- limiting amino acids in ruminally synthesised protein for growing cattle.

Whilst the major emphasis of methionine supply to dairy cows is with respect to milk yield and protein composition, additional importance is also given to the veterinary implications of methionine supply particularly with respect to hepatic metabolism. Supplementary methionine is given to dairy cows when feed intake may be insufficient to meet milk synthesis requirements and there is a risk of ketosis when excessive fat mobilisation occurs. Methionine has been shown to support both in the stimulation of fat mobilisation thus aiding energy metabolism and in overall hepatic function.

Amino acid recommendations of dairy cows

To date, the information with respect to the amino acid requirements of ruminants and dairy cows is not as complete as that which has been accumulated for pigs and poultry. The data presented in Table 19 is based on the estimates of amino acid requirements for dairy cows that have been used in France since 1993. Based on this information, for a dairy cow of 650 kg live weight and producing 30 kg milk/head/day with a composition of 4% fat and 3.4% protein, the daily duodenal lysine and methionine requirements are approximately 130 g and 41g respectively.

Amino acids stable in the rumen

The amino acid profile of microbial protein is being of high biological value for dairy cows. However from experiments carried out with high-yielding cows it has been shown that the methionine and lysine content of microbial protein is insufficient to meet optimal performance. Such deficiencies can be corrected via supplementation with methionine and/or lysine in forms that are stable in the rumen. Use of such technologies permits performance

Milk FCM[1] kg/day	600 kg LW			650 kg LW		
	PDI[2]	Lysine	Methionine	PDI[2]	Lysine	Methionine
		g per day		g per day		
0	395	28	9	420	29	9
10	875	61	19	900	63	20
15	1115	78	25	1140	80	25
20	1355	95	30	1380	97	30
25	1595	112	35	1620	113	36
30	1835	129	40	1860	130	41
35	2075	145	46	2100	147	46
40	2315	162	51	2340	164	52
45	2555	179	56	2580	181	57

[1] FCM: Fat corrected milk; adjusted to 4% milk fat

[2] PDI: Protein digestible in the small intestines

Table 19
Amino acid requirements for dairy cows

in milk and the total amount of milk protein produced to be optimised.
Whilst supplementation with rumen protected amino acids produces highly beneficial responses in terms of milk production there are also benefits to be gained in terms of energy metabolism and hepatic function of dairy cows as indicated earlier.
Strategic supplementation with methionine can contribute to the reduction in the level of blood ketone bodies (-hydroxy butyrate and acetone) in cows. Hence rumen stable methionine can make an important contribution to the reduction of the syndrome of ketosis which is prevalent during the first third of lactation in high-yielding dairy cows.
Positive results have also been obtained when sheep have been given supplements of rumen stable methionine. The amino acid profile in the protein of wool is characterised by a high proportion of sulphur containing amino acids and supplementation with rumen stable methionine has resulted in increased wool growth and improvements in wool quality.

Excessive supply of protein

In ruminants feed protein is primarily broken down by deamination in the rumen where ammonia is released and used as a source of nitrogen by the ruminal bacteria for protein synthesis. However, if the ammonia production from deamination of feed protein or non-protein nitrogen compounds is in excess of that which can be metabolised into microbial protein by the bacteria, the ammonia concentration in the rumen rises and ammonia passes into the blood stream of the cow. Excess ammonia in the systemic circulation must then be metabolised in the liver with the formation of urea and for this reason excess protein supply is a burden to high producing dairy cows. The supply of rumen stable amino acids is a highly efficient means of closing the gap between the requirements in terms of protein without exceeding the total nitrogen available for degradation in the rumen.

3.8 Fish

In water of the appropriate temperature and quality fish have the capacity to exhibit extremely high growth rates and very high feed conversion efficiency. High dietary levels of both protein and amino acids are required to exploit the high performance potential since proteins are the most important source of energy for all metabolic processes. However there is little reference material available on the amino acid requirements of fish in relation to the species, age and performance.

Fish have very efficient N-metabolism so that comparatively high protein levels do not represent any particular metabolic burden compared to land animals. Young fish have a higher requirement for nutrient dense diets compared with older specimens, therefore crude protein and also amino acid levels are accordingly higher. In modern fish production systems there is an increasing tendency, for economic reasons, to replace fish meal with plant based proteins in combination with amino acids.

Table 20
Amino acid requirements of fish

Species	Crude protein (%)	Amino acids Lys (%)	M+C (%)	Met (%)	Thr (%)	Trp (%)
Trout	40	2,40	1,30	0,65	1,35	0,20
Carp	30	1,75	0,90	0,45	1,20	0,20
Salmon	40	2,40	1,30	0,65	1,35	0,20
Catfish	24	2,20	-	0,60	0,5	0,12
Eel	38	2,00	-	1,20	1,5	0,40
Perch	28	1,43	0,90	0,75	1,05	0,28

In the absence of supplementary amino acids, the amino acid requirements of the wide range of different species used in agriculture can only be met by significantly raising the crude protein level of the diet. This is because the requirement for the first limiting amino acid relies on the amount of this specific amino acid contained in raw materials alone.

Formulation in the absence of synthetic amino acids results in a major excess of dietary protein. In most instances, supplementation with synthetic amino acids yields economic benefits, is essential from both the nutritional and physiological point of view and, in addition can yield much needed environmental benefits. Amino acid supplementation offers the following benefits:

in a cost effective manner, meets the demand for limiting amino acids

- reduces the crude protein content of the diet
- decreases N-excretion and reduces environmental nitrogen load
- prevents digestive disorders
- improves energy utilisation
- avoids amino acid imbalance
- provides a higher availability of amino acids compared with protein bound amino acids
- balances variations between raw materials
- permits high nutrient density

Using amino acids in compound feed formulation ensures optimum weight gain associated with high levels of feed efficiency, at low total cost. Amino acid supplementation allows the full growth potential of animals, achieved via genetic improvement, to be exhibited. At the same time use of amino acids is totally compatible with the concept of sustainable agriculture with the reductions in environmental nitrogen load that can be obtained and improvements in animal health. Such factors are an increasingly important factor in consumer perception.

The most important issues with respect to the use of amino acids and their analogues in animal nutrition are indicated in the following.

4.1 Lysine (Lys)

General occurrence	All animal proteins are rich in lysine. In plant proteins, cereal sources are low whilst soybean meal is rich in lysine (see tables 9, 10,11).
Significance in metabolism	Building block of proteins, component of enzymes, is present in almost all tissues of the animal organism. Particularly important in the development of collagen and in ossification. As a component of the nucleotides in the nucleus, stimulates cell division.
Production	Produced by fermentation using micro-organisms; raw material requirement, molasses, sugar, products rich in starch and their hydrolysates plus sources of N
Utilisation	L-amino acids are completely utilised whereas the Dform is not 100% biologically available.

4.1.1 Commercially available types

4.1.1.1 L-lysine monohydrochloride (L-lysine (HCE)

Specification	L-lysine monohydrochloride, technically pure, at least 78% of L-lysine	
Chem. name	L- - diamino n-caproic acid monohydrochloride	
Chem. formular	NH_2-$(CH_2)_4$-CH(NH_2)-COOH*HCl	
Technical Data	Molecular weight:	182.7
	Nitrogen content:	15.3%
	Crude protein equivalent:	95.8%
	Purity:	min. of 98.0% (equivalent to 78%
	ME (pig):	17.8 MJ/kg (4250 kcal/kg)
	ME (poultry):	16.7 MJ/kg (3990 kcal/kg)
	Solubility:	64.2 g/100 ml water at 20C
	Stability:	Stable in premixes and mixed feeds

4.1.1.2 L-lysine concentrate (liquid)

Specification	Basic L-lysine concentrate, liquid, from the fermentation of sucrose, molasses, starch products and their hydrolysates; min. of 50% L-lysine	
	L- - diamino n-caproic acid	
Chem. name	NH_2-$(CH_2)_4$-CH(NH_2)-COOH	
Chem. formular	Molecular weight:	146.2
Technical Data	Nitrogen content:	10.17%
	Crude protein equivalent:	63.6%
	Purity:	min. of 50.0% (equivalent to 50% Llysine)
	ME (pig):	11.7 MJ/kg (2794 kcal/kg)
	ME (poultry):	11.0 MJ/kg (2623 kcal/kg)
	Solubility:	Freely soluble in water
	Stability:	Stable in premixes and mixed feeds

4.1.1.3 L-lysine monohydrochloride, liquid

Specification	L-lysine monohydrochloride concentrate, liquid, from the fermentation of sucrose, molasses, starch products and their hydrolysates min. of 22.4% L-lysine	
Chem. name	L- - diamino n-caproic acid monohydrochloride	
Chem. formular	NH_2-$(CH_2)_4$-CH(NH_2)-COOH*HCl	
Technical Data	Molecular weight:	182.7
	Nitrogen content:	5.0%
	Crude protein equivalent:	31.25%
	Purity:	min. of 22.4% (equivalent to 22.4% Llysine)
	ME (pig):	5.1 MJ/kg (1220 kcal/kg)
	ME (poultry):	4.8 MJ/kg (1150 kcal/kg)
	Solubility:	Freely soluble in water
	Stability:	Stable in premixes and mixed feeds

4.1.1.4 L-lysine sulphate

Specification	L-lysine sulphate and its by-products from the fermentation of sugar syrup, molasses, cereal, starch products and their hydrolysates with Corynebacterium glutaminum; minimum of 40% L-lysine	
Chem. name	L- - diamino n-caproic acid sulphate	
Chem. formular	$(NH_2\text{-}(CH_2)_4\text{-}CH(NH_2)\text{-}COOH)_2{*}H_2SO_4$	
Technical Data	Molecular weight:	200.9
	Nitrogen content:	10.7 - 11.7%
	Crude protein equivalent:	67 - 73%
	Purity:	min of 40% L-lysine
	ME (pig):	16.5 MJ/kg (3940 kcal/kg)
	ME (poultry):	15.7 MJ/kg (3740 kcal/kg)
	Solubility:	Partially soluble in water
	Stability:	Stable in premixes and mixed feeds

4.2 Methionine (Met)

General occurrence	Methionine is present in relatively high amounts in animal proteins but is low in plant proteins such as soybean (see tables 9, 10, 11).
Significance in metabolism	Building blocks of proteins, component of enzymes and is present in almost all tissues in animals. Additional metabolic functions, particularly as a precursor of cysteine/cystine and thus also of peptides such as glutathione as an initiator of protein biosynthesis, participates in methyl group donor (S-adenosyl methionine)
Production	By chemical synthesis, starting with propylene, methyl mercaptan, methane and ammonia. The amino acid in its DL-form is 100% availa-
Utilisation	ble since the D-isomer is converted into the L-form by deamination and reamination

4.2.1 Commercially available types

4.2.1.1 DL-methionine

Specification	DL-methionine, technically pure, minimum of 98% DL-methionine	
Chem. name	DL- amino-CH_3-S-$(CH_2)_2$-methyl mercapto butyric acid	
Chem. formular	CH_3S-$(CH_2)_2$-$CH(NH_2)$-COOH	
Technical Data	Molecular weight:	149.2
	Nitrogen content:	9.4%
	Crude protein equivalent:	58.6%
	ME (pig):	22 MJ/kg (5280 kcal/kg)
	ME (poultry):	21 MJ/kg (5020 kcal/kg)
	Purity:	min. of 98.0% DL-methionine
	Solubility:	3.3 g/100 ml water at 20°C
	Stability:	Stable in premixes and mixed feeds

4.2.1.2 DL-methionine-sodium concentrate, liquid

Specification	DL-methionine-sodium concentrate, liquid, technically pure, minimum of 40% DL-methionine minimum of 6.2% sodium	
Chem. formular	CH_3-S-$(CH_2)_2$-CH(NH_2)-COOH	
Technical Data	Molecular weight:	149.2
	Nitrogen content:	3.76%
	Crude protein equivalent:	23.5%
	ME (pig):	8.8 MJ/kg (2110 kcal/kg)
	ME (poultry):	8.5 MJ/kg (2030 kcal/kg)
	Purity:	min. of 40.0% DLmethionine
	Solubility:	Soluble in water
	Stability:	Stable in premixes and mixed feeds

4.2.1.3 Other commercially available types

Protected methionine

For cattle, sheep and goats with a functional rumen chemically or physically protected forms of DL-methionine are available. (See chapter 3.7)

4.3 Threonine (Thr)

General occurrence	Animal proteins are relatively rich in threonine, whilst plant pr oteins tend to be deficient in threonine (see tables 9, 10, 11).
Significance in metabolism	Important building block in the synthesis of proteins, component of digestive enzymes and immuno substances, plays an important role in energy metabolism, precursor for glycine synthesis.
Production	Produced by microbial fermentation.
Utilisation	L-threonine is completely utilised but D-threonine is biologically not available
Specification	L-threonine, technically pure, minimum of 98% L-threonine
Chemical name	L- amino--hydroxy butyric acid
Chemical formula	CH_3-CH(OH)-CH(NH_2)-COOH

Technical Data	Molecular weight	119.1
	Nitrogen	11.8%
	Crude protein equivalent:	73.7%
	Purity:	min. of 98.0% Lthreonine
	ME (pig)	15.5 MJ/kg (3700 kcal/kg)
	ME (poultry)	14.6 MJ/kg (3490 kcal/kg)
	Solubility:	9 g/100 ml water at 20C
	Stability:	Stable in premixes and mixed feeds

4.4 Tryptophan (Trp)

General occurrence Most plant proteins, above all soybean protein are rich in tryptophan. Corn protein in addition to animal meal and meat and bone meal are extremely low (see tables 9, 10, 11).

Significance in metabolism Protein building blocks involved in the formation of the precursors of NAD (nicotinic acid amide-adenine-dinucleotide) plus many metabolic processes via the tissue hormone serotonin and tryptamine. Tryptophan promotes feed intake.

Production Produced via fermentation by micro-organisms; raw materials molasses, sugar, products containing starch and their hydrolysates plus sources of N.

Utilisation L-tryptophan is completely utilised.

Specification L-tryptophan, technically pure, minimum of 98% L- tryptophan

Chemical name L- - amino-indolyl propionic acid

Chemical formula $(C_8H_5\text{-}NH)\text{-}CH_2(CH)\text{-}NH_2\text{-}COOH$

Technical Data

Molecular weight:	204.2
Nitrogen content:	13.7%
Crude protein equivalent:	85.7%
Purity:	min. of 98.0% Ltryptophan
ME (pig):	25.0 MJ/kg (5970 kcal/kg)
ME (poultry):	23.9 MJ/kg (5710 kcal/kg)
Solubility:	1 g/100 ml water at 20C
Stability:	Tryptophan is sensitive to exposure to light and oxidation as well as to acid. Stable when protected against exposure to light and air.

4.5 Hydroxy analogue of methionine

General occurrence	Metabolic product of micro-organisms in fermentation processes, for example in silage as well as in by-products of the fermentation industry.
Production	By chemical synthesis starting with acrolein, methyl mercaptan and hydrogen cyanide.
Chemical name	DL-2-hydroxy-4-methyl mercaptan butyric acid (monomeric acid)
Chemical formula	CH_3-S-$(CH_2)_2$-CH(OH)-COOH
Commercially available type	DL-2-hydroxy-4-methyl mercapto butyric acid, liquid

Technical Data

Molecular weight:	150.2
Purity:	Total acid min. of 88%; monomeric acid a min. of 65%
ME (pig):	16.9 MJ/kg (4039 kcal/kg)
ME (poultry):	16.1 MJ/kg (3847 kcal/kg)
Solubility:	Completely soluble in water
Stability:	Stable in premixes and mixed feeds

Significance and utilisation

The hydroxy analogue of methionine is a precursor of methi onine. The DL-proportions of the hydroxy analogue of methionine are converted into the L-form of methionine. Details with respect to the biological availability are available in specific literature and from the manufacturers.

5. Ecological aspects of amino acid usage

In the past, increasing productivity has been the prime issue with respect to animal production. However more recently aspects related to sustainability and environmental issues have taken a more prominent role. In regions and countries where there is high density of animal production the need to reduce emissions from agriculture cannot be disputed. The use of amino acids in diets of livestock can make a considerable positive contribution to promoting environmentally sensitive feed management practices. Furthermore the use of phase feeding using amino acid supplementation offers the opportunity to match nutritional requirements to livestock production needs thereby in a cost effective manner offering positive benefits for the environment.

Reduction of nitrogen pollution

Feed management using amino acid supplementation can potentially reduce nitrogen excretion by up to 40% without any detrimental effects on the yield of livestock products.

Inproved animal health

A diet, well balanced to the animals needs reduces the metabolic burden on the animal. In young animals this can result in a lower incidence of digestive malfunction and lower incidence of diarrhoea. Older animals may also be healthier and less susceptible to infection using feeding regimens accurately formulated to needs.

Reduction of noxious gases

Trials have demonstrated major improvements in air quality in animal housing from using feed management programs in which the nutrient supply from amino acids and protein accurately meets livestock needs. The ammonia concentration in the air of livestock housing can be reduced and furthermore the release of ammonia during the storage of slurry and nitrogen losses during and following the removal of slurry can be reduced.

Improved working conditions

Reduction in the concentration of noxious gases such as ammonia, in livestock housing can have major benefits for the health of farmers and employees who work in such environments.

Protection of human food resources

The use of amino acids in feed formulation results in the saving of fish stock in the oceans (1kg methionine corresponds to 230 kg fish). To cover the requirements in terms of methionine by using fishmeal, more than 50% of the fish caught world-wide would beneeded. A decision not to use synthetic lysine in a region such as the European Union would result in an increase in the demand for soybean meal of approximately 3 million tons per annum. For the production of this amount of soybeans about 1.4 million ha

arable land would be required.
There is no physiological difference in animal nutrition between the supply of amino acids produced from either microbiological or chemical processes or from amino acids supplied from dietary protein. The provision of amino acids in sufficient quantity and in standardised concentration permits the adjustment of the spectrum of dietary amino acids in the ration to the nutritional requirements of the animal in a cost effective manner.

Amino acids are stable during normal feed processing and are not adversely effected by the normal moisture content, heat, light and pH of the feed. There is no major affect on shelf life from association with other feed ingredients, such as minerals and trace elements or from processing such as ›molassing‹, compacting, pelleting and preserving. The feed regulatory issues concerning industrially produced amino acids are covered by EU legislation for feed additives.

The particle size distribution and the surface structure of amino acids permits ease of handling during feed production and additions of amino acids to complete diets can normally be made directly. Small volume additions should be made via incorporation in premixes or mineral supplements such that the amino acid at the intended dose level can be evenly distributed in the mixed feed. Liquid products can generally be dosed directly into the feed.
Two additional aspects need to be considered when characterising feedstuffs according to German feedstuff legislation. Industrially produced amino acids are considered as individual feedstuffs and as such must be identified according to their composition. When declaring the composition of feedstuffs the proportions that are present naturally in the diet and those which are added as supplements must be given as a sum total. In the case of the methionine hydroxy analogue it should be noted that the proportion of monomeric acid and the total acid must be indicated in addition to stating the methionine content from the raw material.

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