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# PROTEIN AND AMINO ACIDS

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The daily protein requirement of the horse is a function of the endogenous nitrogen loss and the nitrogen deposition and secretion by the animal. Since the horse has only minimal ability to utilize non-protein nitrogen, most of the animal's nitrogen needs must be provided in the form of protein or amino acids. Like other animals, the horse has a need for specific quantities of essential amino acids and a need for a pool of nonessential amino acids. The dietary protein requirement of the animal is a function of the amino acid needs of the animal, the amino acid composition of the dietary protein, the digestibility of the protein and perhaps the restructuring of the amino acid profile of some of the protein in the hindgut of the animal.

# NRC (1989) Recommendations

With the exception of the lysine requirements of the growing horse, the protein requirements are expressed as crude protein requirements. In most situations, the crude protein needs of the animal were determined directly from the literature or from calculations from information on the digestible protein needs.

*Maintenance* - The daily protein requirement for a horse at maintenance is primarily a function of endogenous nitrogen loss. Data in the literature indicate that this is 0.49 to 0.68 g of digestible protein (DP)/kg BW daily. The committee selected 0.60 g DP/kg BW as appropriate for most horses. If we assume that maintenance horses are likely going to sustain themselves on grass pasture or grass hay and that the digestibility of that forage is 46%, the crude protein requirement of the animal is calculated by dividing the 0.60 g DP by 0.46. This gives a value of 1.30 g CP/kg BW or 650 g CP daily for a 500 kg horse. The requirement can also be expressed as a function of the energy needs of the animal and would be 40 g CP/Mcal DE/daily. There was no information available on the amino acid requirements for maintenance although Slade et al. (1970) demonstrated that the nitrogen requirements of horses at maintenance could be satisfied with a lower nitrogen intake from fish meal than from corn gluten meal. He attributed this primarily to the increased digestibility of the fish meal but the response could have also been due to the amino acid composition of the protein.

*Reproduction* - The protein requirements for maintenance were considered adequate for early gestation. This is because 60 to 65% of the fetal development takes place during the last 90 days of gestation. This means that only 35 to 40% of the development takes place during the first 8 months. This would mean that a 500 kg mare with a requirement of 300 g DP/d would deposit about 5 g DP in the fetus daily during early gestation. It would seem that protein deposition during early gestation is of little significance to the mare, especially when we consider



237

that the typical feeding program of 10 kg grass forage containing 8% CP would provide at least 368 g DP daily without supplementation. During late gestation the protein requirement increases due to the more rapid deposition of fetal tissues. During this time the mare will deposit about 26 g DP daily or about a 9% increase in her needs. This was handled by increasing the CP/Mcal DE from 40 to 44 and is probably quite liberal.

The protein requirement of the lactating mare is maintenance plus the protein required for milk production. This was calculated by dividing the protein content of the milk by the efficiency with which the animal converted dietary protein to milk protein and multiplying that value by the milk production of the animal. The efficiency with which dietary protein is converted to milk protein was estimated from dairy cattle data. Production figures were obtained from the literature and were blocked into early (3% BW/d) and late (2% BW/d) lactation even though the lactation curve is fairly well documented.

*Growth* - Based on numerous feeding trials where energy and protein intakes could be calculated, the DE requirements of weanlings and yearlings were estimated by the equation:

DE (Mcal/d) = (1.4 + 0.03 BW) + (4.81 + 1.17x - 0.023x2)(ADG)where x = age in months and ADG is in kg/d

Based on data from at least ten feeding trials with growing horses it was concluded that there was a relationship between protein and energy needs for growth. This relationship was calculated to be 50 and 45 g CP/Mcal DE daily, for the weanlings and yearlings, respectively. Some of these same experiments allowed the calculation of the lysine requirements which were estimated to be 2.1 and 1.9 g lysine/Mcal DE daily, respectively. Based on these recommendations lysine needs to be at least 4.2% of the protein for growing horses for maximum efficiency.

*Work* - The protein requirement of the working horse is influenced by muscle hypertrophy as the animal trains, increased muscle protein content and perspiration loss. Sweat contains 1.0 to 1.5 g N/kg sweat and horses can lose as much as 5 kg sweat/100 kg BW daily; therefore a 500 kg horse could lose 234 g protein daily in the sweat (25 kg x 1.5 g/kg x 6.25). The recommendation for the working horse is to keep the CP/Mcal DE ratio constant at 40 g CP/Mcal DE. Doubling the energy expenditure would double both the energy requirement and the CP intake. A 500 kg horse would therefore consume 656 g CP at maintenance and 1312 g CP at intense work. This increased intake should more than meet the added protein needs of the working horse.

# **Research since the NRC (1989) Publication**

*Maintenance* - There is little new information on the protein-amino acid requirements of horses at maintenance. This suggests that there is a general



acceptance of the recommendations at the DP level. Some may question whether the 46% digestibility assumed by the NRC (1989) is applicable to all maintenance situations. Additional information is available on the digestibility of forages. Lieb et al. (1993) determined the digestibility of four hays: rhizoma peanut (15.9% CP), alfalfa (19.7% CP), Coastal bermudagrass (14.4% CP), and bahiagrass (7.1% CP). The apparent digestibilities of the four were 70, 79, 63 and 25%, respectively. Dugan et al. (1993) compared Coastal bermudagrass (14.8% CP) and flaccidgrass (8.1% CP) and found digestibilities of 68.4 and 53.9%, respectively. In a study of four bermudagrass varieties containing 10.3 to 11.1% CP (McCann et al., 1995) the protein digestibility was 63.5 to 66.7%. Comparisons of the protein digestibilities of Coastal bermudagrass (8.28% CP), Matua (10.90% CP), and alfalfa (16.44% CP) by Sturgeon et al. (2000) found digestibilities of 60.56, 64.11, and 76.38%, respectively. These studies verify that low protein forages have low digestibility but suggest that moderate protein forages are much better protein sources than NRC (1989) assumed. The above data are plotted in Figure 1 to illustrate the relationship between protein content of the forage and its apparent protein digestibility.

Figure 1. Relationship of hay protein to protein digestibility Y=2.01x+33.15, r=.51



The apparent total tract digestibility does not tell the whole story. The amount of protein that is digested in the foregut greatly influences the amino acid availability to the animal. Thus feeding programs that provide protein in a form that is readily digested in the foregut will have higher value to the animal than those programs where a considerable portion of the protein is digested in the hindgut. This may be especially true for combinations of forage and concentrate. If the concentrate provides sources of highly digestible protein, the digestibility will be considerably higher than shown above. Farley et al. (1995) have demonstrated that for semi-purified diets providing 5, 9.5,14 and 16.5% CP where soybean meal provided all of the supplemental protein, the apparent digestibilities were 62.9, 78.8, 85.8, and 87.4% and the precedult true digestion was 72%.



Gestating and lactating mares - The requirements for protein for the gestating and lactating mare have been calculated by the NRC (1989) based on tissue deposition during gestation and milk composition and production during lactation. Not many feeding trials are available to verify or modify those calculations. Glade and Luba (1990) fed mares one of two programs during the last week of gestation and for seven weeks of lactation. One group of mares was fed a complete pelleted feed containing 17.92% CP at 1.25% BW/d or a mixture of 92% of the complete pellet and 8% soybean meal fed at 1.20% BW/d. The two diets provided similar energy intakes but the soybean meal supplemented diet provided slightly more protein and a different amino acid mixture. Milk from the mares collected on day seven was higher in alanine, aspartic acid, glutamic acid, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, proline, serine, threonine, tyrosine, and valine than milk from unsupplemented mares. Plasma from the foals from mares fed soybean meal was higher for histidine, leucine, lysine, methionine, and valine than the plasma from unsupplemented mares, resulting in increased foal growth based on increased height gain. This work suggests that the lactating mare will respond to amino acid availability by altering milk composition and that the foal will respond to this improved amino acid balance by increasing growth.

Growing horses - Although it is obvious that the protein requirement of the growing horse is influenced by the amino acid content of the protein, we still have very little information on the amino acid requirements. Saastamoinen and Koskinen (1993) demonstrated that weanling foals 7 to 12 months of age would respond to protein quality. They supplemented a basal diet with milk protein or barley protein and measured increased weight gain (P < .01) and heart girth (P < .05) by the milk supplemented foals. Graham et al. (1994) provided evidence that for diets composed of 40% Coastal bermudagrass hay and 60% concentrate, threonine is the second limiting amino acid for yearling horses. This work verified that the yearling needs 1.9 g lysine/Mcal DE and demonstrated that 1.7 g threonine/Mcal gave greater growth than 1.5 g threonine/Mcal. Similar response was reported by Staniar et al. (1999). They supplemented mares and foals with either a 14% CP concentrate or a 9% CP concentrate with 0.6% added lysine and 0.4% added threonine. The foals on both programs grew at the same rate. The NRC (1989) recommendations for protein and lysine were verified in a study by Coleman et al. (1997). They fed weanlings a 60:40 alfalfa hay:concentrate diet providing NRC (1989), 109% NRC and 112% NRC recommendations for protein and lysine. The diets provided 48.5, 52.4 and 57.3 g protein/Mcal and 1.99, 2.24, and 2.58 g lysine/Mcal. They concluded that the NRC (1989) provided adequate protein and lysine to support maximum growth under this type of feeding system. The foals grew at 0.72 kg/d. A similar trial by Wall et al. (1997) also found no difference in growth response of yearlings fed 40:60 ratios of hay to concentrate using bermudagrass hay supplemented with soybean meal vs alfalfa based diets. The rate of gain on these animals was 0.44 kg/d, probably because the animals were older and they had limited access to the feed. In this study the animals consumed only 1.9 % BW of DM daily. The results suggest that even though the animals on



the soybean meal supplemented diet had greater nitrogen retention, the two protein sources were similar in their ability to support growth.

*Working horses* - The effect of exercise on protein requirements has been investigated by several groups. Pagan et al. (1997) demonstrated that exercise would reduce DM digestibility but had no effect on protein digestibility. Graham-Thiers et al. (1999) compared a 7.5% CP diet with added lysine and threonine to a 14.4% CP diet for working horses. The low protein diet seemed to have little or no adverse effect on the physiological factors related to energy or protein metabolism, suggesting that the amino acid supplemented low protein diet was adequate to meet the needs of the animals. High protein intakes appear to have little effect on the metabolism of the working horse. The extra amino acids are apparently deaminated as blood urea-N was higher in the animals receiving the high protein diet but blood ammonia levels were not affected. Blood lactate levels were depressed by the high protein diet suggesting that the high protein diet depressed glycogen availability or that glycogen was spared by the availability of amino acids that were used directly in energy metabolism. Blood alanine was lower in the horses on the high protein diet.

Amino acids are known to play an important role in energy metabolism in working muscle. The branched chain amino acids leucine, isoleucine and valine provide a source of energy that should reduce lactic acid production during strenuous work. Glade (1989) demonstrated that the administration of 25 g of a mixture of leucine, isoleucine, valine, glutamine and carnitine in a ratio of 7:5:5:0.2:0.1 before and after strenuous exercise for 9 weeks resulted in a measurable decrease in plasma lactate (P<0.05) and decreased heart rate for horses doing treadmill work. Horses exercised to fatigue were shown to have a small increase in muscle leucine (P < 0.05) and lysine (P < 0.05), a larger increase in alanine (P < 0.001) and a decrease in glutamate (P < 0.001)(Miller-Graber et al., 1990). Glutamate plays a key role in intermediary metabolism by providing  $\alpha$ -ketogluterate for the tricarboxylic acid cycle and it can be transaminated to alanine. Alanine can be transported to the liver where it is converted to pyruvate and ultimately glucose. It is therefore likely that as animals approach fatigue, amino acids may play a key role in meeting substrate needs.

This need may, however, be met by adequate amounts of high quality protein. The administration of supplemental branched chain amino acids, L-alanine, L-leucine, isoleucine, and L-valine one hour before training of Standardbred racehorses had no measurable beneficial effect on energy metabolism in the animals (Stefanon et al., 1999). Likewise, horses fed branched chain amino acids before and after treadmill training, three times each week provided no evidence of benefit when compared to unsupplemented animals (Casini et al., 1999).

#### Formulating to Meet the Protein and Amino Acid Needs of the Horse

It is evident from the above data that the protein requirement of the horse is influenced by how well the protein provides for the amino acid needs of the animal. It appears that lysine and threonine are the first limiting amino acids in



grass-based feeding programs, but we have little or no data on the horse's needs for, or the availability of, the other essential amino acids. This presents a special challenge when formulating diets since the amino acid composition of typical feed ingredients used in horse feed varies considerably. If we examine the amino acid requirements of growing swine (NRC, 1999) we find a recommended relationship between lysine and the other essential amino acids. It should be noted that the amino acid content of the typical swine diet may vary considerably from the ideal diet. A comparison of this relationship with the same information for the growing foal gives us an insight into some of the challenges facing scientists as we seek to refine the protein and amino acid requirements of the horse. In general, the current recommendation for lysine is considerably lower, as a percent of the total protein, for horses than for swine. This leads to considerable differences in the lysine to other essential amino acid ratios in typical horse diets compared to swine recommendations. This variation can either frustrate us or challenge us to gain more information on the amino acid needs of the horse.

It appears from available data that although all horses may have specific requirements for amino acids, the growing foal is most sensitive to this balance. Using the NRC (1989) recommendations, the yearling (12 months) weighing 325 kg and growing at 0.65 kg/d will require 21.3 Mcal DE, 956 g protein and 40 g lysine. If we feed the yearling 40% Coastal bermudagrass hay (1.8 Mcal DE.kg, 8.5% CP, and 0.3% lysine, as fed) and 60% concentrate (3.0 Mcal DE, 14% CP and 0.60% lysine, as fed), we will meet the animal's requirements for all three nutrients if he consumes 8.45 kg/d (2.6% BW). This program exceeds his protein requirement but just provides his lysine needs. However, by adding L-lysine to the concentrate we could provide this animal with a concentrate containing the same energy and lysine content but only 12% CP (Table 1). This would drop his CP intake to 66 g below his requirement (NRC, 1989) and still provide adequate lysine and enough protein and lysine to achieve the same gain as the 14% protein product will support. The advantage of this approach includes 1) more accurate formulation to meet the animal's amino acid needs, 2) lower protein content for those horsemen concerned about high protein diets, 3) possible economic advantage, and 4) lower nitrogen excretion and less nitrogen load on the environment.

Based on the data available on working horses cited above, we may be able to apply this same approach to mature horses.



ay:Conc.	DE	СР	Lysine	Threonine
	Mcal/kg	%	%	%
33:67	3.0	16.7	0.79	0.60
33.67	3.0	14.0	0.79	0.60
40:60	3.3	15.5	0.88	0.67
40:60	3.0	14.3	0.62	0.52
40:60	3.0	12.0	0.62	0.52
46:54	3.3	13.3	0.69	0.58
	33:67 33.67 40:60 40:60 40:60 46:54	Ay:Conc.         DE Mcal/kg           33:67         3.0           33.67         3.0           40:60         3.3           40:60         3.0           40:60         3.0           40:60         3.0           46:54         3.3	ay:Conc.DE Mcal/kgCP % $33:67$ $3.0$ $16.7$ $33.67$ $3.0$ $14.0$ $40:60$ $3.3$ $15.5$ $40:60$ $3.0$ $14.3$ $40:60$ $3.0$ $12.0$ $46:54$ $3.3$ $13.3$	Ay:Conc.DE Mcal/kgCP %Lysine % $33:67$ $3.0$ $16.7$ $14.0$ $0.79$ $0.79$ $15.5$ $40:60$ $3.3$ $15.5$ $0.88$ $40:60$ $3.0$ $12.0$ $14.3$ $0.62$ $0.62$ $13.3$ $0.62$ $0.69$

 Table 1. Nutrient recommendations for concentrates for growing horses (as fed).

<sup>1</sup>Alternate Conc. 1 has 0.2% added lysine and 0.1% added threonine.

<sup>2</sup>Alternate Conc. 2 has 5% added fat and will need to have about 10% higher concentrations of nutrients.

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