

## Amino acid requirements of the breeding sow: the dietary lysine requirement during pregnancy

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*(Received 10 June 1977 – Accepted 13 July 1977)*

1. Four pregnant sows were used to study lysine requirements by means of the interpretation of values for plasma amino acid and blood urea. Diets containing graded levels of dietary lysine were fed at the rate of 1.82 kg/d.
2. Plasma lysine remained at a low level up to 6.4 g dietary lysine/kg (dry matter basis) and then increased sharply with further increases in lysine intake.
3. Blood urea levels decreased when the dietary lysine content was increased to 6.4 g/kg and then increased at higher dietary lysine concentrations.
4. Both criteria of response indicated that the lysine requirement of the pregnant sow given 1.82 kg diet/d during the later stages of pregnancy does not exceed 10.00 g/d.

Amino acid nutrition of the breeding sow has been the subject of little research work. As a consequence of the move to reduce food and protein allowances for breeding sows there is a need to identify clearly the requirements for individual amino acids.

The lack of information on the amino acid requirements of the breeding sow, compared with the growing pig, is probably a reflexion of the difficulty of the problem. Criteria of performance in the growing pig, i.e. growth rate, food utilization and carcass quality are easily measured. In the breeding sow the end-products of protein nutrition are less clearly defined and less easily measured. In an attempt to achieve progress in knowledge concerning the amino acid nutrition of the breeding sow, it would be desirable to employ techniques of short duration that would indicate the requirements of an amino acid with an extent of reliability. Examples of such techniques are the measurement of plasma amino acids and blood urea.

The concept of the use of plasma amino acid concentration is that the blood is an 'amino acid pool' with inputs resulting from dietary amino acid and outputs being a consequence of the requirements of the animal. The relationship between dietary level and requirement is the determinant of the level of a particular amino acid in the plasma. Thus under normal physiological conditions, when the dietary level of an amino acid exceeds the requirement of the animal its level in the plasma would be greatly increased, while a deficiency of a dietary amino acid would be reflected by its reduced concentration. Thus, use of sequential increments of a dietary amino acid should allow the requirement to be estimated through changes in plasma concentration. A point of inflexion could be expected in the plasma amino acid curve as the level of dietary need is exceeded. This approach has been used successfully by Bravo, Meade, Stockland & Nordstrom (1970), Keith, Christensen & Owen (1972), Holden, Ewan & Speer (1971), Mitchell, Becker, Jensen, Harmon & Norton (1968).

A further approach is the measurement of blood urea levels. At a fixed intake of dietary protein an improvement of amino acid balance would result in a better utilization of dietary protein with a consequent decrease in blood urea levels. Further increases of an amino acid

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Table 1. *Composition and analysis of the diet fed to pregnant sows*

Ingredient (g/kg)		Chemical analysis (g/kg dry matter (DM))	
Ground barley	496.3	Crude protein (nitrogen $\times$ 6.25)	120.0
Ground wheat	496.3	Crude fibre	30.4
Mineral - vitamin supplement*	7.4	Diethyl ether extract	14.9
		Ash	27.1
		Phosphorus	4.4
		Calcium	3.2

Amino acids (g/kg DM): aspartic acid 6.2, threonine 3.5, serine 5.0, glutamic acid 22.9, proline 9.9, glycine 4.4, alanine 4.1, valine 4.7, cystine 2.9, methionine 1.4, isoleucine 3.9, leucine 6.9, tryptophan 1.1, phenylalanine 5.2, lysine 3.2, histidine 2.6, arginine 5.2, tryptophan 1.1.

\* Supplement provided (per kg diet): retinol 1.8 mg, cholecalciferol 30 mg,  $\alpha$ -tocopherol 6 mg, menaphthone 1.2 mg, riboflavin 2.4 mg, cyanocobalamin 6 mg, nicotinic acid 7.2 mg, pantothenic acid 6 mg, iron 60 mg, cobalt 0.6 mg, manganese 24 mg, copper 6 mg, zinc 4.2 mg, iodine 1.8 mg, Ca 2.1 g, sodium chloride 1.5 mg.

would eventually lead to an excess, a worsening of the amino acid balance and as a result the excess would be catabolized and excreted as urea and a small increase in the plasma levels might result. Both these techniques are of value only when used at levels of dietary input where a response may be achieved.

The object of this study was to determine the lysine requirement of the sow, in later stages of pregnancy, by means of interpretation of values for plasma amino acid and blood urea level.

The approach taken was to use a small number of animals because of techniques involved, but to use an experimental design which allowed each treatment to be applied to each animal.

#### MATERIALS AND METHODS

*Animals and treatments.* Four Landrace  $\times$  (Landrace  $\times$  Large White) pregnant sows, of 3rd and 4th parity, were selected on the 75th day of pregnancy and housed in metabolism crates. They were offered a basal diet, the composition of which is given in Table 1, which contained 3.4 g lysine/kg (dry matter (DM) basis). The basal diet was supplemented with L-lysine hydrochloride to achieve seven levels of dietary lysine (g/kg): 3.4, 4.4, 5.4, 6.4, 7.4, 8.4 and 9.4, which resulted in daily lysine intakes of (g): 5.31, 6.87, 8.44, 10.00, 11.56, 13.12 and 14.68 respectively. The sows were fed once daily in the morning and each sow received 1.82 kg diet/d. Water was provided *ad lib*.

Each treatment was offered for a period of 3 d and each sow received each treatment in a randomized manner. Blood samples were obtained on day 1 and day 3 of each treatment period, at 1 h and 4 h after feeding. The experiment lasted for 21 d from day 85 to day 106 of pregnancy.

The results were evaluated by analysis of variance for a 'split-split plot' design: dietary treatments being 'main plots', period of treatment being 'subplots' and sampling period being 'sub-subplots'. A missing value was calculated for one sow on one treatment.

*Collection of blood samples.* Blood samples were withdrawn from the jugular vein by means of an intravenous cannula through the medial ear vein. The cannula was inserted by the method of Seldinger (1953) using a 'guide' wire of the type described by Verel (1966, 1967). Samples were collected into a cooled heparinized glass vial and centrifuged (MSE Super Minor; Measuring and Scientific Equipment, London SW1) immediately at 4000 rev./min for 10 min at 5°. The separated plasma was then deproteinized with sulphosalicylic acid (Hamilton, 1962).

*Blood urea.* Urea was measured by a microdiffusion method as described by Conway & O'Malley (1942) and revised by Conway (1962).

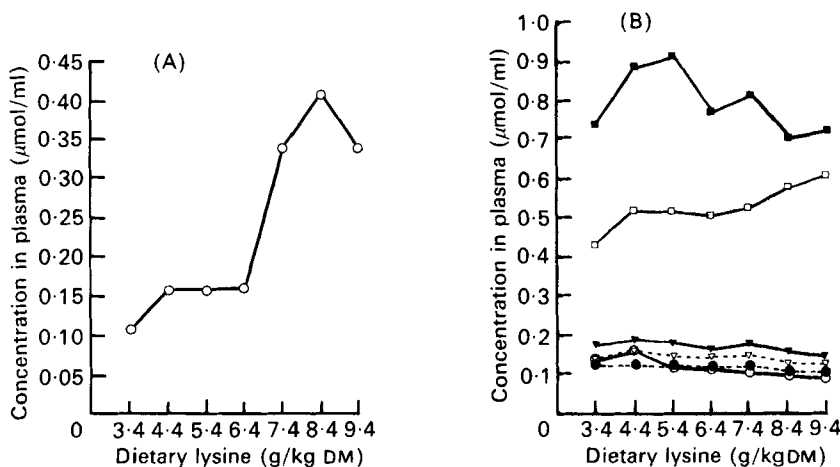


Fig. 1. The effect of graded levels of dietary lysine (g/kg dry matter (DM)) on the concentration of amino acids ( $\mu\text{mol/ml}$ ) in the blood plasma of pregnant sows. (A):  $\circ$ — $\circ$ , lysine; (B):  $\blacksquare$ — $\blacksquare$ , glycine;  $\square$ — $\square$ , alanine;  $\blacktriangledown$ — $\blacktriangledown$ , leucine;  $\nabla$ --- $\nabla$ , serine;  $\circ$ — $\circ$ , threonine;  $\bullet$ --- $\bullet$ , isoleucine.

*Amino acid analysis.* Amino acids in the plasma, with the exception of tryptophan, were analysed by the method of Moore, Spackman & Stein (1958) and Spackman, Stein & Moore (1958) using ion-exchange column chromatography in the form of an amino acid analyser (EEL-294 Amin-AL; Evans Electroelenium Ltd, Halstead, Essex). Evaluation of chromatograms was carried out by the method of Back, Buttery & Gregson (1972)

The determination of tryptophan in the plasma was carried out according to the method of Denckla & Dewey (1967), with minor modifications (Buttery & Soar, 1975).

## RESULTS

### *Plasma amino acids*

There were no significant differences between values obtained from samples on day 1 or day 3. There were significant differences between sampling times; the sample taken 1 h after feeding giving higher plasma amino acid levels than the sample taken 4 h after feeding. Interpretation of the results was the same regardless of which sample was taken or if the mean of all samples was used.

The effect of dietary lysine on the concentration of plasma amino acids is shown in Fig. 1. On the basis of the mean values of both sampling times on both days, plasma lysine maintained an almost constant level as dietary lysine was increased to 6.4 g/kg DM and then increased sharply with increase in lysine intake (Fig. 1). Only three other essential amino acids (threonine, leucine and isoleucine) were affected by the level of dietary lysine intake; they all decreased slightly as dietary lysine increased (Fig. 1)

### *Blood urea*

Urea levels in samples of blood taken 1 h after feeding were significantly ( $P < 0.001$ ) lower than samples taken 4 h after feeding (Table 2 and Fig. 2). However, the pattern of blood urea level with change in dietary lysine intake was similar for both sampling times. There was a decrease in blood urea level as dietary lysine was increased to 6.4 g/kg DM. As dietary lysine intake was increased above this, blood urea level increased.

Table 2. *Effect of graded levels of dietary lysine on blood urea levels (mmol/l) in pregnant sows*

(Mean values for four sows/treatment)

Dietary lysine (g/kg dry matter)	Sampling interval (h)		Treatment mean
	1	4	
3.4	2.82	3.28	3.05
4.4	2.59	3.00	2.80
5.4	2.49	2.85	2.67
6.4	2.31	2.74	2.53
7.4	2.74	3.06	2.90
8.4	3.18	3.39	3.28
9.4	3.04	3.54	3.29
Interval mean	2.74	3.12	

SE for comparison within treatment 0.066, SE for comparison between treatments 0.145.

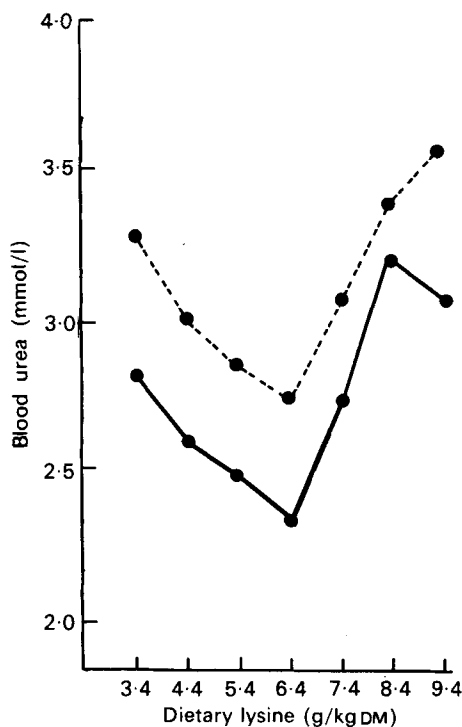


Fig. 2. The effect of graded levels of dietary lysine (g/kg dry matter (DM)) on the concentration of blood urea (mmol/ml) in pregnant sows. Blood samples were taken at 1 h (●—●) and 4 h (●---●).

#### DISCUSSION

If a diet containing all essential amino acids, except one, in adequate amounts is supplemented with increments of the limiting amino acid the plasma level of the supplemented amino acid could be expected to increase markedly when the requirement of that amino acid was exceeded. The point of inflexion in the response curve provides a measure of the needs of that dietary amino acid. The inflexion point of the plasma lysine response curve

Table 3. Estimates of lysine requirements of pregnant sows based on various criteria

Author	Lysine (g/d)	Food intake (kg/d)	Criteria
Allee & Baker (1970)	9.80	2.00	Nitrogen retention
Hesby, Conrad, Plumlee & Martin (1970)	10.88	2.22	N retention and reproductive performance
Hesby, Conrad, Plumlee & Harrington (1970)			
Miller, Becker, Jensen, Harmon & Norton (1969)	12.16	1.90	N retention
Ripple, Harmon, Jensen, Norton & Becker (1965)	7.64	1.82	N retention
Duée (1972)	8.36	1.90	N retention
Present study	10.00	1.82	Plasma lysine and blood urea level

(Fig. 1) showed a close agreement with the trough in blood urea (Fig. 2), which indicated that the lysine need of the pregnant sow does not exceed 10.00 g/d (6.4 g/kg dietary DM).

A limited number of studies have been undertaken to determine the lysine requirement of the pregnant sow (e.g. Ripple, Harmon, Jensen, Norton & Becker, 1965; Salmon-Legagneur & Duée, 1972). These studies have used mainly nitrogen balance to estimate lysine requirement, i.e. requirement is represented by maximum N retention. The estimated daily requirements obtained are shown in Table 3. These requirements range from 7.64 to 12.16 g lysine/d and, considering the differences in, for example, diet and feeding level, give reasonable support to the estimate of 10 g lysine/d for the pregnant sow based on plasma lysine and blood urea response in this study.

Among other essential amino acids, the decrease in plasma threonine, isoleucine and leucine with increasing levels of dietary lysine could not be explained with certainty. A similar decrease in plasma threonine was also reported by Zimmerman & Scott (1965) when diets containing graded levels of lysine were fed to chicks. In growing pigs, Pick & Meade (1970) reported that a decrease in plasma threonine was attributed to increased protein synthesis associated with rapid live-weight gains causing a withdrawal of the amino acid from the plasma. In the present study the decrease in threonine, leucine and isoleucine concentrations could be due to the increased demand of the rapidly-growing foetuses for these amino acids.

The authors gratefully acknowledge the support by the Agricultural Research Council during the course of this study. Appreciation is also expressed to Mr H. L. Back for advice on statistical analysis and Mr J. Coote for technical assistance.

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