

FACTORS AFFECTING AMINO ACID  
AVAILABILITIES FOR CEREAL GRAINS  
AND THEIR COMPONENTS FOR GROWING  
MONOGASTRIC ANIMALS

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by  
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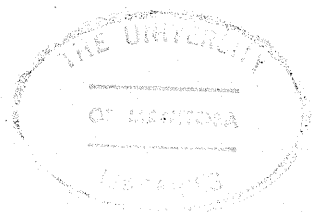
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DEDICATED TO MY DAUGHTER,  
MARGRIET VALENTINA

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## ABSTRACT

The apparent ileal and faecal amino acid availabilities from corn, wheat and barley were determined with barrows ranging in weight from 45 to 75 kg, that were fitted with ileocaecal re-entrant cannulae. The average ileal versus (vs) faecal availabilities were 85.6 vs 89.9, 83.3 vs 89.1 and 77.7 vs 85% for corn, wheat and barley respectively. For the indispensable amino acids, ARG, HIS and THR disappeared to the greatest extent in the large intestine ( $P < .05$ ). For the dispensable amino acids, GLY and PRO disappeared to the largest extent in the large intestine ( $P < .05$ ).

Of the indispensable amino acids, LYS and THR were approximately equal in being least available at the end of the ileum. The ileal LYS vs THR availabilities decreased significantly ( $P < .05$ ) from corn to wheat to barley and were 82.0 vs 78.9, 75.7 vs 76.5 and 73.3 vs 71.2% respectively. Lysine was the least available amino acid when determined by the faecal analysis method. Its availability decreased significantly ( $P < .05$ ) from corn to wheat and to barley and was 83.0, 80.7 and 77.5% respectively.

Faecal amino acid availabilities obtained from rats compared reasonably well with those obtained from pigs for cereal grains of approximately the same protein content of the same variety.

Similarly, apparent ileal and faecal amino acid availabilities from wheat, flour and a diet consisting of

45% bran, 45% shorts and 10% middlings (B+S+M) were determined in pigs. The average ileal vs faecal availabilities were 90.0 vs 94.1, 85.4 vs 92.3 and 72.1 vs 80.4% for flour, wheat and B+S+M respectively. Generally speaking, ARG, HIS THR, GLY and PRO disappeared to the largest extent in the large intestine.

Of the indispensable amino acids, LYS and THR were equal in being least available from flour and wheat. The ileal LYS vs THR availabilities were 84.2 vs 85.4 and 79.5 vs 78.4% from flour and wheat respectively. Lysine was the least available amino acid when determined by the faecal analysis method, namely 86.1 and 86.0% for flour and wheat respectively. Ileal and faecal availabilities of THR in B+S+M were lowest with values of 54.0 and 71.3% respectively.

The apparent ileal and faecal amino acid availabilities from finely ground and cracked wheat were determined with barrows that weighed approximately 75 kg. The average ileal vs faecal availabilities were 87.7 vs 91.6 and 82.8 vs 90.8% for finely ground and cracked wheat respectively. The ileal availabilities of most amino acids were significantly higher ( $P < .05$ ) for finely ground than for cracked wheat, but were not significantly different when determined by the faecal analysis method.

Ileal amino acid availabilities are more biologically meaningful than faecal availabilities. Caecally infused LYS (as free LYS or as part of isolated soy protein) did not significantly

improve the protein retention of barrows, ranging in weight from 25 to 40 kg, that were fed diets low in LYS.

Metabolic ileal and faecal amino acid levels were determined with barrows, ranging in weight from 45 to 75 kg, that were fed 3 protein-free diets containing 5, 10 and 15% Alphafloc respectively. As the level of Alphafloc was increased from 5 to 10 and to 15%, the average ileal vs faecal amino acid levels, expressed as grams per 100 grams of dry matter intake, increased from .072 vs .032 to .091 vs .046 and to .093 vs .050. The average ileal vs faecal nitrogen levels increased from .205 vs .101 to .256 vs .139 and to .271 vs .161. The dispensable amino acids made up 75 to 80% of the total amount of metabolic ileal amino acids, of which PRO and GLY made up 55 and 16% respectively. Arginine, THR, GLY and PRO (especially the last 2 amino acids) disappeared extensively in the large intestine. The levels of ILE, LEU, LYS, MET and ASP increased between the end of the ileum and the anus of pigs fed the protein-free diets.

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LIST OF ABBREVIATIONS  
FOR THE AMINO ACIDS

ALA	Alanine
ARG	Arginine
ASP	Aspartic acid
CYS	Cystine
GLU	Glutamic acid
GLY	Glycine
HIS	Histidine
ILE	Isoleucine
LEU	Leucine
LYS	Lysine
MET	Methionine
PHE	Phenylalanine
PRO	Proline
SER	Serine
THR	Threonine
TRY	Tryptophan
TYR	Tyrosine
VAL	Valine

## INTRODUCTION

The nutritive value of a protein is not only determined by its amino acid composition but also by the availability of the individual amino acids of the protein to the monogastric animal. For more accurate and economical formulation of dietary amino acid levels in relation to requirements, available instead of total amino acid levels, from specific protein sources and from energy feeds that also supply protein, should be taken into account.

The term availability has been defined by de Muelenaere et al. (1967) as that portion of amino acids present in a protein which is used for growth, development and maintenance of an animal insofar as it is dependent on the digestibility of the protein, the presence of enzyme inhibitors and enzyme-resistant peptide linkages, and rate of release of amino acids in the intestinal tract. Amino acid availability defined as such may be determined according to the faecal analysis method, which measures the amounts of ingested amino acids excreted in the faeces.

The faecal analysis method was developed by Kuiken and Lyman in 1948 and has since gained general acceptance in nutritional research. However, in recent years, this method has been often criticized because of possible microbial alterations of undigested and unabsorbed nitrogenous residues in the large intestine.

Holmes et al. (1974) determined the ileal and faecal amino acid availabilities from specific protein sources, namely soybean meal and rapeseed meal using growing pigs fitted with re-entrant cannulae placed at the end of the ileum.

Apart from specific protein sources, cereal grains supply a large proportion of the total dietary protein in practical diets for swine and poultry. In addition, cereal grains are the major source of protein for the human population in many developing countries. Data regarding faecal amino acid availabilities from cereal grains can be found in the literature. However, essentially no information on ileal availabilities, which may be more accurate indices, is available.

The main objective of the studies that were carried out was to determine to what extent ileal and faecal amino acid availabilities from cereal grains differed. It was also attempted to determine which physical and/or chemical fractions were responsible for the discrepancy between total and available amino acid content from the cereal grains. The latter knowledge may be of importance to plant breeders in their quest for improving the protein quality of cereal grains.

The pig was chosen as experimental animal. First of all, this animal can be fitted with re-entrant cannulae. Secondly, it is generally felt that information obtained from the pig is applicable to most other monogastric species.

## REVIEW OF THE LITERATURE

The quality of cereal protein is poor because of the low content of some amino acids that are essential for growth and other forms of production. The limiting amino acids of the major cereal grains fed to pigs and rats are shown in Table 1. References were selected in which both the first and second-limiting (and sometimes third-limiting amino acids) were determined. Lysine and THR were found to be the first and second-limiting amino acids respectively for barley, oats, rice, rye, triticales and wheat. Tryptophan and LYS were found to be the first and second-limiting amino acids respectively for corn fed to growing or finishing pigs (Baker et al., 1969; Gallo and Pond, 1968). The order of limitation of these amino acids was reversed when corn was fed to baby pigs (Gallo et al., 1971). Due to different amino acid requirements for growth and maintenance (McLaughlan, 1972), factors such as the level of protein tested and the age of the animal may determine which amino acid of 2 nearly equally limiting amino acids will be the most limiting. The sulfur containing amino acids have been shown to be third-limiting for corn, oats, and triticales (Table 1).

Lysine, in addition to being usually the most limiting amino acid for pigs and rats, was also found to be the least

TABLE 1.

The limiting amino acids for rats and pigs when cereal grain supply the only protein.

ITEMS	First-Limiting Amino Acid	Second-Limiting Amino Acid	Third-Limiting Amino Acid	Species	References
<u>CEREAL GRAIN</u>					
Barley	LYS.	THR.		rats	Howe <u>et al.</u> , 1965.
Corn	TRY.	LYS.		pigs	Baker <u>et al.</u> , 1969;
	TRY.	LYS.	MET.(CYS)	pigs	Gallo and Pond, 1968;
					Oestemer <u>et al.</u> , 1970.
Oats	LYS.	TRY.		pigs	Gallo <u>et al.</u> , 1968.
	LYS.	TRY.		rats	Pond <u>et al.</u> , 1971.
	LYS.	TRY.	MET.(CYS),THR., ILE., VAL.	rats	Rosenberg <u>et al.</u> , 1960.
	LYS.	THR.	MET. (CYS)	rats	Tang <u>et al.</u> , 1958.
	LYS.	THR.		rats	Howe <u>et al.</u> , 1965.
Rice	LYS.	THR.		rats	Rosenberg <u>et al.</u> , 1959.
Rye	LYS.	THR.		rats	Howe <u>et al.</u> , 1965;
Triticale	LYS.	THR.	MET. (CYS)	rats	Kihlberg and Ericson, 1964; McLaughlan <u>et al.</u> , 1967.
	LYS.	THR.		pigs	Shumada and Cline, 1974.
Wheat	LYS.	THR.		rats	Shumada and Cline, 1974.
Rye					Howe <u>et al.</u> , 1965;
	LYS.	THR.		rats	McLaughlan <u>et al.</u> , 1967; Rosenberg <u>et al.</u> , 1960.
					Howe <u>et al.</u> , 1965;
					Kihlberg and Ericson, 1964.

available indispensable amino acid from cereal grains when determined by the faecal analysis method (Eggum, 1973; Olsen et al., 1968; Poppe and Meier, 1971; Sauer et al., 1974). The availabilities of THR and MET were also found to be relatively low in comparison to the availabilities of the other amino acids of cereal grains.

However, the validity of the faecal analysis method may be questioned on grounds of the more or less unknown effects of the microflora of the large intestine on protein metabolism and this aspect will be discussed in Part 2 of this literature review. Part 3 will deal with alternate methods for determining amino acid availabilities. Factors in cereal grains, and also of other protein sources that may affect the availabilities of amino acids to the monogastric animal will be discussed in Part 1 of this literature review.

True amino acid availabilities are derived from their apparent availabilities by correcting the latter estimates for the metabolic faecal amino acid excretion. The metabolic faecal amino acid excretion is usually determined from faeces of animals fed protein-free diets. Factors affecting the metabolic faecal amino acid excretion have been extensively reviewed previously (Sauer, 1972) and are summarized as follows: 1) Endogenous protein derived from undigested residues from sloughed-off cells make up a much larger proportion of metabolic faecal protein than protein derived from undigested digestive secretions; 2) The level of sloughed-off cells is mainly

determined by the amount of undigestible dry matter in the diet. It would seem to be advisable to formulate a protein-free diet in such a manner that it contains the same amount of undigestible dry matter as the diet for which one determines the true amino acid availabilities. The latter may be achieved by including a certain amount of fiber in the protein-free diets. The test animals should also be pair-fed the test and protein-free diet in order to guarantee equal intake of undigestible dry matter; 3) The level and type of protein is not thought to affect the metabolic faecal amino acid excretion. Factors affecting the metabolic faecal amino acid excretion will not be discussed in this review of literature although they are frequently referred to in the discussion part of this thesis.



## PART 1.

### Cereal grain composition and factors affecting amino acid availabilities.

Cereal protein can be separated into four major protein types by successive solvent extractions on ground material: 1) water, albumin; 2) salt solutions, globulin; 3) 70% ethanol, prolamins; and 4) dilute alkali (or acid), glutelin. The nutritive value of cereal protein is primarily determined by the amount of prolamins. Prolamins contain only small amounts of LYS. Prolamins make up approximately 50-55, 40-50, 35-45 and 10-15% of the total protein from corn, wheat, barley and oats respectively (Neurath and Bailey, 1954). The protein efficiency ratio of cereal protein in diets fed to rats was 2.1 for oats (which has the lowest content of prolamins) and ranged from 1.4 to 1.7 for the other cereals (Jones et al., 1948).

Prolamins and glutelin constitute the protein from the endosperm (inner) part of the seed. The albumin and globulin fractions are found primarily on the outer part of the kernel. More specifically, these are found in the aleurone cell layer whose cells have a thick fibrous cell wall.

The total protein content of a cereal grain will largely be determined by the climatic conditions under which

it is grown. It is further dependent on the particular variety of cereal grain and on other factors such as, the level of fertilizer application. Unfavourable climatic conditions, such as, inadequate moisture usually result in a decrease in the total amount of endosperm protein. Favourable conditions result in an increase of endosperm protein. Regardless of climatic conditions, the total amount of protein derived from albumin and globulin remains relatively constant. Consequently, protein from albumin and globulin contributes relatively more protein to the total amount of protein from cereal grains when these are grown under unfavourable rather than favourable climatic conditions. Protein derived from albumin and globulin generally makes up a larger percentage of the total protein of a low protein cereal variety than of a high protein cereal variety when grown under similar climatic conditions (Draper, 1973; Sodek and Wilson, 1971). Fertilization increases mostly the endosperm proteins, resulting in a relative decrease in the proportions of albumin and globulin (Abrol et al., 1970)

There are considerable differences between the amino acid composition of the cereal protein fractions. In general, the amino acid composition of a fraction in different species of cereal grains tends to be similar. Therefore, the amino acid content of a cereal grain will largely be determined by

the proportion of each protein fraction it contains. The amino acid composition of the protein fractions from wheat and barley are shown in Table 2. The composition of aleurone protein from wheat would be indicative of the combined composition of albumin and globulin. Albumin and globulin protein from both wheat and barley contain much higher levels of the basic amino acids (ARG., HIS. and LYS.), ALA. and ASP. and much lower proportions of GLU. and PRO. than prolamins and glutenins. Of the endosperm proteins, glutenin contained considerably more LYS. than prolamins especially with regard to barley. Hiproly barley (high LYS. barley) contains relatively more glutenin than prolamins compared to other barley varieties.

The main differences in amino acid composition of the different protein fractions from wheat and barley can also be detected in the protein fractions of corn (Sodek and Wilson, 1971). Robbins et al. (1971) postulate similar differences between the protein fractions from oats.

The disproportionate contribution of amino acids by the different protein fractions is also shown by recalculated data from Horn et al. (1958). They determined the composition of amino acids in wheat products. Hard red winter wheat was experimentally milled. Straight grade flour of 71.5% extraction, 16.9% bran and 7.9% shorts was produced. The data were recalculated in order to obtain some idea about the quantitative distribution of amino acids in the wheat products (Table 3). Nitrogen from bran and shorts made up about 30% of the total nitrogen from wheat but 45%

TABLE 2.

The amino acid composition of aleurone protein, glutenin and prolamin from wheat and those of albumin, globulin, glutenin and prolamin from barley.

CEREAL GRAIN	WHEAT			BARLEY			
	1	2	2	3	3	3	3
FRACTIONS	ALEURONE	GLUTENIN	PROLAMIN	ALBUMIN	GLOBULIN	GLUTENIN	PROLAMIN
AMINO ACIDS <sup>4</sup>							
<u>Essential</u>							
ARG.	7.3	4.1	2.7	13.0	22.0	12.0	6.0
HIS.	3.6	2.3	2.2	4.3	3.1	4.3	2.2
ILE.	2.8	3.7	4.3	4.1	2.2	3.5	3.6
LEU.	5.5	6.6	6.9	5.8	4.1	4.9	3.5
LYS.	4.1	2.3	0.6	7.9	6.3	4.8	0.8
MET.	1.2	1.6	1.4	1.4	0.9	1.1	0.8
PHE.	3.8	4.8	5.5	3.0	2.1	2.7	3.6
THR.	2.9	3.1	2.2	3.4	2.4	3.1	1.9
VAL.	4.9	4.2	4.1	5.8	4.1	4.9	3.5
<u>Non-Essential</u>							
ALA.	4.7	2.8	2.0	7.2	0.7	6.6	2.2
ASP.	7.3	3.7	2.9	8.0	5.6	4.7	1.2
GLU.	15.0	33.2	38.9	8.7	6.8	11.6	23.0
GLY.	--	3.8	1.5	6.7	10.7	5.2	1.7
PRO.	3.6	10.3	13.8	4.2	2.7	6.6	15.3
SER.	4.1	5.4	4.7	4.1	3.9	4.2	3.2
TYR.	2.7	3.6	2.6	2.7	1.5	1.9	1.6

1 Stevens et al., (1963)

2 Ewart (1967)

3 Folkes and Yemm (1956)

4 Expressed as grams per 16 grams of nitrogen.

TABLE 3.

The amino acid levels (%) of wheat and wheat by-products and the distribution of amino acids in wheat by-products.

CEREAL COMPONENTS	WHOLE WHEAT	PRODUCTS FROM 71.5% FLOUR EXTRACTION						D <sup>2</sup>
		BRAN	SHORTS		FLOUR			
	%	%	<sup>1</sup> A	%	<sup>1</sup> B	%	<sup>1</sup> C	%
AMINO ACIDS								
<u>Essential</u>								
ARG.	0.63	1.11	0.19	1.23	0.10	0.51	0.36	46.0
HIS.	0.32	0.44	0.07	0.45	0.04	0.27	0.19	34.4
ILE.	0.62	0.64	0.11	0.75	0.06	0.59	0.42	27.4
LEU.	0.90	0.95	0.16	1.06	0.08	0.84	0.60	26.7
LYS.	0.38	0.64	0.11	0.77	0.06	0.28	0.20	44.7
MET.	0.20	0.21	0.04	0.24	0.02	0.18	0.13	30.0
PHE.	0.71	0.72	0.12	0.78	0.06	0.68	0.49	25.4
THR.	0.43	0.55	0.09	0.63	0.05	0.37	0.27	32.6
VAL.	0.59	0.73	0.12	0.83	0.07	0.51	0.38	32.2
<u>Semi-Essential</u>								
CYS.	0.33	0.29	0.05	0.38	0.03	0.30	0.21	24.2
TYR.	0.36	0.37	0.06	0.44	0.03	0.32	0.23	25.0
NITROGEN	2.19	2.57	0.43	2.82	0.22	2.02	1.44	29.7
DRY MATTER	100.0	100.0	16.9	100.0	7.9	100.0	71.5	24.8

<sup>1</sup> Column A, B and C indicate the contribution of amino acids (grams) from bran, shorts and flour respectively to whole wheat (100 grams).

<sup>2</sup> Column D shows the percentage of amino acids in whole wheat that are contained in bran and shorts.

of the total LYS and ARG was present in these fractions (Table 3; Column D). The relatively high levels of LYS and ARG in bran and shorts reflected the high concentration of aleurone protein in bran and shorts, since most, if not all aleurone protein is found in these fractions (Hinton et al., 1953; Shetler et al., 1947).

Protein in aleurone cells (albumin and globulin) may be of limited digestibility since the thick cellulosic cell wall of these cells is thought to interfere with digestion of the protein that is contained in these cells. A certain amount of this protein may also be tightly bound to the cellulosic matrix of the aleurone cells (Saunders and Kohler, 1972). Saunders et al. (1969) fed bran from hard red spring wheat to chicks. The contents of the lower intestine contained intact cells from the aleurone layer. Digestion of bran consists essentially of the breakdown and absorption of the contents from the aleurone cells which occurs only when the cell walls are ruptured. The number of intact aleurone cells remaining in the lower intestine and in the faeces seemed to be affected by the variety of wheat from which bran was derived and by the physical form in which bran was fed. Pelleted feed resulted in a larger number of ruptured aleurone cells than mash based on microscopic examination of intestinal contents and faeces.

The true amino acid availabilities from corn, wheat and barley fed to growing pigs are shown in Table 4. In general,

TABLE 4.

True amino acid availabilities from corn, wheat and barley  
for growing pigs.

CEREAL GRAINS	CORN		WHEAT		BARLEY	
REFERENCE	1 A	2 B	2 B	3 C	2 B	3 C
AMINO ACIDS (%)						
<u>Essential</u>						
ARG.	93.7	95.2	95.0	94.1	88.9	91.4
HIS.	94.7	93.5	95.9	94.7	87.6	93.2
ILE.	92.0	88.5	90.3	89.2	79.2	84.7
LEU.	94.2	92.8	93.2	91.2	84.1	88.7
LYS.	90.5	89.3	84.1	80.8	72.3	77.1
MET.	92.9	93.6	88.6	89.8	77.5	87.3
PHE.	92.4	92.2	92.0	92.4	80.7	90.1
THR.	90.5	89.8	88.4	87.3	77.8	85.5
VAL.	92.5	89.8	90.8	89.0	82.5	87.6
<u>Non-Essential</u>						
ALA.	91.7	93.0	86.5	84.1	75.5	80.6
ASP.	91.5	91.1	87.1	84.6	77.8	85.4
GLU.	95.3	92.2	97.4	97.0	90.4	93.3
GLY.	88.2	89.6	89.7	88.3	79.1	84.3
PRO.	95.5	--	--	96.3	--	93.2
SER.	93.1	96.8	96.5	92.8	88.1	89.9
TYR.	93.0	93.0	92.5	90.3	83.8	87.6
NITROGEN	--	90.2	91.8	90.9	82.4	85.8

1 Easter (1972)

2 Eggum (1973)

3 Sauer et al., (1974)

the availabilities decrease from corn to wheat and to barley and is inversely related to the increase in the proportions of albumin and globulin from corn to wheat to barley. Protein from albumin and globulin makes up approximately 5-6; 9-15 and 13-24% of the total protein of corn, wheat and barley respectively (Neurath and Bailey, 1954). Postel (1957) suggested that since barley has a multicellular aleurone layer, it should be higher in aleurone protein as a percentage of total protein compared with wheat, which has a single aleurone layer. Several workers suggested the relatively high concentration of LYS in aleurone protein to be responsible for its low availability from cereal grains (Eggum, 1973; Munck, 1964; Sauer et al., 1974). The above hypothesis explains also the low availabilities of ASP and ALA and the high availabilities of GLU and PRO. However, the high availabilities of ARG and HIS and the relatively low availability of THR can not be explained in the same manner (Tables 2 and 4).

Other factors may affect protein digestibility and therefore amino acid availability from cereal grains. Trypsin plays an important role in the digestion of protein. The extent of digestion by trypsin can be influenced by the amino acid sequence near the catalytic site. Trypsin exhibits a strict specificity for arginyl and lysyl peptide bonds. The number of peptide bonds split by trypsin will be dependent on the total amount of ARG and LYS present in the protein (Milhalyi, 1972).



Lysylprolyl and arginyl prolyl linkages are completely resistant to trypsin (Bell, 1954). Thus, a high PRO content coupled with a low ARG and LYS content might result in a less efficient digestion of a particular protein. This hypothesis would favor a relatively high level of digestion of aleurone protein (if it would be released from the aleurone cells) and a relatively low level of digestion of glutelin and prolamin. The ratio of PRO to ARG and LYS is particularly high in prolamin (Table 2).

The number of peptide bonds split by chymotrypsin would be dependent on the total amount of LEU and the aromatic amino acids , namely PHE and TYR.

Protein digestion would also depend on the distribution of amino acid residues which are compatible with enzyme specificity. Plant proteins are highly organized folded compact structures. The relative distribution of the "key" amino acids (ARG, LYS, LEU, PHE and TYR) from the inner to outer part of a protein molecule may influence its efficiency of digestion by trypsin and chymotrypsin.

Heating pure proteins gives rise to the formation of cross-linkages between the E-amino group of LYS and the carboxyl groups of the dicarboxylic amino acids or their amides (Bjarnason and Carpenter, 1970). These compounds are not absorbed (Valle-Riestra and Barnes, 1970). The possibility of such linkages occurring naturally in cereal protein should not be overlooked. Such a cross-link has been isolated from hair (Harding and Rogers, 1971).

Of the cereal grains, barley and especially sorghum contain significant amounts of tannins (Chang and Fuller, 1964; Eggum, 1968). The true nitrogen digestibilities of 29 sources of barley, ranging in tannin content from 0.55 to 1.23% were measured on rats (Eggum and Christensen, 1975). The tannin content was measured according to AOAC (1965) procedures. The nitrogen content of the 29 barley samples varied from 1.55 to 3.22%. Therefore, the relationship between nitrogen digestibility and content of nitrogen (which also affects nitrogen digestibility) and tannin was determined by use of a multiple regression equation, which was as follows:  $TD^1 = 82.60 + 3.89 \times N (\%) - 6.27 \times \text{tannin} (\%)$  (Eggum and Christensen, 1975). For example, barley containing 2% nitrogen and 0.55% tannin and barley containing 2% nitrogen and 1.23% tannin will result in true nitrogen digestibilities of 86.9 and 82.7% respectively.

Wheat and rye, and to a lesser extent rice, oats and corn were found to contain a trypsin inhibitor (Laporte and Tremolieres, 1962). The anti-trypsin factor seems to

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<sup>1</sup>TD: true nitrogen digestibility; N (%): percentage of nitrogen in barley; tannin (%): percentage of tannin in barley.

be concentrated in the germ part of the cereal grains (Moran et al., 1968).

In conclusion, differences in amino acid composition between cereal grains can be partly attributed to the different amounts of the 4 major protein fractions. The amount of aleurone protein contained within each cereal grain is dependent on the climatic conditions under which it is grown, the variety and other factors such as the level and type of fertilizer application. The amount of aleurone protein could be an important factor in determining amino acid availabilities. The type of physical processing before feeding in turn would affect the availability of aleurone protein for digestion. Other limiting factors in the utilization of cereal protein may be the presence of undigestible peptides in the endosperm fraction (in relation to PRO levels), the levels and distribution of "key" amino acids necessary for enzymatic cleavage, the possibility of naturally occurring cross-linkages and the presence of tannins and anti-trypsin factors.

## PART 2.

### Protein metabolism in the large intestine of monogastric animals.

When protein is ingested by the monogastric animal it is subjected to digestion in, basically, two steps. In the first step, protein is subjected to hydrolysis by the proteolytic enzymes that are secreted by the stomach and small intestine. The second step consists of fermentation of undigested protein by the micro-organisms of the large intestine.

A very low level of bacterial activity is found in the stomach, duodenum and jejunum. Beginning with the distal part of the ileum important activity can be detected. Bacterial activity reaches its maximum in the caecum and decreases from there to the end of the large intestine (Larson and Hill, 1960; Michel 1961, 1966).

Protein, both of exogeneous and endogeneous nature, that is not digested by the end of the small intestine will either be excreted in the faeces or subjected to fermentation. Protein digestion by the micro-organisms is brought about by extracellular enzymes through peptides of decreasing length to free amino acids to ammonia and short-chain fatty acids. Not necessarily all the free amino acids produced will be

degraded to ammonia and short-chain fatty acids. A certain proportion may be absorbed by the host animal (if amino acid transport systems are present in the epithelial cells of the large intestine). Michel (1961, 1966, 1968) incubated a test amino acid medium that had been inoculated with caecal bacteria. All amino acids were broken down by deamination, decarboxylation or desulfhydration. However, of the amino acids ARG, HIS, ASP, GLU, SER and THR were degraded the fastest. In addition, catabolic activity varied considerably from pig to pig and seasonal variations were detected.

The final nitrogenous end product of protein digestion by the micro-organisms is ammonia. Ammonia may also be derived from peptides, free amino acids and urea that enter the large intestine as such. The order of preferential fermentation of nitrogenous compounds by the flora to yield ammonia may probably be classified as follows: 1) free amino acids and urea, 2) peptides, 3) soluble protein and 4) insoluble protein.

Ammonia produced from the nitrogenous compounds in the large intestine may be taken up by the micro-organisms and be used for de novo microbial protein synthesis. In short, a certain amount of protein restructuring and corresponding changes in the proportions of amino acids will take place in the large intestine. On the other hand, a certain amount of the ammonia that is produced in the large intestine may be directly absorbed by the host animal. Part of the bacterial protein

will be excreted as such in the faeces. Part will undergo proteolysis during autodegradation of the micro-organisms. Degradation, as opposed to synthesis, will take over as the micro-organisms move down the large intestine. This is probably partly due to a decrease in nutrients available for the micro-organisms. The major end product of proteolysis of bacterial protein is ammonia. Blood draining the colon, especially that of the distal part, contains very high levels of ammonia (Michel, 1966).

The level of bacterial fermentation in the large intestine has been shown to be affected by the nature of the carbohydrate portion of the diet. Mason and Palmer (1973) fed a diet to rats that contained egg albumin as the protein source (14%) and rice starch as the carbohydrate source (50%). Replacement of rice starch by cornstarch did not increase the faecal nitrogen excretion but a significant increase was found when rice starch was replaced by raw potato starch. The authors suggested that raw potato starch was digested to a lesser extent in the small intestine than corn or rice starch. Therefore, more carbohydrate was available from potato starch than from rice or cornstarch to the flora of the large intestine for fermentation of undigested exogeneous or endogeneous protein residues. The increase in faecal nitrogen excretion due to dietary inclusion of raw potato starch was associated with a parallel increase in diaminopimelic acid (DAPA) excretion.

DAPA is an amino acid that is only found in some species of bacteria and the amount present in faeces would roughly indicate the relative amount (not absolute amount) of microbial synthesis occurring in the large intestine. It should be kept in mind that a certain amount of DAPA is also degraded in the large intestine (Mason and Milne, 1971).

Freeze-dried cod muscle protein was found to have an apparent nitrogen digestibility of 90 and 89% in intact and caeectomized chicks respectively. Heat-damaged cod muscle protein had a nitrogen digestibility of 77 and 68% for intact and caeectomized chicks (Nesheim and Carpenter, 1966). In other words, caeectomy only affected the nitrogen digestibility of the heat-damaged cod muscle protein. Fermentation by the micro-organisms in the large intestine of the chick for the utilization of poorly digestible protein was also suggested by Nitsan (1965) in the digestion of raw soybeans and by Payne et al. (1968) in the digestion of deteriorated fish concentrates. Therefore, the nature and or level of undigestible protein, in addition to carbohydrate digestibility, will also be a factor that determines the level of bacterial fermentation that takes place in the large intestine.

With germfree (GF) animals it is possible to study protein metabolism in the animal itself and not the combined metabolism of the host animal and its flora. Combe et al.

(1965) found twice as much soluble nitrogen and 50 to 100 times as much free amino acids in the caecum of GF rats than in the caecum of conventional (CV) rats. The ratio of urea to ammonia was also much higher in GF than in CV rats. The latter is due to the absence of bacterial urease in the caecum of GF rats. Urea accounted for at least 25% of the faecal nitrogen in GF rats, whereas only traces were found in the faeces of CV rats (Evrard et al., 1964). Similar observations were made on GF and CV chicks (Salter and Coates 1970, 1971).

Most reports show that GF animals excrete more faecal nitrogen than CV animals. Hoet et al. (1964) and Levenson and Tennant (1963) found twice as much faecal nitrogen excretion for GF rats. Combe et al. (1965) found differences of a smaller magnitude, namely 30%. Miller (1967) and Salter and Coates (1971) obtained similar results for GF and CV chicks. However, Luckey (1963) found no differences in faecal nitrogen excretion between GF and CV rats. As was explained previously, the level of bacterial fermentation may be affected by the digestibility coefficients and/or nature of the dietary carbohydrate and protein. These factors may partially explain the variable results obtained between the faecal nitrogen excretion from GF and CV animals. For example, a highly digestible diet fed to GF and CV animals may only result in



minimal differences between the levels of faecal nitrogen excretion. And indeed, the diet used by Luckey (1963) who found no differences between the faecal nitrogen excretion from GF and CV rats, was of a highly digestible nature.

The higher levels of nitrogen in the caecum of GF rats contribute to the higher levels of faecal nitrogen excretion. However, there still is a net disappearance of nitrogen between the end of the ileum and the rectum (Loesche, 1968). The levels of digestive enzymes are much higher in the contents of the caecum of GF rats than in the caecum of CV rats (Loesche, 1968). Normal digestion may thus proceed longer in GF animals. In addition, the free amino acids produced will be available for absorption by the animal since they are not subjected to deamination by the micro-organisms to yield ammonia and short-chain fatty acids which can be used for de novo microbial protein synthesis.

The micro-organisms of the large intestine have been strongly implicated in the recovery of endogeneous protein. The levels of digestive enzymes and mucoproteins in caecal contents and faeces were found to be much higher in GF than in CV animals (Eorgström et al., 1959; Lindstedt et al., 1965; Loesche 1968). Salter and Fulford (1974) concluded that the micro-organisms are only important with respect to the degradation and recycling of endogeneous protein and that they are

of minor importance in the degradation of dietary protein. A protein-free diet and a diet containing poorly digestible protein (heat-damaged egg albumin) were fed to GF and CV chicks. The endogeneous nitrogen excretion was higher for the GF than CV chicks. They found no difference between the GF and CV chicks that were fed the heat-damaged egg albumin diet. However, as was pointed out by the authors' themselves, they may have heat damaged the protein too severely. Even micro-organisms can not completely breakdown too severely heated proteins (Erbersdobler and Riedel, 1970). In contrast some bacterial degradation of poorly digestible protein (from heat-damaged fish protein) was shown by work by Carpenter and Nesheim (1966) and by Payne et al. (1968).

The form in which nitrogen is absorbed in the large intestine will determine the possible benefit that the host animal might derive from the micro-organisms. If nitrogen were absorbed as ammonia it would generally be of limited value to the animal. In certain instances, ammonia may improve the protein status of animals fed low protein diets because more non-essential amino acids can be produced and this in turn may have a "sparing effect" on the essential amino acids in the body.

Salter and Coates (1971) fed freeze-dried and autoclaved  $^{14}\text{C}$ -labelled egg white to GF and CV chicks. No

difference was found between the  $^{14}\text{C}$  : N ratios of the digesta from GF and CV chicks in the upper gut. However, the ratios were consistently higher for CV animals in the lower gut. Their findings can be explained by bacterial proteolysis and deamination of amino acids with subsequent absorption of ammonia. They also reported that the level of ammonia in caecal contents was five times greater in the CV than in the GF chicks. In addition they found a higher excretion of uric acid by the CV animals. Warren and Newton (1959) detected a fourfold increase in ammonia concentration in the portal blood of CV as compared to GF guinea pigs.

Salter et al. (1974) fed several poor quality proteins to GF and CV chicks and determined the net protein utilization (NPU) by nitrogen balance methods. NPU determination would be a good indicator of whether the host animal derives any nutritional benefit from the action of the microflora. Poor quality proteins were represented by sesame protein, which is deficient in LYS, and by heat-damaged egg albumin and cod muscle. A relatively large proportion of the latter proteins would remain undigested in the small intestine and would be available for microbial fermentation in the large intestine (unless too severely heated). They found no differences in NPU between GF and CV chicks for either the heat-damaged egg albumin or cod muscle. There was a slight

but significant difference between GF and CV chicks fed sesame protein but this could not be confirmed in later studies. With the heat-damaged proteins, the excretion of uric acid tended to be higher in CV than GF chicks. The authors conclude that most of the nitrogen that disappears from the large intestine is in the form of ammonia and that the microflora merely change the route of nitrogen excretion. The latter could be of help to the protein status of animals fed low protein diets.

Wysocki and Baker (1972) studied bacterial protein digestion in the equine lower gut.  $^{14}\text{C}$ -labelled bacteria were infused into the caecum of a caecal-fistulated pony that was anaesthetized. Negligible amounts of  $^{14}\text{C}$  were detected in the deproteinized plasma amino acid fraction of portal blood. They suggest the conversion of bacterial protein to ammonia upon autodegradation of the micro-organisms. However, in a similar type of experiment, Slade et al. (1971) showed that microbial protein can be degraded to yield amino acids which are absorbed in the caecum of the horse. However, the magnitude of absorption and therefore the nutritional significance was not established. Hoover and Heitman (1975) found considerable fermentation of  $^{14}\text{C}$ -alanine in both the caecum and upper colon contents of rabbits, but there was little absorption of  $^{14}\text{C}$ -alanine

into the blood.

Amino acid availability estimates determined by growth methods are generally lower than those determined by the faecal analysis method (Calhoun et al., 1960; Gupta et al., 1958; de Muelenaere et al., 1967; Nesheim and Carpenter 1967). Bacterial degradation of amino acids to ammonia in the large intestine may be the reason for overestimation of the actual amino acid availabilities as determined by the faecal analysis method.

To summarize what was discussed, bacterial degradation of undigested protein in the large intestine results in the formation of ammonia as the major nitrogenous end product. Ammonia may then be absorbed by the animal or may be taken up by the micro-organisms for de novo microbial protein synthesis. Part of the microbial protein will be excreted as such in the faeces while part of the micro-organisms will undergo autodegradation. Autodegradation of microbial protein results in the production of ammonia as the major endproduct, which is absorbed by the host animal. Thus, a certain amount of protein restructuring will take place in the large intestine from undigested exogeneous and endogeneous protein with resultant changes in the proportions of amino acids. Consequently, amino acid availabilities determined with the faecal analysis method could be inaccurate. As was pointed out, the degree of inaccuracy may be dependent on dietary factors such as carbohydrate and protein digestibility.

### PART 3.

Determination of amino acid availabilities  
by ileal sampling from cannulated animals,  
by ileal sampling from sacrificed animals,  
by removal of the caecum and  
by aid of germ-free and antibiotic treated animals.

Several approaches can be made to determine amino acid availabilities that may or may not be partially confounded by protein metabolism in the lower gut. Each approach has its particular advantages and disadvantages.

Ileal sampling by cannulation: More valid amino acid availability estimates will be obtained when the digesta are collected at the end of the ileum as compared to faecal collections.

Continuous collection of ileal digesta at the end of the ileum can be achieved by the use of ileal re-entrant cannulae (Holmes et al., 1974) or by ileal-caecal re-entrant cannulae (Easter, 1972). Ileal-caecal cannulae were found to cause fewer problems in maintaining an unrestricted flow (Easter, 1972). Use of the ileal re-entrant system will give a closer approximation to the digestive processes in situ as compared to the use of the ileal-caecal re-entrant system. The latter system bypasses the ileal-caecal valve and all digesta passes through the caecum first before entering the colon. About one-third to two-thirds of the ileal flow enters

into the colon directly under normal conditions, while the remainder enters into the caecum (Farrell and Johnson, 1972; Holmes et al., 1974).

Surgical modifications of the digestive tract, such as the incorporation of re-entrant cannulae will in all likelihood slow down the rate of passage of digesta. Pulse et al. (1973) studied the effects of caecal fistulation upon nutrient digestion and rate of passage in horses. They found significant increases in crude fiber and ether extract digestion following fistulation. Dry matter, gross energy and crude protein digestibilities did not change. Retention times of chromic oxide and polyethelene increased significantly.

Easter (1972) determined the apparent ileal and faecal amino acid availabilities of corn and sorghum grain in pigs, using ileal-caecal re-entrant cannulae. Faecal availability values were significantly higher than ileal estimates. The difference varied for each amino acid, ranging from 10 percentage units for TYR to 30 percentage units for GLY for both cereals. Lysine is often the most limiting amino acid for pigs fed corn or sorghum grain. The ileal and faecal LYS availabilities from corn were 65 and 85% respectively. In the same order, they were 59 and 78% for sorghum grain. Therefore, the faecal analysis method may overestimate the actual LYS availabilities from corn and sorghum grain to a relatively large extent.

Holmes et al. (1974) determined the ileal and faecal amino acid availabilities from soybean meal (SBM) and rapeseed meal (RSM) using pigs fitted with ileal re-entrant cannulae. The disappearance of nitrogen in the large intestine was 9 and 16% for SBM and RSM respectively. With the exception of ARG and MET, the faecal availabilities of the essential amino acids were higher than their corresponding ileal availabilities. The difference varied from 5.7 for VAL to 1.2% for LYS from SBM and from 13.6 for VAL to 1.2% for HIS from RSM. Methionine is often a limiting amino acid for pigs fed SBM or RSM and the ileal and faecal availabilities were 96.7 and 79.1% respectively for SBM and 92.5 and 81.0% respectively for RSM. In other words, there was more bacterial synthesis than degradation of MET. and its availability was underestimated by the faecal analysis method. With the exception of CYS and TYR, the faecal availabilities of the non-essential amino acids were higher than their corresponding ileal availabilities. The disappearance of PRO was the most extensive and was approximately 25 percentage units for both SBM and RSM.

Collection of ileal digesta from sacrificed animals.

Cho et al. (1971), using pigs fitted with ileal re-entrant cannulae, detected marked differences between the amino acid composition of ileal digesta that was collected



during different time intervals following ingestion of the test diets (SEM and RSM) by the animals. Fractionation of dietary components in the stomach and to a lesser extent in the small intestine is probably responsible for the variation in the composition of ileal digesta. The work by Cho et al. (1971) suggests the necessity for a continuous collection of the digesta from the ileum as can be performed by re-entrant cannulae. Consequently, amino acid availabilities, determined from samples taken from the end of the small intestine from sacrificed animals may be inaccurate since these are dependent on the time at which the animal is killed following ingestion of a test meal. In addition, it is often very difficult to obtain sufficient sample for analysis when only the digesta near the very end of the small intestine from sacrificed animals is collected. One often has to take all the digesta from a relatively large section of the ileum.

#### Removal of the caecum.

This surgical procedure results only in a partial decrease of total bacterial activity in the large intestine. Bacterial activity in the colon remains.

Lloyd et al. (1958) studied protein digestion in whole and caeectomized pigs, ranging in age from 8 to 28 weeks. The removal of the caecum resulted only in a slight decrease in protein digestibility. Caecectomy in chicks

decreased the protein digestibility of heat-damaged cod muscle from 77 to 68% (Nesheim and Carpenter, 1967). Specific amino acid availabilities have not been studied with caeectomized animals.

Germfree animals: Diets fed to GF animals must be sterilized by either heat, radiation or chemical agents. Even well-defined diets may undergo changes of uncertain character. The use of GF animals is limited to small laboratory species only.

Gordon and Wostmann (1960) reported lower weights of the small intestine of GF than in CV rats. The rate of renewal of the mucosa of the small intestine was reduced in the GF rats (Abrams et al., 1963). The surface area of the small intestine was reduced in the GF rat and was 30% less than in CV rats (Gordon and Bruckner-Kardoss, 1961). The changes described may have an effect on protein digestion and amino acid absorption in the small intestine. Thus, digestion data obtained from GF animals should be carefully interpreted.

#### Treatment with antibiotics.

The characteristics of the small intestine of animals receiving relatively large amounts of antibiotics approximate these of GF animals, particularly with regard to the reduction in thickness of the small intestine (Fauconneau and Michel, 1970).

Kuiken (1952) determined the faecal amino acid availabilities from cottonseed meal with or without sulfathiazole (at a level of 2% in the ration). Faecal amino acid availabilities were not changed by supplementation of the antibiotic. Faecal amino acid availabilities obtained with rats fed barley supplemented with chlortetracycline (20 ppm) or sulfathiazole (2% in the diet) were slightly higher than those obtained from rats that were fed barley without the antibiotics (Eggum, 1973). The differences for the individual amino acids ranged from 1 to 5 percentage units.

It is reasonable to assume that a reduction occurs in total bacterial activity upon dietary antibiotic supplementation. The extent to which this occurred (i.e. by bacterial counts on faecal material) was not measured by Kuiken (1952) and Eggum (1973).

## MATERIALS AND METHODS

### Study 1.

High and low protein strains of wheat (18.3 and 11.2%), and barley (11.4 and 8.5%) were selected for test purposes. It was not possible to obtain high and low protein strains of the same variety. The high protein wheat was of the Glenlea variety while the low protein wheat was of an unknown variety. The high and low protein barley were of the Fergus and Herta variety respectively. All grains were purchased directly from farmers.

The kernels were abraded by aid of a small laboratory pearling machine to yield the "endosperm" and "bran" fractions. The abraded kernels were named the endosperm fraction. The highly fibrous feed left after pearling was named the bran fraction. It was attempted by pearling to abrade the kernels in such a manner that all aleurone protein would be contained in the bran fraction. In addition, it was attempted to minimize carry-over of endosperm protein into the bran fraction. The kernels, 50 g at a time, were pearled at intervals of 4 consecutive seconds. This was repeated 5 times for each time of abrasion. Due to the hardness of its kernels, the low

protein wheat was abraded at intervals of 8 instead of 4 consecutive seconds. The abraded parts were pooled together, weighed and analyzed for nitrogen. The concentration of nitrogen in the fractions obtained during each time interval of abrasion are shown in Table 5.

As was pointed out in the review of literature, the outer layers of the cereal grains contain the aleurone cell layer. Of these layers, the aleurone cell layer has the highest protein content. There was a marked depression in the nitrogen content at the 40-48, 24-28 and 20-24 second interval for low protein wheat, high and low protein barley respectively (Table 5). In the same order, the kernels of these grains were routinely abraded for 48, 28 and 24 seconds until sufficient amounts were obtained of the bran and endosperm fractions for test purposes. For the high protein wheat there was a depression in the nitrogen content of the fraction obtained in the 16-20 second interval. However, the nitrogen content of the fractions that followed increased until it was markedly depressed again at the fraction collected in the 32-36 second interval. Based on these data, it was decided to abrade the high protein wheat for 20 seconds.

As will be discussed later, the high protein wheat should perhaps have been abraded for 36 seconds in order to

TABLE 5. Percent nitrogen of wheat and barley fractions .  
obtained at different time intervals of abrasion.

CEREAL GRAIN	WHEAT		BARLEY	
LEVEL OF PROTEIN (%)	18.3	11.2 <sup>1</sup>	11.4	8.1
<u>TIME INTERVAL OF</u> <u>ABRASION</u> (seconds)				
0-4	0.96		1.14	1.29
4-8	3.19	1.07	1.95	2.50
8-12	4.14		2.59	3.46
12-16	4.47	2.17	3.25	2.18
16-20	3.10		3.14	1.59
20-24	4.45	2.69	2.94	1.20
24-28	4.36		2.35	
28-32	3.64	2.56	2.15	
32-36	3.14			
36-40	3.06	2.24		
40-44	2.86			
44-48		1.84		
48-52				
52-56		1.59		

<sup>1</sup> Values in this column were determined at intervals of  
8 consecutive seconds (e.g. 1.07%N in 0-8 second fraction).

obtain a more complete separation of the aleurone protein from the endosperm protein.

The whole kernels and the fractions obtained were ground through a 2.00 mm screen, fortified with vitamins and minerals (Table 6), mixed and made into crumbles. The crumbles were prepared as follows: the mixed diets were spread out into a thin layer of approximately 0.25 cm. thickness, sprinkled with water, kneaded loosely into small "pellets" of approximately 0.5 cm. diameter and dried at 50 °C in a forced draft oven. The diets took on the appearance of crumbles.

The test diets (Table 6) were fed for 4 days to 60 male growing Wistar rats. Their initial average weight was 98 g the day prior to the start of the test period. Each diet was individually fed to 5 rats each. The experiment was repeated when the rats weighed 170 grams.

Two protein-free diets containing 10 and 20% alphafloc were fed to the same rats for a period of 2 days for the determination of the metabolic faecal amino acid excretion (Table 6). Faecal collections from 2 rats, fed the same protein-free diet, were pooled on the basis of similar (or nearly similar) dry matter intake. The protein-free diets were fed when the rats weighed 71 g and later when they weighed 207 g.

TABLE 6. Formulation of test diets for study 1.

DIET	HIGH PROTEIN WHEAT (18.3%) <sup>1</sup>			LOW PROTEIN WHEAT (11.2%) <sup>1</sup>			HIGH PROTEIN BARLEY (11.4%)			LOW PROTEIN BARLEY (8.1%)			PROTEIN-FREE	
CEREAL COMPONENT	WHOLE WHEAT	BRAN	ENDO- SPERM	WHOLE WHEAT	BRAN	ENDO- SPERM	WHOLE BARLEY	BRAN	ENDO- SPERM	WHOLE BARLEY	BRAN	ENDO- SPERM	ALPHA- FLOC	ALPHA- FLOC
Ingredients (%):														
WHEAT														
Whole	93.5			93.5										
Bran		91.5			91.5									
Endosperm			92.5			92.5								
BARLEY														
Whole							93.5							
Bran								91.5						
Endosperm									92.5					
Sucrose													81.0	71.0
Alphafloc													10.0	20.0
Soybean oil	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	5.0	5.0
Calcium carbonate	1.3	4.0	2.0	1.3	4.0	2.0	1.3	4.0	2.0	1.3	4.0	2.0	0.5	0.5
Dicalcium phosphate	0.7		1.0	0.7		1.0	0.7		1.0	0.7		1.0	2.0	2.0
Vitamin premix <sup>2</sup>	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0
Mineral premix <sup>3</sup>	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5

<sup>1</sup> Values in parentheses indicate the level of protein as a percent of dry matter.

<sup>2</sup> Supplied (kg diet): vitamin A 7150 IU; vitamin D 818 IU; vitamin E 5.5 IU; vitamin B<sub>12</sub> 11.0 ug.; riboflavin 5.5 mg; pantothenic acid 11.0 mg; nicotinic acid 16.5 mg; choline chloride 275 mg; menadione 1.1 mg; santoquin 1.1 mg; penicillin-streptomycin 26.4 mg.

<sup>3</sup> Supplied (kg diet): Manganese 81.4 mg; zinc 11 mg; iron 35.2 mg; copper 6.6 mg; iodized salt 4.7 g.



A basic diet (Giovannetti et al., 1970) was fed when the rats were not on test. One percent ferric oxide was incorporated into this diet in order to permit easier identification and collection of faeces resulting from the test and protein-free diets. The basic and protein-free diets were fed as pellets of 1.6 cm. diameter.

The rats were housed individually in suspended wire-bottom cages, in an environmentally controlled room at a temperature of 23°C. Feed and water were supplied ad libitum at all times.

Originally, during the first collection when the rats were fed the protein-free diets faeces were collected in a tray. Filter paper was placed on the bottom of the tray. This was thought to result in only a small amount of contamination of faecal pellets with urine. For improvement of the collection method, faeces were gathered on a screen on top of the trays during the following collections.

The total amount of feed consumed and faeces produced from the test diets were recorded. Apparent amino acid availability estimates were determined by measuring the total amount of each amino acid ingested and lost in the faeces. Metabolic faecal amino acid losses were considered for the estimation of true amino acid availabilities.

## Study 2.

This study was subdivided into study 2a and 2b.

Study 2a dealt with the determination of ileal and faecal amino acid availabilities from selected cereal grains by use of pigs fitted with ileocaecal re-entrant cannulae.

Study 2b was carried out to determine if faecal availability estimates obtained from cannulated pigs were representative of those of normal pigs.

### Study 2a.

The apparent ileal and faecal amino acid availabilities from barley (Herta), commercial corn and wheat (Glenlea) were determined with 6 Managra barrows. Metabolic ileal and faecal amino acid levels were estimated with 2 purified diets containing 2 levels of fiber (Table 7).

Ileocaecal re-entrant cannulae were fitted to barrows weighing 39 to 43 kg. The animals weighed 75 to 82 kg. at the end of the experiment.

The technique for re-entrant ileocaecal cannulation developed by Easter and Tanksley (1973) for pigs was used.

Type and size of cannulae used: a diagram of the type and size of the cannulae used is shown in Fig. 1. The cannulae were made from polyvinylchloride plastisol. The thickness of the cannulae is indicated by the dotted lines in the side view of the diagram. The top view shows the size of the

TABLE 7. Formulation of diets for study 2.

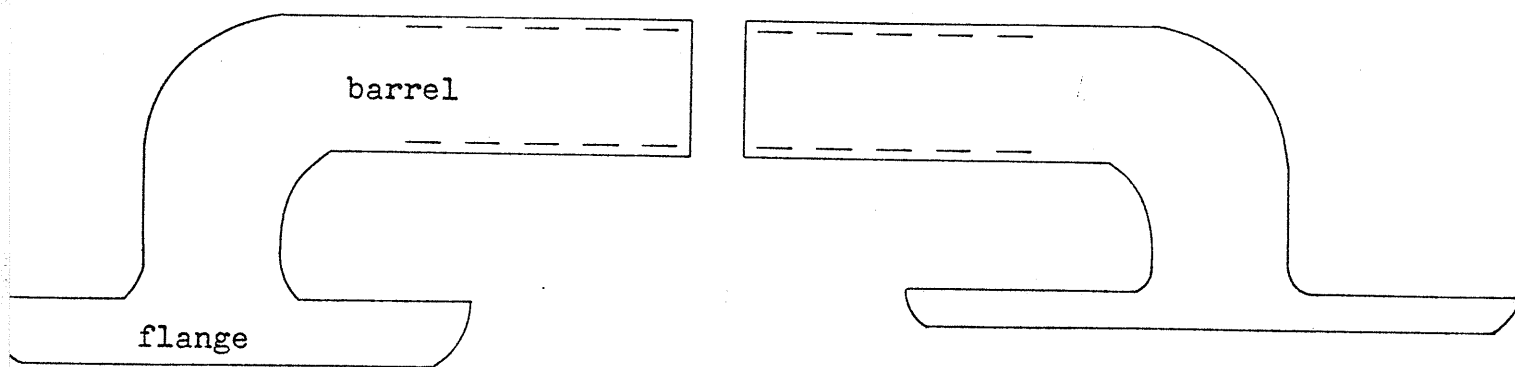
DIETS	GROWER	BARLEY	CORN	WHEAT	MAINTENANCE	
LEVEL OF ALPHAFLOC (%)					7	14
Ingredients (%):						
Barley	82.50	97.00				
Corn			97.00			
Wheat				97.00		
Soybean meal	14.50					
Casein					4.00	4.00
Cornstarch					63.00	63.00
Alphafloc					7.00	14.00
Sucrose					21.50	14.50
Soybean oil					1.00	1.00
Salt <sup>1</sup>	0.50	0.50	0.50	0.50	0.50	0.50
Calcium carbonate	1.25	1.25	1.25	1.25	0.50	0.50
Dicalcium phosphate	0.75	0.75	0.75	0.75	2.00	2.00
Vitamin-antibiotic premix <sup>2</sup>	0.50	0.50	0.50	0.50	0.50	0.50
Chromic oxide	+	+	+	+	+	+

<sup>1</sup> Supplied (kg diet): sodium chloride 4.83 g; zinc 20 mg; iron 30 mg; manganese 6 mg, copper 165 mg; iodine .33 mg; cobalt .20 mg.

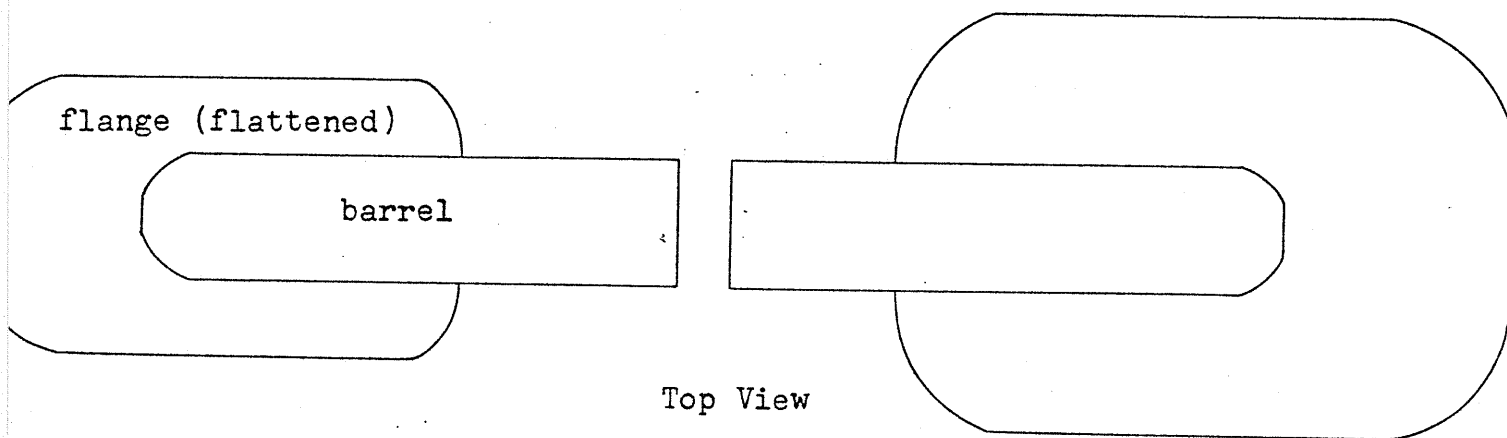
<sup>2</sup> Supplied (kg diet ): vitamin A 2200 IU; vitamin D 330 IU; vitamin B<sub>12</sub> 11.0 ug; DL- $\alpha$ -tocopheryl acetate 11 mg; thiamine hydrochloride 1.1 mg; riboflavin 2.2 mg; calcium-D-pantothenate 100 mg; pyridoxine hydrochloride 11.0 mg; nicotinic acid 10.0 mg; ASP-250 1g.

FIGURE 1. Type and size of cannulae used.

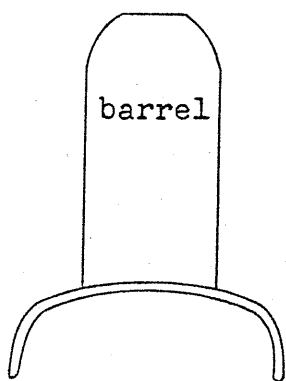
42



Side View



Top View

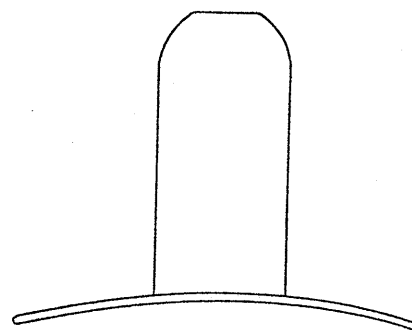


barrel

flange

Front View

Ileal Cannula



Caecal Cannula

SCALE 1:1



flanges when these are flattened out. The flanges were made in such a manner as to conform to the lumen of the ileum and the caecum (front view). The ileal and caecal cannulae were connected by aid of a 5 cm. polyvinylchloride tube.

Pre and post-operative care: the barrows were housed in individual metabolism cages for 5 days before surgery was performed. The pigs were starved the day before the operation and were given therapeutic levels of antibiotics (Pen-Di-Strep) by intramuscular injection. Administration of antibiotics was continued everyday after the operation until the animals seemed to be recovered. Recovery was indicated by healing of the tissue around the cannulae, normal rectal temperature and return to normal feed consumption.

The pigs were given access to a liquid diet for 3 days after the operation to minimize blockage by digesta. The liquid diet consisted of 9 parts water and 1 part milk replacer. Blockage of digesta during the first few days after the operation could be very critical. At this stage, there is no firm fibrous adhesion between the visceral and parietal layers of the peritoneum. Displacement and loss of cannulae is usually irreversible during this time, unless the animal is subjected to further surgery.

Following the liquid diet, the barrows were given access to the grower ration (Table 7) until placed on test. Readiness for test was judged by normal dietary intake of the grower ration for 4 to 5 consecutive days by all pigs. The barrows were switched to their respective test diets 12 days after the operation.

Problems associated with cannulation of pigs: blockage of digesta was shown by abnormal leakage around the ileal cannula, concomitant with an acute drop in feed intake. The pigs were fed 3 times daily and usually consumed all the feed that was supplied within one hour after feeding. None or partial consumption usually indicated blockage. The interconnecting tube was removed and warm water was flushed into the ileum until normal flow was reestablished. Thereafter, pigs went on feed again within a couple of hours. The frequency of blockage was very dependent on the type of diet fed. Blockage of digesta was a serious problem when the pigs were fed the M-14 diet (Fig. 2).

Blockage was also caused by the "curling up" of the flanges of the ileal cannula and was corrected by removing the cannula and replacing it with one that had a less flexible (thicker) flange.

Another cause of blockage was the relocation of the flange of the ileal cannula between the muscle layers under the skin. Subsequently, the inner muscle layer contracted

and the flow of digesta was often cut off. This condition was corrected by removing the cannula and replacing it with one that had a larger and thicker flange.

Formation of a "pocket" (hypertrophy of the distal end of the ileum) occurred in one instance, namely to pig number 5. This condition resulted in accumulation of relatively dry digesta in the "pocket". Peristalsis squeezed out the liquid part of the digesta and intestinal flow came to a halt. This condition could not be corrected and pig number 5 was replaced by number 7 (Fig. 2). However, the first test period for pig number 7 (diet M-7) did not start until the other 5 pigs were on the last test period for the cereal grains (days 34-40).

Build-up of pressure due to blockage and rubbing of the cannulae against the steel bars of the cages by the pigs sometimes resulted in disconnection of the ileal and caecal cannulae and large amounts of digesta were lost. About 200 ml. of 0.9% sodiumchloride was infused into the caecum before reconnecting the cannulae.

At times, the caecal cannulae were lost in the lumen of the large intestine and could not be retrieved. A spare caecal cannula was inserted. The lost cannula appeared in the faeces 48 to 72 hours later.

The pigs had to be prevented from laying on their right side (where the cannulae were inserted) during the

24-hour collection. Mild physical restraint was applied and the pigs became gradually trained to lie on their left side during collection.

Feed preparation: barley, corn and wheat were ground through a 4/16, 3/16 and 1 inch screen respectively. The ground material was mixed with vitamins and minerals and made into pellets of 0.5 cm. diameter in a steam pellet mill (California pellet mill). The maintenance diets were made into pellets of 0.25 cm. in a small laboratory pellet mill (Superior-Templewood dry pellet mill).

Feeding regimen: equal amounts of feed were offered 3 times daily at 6:00 A.M., 2:00 P.M. and 10:00 P.M. The average daily intake of the grower ration (average of all pigs) was recorded and determined the daily allowable intake of the test diets for the period following. The allowable intake was calculated to the nearest 50 or 100 grams (Fig. 2). Water was supplied ad libitum at all times.

Experimental design: the experiment was conducted as a simple cross-over design in which two barrows were fed the 3 cereal diets at 3 different periods during 7 consecutive days (Fig. 2). The 7% alphafloc maintenance diet (M-7) was fed prior to and after the cereal test diet sequence. The 14% alphafloc maintenance diet (M-14) was fed for only one test period, following the M-7 diet fed after the cereal test



FIGURE 2. Experimental design and allowable dietary intake for study 2a.

DAY <sup>1</sup>	1-7	8-11	12-18	19-22	23-29	30-33	34-40	41-44	45-61	62-65	66-72
PIG NO. 2	M-7 <sup>2</sup>	GR <sup>2</sup>	W <sup>2</sup>	GR	B <sup>2</sup>	GR	C <sup>2</sup>	GR	M-7	GR	M-14 <sup>2</sup>
5 (7)	M-7	GR	W	GR	B	GR	C	GR	M-7	GR	M-14
1	M-7	GR	C	GR	W	GR	B	GR	M-7	GR	M-14
6	M-7	GR	C	GR	W	GR	B	GR	M-7	GR	M-14
3	M-7	GR	B	GR	C	GR	W	GR	M-7	GR	M-14
4	M-7	GR	B	GR	C	GR	W	GR	M-7	GR	M-14
ALLOWABLE DIETARY INTAKE <sup>3</sup>	1800		1950		2100		2100		2100		2100

<sup>1</sup> Day 1 indicates the 12th day after the operation.

<sup>2</sup> Maintenance diet, 7% alphafloc (M-7); Grower (GR); Wheat (W); Corn (C); Maintenance diet, 14% alphafloc (M-14).

<sup>3</sup> Grams per day, as is.

diet sequence. The grower ration was fed for 4 days between the cereal and/or maintenance diet test periods.

Collection procedures: faeces were collected for 24 hours from 2:00 P.M. on day 5 to 2:00 P.M. on day 6 during each test period. A 24-hour continuous ileal collection was carried out from 2:00 P.M. on day 6 to 2:00 P.M. on day 7. A plastic bag was tied to the ileal cannulae during collection and its contents were emptied into a beaker until a total of 400 ml. was collected. After stirring a 10% aliquot was taken by aid of a 50 cc syringe with a catheter tip. The remainder was made up to 400 ml. again with distilled water, reheated to pig body temperature and infused within half an hour into the caecal cannula by aid of a 50 cc syringe. The aliquots taken for analyses were put into a container placed in ice. These containers were changed every eight hours after the start of the ileal collection. Therefore, for each pig 3 pooled 8-hour collections were obtained.

Calculations: apparent availabilities of the nutrients were determined by aid of calculations based on the levels of chromic oxide in feed, ileal digesta and faeces (Crampton and Harris, 1969). Ileal nutrient availabilities were also determined by total collection. True availabilities were determined by correcting the apparent nutrient availabilities

for the levels of endogenous nutrients, which were obtained by feeding the maintenance diets.

#### Study 2b.

Six non-cannulated Managra barrows were subjected to essentially the same experimental conditions as the cannulated pigs in study 2a. They ranged in weight from 36 to 41 kg. at the start of the experiment. The same test diets were used as for study 2a (Table 7).

In this study, M-14 was also fed prior to the cereal test diet sequence. Three pigs were fed M-7 and 3 pigs were fed M-14 during each period that the maintenance diets were fed. Pigs receiving M-7 during the first test diet period would receive M-14 the following test period and vice versa, prior to and after the cereal test diet sequence.

#### Study 3.

The fate of caecally infused isolated soy protein and LYS was studied with 6 growing Managra barrows.

Caecal fistulae were fitted to 5 barrows weighing approximately 20 to 23 kg. and to one barrow that weighed 14 kg. With the exception of the surgery including the fitting of ileal cannulae, the procedure for insertion of caecal fistulae was similar to that cited under Study 2a (Easter and Tanksley, 1973).

The caecal fistula was identical in size and shape to the caecal cannula used in study 2a with the exception that the barrel was 2 cm. longer (Fig. 1). The fistula was closed with a plug and firmly taped to the skin with the barrel pointing dorsally.

Management procedures, prior to and after the operation, were similar to those described under study 2a.

Study 3 was subdivided into 2 separate experiments. Experiment 3a dealt with the infusion of isolated soy protein into the caecum. Isolated soy protein (ISP) was dissolved in warm water so that the concentration of ISP of the infused solution was approximately 30%. Experiment 3b involved the infusion of lysine monohydrochloride. One part of LYS was mixed with two parts of cornstarch and dissolved in warm water so that the LYS concentration of the infused solution was approximately 10%. To facilitate caecal administration, cornstarch was mixed with LYS to increase the viscosity of the infusion solution.

The formulations of the test diets are shown in Table 8. The E+2ISP diet was formulated to contain twice as much ISP (isolated soy protein) as the E+ISP diet. The B+ISP+LYS diet was formulated to contain as much LYS as the B+2ISP diet. The diets were made into pellets of 0.5 cm. diameter in a commercial type pellet mill (California pellet mill) used by our university.

TABLE 8. Formulation of test diets in study 3.

DIETS	B+2ISP <sup>1</sup>	B+ISP <sup>1</sup>	B+ISP+LYS <sup>1</sup>
Ingredients (%)			
Barley	84.72	84.72	84.72
Isolated soy protein	6.78	3.39	3.39
Supplemental Lysine <sup>2</sup>	0.00	0.00	0.20
Cornstarch	3.00	6.39	6.39
Soybean oil	2.00	2.00	2.00
Calcium carbonate	1.25	1.25	1.25
Dicalcium phosphate	0.75	0.75	0.75
Chromic oxide	+	+	+
Salt <sup>3</sup>	0.50	0.50	0.50
Vitamin-mineral <sup>4</sup> antibiotic premix	1.00	1.00	1.00

<sup>1</sup> B+2ISP (Barley plus 2 times 3.39 units of isolated soy protein); B+ISP (Barley plus 3.39 units of isolated soy protein); B+ISP+LYS (Barley plus 3.39 units of isolated soy protein plus supplemental lysine).

<sup>2</sup> Lysine monohydrochloride.

<sup>3</sup> As in Table 7 .

<sup>4</sup> Supplied (kg diet): vitamin A 2200 IU; vitamin D 330 IU; vitamin B<sub>12</sub> 11.0 ug; zinc 123 mg; mecadox 22 g.

Both experiments were set up as 3X3X3 Latin Square designs. The test diets were fed for 8 days. Equal amounts of feed were offered 3 times daily at 6:00 A.M., 2:00 P.M. and 10:00 P.M. The daily amounts of feed offered were 1800, 1950 and 2040 grams (on an as is basis) during periods I, II and III respectively. Approximate daily intakes per unit of metabolic body weight ( $W_{kg}^{0.75}$ ) were 1320, 1370 and 1390 grams for periods I, II and III respectively. Water was offered freely just prior to and after each feeding. The barrows were fed a pig grower ration for 4 days between test periods (Table 7). The grower ration was fed at the same level of intake as the test diets during the following test period.

Starting on day 1 of each test period at 6:00 A.M. to day 8 at 2:00 P.M., ISP or LYS was infused 3 times daily into the caecum of the pigs. The daily amounts of nitrogen from ISP and LYS that were infused into the caecum of the pigs are shown in Table 9 and Table 10 respectively. The infusions were performed during the time the pigs were eating. The test diets were usually consumed within 30 minutes after they were offered.

Starting on day 5 of each test period a 3-day nitrogen balance study was initiated. Three consecutive 24-hour collections of urine and faeces were carried out beginning at 2:00 P.M. each collection day.

TABLE 9. The experimental design and the daily intake of dry matter and orally and caecally supplied nitrogen in study 3a.

PERIOD	I	II	III
<u>Pig No. 1</u>	(B+2ISP) <sup>1</sup>	(B+ISP) <sup>1</sup>	(B+ISP+C-ISP) <sup>1</sup>
Dry matter intake (g)	1687.50	1802.78	1885.98
Nitrogen intake (g):			
Orally	41.95	37.79	39.53
Caecally	0.00	0.00	10.85
Total	41.95	37.79	50.38
<u>Pig No. 2</u>	(B+ISP)	(B+ISP+C-ISP)	(B+2ISP)
Dry matter intake (g)	1664.10	1802.78	1912.50
Nitrogen intake (g):			
Orally	34.88	37.79	47.54
Caecally	0.00	10.37	0.00
Total	34.88	48.16	47.54
<u>Pig No. 3</u>	(B+ISP+C-ISP)	(B+2ISP)	(B+ISP)
Dry matter intake (g)	1664.10	1828.13	1885.98
Nitrogen intake (g):			
Orally	34.88	45.45	39.53
Caecally	9.57	0.00	0.00
Total	44.45	45.45	39.53

<sup>1</sup> Abbreviations in parentheses indicate the dietary treatments. B+ISP+C-ISP represents the treatment in which ISP is infused into the caecum.

TABLE 10. The experimental design and the daily intake of dry matter and nitrogen and orally and caecally supplied lysine in study 3b.

PERIOD	I	II	III
<u>Pig No. 4</u>	(B+ISP+C-LYS) <sup>1</sup>	(B+ISP+LYS) <sup>1</sup>	(B+ISP) <sup>1</sup>
Dry matter intake (g)	1664.10	1815.45	1885.98
Nitrogen intake (g)	34.88	37.96	39.53
Lysine intake (g):			
Orally	9.32	12.89	10.56
Caecally	3.17	0.00	0.00
Total	12.49	12.89	10.56
<u>Pig No. 5</u>	(B+ISP)	(B+ISP+C-LYS)	(B+ISP+LYS)
Dry matter intake (g)	1664.10	1802.78	1899.24
Nitrogen intake (g)	34.88	37.79	39.71
Lysine intake (g):			
Orally	9.32	10.10	13.48
Caecally	0.00	3.43	0.00
Total	9.32	13.53	13.48
<u>Pig No. 6<sup>2</sup></u>	(B+ISP+LYS)	(B+ISP)	(B+ISP+C-LYS)
Dry matter intake (g)	1675.08	1802.78	
Nitrogen intake (g)	35.04	37.79	
Lysine intake (g):			
Orally	11.89	10.10	
Caecally	0.00	0.00	
Total	11.89	10.10	

<sup>1</sup> Abbreviations in parentheses indicate the dietary treatments. B+ISP+C-LYS represents the treatment in which LYS is infused into the caecum.

<sup>2</sup> Data from pig number 6 on treatment B+ISP+C-LYS were not available.



Data from pig number 6 for period III are not available (Table 10). This pig was about 8 kg. lighter than the other pigs when surgery was performed and the first test period for this pig was not started until the other pigs were on test period III. Inadvertently, insufficient B+ISP diet was prepared to carry this pig through period III.

The amount of nitrogen from ISP to be infused was to be the difference between the nitrogen intake of the pig receiving the B+2ISP diet and the pig receiving the B+ISP diet during each test period. For example, pig number 3 should have received  $7.07^1$  grams of nitrogen per day by caecal infusion (Table 9). However, 9.57 instead of 7.07 grams of nitrogen from ISP was infused daily into the caecum. This value was arrived at as follows: the difference between the B+2ISP and the B+ISP diet was 3.39 grams of ISP per 100 grams of diet (Table 8). For 1800 grams, the daily intake (as is basis) during period I, the difference would be equivalent to  $61.02^2$  grams of ISP. Because of calculation errors, the percentage of dry matter of the test diets was not taken into account. Ten percent

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$^1 41.95 - 34.88 = 7.07.$   $^2 18 \times 3.39 = 61.02.$

allowance was made to account for losses during infusion. Isolated soy protein contained 14.26% nitrogen. Consequently, the total amount of nitrogen from ISP infused daily was 9.57 grams<sup>1</sup>. If the percentage of dry matter (93.0%) had been considered and the allowance had not been made, the amount of infused nitrogen would have been 8.09 grams<sup>2</sup> per day. As was mentioned previously, the diets were pelleted in a commercial type pellet mill. Contamination from other diets, present in the mixing and or pelleting system, could have taken place and accounted for some of the remainder of the difference which was 1.02 grams<sup>3</sup> of nitrogen.

The difference between LYS that was infused and that should have been infused may be explained in the same manner as for ISP infusion (Table 8 and 10).

#### Study 4.

This study was subdivided into study 4a and 4b. Study 4a dealt with the determination of ileal and faecal amino acid availabilities from wheat and wheat derived fractions, in addition to the determination of metabolic ileal and faecal amino acid levels. Study 4b was carried out to determine the differences in ileal and faecal amino acid availabilities from

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$$^1 61.02 \times \frac{110}{100} \times \frac{14.26}{100}. \quad ^2 61.02 \times \frac{93}{100} \times \frac{14.26}{100}.$$

$$^3 8.09 - 7.07 = 1.02.$$

finely ground and cracked whole wheat.

Study 4a.

Whole wheat was milled to give 75% flour, 11.25% bran, 11.25% shorts and 2.5% middlings. The bran, shorts and middlings were recombined in the same proportion as they were obtained by milling to give the E+S+M diet.

The ileal and faecal amino acid availabilities from whole wheat, flour and the E+S+M diet were determined with 6 Managra barrows that were fitted with ileocaecal re-entrant cannulae (Table 11). Three protein-free diets containing 5, 10 and 15% alphafloc respectively were employed to determine the levels of metabolic ileal and faecal amino acids (Table 11).

Re-entrant cannulae were fitted to barrows weighing 37 to 45 kg. The pigs weighed from 81 to 91 kg. at the end of the experiment.

The surgical procedure, type and size of cannulae used, pre and post-operative care, housing, collection procedure and the duration and times the test diets were fed were similar to those described under study 2a with one exception. Ileal digesta was collected continuously for only 16 hours, starting at 6:00 A.M., 7 days after the pigs were switched to their respective test diets. Faeces were collected for 24 hours prior to the start of the ileal collection.

All diets were made into pellets of 0.25 cm. diameter. Whole wheat was ground through a 2.00 mm screen prior to

TABLE 11. Formulation of diets for study 4.

DIETS	WHOLE WHEAT	FLOUR	B+S+M	PROTEIN-FREE		
LEVEL OF ALPHAFLOC (%)				5	10	15
Ingredients (%):						
Whole wheat	97.00					
Flour		96.00				
B+S+M <sup>1</sup>			95.00			
Cornstarch				63.00	63.00	63.00
Alphafloc				5.00	10.00	15.00
Sucrose				27.50	22.50	17.50
Soybean oil				1.00	1.00	1.00
Salt <sup>2</sup>	0.50	0.50	0.50	0.50	0.50	0.50
Calcium carbonate	1.25	2.00	4.00	0.50	0.50	0.50
Dicalcium phosphate	0.75	1.00	0.00	2.00	2.00	2.00
Vitamin-antibiotic premix <sup>3</sup>	0.50	0.50	0.50	0.50	0.50	0.50
Chromic oxide	+	+	+	+	+	+

<sup>1</sup> Consists of 45% bran, 45% shorts and 10% middlings.

<sup>2</sup> As for Table 7 .

<sup>3</sup> As for Table 7 .

pelletting. The E+S+M diet was not ground prior to pelletting.

The experiment was set up in such a manner that the effect of level of fiber on metabolic ileal and faecal amino acid levels and the differences in availabilities of amino acids from whole wheat, flour and the E+S+M diet could be analyzed statistically by 2 separate single cross-over designs. The periods during which the protein-free diets were fed were alternated with periods during which the wheat or wheat derived diets were fed (Fig. 3).

Two cannulated pigs (pig no. 6 and 9) were kept on standby and served as replacement test animals in case complications arose with the pigs that were actually tested (Fig. 3). Pigs number 1, 2, 4 and 7 did not consume the 15% alphafloc protein-free diet nor the E+S+M diets. Pigs number 1 and 2 also refused the 10% alphafloc protein-free diet. Replacement pig number 6 consumed both the 15% alphafloc protein-free diet and the E+S+M diet. Replacement pig number 9 did not consume the E+S+M nor the 15% alphafloc protein-free diet (Fig. 3).

Only three observations (including data from pig number 6) were obtained from diet E+S+M. The original E+S+M diet was diluted with cornstarch so that cornstarch made up 60% of the diluted E+S+M diet. The diluted E+S+M diet (E+S+M-D) was fed to the test animals from day 67 to 73 (Fig. 3).

FIGURE 3. Experimental design and allowable daily dietary intake for study 4.

DAY <sup>1</sup>	1-7	8-11	12-18	19-22	23-39	30-33	34-40	41-44	45-51	52-55	56-62	63-66	67-73	74-77	78-84
<u>FIG NUMBER</u>															
1	PF-5 <sup>2</sup>	GR	FL	GR	(PF-15) <sup>3</sup>	GR	WW	GR	(PF-10)	GR	(B+S+M)	GR	B+S+M-D	GR	WW-G
2	PF-5	GR	FL	GR	(PF-15)	GR	WW	GR	(PF-10)	GR	(B+S+M)	GR	B+S+M-D	GR	WW-CR
3	PF-10	GR	B+S+M	GR	PF-5	GR	FL	GR	PF-15	GR	WW	GR	B+S+M-D	GR	WW-G
4	PF-10	GR	(B+S+M)	GR	PF-5	GR	FL	GR	(PF-15)	GR	WW	GR	B+S+M-D	GR	WW-CR
5	PF-15	GR	WW	GR	PF-10	GR	B+S+M	GR	PF-5	GR	FL	GR	B+S+M-D	GR	WW-G
7	(PF-15)	GR	WW	GR	PF-10	GR	(B+S+M)	GR	PF-5	GR	FL	GR	B+S+M-D	GR	WW-G
6	GR	GR	PF-15	GR	B+S+M	GR	PF-15	GR	GR	GR	(B+S+M)	GR	GR	GR	WW-CR
9	GR	GR	PF-10	GR	(B+S+M)	GR	PF-10	GR	(PF-15)	GR	PF-10	GR	GR	GR	WW-CR
ALLOWABLE DIETARY INTAKE (g/day)	1500		1500		1800		1800		2100		2100		2100		2400

<sup>1</sup> Day 1 is the eleventh day after the operation.

<sup>2</sup> Protein-free, 5% alphafloc (PF-5); Protein-free, 10% alphafloc (PF-10); Protein-free, 15% alphafloc (PF-15); Grower (GR); Flour (FL); Whole wheat (WW); Bran and Shorts and Middlings (B+S+M); Bran and Shorts and Middlings diluted with cornstarch (B+S+M-D); Whole wheat finely ground (WW-G); Whole wheat cracked (WW-CR).

<sup>3</sup> Diets in parentheses indicate missing observations.

<sup>4</sup> Allowable daily dietary intake (g), as is basis.

#### Study 4b.

Finely ground wheat was prepared by grinding the whole wheat three times through a hammer mill with a 1.5 mm screen. The cracked wheat was prepared by grinding whole wheat two times through a 6 mm screen. Both diets were made into pellets of 0.5 cm diameter.

Four pigs were assigned at random to each of the 2 whole wheat diets (Fig. 3). The whole wheat used in this study was of the same source as that used in study 4a.

#### Analytical Procedures

The test diets and faeces were dried in a forced-draft oven at 60-70°C. Ileal digesta were freeze-dried. Prior to analyses, the samples were ground through a 1 mm mesh screen in a Wiley mill.

Analyses for ash, crude fiber, dry matter, ether extract and nitrogen were carried out according to A.O.A.C. (1971) methods. Acid and neutral-detergent fiber were determined according to the method outlined in Agriculture Handbook no. 379 (1970).

Total starch was determined according to the method of R. A. MacGregor (1976, unpublished). This method involves dispersion of the ground sample, autoclaving, followed by digestion with glucoamylase. The total glucose released was measured using a Glucostat reagent kit (glucose oxidase method)

on a autoanalyzer.

The levels of chromic oxide in feed, digesta and faeces were determined according to the atomic absorption spectrophotometry method of Williams et al. (1962).

Amino acid analyses were carried out according to the method of Bragg et al. (1966) with modifications as described by Giovannetti et al. (1970). Methionine, CYS and DAPA ( $\alpha$ - $\epsilon$ -diaminopimelic acid) were determined according to the method of Hirs (1967). Tryptophan analyses were carried out according to the method of Hugli and Moore (1972). The samples were analyzed on a model 116-Beckman amino acid analyzer.



## RESULTS

### Study 1.

With the exception of GLU and PRO from low protein wheat, bran from high and low protein wheat contained higher levels (%) of all amino acids than endosperm from high and low protein wheat (Table 12). The higher levels (%) of amino acids in bran than in endosperm were particularly evident for ARG, LYS and THR (essential amino acids) and for ALA, ASP and GLY (non-essential amino acids). These data were also reflected by the higher concentration (as grams per 16 grams of nitrogen) of these amino acids in bran than in endosperm protein. The concentrations (g/16g N) of all other amino acids in protein from bran were lower than those in protein of endosperm from both high and low protein wheat. The differences in amino acid concentration (g/16g N) were more marked for bran and endosperm derived from low protein wheat than for bran and endosperm derived from high protein wheat.

With the exception of LEU, MET, PHE, GLU, PRO, SER, and TYR, bran from both high and low protein barley contained higher levels (%) of all amino acids than endosperm from high and low protein barley (Table 13). However, only ARG, LYS,

TABLE 12. The amino acid and nitrogen content of high and low protein wheat and their fractions.

DIET	HIGH PROTEIN WHEAT							LOW PROTEIN WHEAT						
	WHOLE WHEAT		BRAN			ENDOSPERM		WHOLE WHEAT		BRAN			ENDOSPERM	
	% <sup>1</sup>	g/16gN <sup>2</sup>	%	g/16gN	% <sup>3</sup>	%	g/16gN	%	g/16gN	%	g/16gN	%	%	g/16gN
AMINO ACIDS														
<u>Essential</u>														
ARG	0.74	4.05	1.14	4.86	22.7	0.65	3.81	0.46	4.11	0.73	5.19	31.1	0.41	3.81
HIS	0.35	1.92	0.43	1.83	17.7	0.33	1.93	0.22	1.97	0.26	1.85	23.2	0.21	1.95
ILE	0.56	3.09	0.69	2.94	18.1	0.58	3.40	0.36	3.22	0.40	2.84	22.0	0.37	3.44
LEU	1.24	6.80	1.51	6.44	18.1	1.23	7.21	0.79	7.06	0.89	6.32	21.9	0.80	7.44
LYS	0.39	2.11	0.55	2.35	21.2	0.36	2.11	0.29	2.59	0.46	3.27	30.7	0.25	2.33
MET <sup>4</sup>	0.32	1.77	0.35	1.49	16.3	0.29	1.70	0.23	2.06	0.22	1.56	18.6	0.21	1.95
PHE	0.89	4.86	1.04	4.44	17.4	0.87	5.10	0.53	4.74	0.55	3.91	20.1	0.55	5.12
THR	0.50	2.74	0.67	2.86	19.8	0.41	2.40	0.33	2.95	0.44	3.13	25.7	0.32	2.98
VAL	0.71	3.89	0.94	4.01	19.5	0.71	4.16	0.47	4.20	0.59	4.20	24.5	0.48	4.47
<u>Non-Essential</u>														
ALA	0.65	3.54	0.97	4.14	22.4	0.60	3.52	0.45	4.02	0.70	4.98	30.4	0.41	3.81
ASP	0.93	5.09	1.34	5.72	21.4	0.84	4.92	0.59	5.27	0.89	6.33	29.2	0.56	5.21
GLU	6.49	35.60	7.04	30.04	16.1	6.50	38.10	3.52	31.46	3.08	21.90	17.1	3.78	35.16
GLY	0.77	4.23	1.11	4.74	21.5	0.73	4.28	0.50	4.47	0.73	5.19	28.4	0.47	4.37
PRO	2.15	11.77	2.33	9.94	16.0	2.12	12.42	1.27	11.35	1.02	7.25	15.6	1.32	12.28
SER	0.85	4.69	1.05	4.48	18.3	0.84	4.92	0.54	4.83	0.61	4.34	21.9	0.52	4.84
TYR	0.58	3.20	0.69	2.94	17.4	0.55	3.22	0.32	2.86	0.36	2.56	21.6	0.32	2.98
NITROGEN	2.92	16.00	3.75	16.0	19.0	2.73	16.0	1.79	16.00	2.25	16.0	24.5	1.72	16.0

<sup>1</sup> Expressed as percentage of dry matter. <sup>2</sup> Expressed as grams per sixteen grams of nitrogen. <sup>3</sup> The percentage of amino acids from whole wheat present in the bran fraction. <sup>4</sup> Determined by acid hydrolysis.

TABLE 13. The amino acid and nitrogen content of high and low protein barley and their fractions.

DIET	HIGH PROTEIN BARLEY							LOW PROTEIN BARLEY						
CEREAL COMPONENT	WHOLE BARLEY			BRAN		ENDOSPERM		WHOLE BARLEY			BRAN		ENDOSPERM	
	% <sup>1</sup>	g/16gN <sup>2</sup>	%	g/16gN	% <sup>3</sup>	%	g/16gN	%	g/16gN	%	g/16gN	% <sup>3</sup>	%	g/16gN
AMINO ACIDS														
<u>Essential</u>														
ARG	0.51	4.5	0.67	5.1	26.9	0.45	4.2	0.36	4.2	0.54	5.2	33.9	0.37	4.6
HIS	0.20	1.7	0.28	2.1	28.7	0.20	1.9	0.14	1.6	0.22	2.1	35.5	0.14	1.7
ILE	0.39	3.4	0.45	3.4	23.7	0.39	3.7	0.28	3.3	0.36	3.5	29.1	0.30	3.7
LEU	0.38	7.3	0.81	6.2	20.0	0.84	7.9	0.62	7.3	0.65	6.3	23.7	0.64	7.9
LYS	0.35	3.1	0.57	4.4	33.4	0.32	3.0	0.29	3.4	0.51	4.9	39.7	0.28	3.5
MET <sup>4</sup>	0.20	1.7	0.13	1.0	13.3	0.22	2.1	0.15	1.8	0.09	0.9	13.6	0.17	2.1
PHE	0.58	5.1	0.54	4.1	19.1	0.60	5.6	0.40	4.7	0.39	3.8	22.0	0.40	5.0
THR	0.40	3.5	0.44	3.4	22.6	0.37	3.5	0.30	3.5	0.37	3.6	27.9	0.28	3.5
VAL	0.58	5.1	0.68	5.2	24.0	0.58	5.5	0.41	4.8	0.51	4.9	28.1	0.41	5.1
<u>Non-Essential</u>														
ALA	0.52	4.5	0.64	4.9	25.2	0.46	4.3	0.42	4.9	0.55	5.3	29.6	0.37	4.6
ASP	0.72	6.3	0.92	7.0	26.2	0.66	6.2	0.60	7.1	0.81	7.8	29.4	0.55	6.8
GLU	2.90	25.4	2.34	17.9	16.5	3.09	29.1	1.87	22.0	1.64	15.8	19.8	1.92	23.8
GLY	0.50	4.4	0.62	4.7	25.4	0.46	4.3	0.40	4.7	0.52	5.0	29.4	0.37	4.6
PRO	1.27	11.1	0.85	6.5	13.7	1.33	12.5	0.82	9.6	0.59	5.7	16.3	0.90	11.2
SER	0.53	4.6	0.48	3.7	18.6	0.51	4.8	0.37	4.4	0.39	3.8	23.8	0.35	4.3
TYR	0.37	3.2	0.27	2.1	15.0	0.38	3.6	0.27	3.2	0.18	1.7	15.1	0.26	3.2
NITROGEN	1.83	16.0	2.09	16.0	23.4	1.70	16.0	1.36	16.0	1.66	16.0	27.6	1.29	16.0

<sup>1</sup> Expressed as percentage of dry matter. <sup>2</sup> Expressed as grams per sixteen grams of nitrogen. <sup>3</sup> The percentage of amino acids from whole barley present in the bran fraction. <sup>4</sup> Determined by acid hydrolysis.

ALA, ASP and GLY were higher in bran than in endosperm when these were expressed as grams per 16 grams of nitrogen. The concentration (g/16g N) of the other amino acids, especially LEU, MET, PHE, GLU, PRO and TYR were lower in bran than in endosperm protein. Differences in amino acid concentration (g/16g N) were more marked for bran and endosperm protein from low than from high protein barley.

The rats did not consume the bran diets from barley in its complete form. The crumbles were broken up and sorted. In addition, faeces from barley bran were of a very moist nature and became contaminated with sorted and spilled food. Due to these difficulties and the biased data that would be obtained, feeding bran from barley to the rats was discontinued. Therefore, no amino acid availability estimates from barley bran are available.

The average daily dry matter intake of the test diets by the rats are shown in Table 14. The wheat bran diet was consumed to a slightly lesser extent than the whole wheat or endosperm diet.

Apparent amino acid availabilities, nitrogen and dry matter digestibilities are given in Table 15. Mean squares of the analyses of variance are given in the appendix, Table 1. With the exception of HIS, LEU, ALA, and GLU, there were no significant differences in apparent availabilities of

TABLE 14. Means and standard deviation of daily dry matter intake for the test and protein-free diets for study 1.

DIET		HIGH PROTEIN WHEAT			LOW PROTEIN WHEAT			HIGH-PROTEIN BARLEY		LOW PROTEIN BARLEY		PROTEIN-FREE	
CEREAL COMPONENT	WHOLE WHEAT	BRAN	ENDO-SPERM	WHOLE WHEAT	BRAN	ENDO-SPERM	WHOLE BARLEY	ENDO-SPERM	WHOLE BARLEY	ENDO-SPERM	10% FILLER	20% FILLER	
Weight of rats (g) <sup>1</sup>													
71													
98	14.3±1.3	12.7±0.8	14.0±1.5	13.7±1.4	12.4±1.4	13.2±2.5	14.5±1.6	13.2±1.4	13.0±1.0	12.0±0.7	8.0±0.7	8.8±0.9	
170	21.7±2.7	16.8±2.9	20.5±1.9	19.5±2.3	18.7±3.2	19.5±3.9	22.7±1.1	21.8±1.3	21.5±3.0	21.8±2.1			
207											18.5±2.8	20.1±2.4	

<sup>1</sup> Weight of the rats on the day prior to each test period.

amino acids due to weight. For the amino acids mentioned, apparent availabilities were only 1 to 2% higher for the heavier than for the lighter rats. Only one weight x test diet interaction was found, namely for HIS. The dry matter digestibility was significantly higher (by 0.9%) for the heavier than for the lighter rats.

Nitrogen and dry matter digestibilities were significantly higher from wheat endosperm than from wheat bran (Table 15). Those of whole wheat were between those of endosperm and bran. Apparent amino acid availabilities from whole wheat and its derived fractions followed the same pattern. However, the differences in percentage units even if significant were of a small magnitude. Lysine was always the least available amino acid and ranged from 78.9 to 82.2% for the high protein wheat and its fractions and from 71.4 to 73.4 for the low protein wheat and its fractions. Threonine was always the second least available essential amino acid. Isoleucine and MET were also of low availability (less than 80%) for bran from the low protein wheat. For the non-essential amino acids, ALA and ASP were always the least available.

Dry matter digestibility, apparent nitrogen digestibility and amino acid availabilities were higher for endosperm of barley than of whole barley (Table 16). The apparent availabilities of lysine were very low: they were

TABLE 15.1 Means<sup>1</sup> and standard error of the mean of the apparent availabilities of amino acids from wheat and its fractions.

DIET	HIGH PROTEIN WHEAT			LOW PROTEIN WHEAT			$S_{\bar{x}}^2$
CEREAL COMPONENT	WHOLE WHEAT	BRAN	ENDO-SPERM	WHOLE WHEAT	BRAN	ENDO-SPERM	
AMINO ACIDS (%)							
<u>Essential</u>							
ARG	91.2 <sup>B</sup>	92.4 <sup>A</sup>	92.9 <sup>A</sup>	87.8 <sup>C</sup>	87.8 <sup>C</sup>	88.7 <sup>C</sup>	0.49
HIS	93.7 <sup>A</sup>	93.1 <sup>AB</sup>	94.6 <sup>A</sup>	90.8 <sup>C</sup>	87.9 <sup>D</sup>	91.9 <sup>BC</sup>	0.48
ILE	90.4 <sup>A</sup>	87.0 <sup>CD</sup>	92.8 <sup>A</sup>	85.3 <sup>DE</sup>	77.7 <sup>F</sup>	88.1 <sup>C</sup>	0.62
LEU	92.7 <sup>A</sup>	89.8 <sup>B</sup>	94.3 <sup>A</sup>	89.4 <sup>B</sup>	83.0 <sup>D</sup>	91.0 <sup>B</sup>	0.57
LYS	78.9 <sup>B</sup>	76.9 <sup>B</sup>	82.2 <sup>A</sup>	73.4 <sup>C</sup>	71.4 <sup>CD</sup>	72.7 <sup>C</sup>	1.02
MET <sup>3</sup>	91.0 <sup>A</sup>	86.5 <sup>B</sup>	92.1 <sup>A</sup>	87.9 <sup>B</sup>	77.0 <sup>E</sup>	87.9 <sup>B</sup>	0.75
PHE	94.0 <sup>A</sup>	91.7 <sup>CD</sup>	95.4 <sup>A</sup>	90.8 <sup>D</sup>	84.8 <sup>G</sup>	92.2 <sup>C</sup>	0.40
THR	86.8 <sup>B</sup>	83.5 <sup>C</sup>	89.7 <sup>A</sup>	81.9 <sup>CD</sup>	74.9 <sup>E</sup>	83.7 <sup>C</sup>	0.73
VAL	90.4 <sup>A</sup>	87.6 <sup>CD</sup>	92.7 <sup>A</sup>	86.3 <sup>D</sup>	80.7 <sup>E</sup>	88.6 <sup>C</sup>	0.53
<u>Non-Essential</u>							
ALA	85.2 <sup>B</sup>	82.4 <sup>C</sup>	88.0 <sup>A</sup>	80.9 <sup>C</sup>	75.8 <sup>D</sup>	81.2 <sup>C</sup>	0.68
ASP	85.3 <sup>A</sup>	82.5 <sup>B</sup>	87.7 <sup>A</sup>	79.1 <sup>C</sup>	74.1 <sup>DE</sup>	80.5 <sup>BC</sup>	0.83
GLU	97.4 <sup>A</sup>	96.0 <sup>B</sup>	98.0 <sup>A</sup>	95.7 <sup>B</sup>	91.0 <sup>D</sup>	96.5 <sup>B</sup>	0.26
GLY	89.8 <sup>B</sup>	86.0 <sup>C</sup>	92.2 <sup>A</sup>	84.8 <sup>C</sup>	77.1 <sup>E</sup>	86.8 <sup>C</sup>	0.55
PRO	97.3 <sup>AB</sup>	95.5 <sup>D</sup>	98.0 <sup>A</sup>	95.9 <sup>CD</sup>	89.0 <sup>G</sup>	96.8 <sup>BC</sup>	0.29
SER	92.6 <sup>B</sup>	89.1 <sup>C</sup>	94.4 <sup>A</sup>	89.4 <sup>C</sup>	82.0 <sup>E</sup>	90.7 <sup>C</sup>	0.49
TYR	91.8 <sup>A</sup>	88.4 <sup>B</sup>	93.3 <sup>A</sup>	85.6 <sup>C</sup>	78.1 <sup>E</sup>	87.4 <sup>B</sup>	0.59
NITROGEN (%)	89.7 <sup>B</sup>	83.7 <sup>C</sup>	92.2 <sup>A</sup>	84.4 <sup>C</sup>	75.6 <sup>E</sup>	87.5 <sup>B</sup>	0.66
DRY MATTER (%)	90.3 <sup>C</sup>	73.3 <sup>F</sup>	94.1 <sup>A</sup>	88.8 <sup>D</sup>	69.0 <sup>G</sup>	94.0 <sup>A</sup>	0.29

<sup>1</sup> Means in the same row with the same superscript do not differ significantly ( $P < .05$ )

<sup>2</sup> Standard error of the mean.

<sup>3</sup> Determined by acid hydrolysis.

TABLE 16. Means<sup>1</sup> and standard error of the mean of the apparent availabilities of amino acids from whole barley and endosperm from barley.

DIET	HIGH PROTEIN BARLEY		LOW PROTEIN BARLEY		$S_{\bar{x}}^2$
CEREAL COMPONENT	WHOLE BARLEY	ENDO- SPERM	WHOLE BARLEY	ENDO- SPERM	
AMINO ACIDS (%)					
<u>Essential</u>					
ARG	82.3 <sup>E</sup>	85.2 <sup>E</sup>	74.7 <sup>G</sup>	81.0 <sup>F</sup>	0.49
HIS	83.6 <sup>E</sup>	88.4 <sup>D</sup>	78.0 <sup>F</sup>	83.5 <sup>E</sup>	0.48
ILE	79.0 <sup>F</sup>	83.5 <sup>E</sup>	70.6 <sup>G</sup>	79.6 <sup>F</sup>	0.62
LEU	82.5 <sup>D</sup>	87.3 <sup>C</sup>	77.3 <sup>E</sup>	83.4 <sup>D</sup>	0.57
LYS	67.7 <sup>D</sup>	70.8 <sup>CD</sup>	60.1 <sup>E</sup>	68.2 <sup>D</sup>	1.02
MET <sup>3</sup>	81.0 <sup>CD</sup>	82.8 <sup>C</sup>	71.5 <sup>F</sup>	79.3 <sup>D</sup>	0.75
PHE	86.0 <sup>F</sup>	89.3 <sup>E</sup>	78.6 <sup>I</sup>	83.5 <sup>H</sup>	0.40
THR	76.0 <sup>E</sup>	79.6 <sup>D</sup>	67.8 <sup>F</sup>	73.7 <sup>E</sup>	0.73
VAL	82.0 <sup>E</sup>	86.5 <sup>D</sup>	73.7 <sup>F</sup>	80.5 <sup>E</sup>	0.53
<u>Non-Essential</u>					
ALA	73.2 <sup>E</sup>	77.3 <sup>D</sup>	67.0 <sup>F</sup>	71.3 <sup>E</sup>	0.68
ASP	72.8 <sup>DE</sup>	75.6 <sup>D</sup>	67.2 <sup>F</sup>	71.5 <sup>E</sup>	0.83
GLU	89.8 <sup>E</sup>	93.5 <sup>C</sup>	84.1 <sup>G</sup>	87.6 <sup>F</sup>	0.26
GLY	76.0 <sup>E</sup>	81.4 <sup>D</sup>	70.0 <sup>G</sup>	74.9 <sup>F</sup>	0.55
PRO	90.5 <sup>F</sup>	94.3 <sup>E</sup>	83.6 <sup>H</sup>	89.0 <sup>G</sup>	0.29
SER	82.8 <sup>E</sup>	86.0 <sup>D</sup>	74.2 <sup>G</sup>	80.2 <sup>F</sup>	0.49
TYR	82.8 <sup>D</sup>	84.5 <sup>CD</sup>	74.7 <sup>F</sup>	79.0 <sup>E</sup>	0.59
NITROGEN (%)	78.7 <sup>D</sup>	83.1 <sup>C</sup>	72.8 <sup>F</sup>	76.1 <sup>E</sup>	0.66
DRY MATTER (%)	83.7 <sup>C</sup>	93.2 <sup>A</sup>	83.5 <sup>E</sup>	91.3 <sup>B</sup>	0.29

<sup>1</sup> Means in the same row with the same superscript do not differ significantly ( $P < 0.05$ ).

<sup>2</sup> Standard error of the mean.

<sup>3</sup> Determined by acid hydrolysis.



67.7 and 60.1% for the high and low protein barley respectively. Lysine availabilities from endosperm were slightly higher. Threonine was always the second least available essential amino acid. For the non-essential amino acids, ASP, ALA and GLY were of low apparent availability.

Apparent amino acid availabilities of high and low protein wheat and barley are compared separately in Table 17. Apparent availabilities of whole wheat are significantly higher than those of whole barley. The availabilities of amino acids from high protein wheat were higher than those from low protein wheat. The same relationship was found for high and low protein barley.

The metabolic faecal amino acid excretion increased as the level of fiber of the protein-free diets was increased from 10 to 20% (Table 18). Significant differences in amino acid excretions due to weight, in addition to a significant weight x test diet interaction for nitrogen were found (Table 18; Appendix, Table 2). For both protein-free diets, the excretion of ARG was significantly higher for the heavier than for the lighter rats. The excretions of LEU, MET and VAL were significantly higher for the lighter than for the heavier rats when these were fed the 10% alphafloc protein-free diet. Leucine, MET, VAL, ALA and ASP excretions were significantly higher for the

TABLE 17. Means<sup>1</sup> and standard error of the mean of apparent amino acid availabilities of high and low protein wheat and barley.

CEREAL GRAIN	WHEAT		BARLEY		$S_{\bar{x}}^2$
LEVEL OF PROTEIN (%)	18.3	11.2	11.4	8.1	
AMINO ACIDS (%)					
<u>Essential</u>					
ARG	91.2 <sup>B</sup>	87.8 <sup>C</sup>	82.3 <sup>E</sup>	74.7 <sup>G</sup>	0.49
HIS	93.7 <sup>A</sup>	90.8 <sup>C</sup>	83.6 <sup>E</sup>	78.0 <sup>F</sup>	0.48
ILE	90.4 <sup>A</sup>	85.3 <sup>DE</sup>	79.0 <sup>F</sup>	70.6 <sup>G</sup>	0.62
LEU	92.7 <sup>A</sup>	89.4 <sup>B</sup>	82.5 <sup>D</sup>	77.3 <sup>E</sup>	0.57
LYS	78.9 <sup>B</sup>	73.4 <sup>C</sup>	67.7 <sup>D</sup>	60.1 <sup>E</sup>	1.02
MET <sup>3</sup>	91.0 <sup>A</sup>	87.9 <sup>B</sup>	81.0 <sup>CD</sup>	71.5 <sup>F</sup>	0.75
PHE	94.0 <sup>A</sup>	90.8 <sup>D</sup>	86.0 <sup>F</sup>	78.6 <sup>I</sup>	0.40
THR	86.8 <sup>B</sup>	81.9 <sup>CD</sup>	76.0 <sup>E</sup>	67.8 <sup>F</sup>	0.73
VAL	90.4 <sup>A</sup>	86.3 <sup>D</sup>	82.0 <sup>E</sup>	73.7 <sup>F</sup>	0.53
<u>Non-Essential</u>					
ALA	85.2 <sup>B</sup>	80.9 <sup>C</sup>	73.2 <sup>E</sup>	67.0 <sup>F</sup>	0.68
ASP	85.3 <sup>A</sup>	79.1 <sup>C</sup>	72.8 <sup>DE</sup>	67.2 <sup>F</sup>	0.83
GLU	97.4 <sup>A</sup>	95.7 <sup>B</sup>	89.8 <sup>E</sup>	84.1 <sup>G</sup>	0.26
GLY	89.8 <sup>B</sup>	84.8 <sup>C</sup>	76.0 <sup>E</sup>	70.0 <sup>G</sup>	0.55
PRO	97.3 <sup>AB</sup>	95.9 <sup>CD</sup>	90.5 <sup>F</sup>	83.6 <sup>H</sup>	0.29
SER	92.6 <sup>B</sup>	89.4 <sup>C</sup>	82.8 <sup>E</sup>	74.2 <sup>G</sup>	0.49
TYR	91.8 <sup>A</sup>	85.6 <sup>C</sup>	82.8 <sup>D</sup>	74.7 <sup>F</sup>	0.59
NITROGEN (%)	89.7 <sup>B</sup>	84.4 <sup>C</sup>	78.7 <sup>D</sup>	72.8 <sup>F</sup>	0.66
DRY MATTER (%)	90.3 <sup>C</sup>	88.8 <sup>D</sup>	83.7 <sup>E</sup>	83.5 <sup>E</sup>	0.29

<sup>1</sup> Means in the same row with the same superscript do not differ significantly ( $P < 0.05$ ).

<sup>2</sup> Standard error of the mean.

<sup>3</sup> Determined by acid hydrolysis.

TABLE 18. The effect of dietary fiber level and weight of the rats on the metabolic faecal amino acid and nitrogen excretion<sup>1</sup>.

LEVEL OF ALPHAFLOC (%)	10		20		$S_{\bar{x}}^2$
WEIGHT (g)	71	207	71	207	
AMINO ACIDS <sup>3</sup>					
<u>Essential</u>					
ARG	.0169 <sup>D</sup>	.0191 <sup>C</sup>	.0226 <sup>B</sup>	.0263 <sup>A</sup>	.0072
HIS	.0071 <sup>B</sup>	.0078 <sup>B</sup>	.0103 <sup>A</sup>	.0106 <sup>A</sup>	.0043
ILE	.0214 <sup>B</sup>	.0194 <sup>B</sup>	.0322 <sup>A</sup>	.0298 <sup>A</sup>	.0076
LEU	.0367 <sup>C</sup>	.0328 <sup>D</sup>	.0541 <sup>A</sup>	.0497 <sup>B</sup>	.0123
LYS	.0294 <sup>B</sup>	.0274 <sup>B</sup>	.0418 <sup>A</sup>	.0411 <sup>A</sup>	.0094
MET <sup>4</sup>	.0104 <sup>C</sup>	.0082 <sup>D</sup>	.0162 <sup>A</sup>	.0123 <sup>B</sup>	.0043
PHE	.0203 <sup>B</sup>	.0192 <sup>B</sup>	.0314 <sup>A</sup>	.0285 <sup>A</sup>	.0078
THR	.0279 <sup>B</sup>	.0247 <sup>B</sup>	.0388 <sup>A</sup>	.0366 <sup>A</sup>	.0091
VAL	.0267 <sup>C</sup>	.0236 <sup>D</sup>	.0399 <sup>A</sup>	.0369 <sup>B</sup>	.0099
<u>Non-Essential</u>					
ALA	.0322 <sup>C</sup>	.0292 <sup>C</sup>	.0476 <sup>A</sup>	.0427 <sup>B</sup>	.0110
ASP	.0523 <sup>C</sup>	.0478 <sup>C</sup>	.0763 <sup>A</sup>	.0707 <sup>B</sup>	.0174
GLU	.0619 <sup>B</sup>	.0569 <sup>B</sup>	.0905 <sup>A</sup>	.0841 <sup>A</sup>	.0209
GLY	.0286 <sup>B</sup>	.0281 <sup>B</sup>	.0417 <sup>A</sup>	.0405 <sup>A</sup>	.0101
PRO	.0234 <sup>B</sup>	.0229 <sup>B</sup>	.0320 <sup>A</sup>	.0326 <sup>A</sup>	.0115
SER	.0262 <sup>B</sup>	.0247 <sup>B</sup>	.0388 <sup>A</sup>	.0366 <sup>A</sup>	.0088
TYR	.0180 <sup>B</sup>	.0164 <sup>B</sup>	.0266 <sup>A</sup>	.0253 <sup>A</sup>	.0077
NITROGEN	.2630 <sup>AB</sup>	.2204 <sup>B</sup>	.3188 <sup>A</sup>	.3309 <sup>A</sup>	.0934

1 Means are expressed as grams excreted per 100 grams of dry matter intake of the protein-free diets.

2 Standard error of the mean.

3 Means in the same row with the same superscript do not differ significantly ( $P < 0.05$ ).

4 Determined by acid hydrolysis.

lighter than for the heavier rats when these were fed the 20% alphafloc protein-free diets.

The differences due to weight, although significant, were of small magnitude. Most variation was due to the level of alphafloc in the protein-free diets as is shown by the large mean squares for test diets (Appendix, Table 2).

The metabolic faecal nitrogen excretion increased as the level of alphafloc was raised. However, there was no significant difference between the nitrogen excretion of the lighter rats fed the 10% alphafloc protein-free diet and for the heavier and lighter rats fed the 20% alphafloc protein-free diet (Table 18). With the exception of contamination from urinary nitrogen, no satisfactory explanation can be given for this observation.

#### Study 2.

##### Study 2a.

The nitrogen content decreased from wheat to barley to corn. The levels of crude and acid-detergent fiber were somewhat higher for corn than for wheat while the level of neutral-detergent fiber was slightly lower. Chemical analysis showed that approximately 70% of alphafloc is actually crude fiber (Table 19).

With the exception of LEU, LYS and MET the levels of essential amino acids decreased from wheat to barley to

TABLE 19. Proximate, acid and neutral-detergent fiber, starch and chromic oxide analyses<sup>1</sup> for diets in study 2.

DIETS	BARLEY	CORN	WHEAT	MAINTENANCE	
LEVEL OF ALPHAFLOC (%)				7	14
<u>ITEMS (%)</u>					
Nitrogen	2.22	2.00	2.61	0.66	0.66
Ether extract	1.89	3.87	1.70	1.30	1.17
Fiber					
Crude	5.37	2.62	2.56	4.95	9.62
Acid-detergent	7.34	3.95	3.57		
Neutral-detergent	21.47	11.73	12.22		
Ash	5.32	4.76	4.29		
Starch	64.35	72.40	66.65		
Chromic oxide	0.47	0.52	0.43	0.53	0.54

<sup>1</sup> Percent expressed on a dry weight basis.

corn. The level of LYS decreased from barley to wheat to corn. The levels of LEU and MET were highest in corn (Table 20). The results obtained for TRY from duplicate analyses on diets and digesta varied extensively. Therefore, TRY was not considered in these studies.

The metabolic ileal and faecal amino acid levels were estimated at 2 levels of dietary fiber by aid of the M-7 and M-14 diets. These diets contained 4% casein. Protein-free diets are usually employed for the estimation of metabolic losses. However, low dietary consumption caused by palatability problems with pigs fed protein-free diets is often a major problem. It was not thought advisable to subject the cannulated pigs to this extra stress. Consequently, a highly digestible protein at a low level (4% casein) was included in the protein-free diets. These diets were named maintenance diets. It was assumed at this time that all amino acids from casein would be absorbed by the end of the small intestine and that the ileal and faecal levels found by feeding the maintenance diets would be representative of the metabolic levels.

In order to avoid complications due to blockage (which could have resulted in permanent problems), the M-14 diet was not fed prior to the cereal test diet sequence but only near the end of the complete experiment (Fig. 2).

TABLE 20. Amino acid composition of diets for study 2.

DIETS	BARLEY		CORN		WHEAT		M-7		M-14	
	% <sup>1</sup>	g/16gN <sup>2</sup>	%	g/16gN	%	g/16gN	%	g/16gN	%	g/16gN
AMINO ACIDS										
<u>Essential</u>										
ARG	0.57	4.10	0.45	3.61	0.58	3.56	0.12	2.95	0.12	2.95
HIS	0.23	1.66	0.25	2.01	0.29	1.78	0.10	2.46	0.10	2.46
ILE	0.39	2.81	0.35	2.81	0.44	2.70	0.19	4.67	0.20	4.92
LEU	0.86	6.19	1.31	10.51	0.97	5.95	0.38	9.34	0.39	9.59
LYS	0.42	3.02	0.34	2.73	0.36	2.21	0.30	7.37	0.29	7.13
MET	0.19 <sup>3</sup>	1.37	0.25 <sup>3</sup>	2.00	0.24 <sup>3</sup>	1.47	0.10 <sup>4</sup>	2.42	0.10 <sup>4</sup>	2.42
PHE	0.60	4.32	0.52	4.17	0.67	4.11	0.21	5.16	0.20	4.92
THR	0.38	2.74	0.36	2.89	0.39	2.39	0.16	3.93	0.16	3.93
VAL	0.54	3.89	0.46	3.69	0.55	3.37	0.24	5.90	0.24	5.90
<u>Non-Essential</u>										
ALA	0.48	3.46	0.76	6.10	0.49	3.00	0.13	3.20	0.14	3.44
ASP	0.69	4.97	0.72	5.77	0.68	4.17	0.32	7.86	0.33	8.11
CYS	0.22 <sup>3</sup>	1.59	0.20 <sup>3</sup>	1.60	0.28 <sup>3</sup>	1.72	--	--	--	--
GLU	2.93	21.10	2.09	16.76	4.44	27.23	0.92	22.61	0.91	22.37
GLY	0.49	3.53	0.40	3.21	0.57	3.50	0.09	2.21	0.09	2.21
PRO	1.22	8.78	0.84	6.74	1.45	8.89	0.39	9.59	0.40	9.83
SER	0.50	3.60	0.51	4.09	0.63	3.86	0.30	4.92	0.19	4.67
TYR	0.36	2.59	0.40	3.21	0.41	2.51	0.17	4.18	0.17	4.18

- 1 Percent expressed on a dry matter basis.
- 2 Grams per sixteen grams of nitrogen.
- 3 Determined by the oxidation method.
- 4 Determined by acid hydrolysis.

Three separate consecutive 8-hour ileal collections were carried out. The samples were frozen and stored. Accidentally, several ileal and faecal collections were removed from the freezer and were spoiled. The ileal samples that were spoiled were the first and third 8-hour collections from pig number 1 fed wheat and the second and third ileal collections from pig number 6 fed wheat. In addition, all ileal and faecal samples from the second time the M-7 diet was fed were spoiled.

The amino acid and chromic oxide composition of every 8-hour ileal collection of pig number 2, 7, 3 and 4 when fed wheat are compared in Table 21. There were only small differences between the individual 8-hour collections for each pig. This finding was not unexpected since the animals were fed 3 times daily, the same amount at each feeding and exactly 8 hours apart. Under the experimental conditions carried out, the data show that the amino acid and chromic oxide content from ileal digesta from wheat obtained during each single 8-hour collection is representative of that obtained during a complete 24-hour ileal collection.

The apparent ileal and faecal amino acid availabilities of the cereal grains are shown in Table 22. The ileal amino acid availabilities were obtained from



TABLE 21. Comparisons<sup>1</sup> of the individual eight-hour ileal collections from wheat for study 2a.

NUMBER OF PIG	2			7			3			4		
PERIOD	I			I			III			III		
TIME OF COLLECTION <sup>2</sup>	0-8	8-16	16-24	0-8	8-16	16-24	0-8	8-16	16-24	0-8	8-16	16-24
AMINO ACIDS (%) <sup>3</sup>												
<u>Essential</u>												
ARG	0.34	0.34	0.33	0.32	0.32	0.29	0.27	0.27	0.27	0.32	0.29	0.33
HIS	0.16	0.18	0.17	0.17	0.18	0.16	0.12	0.16	0.14	0.16	0.14	0.17
ILE	0.26	0.26	0.26	0.30	0.29	0.29	0.23	0.22	0.22	0.26	0.27	0.28
LEU	0.49	0.49	0.49	0.55	0.53	0.53	0.44	0.44	0.44	0.49	0.49	0.51
LYS	0.34	0.34	0.34	0.38	0.38	0.37	0.26	0.29	0.28	0.35	0.31	0.37
PHE	0.31	0.31	0.31	0.36	0.35	0.34	0.28	0.27	0.27	0.28	0.31	0.34
THR	0.36	0.35	0.35	0.39	0.41	0.43	0.32	0.32	0.33	0.34	0.35	0.35
VAL	0.40	0.40	0.39	0.43	0.41	0.42	0.35	0.35	0.36	0.37	0.37	0.37
<u>Non-Essential</u>												
ALA	0.43	0.42	0.42	0.54	0.51	0.52	0.35	0.37	0.37	0.43	0.44	0.44
ASP	0.63	0.62	0.62	0.80	0.76	0.77	0.51	0.52	0.53	0.62	0.63	0.63
GLU	1.22	1.20	1.20	1.50	1.40	1.35	0.97	0.99	0.98	1.22	1.25	1.25
GLY	0.53	0.51	0.51	0.80	0.73	0.74	0.44	0.45	0.45	0.48	0.49	0.49
PRO	0.64	0.61	0.65	0.65	0.63	0.56	0.44	0.48	0.41	0.52	0.59	0.46
SER	0.36	0.35	0.35	0.44	0.41	0.43	0.32	0.33	0.32	0.34	0.35	0.35
TYR	0.17	0.19	0.18	0.21	0.20	0.19	0.17	0.17	0.15	0.18	0.17	0.19
CHROMIC OXIDE(%)	1.66	1.39	1.69	1.29	1.39	1.31	1.79	1.64	1.62	1.64	1.51	1.56

<sup>1</sup> Comparisons for pig number 1 and 6 could not be made since only the second (8-16) and the first 8-hour (0-8) ileal collections respectively were available.

<sup>2</sup> Time interval after the start of the collection.

<sup>3</sup> Percent expressed on a dry matter basis.

TABLE 22. Apparent ileal and faecal amino acid availabilities<sup>1</sup> of the cereal grains in study 2a.

CEREAL GRAIN	CORN		WHEAT		BARLEY		$S_{\bar{x}}^2$
LOCATION	ILEUM	FAECES	ILEUM <sup>3</sup>	FAECES	ILEUM	FAECES	
AMINO ACIDS (%) <sup>4</sup>							
<u>Essential</u>							
ARG	87.4 <sup>C</sup>	92.2 <sup>A</sup>	85.8 <sup>C</sup>	92.7 <sup>A</sup>	81.5 <sup>D</sup>	89.4 <sup>B</sup>	0.595
HIS	88.3 <sup>D</sup>	93.6 <sup>AB</sup>	89.1 <sup>CD</sup>	94.9 <sup>A</sup>	80.4 <sup>E</sup>	91.9 <sup>BC</sup>	0.665
ILE	87.5 <sup>A</sup>	88.1 <sup>A</sup>	85.3 <sup>B</sup>	89.4 <sup>A</sup>	79.1 <sup>C</sup>	83.1 <sup>B</sup>	0.761
LEU	92.5 <sup>A</sup>	93.8 <sup>A</sup>	86.9 <sup>B</sup>	91.5 <sup>A</sup>	81.5 <sup>C</sup>	86.6 <sup>B</sup>	0.630
LYS	82.0 <sup>A</sup>	83.0 <sup>A</sup>	75.7 <sup>B</sup>	80.7 <sup>A</sup>	73.3 <sup>C</sup>	77.5 <sup>B</sup>	0.794
MET <sup>5</sup>	91.9 <sup>A</sup>	89.5 <sup>A</sup>	86.6 <sup>A</sup>	88.9 <sup>A</sup>	80.4 <sup>B</sup>	79.9 <sup>B</sup>	0.877
PHE	90.5 <sup>AB</sup>	91.3 <sup>A</sup>	88.8 <sup>BC</sup>	92.5 <sup>A</sup>	82.2 <sup>D</sup>	87.9 <sup>C</sup>	0.564
THR	78.9 <sup>B</sup>	86.3 <sup>A</sup>	76.5 <sup>C</sup>	86.7 <sup>A</sup>	71.2 <sup>D</sup>	81.4 <sup>B</sup>	0.938
VAL	84.9 <sup>B</sup>	88.2 <sup>A</sup>	82.8 <sup>A</sup>	88.9 <sup>A</sup>	78.0 <sup>C</sup>	84.3 <sup>B</sup>	0.720
<u>Non-Essential</u>							
ALA	88.5 <sup>A</sup>	90.8 <sup>A</sup>	74.0 <sup>D</sup>	84.0 <sup>B</sup>	69.7 <sup>E</sup>	77.4 <sup>C</sup>	0.901
ASP	83.9 <sup>B</sup>	86.6 <sup>A</sup>	75.4 <sup>C</sup>	83.1 <sup>B</sup>	71.2 <sup>D</sup>	77.9 <sup>C</sup>	0.883
CYS <sup>5</sup>	82.1 <sup>AB</sup>	90.2 <sup>AB</sup>	85.2 <sup>AB</sup>	93.7 <sup>A</sup>	77.6 <sup>B</sup>	88.7 <sup>AB</sup>	2.035
GLU	91.8 <sup>C</sup>	94.1 <sup>B</sup>	92.7 <sup>BC</sup>	97.0 <sup>A</sup>	86.6 <sup>D</sup>	92.7 <sup>BC</sup>	0.480
GLY	71.2 <sup>C</sup>	86.2 <sup>A</sup>	73.7 <sup>C</sup>	89.3 <sup>A</sup>	71.2 <sup>D</sup>	82.6 <sup>B</sup>	1.191
PRO	80.4 <sup>C</sup>	93.4 <sup>A</sup>	86.8 <sup>B</sup>	96.7 <sup>A</sup>	80.9 <sup>C</sup>	92.3 <sup>A</sup>	1.495
SER	84.9 <sup>B</sup>	91.0 <sup>A</sup>	84.1 <sup>B</sup>	92.5 <sup>A</sup>	76.3 <sup>C</sup>	86.9 <sup>B</sup>	0.714
TYR	89.0 <sup>A</sup>	90.2 <sup>A</sup>	85.9 <sup>B</sup>	89.5 <sup>A</sup>	79.7 <sup>C</sup>	85.0 <sup>B</sup>	0.680
NITROGEN (%)	82.4 <sup>D</sup>	89.4 <sup>B</sup>	82.9 <sup>D</sup>	91.2 <sup>A</sup>	74.9 <sup>E</sup>	85.9 <sup>C</sup>	0.517
DRY MATTER (%)	80.2 <sup>B</sup>	89.4 <sup>A</sup>	73.3 <sup>C</sup>	89.0 <sup>A</sup>	66.2 <sup>D</sup>	83.6 <sup>B</sup>	0.979

<sup>1</sup> Each value is the mean of 6 observations. <sup>2</sup> Standard error of the mean.

<sup>3</sup> Ileal availabilities from pig number 1 and 6 were based on single 8-hour collections.

<sup>4</sup> Means in the same row with the same superscript do not differ significantly ( $P < .05$ ).

<sup>5</sup> Determined by the oxidation method.

pooling of the 3 individual 8-hour collections that were derived from each of the cereal grains. However, apparent ileal amino acid availabilities from pig number 1 and 6 were based on one single 8-hour collection. Both ileal and faecal amino acid availabilities were determined by the chromic oxide method (Crampton and Harris, 1969). For the determination of MET and CYS availabilities, faeces from the 2 pigs, fed the same diet during each particular test period, were pooled and analyzed (Fig. 2). The individual apparent ileal and faecal amino acid availabilities and analyses of variance are shown in the appendix (Table 3, 4, 5, 6, 7 and 8).

There were no significant differences between the apparent faecal availabilities of essential amino acids from wheat and corn (Table 22). The availabilities of essential amino acids from wheat and corn were significantly higher than those from barley. The differences in LYS availabilities were of a small magnitude. They were 83.0, 80.7 and 77.5% for corn, wheat and barley respectively.

The apparent faecal availabilities of the non-essential amino acids from wheat and corn were higher than those from barley. The availabilities of ALA and ASP from corn were significantly higher than those from wheat. There were no significant differences between the apparent PRO availabilities of the cereal grains tested.

Generally, the ileal availabilities of amino acids decreased from corn to wheat to barley. The LYS availability, in particular, decreased from 82.0 to 75.7 to 73.3% for corn, wheat and barley respectively.

Faecal availabilities were higher than ileal availabilities for all amino acids for each of the cereal grains tested. The differences were largest for ARG, HIS, THR, GLY, PRO and SER. In most cases, there was less difference between ileal and faecal amino acid availabilities from corn than from wheat and barley. For instance, the difference for LYS was 1% for corn and 5 and 4.2% for wheat and barley respectively. The difference for THR was 7.4, 10.2 and 10.2% for corn, wheat and barley respectively.

Threonine was the least available essential amino acid for corn and barley when determined at the end of the ileum. Lysine was the least available amino acid from corn and barley when determined by the faecal analysis method. Lysine was the least available amino acid from wheat regardless of ileal or faecal collection (Table 22).

Significant pig effects were observed for some amino acids (Appendix, Table 6 and 7). The apparent amino acid availabilities from pig number 7 (replacement pig) were generally lower than those of the other pigs, in particular, when determined at the end of the ileum. The latter resulted

also in several significant pig x location interactions.

Dry matter digestibilities based on total ileal dry matter collection and chromic oxide levels were nearly the same for the cereal grains (Table 23). Individual comparisons are shown in the appendix (Table 3, 4 and 5). Dry matter digestibilities determined by chromic oxide levels were lower for M-7 and M-14 than when determined by total ileal dry matter excretion. Data obtained from M-7 represent those from the first 24-hour ileal collection that was carried out. Lack of experience with regard to the collection procedure resulted in unforeseen losses of ileal digesta at that time. Blockage and associated problems occurred sometimes when the pigs were fed the M-7 diets but very often when they were fed the M-14 diets. For the M-14 diet, feed intake was very irregular and meals were often skipped. Some pigs started nibbling instead of consuming the meals within a short time after feeding. The average daily dry matter intakes by the pigs fed M-7 and M-14 were 1486 (87.8% of allowable intake) and 935 grams (48.9% of allowable intake) respectively (Appendix, Table 9 and 10). The cereal test diets in these studies were always consumed within 1 hour after feeding (ie. 100% of allowable intake) and the amount of ileal dry matter collected during 3 consecutive 8-hour collections could be

TABLE 23. Comparison of ileal dry matter digestibilities<sup>1</sup> based on total dry matter excretion and chromic oxide levels.

Measurement	Total	Chromic Oxide
<u>DIETS</u>		
Barley (6) <sup>2</sup>	67.11±2.87	66.17±3.86
Corn (6)	80.06±2.16	80.18±0.77
Wheat (4)	73.11±0.77	73.25±2.05
Maintenance, 7% alphafloc (5)	88.52±2.54	86.56±0.48
Maintenance, 14% alphafloc (6)	84.5±5.72	76.8±0.34

<sup>1</sup> Mean and standard deviation.

<sup>2</sup> Values in parentheses indicate the number of observations.

related back to the total dry matter intake from 3 feedings. Ileal dry matter digestibilities based on total ileal dry matter were calculated on the basis of the latter principle. The higher ileal dry matter digestibilities obtained from M-7 and M-14 by total ileal dry matter levels than by chromic oxide levels in part reflect the lower intake of these diets by the pigs in comparison to their allowable daily dry matter intake.

The average metabolic ileal and faecal amino acid and nitrogen levels, expressed as grams per 100 grams of dry matter intake, are shown in Table 24. The individual metabolic ileal and faecal amino acid levels of the pigs are shown in the appendix (Table 9 and 10). All ileal and faecal collections from pigs fed the M-7 diet following the cereal test diet sequence were spoiled. Pig number 6, fed M-7 prior to the cereal test diet sequence, went off feed the day before the ileal collection was carried out.

Not taking into account the possible effect of body weight on metabolic and faecal amino acid levels, these levels were increased as the level of alphafloc was raised from 7 to 14% (Table 24). For both the M-7 and M-14 diets, there was a net loss (as grams per 100 grams of dry matter intake) of total nitrogen between the end of the ileum and the anus. There was a net disappearance of ARG, GLU, GLY,

TABLE 24. Metabolic ileal and faecal amino acid levels<sup>1</sup> from study 2a.

PERIOD OF COLLECTION	BEFORE CEREAL DIETS <sup>2</sup>		AFTER CEREAL DIETS <sup>3</sup>	
LEVEL OF ALPHAFLOC (%)	7		14	
LOCATION	ILEUM (5) <sup>4</sup>	FAECES (5)	ILEUM (6)	FAECES (6)
AMINO ACIDS				
<u>Essential</u>				
ARG	0.0380±0.0119	0.0288±0.0120	0.0460±0.0056	0.0347±0.0130
HIS	0.0126±0.0033	0.0114±0.0034	0.0177±0.0041	0.0152±0.0422
ILE	0.0326±0.0030	0.0332±0.0085	0.0352±0.0417	0.0442±0.0143
LEU	0.0424±0.0090	0.0502±0.0182	0.0447±0.0033	0.0681±0.0292
LYS	0.0356±0.0061	0.0456±0.0131	0.0428±0.0020	0.0567±0.0202
MET <sup>5</sup>	0.0088±0.0015	0.0162±0.0056	0.0110±0.0009	0.0198±0.0074
PHE	0.0224±0.0058	0.0310±0.0099	0.0238±0.0029	0.0407±0.0171
THR	0.0444±0.0082	0.0342±0.0939	0.0455±0.0034	0.0468±0.0172
VAL	0.0384±0.0063	0.0380±0.0101	0.0418±0.0023	0.0510±0.0176
<u>Non-Essential</u>				
ALA	0.0442±0.0096	0.0462±0.0119	0.0572±0.0117	0.0632±0.0231
ASP	0.0662±0.0071	0.0720±0.0189	0.0732±0.0022	0.0943±0.0309
GLU	0.1238±0.0115	0.0948±0.0208	0.1423±0.0192	0.1250±0.0376
GLY	0.1146±0.0425	0.0376±0.0111	0.1452±0.0690	0.0497±0.0192
PRO	0.3438±0.1243	0.0386±0.0131	0.5548±0.3484	0.0665±0.0212
SER	0.0670±0.0064	0.0366±0.0078	0.0752±0.0140	0.0502±0.0130
TYR	0.0182±0.0042	0.0232±0.0607	0.0178±0.0013	0.0302±0.0121
NITROGEN	0.2148±0.0385	0.1436±0.0368	0.3065±0.0545	0.1933±0.0666

<sup>1</sup> Means, expressed as grams per 100 gram dry matter intake, and standard deviation.

<sup>2</sup> No ileal and faecal collections were available from pig number 6.

<sup>3</sup> Ileal and faecal collections from M-7, fed following the cereal test diet sequence were spoiled.

<sup>4</sup> Numbers in parentheses indicate the number of observations.

<sup>5</sup> Determined by acid hydrolysis.



PRO and SER. The net disappearance was very large for PRO and GLY. Leucine, LYS, MET, PHE, ASP and TYR showed a net increase from the end of the ileum to the anus.

The metabolic ileal levels of PRO and GLY varied extensively from pig to pig. In general, the variation in levels for the other amino acids was much less (Appendix, Table 9). Most variation between the metabolic faecal amino acid levels of the pigs could be attributed to pig number 1. The metabolic faecal amino acid levels of this pig were about twice as high as those of the other pigs (Appendix, Table 10).

#### Study 2b.

The apparent faecal amino acid availabilities determined from 6 normal pigs were very close to those determined from the cannulated pigs. The largest differences were found for barley and ranged from 1.2 to 4.8 percentage units for PRO and THR respectively (Table 22 and 25). Faecal availabilities from corn and wheat were nearly identical for cannulated and normal pigs. The LYS availabilities of normal pigs were 81.8, 79.9 and 73.5% for corn, wheat and barley respectively. In the same order, they were 83, 80.7 and 77.5% for the cannulated pigs (Table 22 and 25).

The apparent amino acid availabilities from pig number 6 were lower than those of the other 5 pigs. These

TABLE 25.

Apparent faecal amino acid availabilities of the cereal grains, in addition to nitrogen and dry matter digestibilities for study 2b.

CEREAL GRAIN	CORN	WHEAT	BARLEY	$S\bar{x}^1$
AMINO ACIDS (%) <sup>2</sup>				
<u>Essential</u>				
ARG	91.2 <sup>A</sup>	92.1 <sup>A</sup>	87.4 <sup>B</sup>	0.620
HIS	92.4 <sup>B</sup>	94.1 <sup>A</sup>	87.8 <sup>C</sup>	0.446
ILE	87.8 <sup>A</sup>	88.9 <sup>A</sup>	79.5 <sup>B</sup>	0.852
LEU	93.6 <sup>A</sup>	91.2 <sup>B</sup>	84.1 <sup>C</sup>	0.605
LYS	81.8 <sup>A</sup>	79.9 <sup>A</sup>	73.5 <sup>E</sup>	1.364
MET <sup>3</sup>	87.4 <sup>A</sup>	87.3 <sup>A</sup>	76.2 <sup>B</sup>	1.198
PHE	91.2 <sup>B</sup>	92.6 <sup>A</sup>	86.2 <sup>C</sup>	0.304
THR	85.9 <sup>A</sup>	86.1 <sup>A</sup>	78.3 <sup>B</sup>	0.849
VAL	87.7 <sup>A</sup>	88.6 <sup>A</sup>	81.1 <sup>E</sup>	0.740
<u>Non-Essential</u>				
ALA	90.2 <sup>A</sup>	83.3 <sup>E</sup>	74.8 <sup>C</sup>	0.838
ASP	86.0 <sup>A</sup>	82.6 <sup>A</sup>	74.6 <sup>A</sup>	1.660
GLU	93.8 <sup>B</sup>	97.0 <sup>A</sup>	91.4 <sup>C</sup>	0.416
GLY	86.0 <sup>B</sup>	88.9 <sup>A</sup>	79.8 <sup>C</sup>	0.878
PRO	93.4 <sup>B</sup>	97.0 <sup>A</sup>	91.1 <sup>C</sup>	0.339
SER	90.6 <sup>A</sup>	92.1 <sup>A</sup>	84.3 <sup>B</sup>	0.518
TYR	90.0 <sup>A</sup>	89.5 <sup>A</sup>	82.5 <sup>B</sup>	0.633
NITROGEN (%)	88.5 <sup>A</sup>	91.1 <sup>B</sup>	82.0 <sup>C</sup>	0.552
DRY MATTER (%)	89.0 <sup>A</sup>	88.5 <sup>A</sup>	81.8 <sup>B</sup>	0.180

<sup>1</sup> Standard error of the mean of 6 observations per treatment.

<sup>2</sup> Means in the same row with the same superscript do not differ significantly ( $P < .05$ ).

<sup>3</sup> Determined by acid hydrolysis.

differences were significant for PHE and ALA (Appendix, Table 11). The individual faecal amino acid availabilities from the pigs are shown in the appendix (Table 12, 13, 14).

The metabolic faecal amino acid excretion increased as the level of alphafloc was raised in the maintenance diets (Table 26).

With the exception of ARG and PHE there were no significant differences due to weight (Appendix, Table 15). The pigs differed approximately 30 kg. in weight between the first and second time they were fed the same maintenance diet. The metabolic faecal ARG and PHE excretion were significantly higher for the lighter than heavier pigs fed the M-14 diet (Table 26). The individual pig data are shown in the appendix (Table 16 and 17).

Metabolic faecal amino acid excretions were approximately 30% higher for the normal than for the cannulated pigs (Table 24 and 26). As stated previously, there was considerable variation in the metabolic faecal amino acid excretion in study 2a. The variation that was observed could be largely attributed to one particular pig.

### Study 3.

This study was subdivided into 2 separate experiments. Study 3a deals with the infusion of isolated soy protein. Experiment 3b involved the infusion of lysine monohydrochloride into the caecum.

TABLE 26. Metabolic faecal amino acid and nitrogen levels<sup>1</sup> for study 2b.

PERIOD OF COLLECTION	BEFORE CEREAL DIETS		AFTER CEREAL DIETS		$S_{\bar{x}}^2$
LEVEL OF ALPHAFLOC (%)	7	14	7	14	
AMINO ACIDS <sup>3</sup>					
<u>Essential</u>					
ARG	.022 <sup>BC</sup>	.032 <sup>A</sup>	.018 <sup>C</sup>	.026 <sup>B</sup>	.00170
HIS	.008 <sup>B</sup>	.011 <sup>A</sup>	.007 <sup>B</sup>	.012 <sup>A</sup>	.00078
ILE	.027 <sup>E</sup>	.039 <sup>A</sup>	.029 <sup>B</sup>	.038 <sup>A</sup>	.00253
LEU	.041 <sup>E</sup>	.060 <sup>A</sup>	.036 <sup>B</sup>	.054 <sup>A</sup>	.00442
LYS	.033 <sup>B</sup>	.047 <sup>A</sup>	.030 <sup>B</sup>	.045 <sup>A</sup>	.00302
MET <sup>4</sup>	.012 <sup>E</sup>	.017 <sup>A</sup>	.012 <sup>B</sup>	.020 <sup>A</sup>	.00149
PHE	.025 <sup>BC</sup>	.036 <sup>A</sup>	.020 <sup>C</sup>	.031 <sup>B</sup>	.00226
THR	.029 <sup>B</sup>	.043 <sup>A</sup>	.026 <sup>B</sup>	.038 <sup>A</sup>	.00321
VAL	.032 <sup>B</sup>	.048 <sup>A</sup>	.031 <sup>B</sup>	.045 <sup>A</sup>	.00392
<u>Non-Essential</u>					
ALA	.040 <sup>B</sup>	.058 <sup>A</sup>	.035 <sup>B</sup>	.053 <sup>A</sup>	.00405
ASP	.062 <sup>B</sup>	.088 <sup>A</sup>	.057 <sup>B</sup>	.088 <sup>A</sup>	.00704
GLU	.080 <sup>B</sup>	.114 <sup>A</sup>	.087 <sup>B</sup>	.112 <sup>A</sup>	.00768
GLY	.032 <sup>B</sup>	.046 <sup>A</sup>	.027 <sup>B</sup>	.040 <sup>A</sup>	.00298
PRO	.032 <sup>B</sup>	.043 <sup>A</sup>	.030 <sup>B</sup>	.040 <sup>A</sup>	.00254
SER	.032 <sup>B</sup>	.043 <sup>A</sup>	.036 <sup>B</sup>	.044 <sup>A</sup>	.00299
TYR	.019 <sup>B</sup>	.026 <sup>A</sup>	.015 <sup>B</sup>	.023 <sup>A</sup>	.00198
NITROGEN	.118 <sup>B</sup>	.173 <sup>A</sup>	.121 <sup>B</sup>	.175 <sup>A</sup>	.00861

<sup>1</sup> Means from 6 observations, expressed per 100 gram of dry matter intake.

<sup>2</sup> Standard error of the mean.

<sup>3</sup> Means in the same row with the same superscript do not differ significantly ( $P < .05$ ).

<sup>4</sup> Determined by acid hydrolysis.

Study 3a.

Dietary amino acid and nitrogen analyses are shown in Table 27. The level of LYS from E+2ISP was 0.15% higher than that of E+ISP. The individual data are given in Table 28 and are summarized in Table 29.

The amount of nitrogen retained per day was significantly higher for E+2ISP than for the other 2 diets, for which the daily nitrogen retentions were almost similar (Table 29; Appendix, Table 18). The retention of nitrogen, expressed per 100 grams of dry matter intake, was approximately 1 g. for the E+2ISP diet and 0.9 g. for the E+ISP and E+ISP+C-ISP diets. The latter was not significant at the 5% level of significance, but was significant at the 10% level of significance. Percent nitrogen retention and biological value were the highest for E+ISP and were significantly higher than those for E+ISP+C-ISP, but not than the values for E+2ISP.

If all caecally infused ISP had been digested, absorbed and utilized to the same extent in the large intestine as in the small intestine, then 18.0 g. of nitrogen would have been retained per day (Table 29). If not utilized at all, it may be expected that 15.8 g. of nitrogen would have been retained from the caecally fed ISP. The amount retained from E+ISP+C-ISP was found to be 15.91 g., which is only 0.11 g. <sup>1</sup> more than

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$$^1 15.91 - 15.80 = 0.11.$$

TABLE 27. Amino acid and nitrogen composition<sup>1</sup> of test diets in study 3.

DIETS	B+2ISP	B+ISP	B+ISP+LYS
AMINO ACIDS (%)			
<u>Essential</u>			
ARG	0.88	0.68	0.66
HIS	0.36	0.27	0.27
ILE	0.73	0.55	0.55
LEU	1.32	1.03	1.02
LYS	0.71	0.56	0.71
MET <sup>2</sup>	0.24	0.21	0.21
PHE	0.96	0.76	0.75
THR	0.57	0.45	0.45
VAL	0.87	0.68	0.68
<u>Non-Essential</u>			
ALA	0.69	0.56	0.57
ASP	1.39	0.95	0.97
CYS <sup>2</sup>	0.21	0.22	0.22
GLU	4.23	3.39	3.44
GLY	0.68	0.55	0.55
PRO	1.54	1.39	1.34
SER	0.71	0.55	0.57
TYR	0.44	0.34	0.33
NITROGEN (%)	2.486	2.096	2.091

<sup>1</sup> Expressed as percent of dry matter.

<sup>2</sup> Determined by the oxidation method.

TABLE 28 Individual daily nitrogen intakes and faecal and urinary losses and some calculated parameters in study 3a.

PERIOD	I	II	III
<u>Pig no. 1</u>	(B+2ISP) <sup>1</sup>	(B+ISP)	(B+ISP+C-ISP)
Nitrogen intake (g/day)	41.95	37.79	50.38
Faecal nitrogen loss (g/day)	7.89	7.09	9.41
Urinary nitrogen loss (g/day)	16.80	14.41	24.61
Nitrogen retained (g/day)	17.26	16.29	16.36
Nitrogen retained <sup>2</sup>	1.023	0.904	0.867
Nitrogen retention (%) <sup>3</sup>	41.14	43.11	32.47
Biological value (%) <sup>4</sup>	50.68	53.06	39.93
<u>Pig no. 2</u>	(E+ISP)	(E+ISP+C-ISP)	(E+2ISP)
Nitrogen intake	34.88	48.16	47.54
Faecal nitrogen loss	7.39	10.47	8.40
Urinary nitrogen loss	13.18	22.27	20.76
Nitrogen retained	14.31	15.42	18.38
Nitrogen retained <sup>2</sup>	0.860	0.855	0.961
Nitrogen retention <sup>3</sup>	41.03	32.02	38.66
Biological value <sup>4</sup>	52.06	40.91	46.96
<u>Pig no.3</u>	(B+ISP+C-ISP)	(E+2ISP)	(E+ISP)
Nitrogen intake	44.45	45.45	39.53
Faecal nitrogen loss	9.98	8.59	7.25
Urinary nitrogen loss	18.52	18.49	15.48
Nitrogen retained	15.95	18.37	16.80
Nitrogen retained <sup>2</sup>	0.958	1.005	0.891
Nitrogen retention <sup>3</sup>	35.88	40.42	42.50
Biological value <sup>4</sup>	46.27	49.84	52.04

1 Abbreviations in parentheses indicate the diet fed.

2 Grams of nitrogen retained per 100 grams of dry matter intake.

3 Percent nitrogen retained.

4 Percent nitrogen retained from nitrogen absorbed; Metabolic faecal and endogenous urinary losses were not taken into account.

TABLE 29

The average<sup>1</sup> amounts of nitrogen retained, nitrogen retentions and biological values for diets in study 3a.

DIET	B+2ISP	B+ISP	B+ISP+C-ISP	$S_{\bar{x}}^2$
<u>ITEMS</u>				
Nitrogen retained (g/day)	18.00 <sup>A</sup>	15.80 <sup>B</sup>	15.91 <sup>B</sup>	.2718
Nitrogen retained <sup>3</sup>	0.996 <sup>A</sup>	0.885 <sup>A</sup>	0.893 <sup>A</sup>	.01526
Nitrogen retention (%) <sup>4</sup>	40.07 <sup>A</sup>	42.21 <sup>A</sup>	33.46 <sup>B</sup>	.6221
Biological value (%) <sup>5</sup>	49.16 <sup>A</sup>	52.39 <sup>A</sup>	42.37 <sup>E</sup>	.7479

<sup>1</sup> Means with the same superscript in the same row do not differ significantly ( $P < .05$ ).

<sup>2</sup> Standard error of the mean.

<sup>3, 4, 5</sup> As shown in Table 28.



that for B+ISP. Therefore, the relative retention of caecally infused ISP was only 5%<sup>1</sup> of that of orally supplied ISP.

The average nitrogen intake, faecal nitrogen loss, urinary nitrogen loss and retained nitrogen were 37.40, 7.24, 14.36 and 15.80 g. per day respectively for the pigs fed B+ISP. In the same order, these values were 47.66, 9.95, 21.80 and 15.91 g. per day respectively for pigs given B+ISP+C-ISP (Table 28 and 29). Of the nitrogen caecally infused which was 10.26<sup>2</sup>g., 2.71<sup>3</sup>g. was lost in the faeces, 7.44<sup>4</sup>g. was lost in the urine and 0.11<sup>5</sup>g. was retained. Expressed on a percentage basis, 26.4% of the nitrogen from caecally infused ISP was excreted in the faeces, 72.5% was excreted in the urine and 1.9% was retained by the pigs.

### Study 3b.

The individual data are given in Table 30 and are summarized in Table 31. Data from pig number 6 on treatment B+ISP+C-LYS were not available. Statistical analyses could be performed with the inclusion of the calculated missing values (Table 31). However, this would be of no use in

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$$^1 0.11 : \{(18.00 - 15.60)\} \times 100.$$

$$^2 (9.57 + 10.37 + 10.85) : 3 = 10.26. \quad ^3 9.95 - 7.24 = 2.71.$$

$$^4 21.80 - 14.36 = 7.44. \quad ^5 15.91 - 15.80 = 0.11.$$

TABLE 30 Individual daily nitrogen intakes and faecal and urinary losses and some calculated parameters in study 3b.

PERIOD	I	II	III
<u>Pig no. 4</u>	(B+ISP+C-LYS) <sup>1</sup>	(B+ISP+LYS)	(B+ISP)
Nitrogen intake (g/day)	34.88	37.96	39.53
Faecal nitrogen loss (g/day)	7.16	6.46	6.84
Urinary nitrogen loss (g/day)	13.04	13.50	17.11
Nitrogen retained (g/day)	14.68	18.00	15.58
Nitrogen retained <sup>2</sup>	0.882	0.991	0.826
Nitrogen retention (%) <sup>3</sup>	42.09	47.42	39.41
Biological value (%) <sup>4</sup>	53.17	57.14	47.21
<u>Pig no. 5</u>	(E+ISP)	(B+ISP+C-LYS)	(E+ISP+LYS)
Nitrogen intake	34.88	37.79	39.71
Faecal nitrogen loss	7.68	7.39	8.34
Urinary nitrogen loss	12.39	14.02	12.76
Nitrogen retained	14.81	16.38	18.61
Nitrogen retained <sup>2</sup>	0.900	0.909	0.980
Nitrogen retention <sup>3</sup>	42.46	43.34	46.86
Biological value <sup>4</sup>	54.45	53.88	59.32
<u>Pig no. 6</u>	(B+ISP+LYS)	(E+ISP)	(B+ISP+C-LYS) <sup>5</sup>
Nitrogen intake	35.04	37.79	
Faecal nitrogen loss	7.62	7.66	
Urinary nitrogen loss	10.80	14.42	
Nitrogen retained	16.62	15.71	15.98
Nitrogen retained <sup>2</sup>	0.992	0.871	0.839
Nitrogen retention <sup>3</sup>	47.43	41.57	40.47
Biological value <sup>4</sup>	60.61	52.14	51.58

1, 2, 3, 4 As shown in Table 28.

5 Missing data were calculated by least square method.

TABLE 31. Average<sup>1</sup> amounts of nitrogen retained, nitrogen retentions and biological values for diets in study 3b.

DIETS	B+ISP+C-LYS	B+ISP+LYS	E+ISP
<u>ITEMS</u>			
Nitrogen retained (g/day)	15.68 (15.53) <sup>2</sup>	17.74	15.36
Nitrogen retained <sup>3</sup>	0.877 (0.896)	0.988	0.866
Nitrogen retention <sup>4</sup> (%)	41.97 (42.72)	47.24	41.15
Biological value <sup>5</sup> (%)	52.88 (53.53)	59.02	51.27

<sup>1</sup> Statistical analyses were not performed.

<sup>2</sup> Values in parentheses do not include the calculated missing values.

<sup>3, 4, 5</sup> As shown in Table 28.

this study since the error degrees of freedom would decrease from 2 to 1 resulting in the increase of F critical from 19.0 to 200.00 (at the 5% level of significance).

All parameters determined were the highest for B+ISP+LYS and the lowest for B+ISP. The values obtained for B+ISP+C-LYS were slightly higher than those of B+ISP (Table 31).

Including the calculated missing value the amount of nitrogen retained per day increased by  $0.32^1$ g. upon LYS infusion into the caecum. Orally given LYS increased the amount of nitrogen retained per day by  $2.38^2$ g. (Table 31). Therefore, the relative improvement in nitrogen retained per day (g.) of caecally infused LYS was only  $13.4^3\%$  of that orally supplied LYS.

Study 4.

Study 4a.

The nitrogen and fiber levels decreased from B+S+M to whole wheat and to flour (Table 32). B+S+M contained much higher levels of ARG, HIS, LYS, ALA, ASP and GLY than flour while those of THR, VAL, SER and TYR were slightly

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$$^1 15.68 - 15.36 = 0.32. \quad ^2 17.74 - 15.36 = 2.32.$$

$$^3 (0.23 : 2.32) \times 100 = 13.4\%.$$

TABLE 32 Proximate, acid and neutral-detergent fiber and chromic oxide analyses<sup>1</sup> for diets of study 4.

DIETS	WHOLE WHEAT	FLOUR	B+S+M	PROTEIN-FREE		
LEVEL OF ALPHAFLOC (%)				5	10	15
<u>ITEMS (%)</u>						
Nitrogen	2.46	2.35	2.75	0.09	0.10	0.11
Ether extract	1.66	1.03	4.48	1.06	1.16	1.00
Fiber:						
Crude	2.35	0.25	8.49	3.86	6.80	10.83
Acid-detergent	5.38	3.21	12.94	6.42	9.90	14.64
Neutral-detergent	12.17	3.43	40.98			
Ash	4.82	4.01	8.44	3.26	3.08	3.15
Chromic oxide	0.52	0.56	0.55	0.59	0.55	0.55

<sup>1</sup> Expressed on a dry weight basis.

higher. The levels of PHE, GLU and PRO were much lower in E+S+M than in flour. As expected, the amino acid levels of whole wheat were between those of E+S+M and flour (Table 33).

Statistical analyses could not be performed, at least not in the form of a single cross-over design on the apparent amino acid availabilities from whole wheat, flour and E+S+M (Table 34). Pigs numbered 1, 2, 4 and 7 refused to consume the E+S+M diet (Fig. 3). Data obtained from replacement pig number 6 (day 23-29) are included in the availabilities of amino acids from E+S+M.

Generally, apparent ileal and faecal amino acid availabilities decreased from flour to whole wheat to E+S+M. Total nitrogen and dry matter digestibilities followed the same pattern (Table 34).

The differences in percentage units between ileal and faecal availabilities were the smallest for flour and the largest for diet E+S+M. The differences for all diets were always the most pronounced for ARG, HIS, THR, GLY, PRO, and SER (Table 34).

Lysine and THR were about equally the least available essential amino acids of whole wheat and flour when determined at the end of the ileum. Lysine was the least available from these 2 diets when determined by the

TABLE 33 Amino acid composition of diets from study 4.

DIETS	WHOLE WHEAT		FLOUR		B+S+M	
	% <sup>1</sup>	g/16gN <sup>2</sup>	%	g/16gN	%	g/16gN
AMINO ACIDS						
<u>Essential</u>						
ARG	0.66	4.27	0.47	3.24	1.05	6.11
HIS	0.32	2.07	0.29	2.01	0.42	2.46
ILE	0.54	3.52	0.55	3.74	0.55	3.18
LEU	1.01	6.57	1.00	6.86	1.03	5.99
LYS	0.37	2.39	0.25	1.74	0.59	3.43
MET <sup>3</sup>	0.21	1.37	0.17	1.16	0.25	1.45
PHE	0.73	4.72	0.76	5.23	0.66	3.87
THR	0.40	2.59	0.36	2.44	0.45	2.64
VAL	0.66	4.31	0.61	4.20	0.77	4.49
<u>Non-Essential</u>						
ALA	0.50	3.25	0.40	2.76	0.79	4.58
ASP	0.76	4.91	0.57	3.39	1.13	6.56
GLU	4.69	30.50	5.19	35.57	3.23	18.82
GLY	0.60	3.92	0.48	3.29	0.64	4.89
PRO	1.49	9.69	1.67	11.45	0.97	5.67
SER	0.60	3.92	0.58	3.99	0.66	3.84
TYR	0.34	2.19	0.30	2.05	0.34	1.95

1 Expressed on a dry weight basis.

2 Grams per 16 grams of nitrogen.

3 Determined by acid hydrolysis.

TABLE 34. Apparent ileal and faecal amino acid availabilities<sup>1</sup>  
of whole wheat, flour and B+S+M and B+S+M-D for study 4.

DIET	WHOLE WHEAT (6) <sup>2</sup>		FLOUR (6)		B+S+M (3)		B+S+M-D (6)	
LOCATION	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES
AMINO ACIDS (%)								
<u>Essential</u>								
ARG	87.1±1.7	94.6±0.5	90.7±2.5	95.6±1.3	84.8±1.5	90.0±2.1	71.5±6.7	90.2±1.0
HIS	88.4±1.4	93.9±0.9	93.9±0.3	96.6±1.1	78.5±2.6	88.8±2.5	73.6±3.1	88.0±1.5
ILE	89.1±2.0	91.6±1.0	93.9±0.6	94.7±1.5	72.9±2.3	74.6±2.0	69.6±1.6	75.5±4.0
LEU	89.9±1.5	93.2±0.5	94.6±0.5	95.5±1.4	74.4±3.0	78.7±2.3	71.7±1.7	80.7±2.9
LYS	79.5±3.3	86.1±1.2	84.2±1.6	86.0±4.5	66.4±4.8	75.5±4.2	57.5±3.9	77.6±3.1
MET	92.4±2.2	93.4±1.8	93.7±1.3	93.4±2.7	77.8±3.8	81.8±1.7	76.5±6.0	73.2±7.6
PHE	91.5±1.1	94.3±0.7	95.5±1.1	96.3±1.1	76.0±2.5	79.5±1.8	73.9±1.3	81.5±2.8
THR	78.4±2.6	89.1±0.9	85.4±1.5	92.3±2.3	53.9±6.5	71.3±3.5	47.8±3.9	74.7±3.2
VAL	86.7±1.7	91.7±1.1	92.7±0.6	94.3±1.6	71.3±2.7	76.0±4.7	66.5±2.6	78.2±3.2
<u>Non-Essential</u>								
ALA	79.6±2.6	88.1±1.5	86.1±0.9	90.8±2.8	70.2±0.7	75.5±3.3	60.7±5.9	76.2±4.0
ASP	80.8±2.5	88.0±1.2	85.5±1.0	89.2±3.0	69.8±1.2	75.6±2.8	64.2±3.3	78.2±2.5
GLU	95.6±0.6	97.9±0.1	97.9±0.1	98.6±0.4	85.8±1.2	89.9±0.9	83.8±1.6	90.9±1.4
GLY	72.6±6.2	90.6±0.6	78.5±7.8	93.6±1.8	57.3±8.9	78.3±1.5	31.9±21.0	79.3±2.5
PRO	79.1±14.7	96.8±1.1	83.0±14.7	98.5±0.4	70.3±11.0	89.5±2.4	-47.5±66.5	78.4±2.8
SER	86.3±1.5	94.3±0.9	91.7±0.4	95.8±1.3	72.2±2.1	83.1±1.8	67.7±2.8	84.2±2.2
TYR	89.2±1.7	92.9±1.2	93.1±0.7	94.4±1.4	71.1±3.0	78.8±2.1	68.5±2.6	78.6±3.8
NITROGEN (%)	85.2±1.7	93.3±0.6	90.5±1.8	95.6±0.9	69.8±1.4	80.9±0.7	59.2±6.3	83.0±1.4
DRY MATTER (%)	78.2±1.6	89.4±0.5	90.2±0.4	95.0±0.8	43.3±2.7	65.0±1.5	69.2±1.4	83.9±0.3

<sup>1</sup> Means and standard deviation. <sup>2</sup> Numbers in parentheses indicate the number of observations per dietary treatment. <sup>3</sup> Determined by acid hydrolysis



faecal analysis method.

The ileal availabilities of essential amino acids from diet B+S+M were very low. They were 66.4 and 53.9% for LYS and THR respectively. Those of the other essential amino acids varied between 71.3 and 84.8%. Isoleucine, LYS, THR and VAL were about equally the least available from diet B+S+M when determined by the faecal analysis method and their availabilities ranged from 71.3 to 76.0% (Table 34).

The apparent ileal availabilities of ARG, GLY and PRO were markedly lower for the cornstarch diluted B+S+M diet (E+S+M-D) than for the B+S+M diet. The ileal availability of PRO from B+S+M-D was even found to be negative i.e. more PRO leaves the end of the ileum than is ingested. The ileal availabilities of the other amino acids were also lower for B+S+M-D than for B+S+M but to a lesser extent. The apparent faecal amino acid availabilities of B+S+M and B+S+M-D, with the exception of PRO and MET, were of the same order (Table 34).

The individual apparent ileal and faecal amino acid availabilities, in addition to nitrogen and dry matter digestibilities and average daily dry matter intake of the pigs fed whole wheat, flour, diet B+S+M and diet B+S+M-D are shown in the appendix (Table 19, 20, 21 and 22).

The levels of the ileal and faecal amino acids,

total nitrogen and dry matter increased as the level of alphafloc of the protein-free diets was increased. The increment in amino acid levels was much larger when the level of alphafloc was raised from 5 to 10% than from 10 to 15% (Table 35).

For all protein-free diets, there was a large disappearance of ARG, THR, GLY, PRO, SER, total nitrogen and dry matter between the end of the ileum and the rectum. The net disappearance was largest for PRO, followed by GLY. Proline and GLY in this order were also the first and second most prominent amino acids in ileal digesta. The levels of these amino acids in ileal digesta varied markedly from pig to pig as shown by the relatively large standard deviations for these amino acids compared to those of the other amino acids (Table 35). Arginine was the most prominent essential amino acid in ileal digesta.

Small but consistent increases were found for ILE, LEU, LYS, MET, PHE, VAL and ASP from the end of the ileum to the rectum (Table 35).

Only 4 of the actual 6 test pigs consumed the PF-10 diet (Fig. 3). The average metabolic ileal and faecal amino acid levels obtained from replacement pig number 9 were included in data for PF-10 in Table 35. Pig number 9 gained approximately 25 kg. from the first time (day 12-18) to the third time (day 56-62) it was fed the PF-10 diet (Fig. 3).

TABLE 35. Metabolic ileal and faecal amino acid levels  
in study 4a.

LEVEL OF ALPHAFLOC (%)	5		10		15	
	ILEUM (6)	FAECES (6)	ILEUM (5)	FAECES (5)	ILEUM (3)	FAECES (3)
AMINO ACIDS <sup>2</sup>						
<u>Essential</u>						
ARG	0.049±0.023	0.022±0.002	0.058±0.014	0.029±0.003	0.061±0.012	0.033±0.004
HIS	0.014±0.003	0.010±0.001	0.017±0.002	0.014±0.002	0.022±0.007	0.015±0.001
ILE	0.021±0.004	0.027±0.004	0.029±0.008	0.039±0.006	0.031±0.005	0.045±0.003
LEU	0.039±0.007	0.042±0.005	0.051±0.011	0.059±0.008	0.057±0.010	0.067±0.004
LYS	0.027±0.003	0.036±0.003	0.036±0.008	0.048±0.007	0.039±0.011	0.054±0.005
MET <sup>3</sup>	0.006±0.001	0.012±0.002	0.008±0.003	0.017±0.003	-----	-----
PHE	0.023±0.004	0.026±0.003	0.030±0.007	0.037±0.005	0.033±0.008	0.040±0.003
THR	0.039±0.005	0.030±0.004	0.051±0.008	0.041±0.007	0.059±0.014	0.045±0.005
VAL	0.031±0.004	0.033±0.004	0.043±0.009	0.049±0.007	0.044±0.010	0.053±0.005
<u>Non-Essential</u>						
ALA	0.041±0.009	0.036±0.005	0.056±0.010	0.053±0.008	0.058±0.008	0.059±0.007
ASP	0.056±0.009	0.061±0.006	0.078±0.013	0.084±0.013	0.081±0.012	0.096±0.010
CYS <sup>3</sup>	0.013±0.002	0.010±0.001	0.016±0.003	0.013±0.002	-----	-----
GLU	0.071±0.012	0.068±0.008	0.094±0.018	0.096±0.015	0.100±0.019	0.108±0.010
GLY	0.139±0.065	0.031±0.004	0.171±0.043	0.043±0.006	0.169±0.015	0.047±0.005
PRO	0.474±0.325	0.026±0.005	0.577±0.272	0.046±0.006	0.585±0.161	0.038±0.004
SER	0.038±0.006	0.025±0.002	0.049±0.007	0.035±0.004	0.053±0.010	0.037±0.002
TYR	0.013±0.002	0.013±0.004	0.015±0.004	0.016±0.002	0.016±0.005	0.018±0.004
NITROGEN	0.205±0.078	0.101±0.009	0.256±0.052	0.139±0.019	0.271±0.034	0.161±0.010
DRY MATTER	11.67 ±0.95	6.48 ±0.33	17.69 ±0.99	10.53 ±0.44	26.32 ±0.85	14.78±0.99

<sup>1</sup> Numbers in parentheses indicate the number of observations.

<sup>2</sup> Means and standard deviation, expressed as grams per 100 grams of dry matter intake.

<sup>3</sup> Determined by the oxidized method.

Generally, there were no substantial differences due to body weight on the metabolic ileal and faecal amino acid levels. The levels of ileal ARG, PRO and GLY varied slightly more than the levels of the other amino acids (Appendix, Table 24).

Pig numbers 3 and 5 were the only ones of the 6 test pigs that consumed the PF-15 diet (Fig. 3). The average metabolic ileal and faecal amino acid levels obtained from replacement pig number 6 are included in data for diet PF-15 in Table 35. Pig number 6 gained about 15 kg. from the first time (day 12-18) to the second time (day 34-40) it was fed the PF-15 diet (Fig. 3). Although there were some differences in ileal and faecal amino acid levels between the 2 collections, they were of a relatively small magnitude (Appendix, Table 25).

The individual metabolic ileal and faecal amino acid levels, in addition to nitrogen and dry matter (as grams per 100 grams of dry matter intake) and daily dry matter intake are shown in the appendix (Table 23, 24 and 25).

#### Study 4b.

Sixty percent of the particles of the cracked wheat did not pass through a 2.00 mm sieve, 80% did not pass through a 1.00 mm screen (Table 36). The particle size distribution was determined after the cracked wheat diet was pelleted. A 250 gram sample was taken, steamed through thoroughly until disintegration occurred, dried and sieved.

TABLE 36. Particle size distribution of finely ground and cracked wheat and of the ileal digesta derived thereof.

PRE-PROCESSING	FINELY GROUND		CRACKED	
	DIET <sup>1</sup>	DIGESTA <sup>2</sup>	DIET	DIGESTA <sup>2</sup>
PARTICLE DISTRIBUTION (%) <sup>3</sup>				
>2.00		1.2	4.5	4.6
1.00 to 2.00 mm		3.1	56.2	22.8
0.50 to 1.00 mm		18.0	19.8	17.7
0.25 to 0.50 mm		10.9	10.8	7.7
0.125 to 0.25 mm		45.0	4.8	28.5
<0.125		22.3	3.9	19.0

<sup>1</sup> Ground through a 1.5 mm mesh screen.

<sup>2</sup> Average from 4 ileal collections.

<sup>3</sup> Determined after the diets were pelleted.

There were no significant differences between the apparent faecal amino acid availabilities, total nitrogen and dry matter digestibilities from finely ground and cracked wheat (Table 37; Appendix, Table 26). With the exception of MET, GLU, SER and PRO, the apparent ileal availabilities of amino acids were significantly higher for ground than for cracked wheat. The ileal availabilities of LYS of ground and cracked wheat were 81.5 and 72.6% respectively.

With the exception of HIS, THR, ASP, GLU, GLY, and PRO, ileal amino acid availabilities were not significantly different from faecal availabilities for ground wheat. The ileal amino acid availabilities from cracked wheat were all significantly lower than their faecal availabilities (Table 37).

The individual apparent ileal and faecal amino acid availabilities, in addition to nitrogen and dry matter digestibilities and average daily dry matter intake from the pigs fed cracked or finely ground wheat are shown in the appendix (Table 27 and 28).

TABLE 37.

Apparent ileal and faecal amino acid availabilities of ground and cracked wheat for study 4b.

DIET	GROUND WHEAT		CRACKED WHEAT		$S_{\bar{x}}^1$
LOCATION	ILEUM	FAECES	ILEUM	FAECES	
AMINO ACIDS (%) <sup>2</sup>					
<u>Essential</u>					
ARG	92.0 <sup>A</sup>	94.4 <sup>A</sup>	86.5 <sup>C</sup>	93.9 <sup>A</sup>	0.615
HIS	91.1 <sup>B</sup>	94.4 <sup>A</sup>	85.7 <sup>C</sup>	93.9 <sup>A</sup>	0.557
ILE	90.2 <sup>A</sup>	91.2 <sup>A</sup>	85.6 <sup>B</sup>	89.9 <sup>A</sup>	0.775
LEU	91.5 <sup>A</sup>	92.9 <sup>A</sup>	87.6 <sup>B</sup>	92.1 <sup>A</sup>	0.551
LYS	81.5 <sup>A</sup>	85.3 <sup>A</sup>	72.6 <sup>B</sup>	83.8 <sup>A</sup>	1.842
MET <sup>3</sup>	88.0 <sup>AB</sup>	88.9 <sup>A</sup>	83.6 <sup>B</sup>	89.6 <sup>A</sup>	1.248
PHE	92.9 <sup>A</sup>	93.5 <sup>A</sup>	89.4 <sup>B</sup>	92.9 <sup>A</sup>	0.425
THR	83.7 <sup>B</sup>	88.7 <sup>A</sup>	78.0 <sup>C</sup>	87.3 <sup>A</sup>	1.054
VAL	88.6 <sup>A</sup>	91.3 <sup>A</sup>	83.1 <sup>B</sup>	90.5 <sup>A</sup>	0.787
<u>Non-Essential</u>					
ALA	82.1 <sup>A</sup>	87.1 <sup>A</sup>	74.5 <sup>B</sup>	86.5 <sup>A</sup>	1.515
ASP	82.8 <sup>B</sup>	87.1 <sup>A</sup>	77.3 <sup>C</sup>	86.2 <sup>A</sup>	1.045
GLU	95.3 <sup>B</sup>	97.8 <sup>A</sup>	94.1 <sup>B</sup>	97.6 <sup>A</sup>	0.531
GLY	81.7 <sup>B</sup>	90.1 <sup>A</sup>	74.4 <sup>C</sup>	89.8 <sup>A</sup>	0.979
PRO	84.5 <sup>B</sup>	95.9 <sup>A</sup>	83.8 <sup>B</sup>	96.3 <sup>A</sup>	1.644
SER	88.2 <sup>AB</sup>	93.3 <sup>A</sup>	84.7 <sup>B</sup>	90.3 <sup>A</sup>	1.456
TYR	89.8 <sup>A</sup>	90.7 <sup>A</sup>	85.3 <sup>B</sup>	90.6 <sup>A</sup>	0.809
NITROGEN (%)	88.4 <sup>B</sup>	93.4 <sup>A</sup>	84.7 <sup>C</sup>	92.3 <sup>A</sup>	0.422
DRY MATTER (%)	80.4 <sup>B</sup>	88.8 <sup>A</sup>	76.8 <sup>C</sup>	88.8 <sup>A</sup>	0.400

<sup>1</sup> Standard error of the mean.

<sup>2</sup> Means in the same row with the same superscript do not differ significantly ( $P < 0.05$ ).

<sup>3</sup> Determined by acid hydrolysis.

## DISCUSSION<sup>1</sup>

### STUDY 2.

#### Comparison of results obtained from study 2a and 2b.

The experiment on the cannulated pigs (study 2a) was carried out during the spring and summer of 1974. The experiment on the normal pigs (study 2b) was carried out during the summer and fall of 1974.

Cannulation did not seem to affect the apparent faecal amino acid availabilities of pigs fed barley, corn or wheat. In general, the apparent availabilities tended to be slightly higher for the cannulated than for the normal pigs (Table 38). The largest differences were found for the amino acid availabilities from barley.

In addition to the insignificant effect of cannulation on apparent faecal amino acid availabilities, the results obtained show the reproducibility of the faecal analysis method.

Faecal collections from the cannulated pigs fed the M-7 diet following the cereal test diet sequence were lost. The M-14 diet was only fed to the cannulated pigs following the cereal test diet sequence. Consequently, a comparison between the metabolic faecal amino acid excretion between the cannulated and normal pigs could only be made for M-7 fed prior to the cereal test diet sequence and for M-14 fed following the cereal test diet sequence.

<sup>1</sup>The studies are discussed in the following sequence: study 2, study 4, study 3 and study 1.



TABLE 38. Comparisons of apparent faecal amino acid availabilities, nitrogen and dry matter digestibilities between cannulated and normal pigs fed corn, wheat and barley.

CEREAL GRAIN	CORN			WHEAT			BARLEY		
STUDY	2a	2b	A <sup>1</sup>	2a	2b	B <sup>1</sup>	2a	2b	C <sup>1</sup>
AMINO ACIDS (%)									
<u>Essential</u>									
ARG	92.2	91.2	1.0	92.7	92.1	0.6	89.4	87.4	2.0
HIS	93.6	92.4	1.2	94.9	94.1	0.8	91.9	87.8	4.1
ILE	88.1	87.8	0.3	89.4	88.9	0.5	83.1	79.5	3.6
LEU	93.8	93.6	0.2	91.5	91.2	0.3	86.6	84.1	2.5
LYS	83.0	81.8	1.2	80.7	79.9	0.8	77.5	73.5	4.0
PHE	91.3	91.2	0.1	92.5	92.6	-0.1	87.9	86.2	1.7
THR	86.3	85.9	0.4	86.7	86.1	0.6	81.4	78.3	3.1
VAL	88.2	87.7	0.5	88.9	88.6	0.3	84.3	81.1	3.2
<u>Non-Essential</u>									
ALA	90.8	90.2	0.6	84.0	83.3	0.7	77.4	74.8	2.6
ASP	86.6	86.0	0.6	83.1	82.6	0.5	77.9	74.6	3.3
GLU	94.1	93.8	0.3	97.0	97.0	0.0	92.7	91.4	1.3
GLY	86.2	86.0	0.2	89.3	88.9	0.4	82.6	79.8	2.8
PRO	93.4	93.4	0.0	96.7	97.0	-0.3	92.3	91.1	1.2
SER	91.0	90.6	0.4	92.5	92.1	0.4	86.9	84.3	2.6
TYR	90.2	90.0	0.2	89.5	89.5	0.0	85.0	82.5	2.5
NITROGEN (%)	89.4	88.5	0.9	91.2	91.1	0.1	85.9	82.0	3.9
DRY MATTER (%)	89.4	89.0	0.4	89.0	88.5	0.5	83.6	81.8	1.8

<sup>1</sup> Columns A, B and C indicate the difference in percentage units between the faecal amino acid availabilities from cannulated and non-cannulated pigs for corn, wheat and barley respectively.

There seemed to be substantial differences between the metabolic faecal amino acid excretion from normal and cannulated pigs (Table 39). A closer look at the individual data obtained showed that the metabolic faecal amino acid excretions from pig number 1 fed either M-7 or M-14 were considerably higher than those obtained from the other pigs (Appendix, Table 10). This pig was always very prone to blockage which resulted in a very low dietary intake (Appendix, Table 9) of the M-7 and M-14 diets and in excessive leakage at times. Proper dietary equilibration with respect to the maintenance diets had probably not taken place at the time the faeces were collected. In other words, the faeces that were collected may have contained undigested residues of the pig grower ration that was fed prior to the feeding of the M-7 and M-14 diets. Closer agreement between the metabolic faecal amino acid excretion from normal and cannulated pigs were obtained when the data from pig number 1 were excluded (Table 39).

Disappearance of amino acids  
in the large intestine.

Apparent amino acid availabilities include the metabolic faecal amino acid losses by the animal which true availabilities do not. The metabolic faecal losses are a function of the particular diet fed and where possible

TABLE 39. Comparisons of the metabolic faecal amino acid and nitrogen excretion<sup>1</sup> between study 2a and 2b.

LEVEL OF ALPHAFLC (%)	7		14	
STUDY	2a	2b	2a	2b
AMINO ACIDS				
<u>Essential</u>				
ARG	.029 (.024) <sup>2</sup>	.022	.032 (.030) <sup>2</sup>	.026
HIS	.011 (.010)	.008	.015 (.014)	.012
ILE	.033 (.030)	.027	.044 (.039)	.038
LEU	.050 (.043)	.041	.068 (.057)	.054
LYS	.046 (.040)	.033	.057 (.049)	.045
MET <sup>3</sup>	.016 (.014)	.012	.020 (.017)	.020
PHE	.031 (.027)	.025	.041 (.034)	.031
THR	.034 (.030)	.029	.047 (.040)	.038
VAL	.038 (.034)	.032	.051 (.044)	.045
<u>Non-Essential</u>				
ALA	.046 (.041)	.040	.063 (.054)	.053
ASP	.072 (.064)	.062	.094 (.083)	.088
GLU	.095 (.086)	.080	.125 (.111)	.112
GLY	.038 (.033)	.032	.050 (.042)	.040
PRO	.039 (.033)	.032	.067 (.058)	.040
SER	.037 (.034)	.034	.050 (.045)	.044
TYR	.023 (.020)	.019	.030 (.025)	.023
NITROGEN	.144 (.129)	.118	.193 (.167)	.175

<sup>1</sup> Expressed as grams per 100 gram dry matter intake.

<sup>2</sup> Values in parentheses exclude data obtained from pig number 1.

<sup>3</sup> Determined by acid hydrolysis.

should be taken into account during the formulation of dietary amino acid levels in order to meet the requirements for optimum response.

The apparent availabilities of LYS especially and those of THR, MET and TRY of cereal grains are of practical importance. As was mentioned previously, the method used for TRY analysis did not yield accurate duplications.

Only small differences were found between the apparent ileal and faecal amino acid availabilities for LYS and MET from corn, wheat and barley (Table 40). The difference between ileal and faecal LYS availabilities were 1.0, 4.0 and 4.2% for corn, wheat and barley respectively. In the same order, these differences were 2.4, 2.3 and 0.5% for MET.

The faecal analysis method might overestimate the actual availability of THR. The difference between ileal and faecal THR availabilities were 7.4, 10.2 and 10.2% for corn, wheat and barley respectively. Overestimation of THR by the faecal analysis method would occur if THR were subjected to microbial fermentation by the flora in the large intestine.

The differences between apparent faecal and ileal amino acid availabilities of the other essential amino acids

TABLE 40. Apparent ileal and faecal amino acid availabilities, in addition to nitrogen, dry matter, starch and crude fiber digestibilities and the disappearance of these substances in the large intestine.

CEREAL GRAIN	CORN				WHEAT				BARLEY			
LOCATION	ILEUM	FAECES	L.I. <sup>1</sup>	△ <sup>2</sup>	ILEUM	FAECES	L.I. <sup>1</sup>	△ <sup>2</sup>	ILEUM	FAECES	L.I. <sup>1</sup>	△ <sup>2</sup>
AMINO ACIDS (%)												
<u>Essential</u>												
ARG	87.4	92.2	37.5	-4.8	85.8	92.7	48.8	-6.9	81.5	89.4	43.4	-7.9
HIS	88.3	93.6	46.7	-5.3	89.1	94.9	53.1	-5.8	80.4	91.9	55.6	-11.5
ILE	87.5	88.1	6.6	-0.6	85.3	89.4	26.2	-4.1	79.1	83.1	19.5	-4.0
LEU	92.5	93.8	17.2	-1.3	86.9	91.5	35.4	-4.6	81.5	86.6	27.2	-5.1
LYS	82.0	83.0	4.9	-1.0	75.7	80.7	20.5	-4.0	73.3	77.5	15.3	-4.2
MET <sup>3</sup>	91.9	89.5	-29.1	+2.4	86.6	88.9	17.4	-2.3	80.4	79.9	-2.4	+0.5
PHE	90.5	91.3	6.1	-0.8	88.8	92.5	33.3	-3.7	82.2	87.9	31.8	-5.7
THR	78.9	86.3	35.1	-7.4	76.5	86.7	44.0	-10.2	71.2	81.4	35.5	-10.2
VAL	84.9	88.2	21.7	-3.3	82.8	88.9	36.2	-6.1	78.0	84.3	29.2	-6.3
<u>Non-Essential</u>												
ALA	88.5	90.8	20.5	-2.3	74.0	84.0	38.6	-10.0	69.7	77.4	25.3	-7.7
ASP	83.9	86.6	16.5	-2.7	75.4	83.1	31.7	-7.7	71.2	77.9	23.0	-6.7
CYS <sup>3</sup>	82.1	90.2	45.2	-8.1	85.2	93.7	57.6	-8.5	77.6	88.7	49.6	-11.1
GLU	91.8	94.1	25.9	-2.3	92.7	97.0	59.0	-4.3	86.6	92.7	45.8	-6.1
GLY	71.2	86.2	57.4	-15.0	73.7	89.3	59.1	-15.6	71.2	82.6	50.0	-11.4
PRO	80.4	93.4	74.3	-13.0	86.8	96.7	74.9	-9.9	80.9	92.3	59.4	-11.4
SER	84.9	91.0	41.6	-6.1	84.1	92.5	53.5	-8.4	76.3	86.9	44.5	-10.6
TYR	89.0	90.2	9.3	-1.2	85.9	89.5	24.6	-3.6	79.7	85.0	25.7	-5.3
NITROGEN (%)	82.4	89.4	39.1	-7.0	82.9	91.2	48.3	-8.3	74.9	85.9	43.7	-11.0
DRY MATTER (%)	80.2	89.4	46.5	-9.2	73.3	89.0	58.8	-15.7	66.2	83.6	51.5	-17.4
STARCH (%) <sup>4</sup>	98.2	99.9	92.5	-1.7	93.1	99.8	97.4	-5.7	92.0	99.8	96.9	-7.8
CRUDE FIBER (%) <sup>5</sup>	4.2	51.1	49.0	-46.9	0.0	22.2	22.2	-22.2	5.0	30.9	27.3	-25.9

<sup>1</sup> Digestibilities in the large intestine.

<sup>2</sup> Differences in percentage units between faecal and ileal estimates.

<sup>3</sup> Determined by the oxidation method. <sup>4</sup> For the determination of starch digestibilities, digesta from the 2 pigs, fed the same diet during each particular test period, were pooled.

<sup>5</sup> For the determination of crude fiber digestibilities digesta from 6 pigs fed the same test diet, were pooled.

varied from 0.8% for PHE to 5.3% for HIS from corn, from 3.7% for PHE to 6.9% for ARG from wheat and from 4.0% for ILE to 11.5% for HIS from barley.

Generally, there was an increased net disappearance of amino acids in the large intestine from corn to wheat to barley. Total nitrogen, dry matter and starch but not crude fiber followed the same relationship (Table 40).

For all the cereal grains tested, certain amino acids always disappeared to the largest extent. These amino acids were ARG, HIS, THR, CYS, GLU, GLY, PRO and SER (Table 40). The net disappearance of most of these amino acids was also extensive in the large intestine of the pigs when these were fed the maintenance or protein-free diets (Table 24 and 35). However, these observations do not necessarily mean that the micro-organisms of the large intestine are mainly active on the metabolic amino acids present in digesta from pigs fed cereal grains. These observations could simply mean that the flora has a particular affinity for the degradation of particular amino acids. On the other hand, a type of flora may have established itself in the large intestine of the pig with a dependence on endogeneous substrates. Endogeneous substrates are a consistent and fairly constant source of potential nutrients for the flora throughout the life span of a pig.

Comparison of results with those  
obtained from other laboratories.

A limited amount of comparative data are available and are shown in Table 41. The ileal availability estimates of amino acids obtained by Easter (1973) from corn were much lower than those obtained in this study. Faecal availabilities were also lower but to a lesser extent. The collection procedure employed by Easter was different than that used in this study. In Easter's study, ileal digesta was collected for each minute of a 24 hour day over a 5 day period. Fifteen equally spaced collections of 96 minutes duration were obtained. The collection procedure used did not permit the determination of ileal amino acid availabilities on the basis of total amino acid intake and total amounts of amino acids passing through the end of the ileum. Easter's estimates are based only on the relative chromic oxide content of feed and digesta and give no indication of the total amount of chromic oxide recovered from ileal digesta. The experiments carried out in this study allowed for the determination of ileal amino acid availabilities by both total collection and chromic oxide levels. Determinations by both methods were found to give nearly similar ileal availability estimates for the cereal grains, indicating total recovery of chromic oxide from ileal digesta (Table 23).

Easter did not return any of the ileal digesta that was collected from the ileal cannula back into the animal via

TABLE 41. Comparison of apparent ileal and faecal amino acid availabilities from corn and barley as obtained from different laboratories.

CEREAL GRAIN	CORN				BARLEY	
LOCATION	ILEUM	FAECES	ILEUM	FAECES	ILEUM	ILEUM
REFERENCE	A <sup>1</sup>	A <sup>1</sup>	B <sup>2</sup>	B <sup>2</sup>	B <sup>2</sup>	C <sup>3</sup>
AMINO ACIDS (%)						
<u>Essential</u>						
ARG	73.0	87.4	88.0	92.2	81.5	82.0
HIS	77.4	88.3	90.7	93.6	80.4	75.0
ILE	68.4	87.5	83.4	88.1	79.1	77.0
LEU	76.8	92.5	88.5	93.8	81.5	78.0
LYS	64.1	82.0	81.2	83.0	73.3	70.0
MET	73.8	91.9	85.1	89.5	80.4	--
PHE	73.2	90.5	85.7	91.3	82.2	80.0
THR	61.1	78.9	80.8	86.3	71.2	67.0
VAL	69.7	84.9	85.3	88.2	78.0	75.0
<u>Non-Essential</u>						
ALA	67.2	88.5	83.8	90.8	69.7	68.0
ASP	61.6	83.9	80.9	86.6	71.2	75.0
GLU	77.8	91.8	90.4	94.1	86.6	84.0
GLY	50.1	71.2	78.3	86.2	71.2	80.0
PRO	76.0	80.4	92.1	93.4	80.9	83.0
SER	71.1	84.9	86.6	91.0	76.3	72.0
TYR	76.5	89.0	86.7	90.2	79.7	78.0

- 1 Reference A indicates Easter (1973).
- 2 Reference B indicates data from this study.
- 3 Reference C indicates Zébrowska (1973 a, b).



the caecal cannula. Phillipson (1952) in studies with sheep found that the amount of duodenal digesta returned to the distal cannula had a considerable effect on the flow of digesta from the proximal cannula. When no digesta was returned within even one hour, the flow rate was increased by 70%. An increase in flow rate above the normal flow rate at the end of the ileum (which could have occurred in Easter's studies) may have resulted in a decrease in protein digestibility and/or amino acid absorption and therefore lower ileal amino acid availabilities.

In general, the ileal amino acid availabilities obtained by Zébrowska (1973 a, b) for barley compare very well with those obtained in this study (Table 41). The data obtained by Zébrowska were also based on a continuous 24 hour ileal collection and ileal digesta was returned to the animal after sampling. Faecal amino acid availabilities were not measured.

#### Microbial fermentation in the large intestine.

The net disappearance of the individual amino acids, nitrogen, dry matter, starch and crude fiber are shown on a quantitative basis (grams per 100 grams dry matter intake) in Table 42. The pigs consumed the same amount of each cereal diet daily, namely their total allowable intake (Fig. 2) and the values shown would therefore reflect the total net

TABLE 42. The disappearance<sup>1</sup> of amino acids, nitrogen, dry matter, starch and crude fiber in the large intestine.

CEREAL GRAIN	CORN	WHEAT	BARLEY
AMINO ACIDS			
<u>Essential</u>			
ARG	.021	.040	.046
HIS	.014	.017	.025
ILE	.003	.017	.016
LEU	.017	.045	.043
LYS	.003	.018	.017
MET <sup>2</sup>	+.006 <sup>3</sup>	.005	+.001 <sup>3</sup>
PHE	.003	.025	.034
THR	.027	.040	.039
VAL	.015	.034	.035
<u>Non-Essential</u>			
ALA	.018	.049	.037
ASP	.019	.053	.046
CYS <sup>2</sup>	.016	.024	.024
GLU	.043	.191	.180
GLY	.074	.088	.084
PRO	.165	.143	.138
SER	.032	.054	.053
TYR	.004	.014	.019
NITROGEN	.136	.215	.244
DRY MATTER	9.2	15.7	17.4
STARCH	1.19	4.16	5.01
CRUDE FIBER	1.23	0.57	1.39

<sup>1</sup> Expressed as grams per 100 grams of dry matter intake.

<sup>2</sup> Determined by the oxidation method.

<sup>3</sup> Plus sign indicates an increase in the quantity of amino acids from the end of the ileum to the faeces.

daily disappearance in the large intestine of the particular nutrients.

Although this observation could be coincidental, the increase in starch disappearance from corn to wheat to barley in the large intestine was directly proportional to total nitrogen disappearance from these cereal grains in the large intestine. Hydrolysis in the rumen takes place through peptides of decreasing chain length to free amino acids. These free amino acids are finally degraded to ammonia and short-chain fatty acids. Presumably, this type of bacterial hydrolysis also takes place in the caecum. The extent of bacterial hydrolysis of undigested exogeneous and/or endogeneous protein will depend on the amount of energy that is available to the microflora of the large intestine.

More undigestible starch from barley or wheat than from corn enters the large intestine and is available to the microflora (Table 40 and 42). As a consequence, relatively more bacterial hydrolysis of undigested protein will take place when barley or wheat, rather than corn, is fed and a relatively larger production of ammonia and short-chain fatty acids will result. A proportion of the ammonia produced in the large intestine will be absorbed. As the amount produced increases, absorption will probably also increase. At the same time, with an increase in ammonia absorption, a proportion of the increase in ammonia produced will be assimilated by the micro-organisms.

Therefore, one would also expect more microbial protein synthesis in the large intestine when barley or wheat is fed and this should be reflected by relatively higher levels of DAPA in the faeces from barley and wheat than from corn. However, it should be kept in mind that as the micro-organisms move down the large intestine, part of these will undergo autolysis, presumably proportional to their number. Ammonia seems to be the major end product of degradation of microbial protein and is absorbed (Michel, 1966). The levels of DAPA in the faeces were measured but the reproducibility of results was poor and therefore did not warrant interpretation.

Disappearance of starch and crude  
fiber in the large intestine.

A substantial amount of starch from wheat and barley disappeared in the large intestine, namely 4.16 and 5.01 grams respectively (Table 42). These values correspond to 5.7 and 7.8% of the total dietary starch from wheat and barley respectively (Table 40). For corn, starch digestion was nearly complete by the end of the small intestine. Only 1.19 grams, corresponding to 1.7% of the total dietary intake, disappeared in the large intestine (Table 40 and 42). Holmes *et al.* (1973) also found starch digestion from corn to be nearly complete by the end of the small intestine. In this study complete digestion of starch was found by the faecal analysis method (Table 40).

If the remaining starch entering the large intestine was fermented in the same manner as in rumen fermentation, then 10% of the energy would be converted into bacterial cell bodies, 15% would be lost as heat and methane and 75% would be absorbed as volatile fatty acids. Volatile fatty acids are utilized with about 85% of the efficiency of carbohydrates. The net yield to the animal of the energy disappearing in the large intestine is then 64%<sup>1</sup> of glucose equivalents (Holmes et al., 1973). The actual amounts of energy lost from starch to the animal from corn, wheat and barley would then be 0.43<sup>2</sup>, 1.50<sup>2</sup>, and 1.80<sup>2</sup> grams per 100 grams of dry matter intake respectively.

Potentially, the flora of the large intestine may play a role in making undigestible carbohydrate available to the animal as volatile fatty acids. The latter may be of some importance to the animal when wheat or barley is fed.

The total amount of disappearance of crude fiber from the cereal grains in the large intestine did not follow the same relationship as that of starch from corn, wheat and

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$$^1(75/100 \times 85/100) = 64\%.$$

$$^21.19 - \left( \frac{64}{100} \times 1.32 \right) = 0.43; \quad 4.16 - \left( \frac{64}{100} \times 4.16 \right) = 1.50;$$

$$5.01 - \left( \frac{64}{100} \times 5.01 \right) = 1.80.$$

barley. Wheat and corn used in this experiment had approximately the same crude fiber content but relatively more crude fiber disappeared from corn in the large intestine than from wheat (Table 40). Due to the relatively low amount of starch from corn entering the large intestine, the flora may have had to obtain relatively more energy from the crude fiber fraction.

Possible effect of dietary particle size  
on ileal amino acid availabilities.

The distribution of particle size of the cereal grain diets and the ileal digesta derived from the cereal grains are shown in Table 43. The percentages of particles larger than 1.00 mm were 37.7, 79.8 and 61.9% for the corn, wheat and barley diets respectively. In the same order they were 12.9, 35.4 and 27.5% for ileal digesta from corn, wheat and barley.

The fractions larger than 1.00 mm were found to contain undigested cracked grain particles and fibrous material. The higher percentages of larger particles in ileal digesta from wheat and barley than from corn reflect the dietary particle size distribution of these grains. Higher ileal amino acid availabilities and starch digestibilities may possibly have been obtained from wheat and barley if these grains had been ground through a finer screen before they were made into pellets (page 46).

The implications of the effect of size of grinding

TABLE 43. The distribution of particle size of the cereal grain diets and of the ileal digesta derived from each diet.

CEREAL GRAIN	CORN		WHEAT		BARLEY	
	DIET <sup>1</sup>	DIGESTA <sup>2</sup>	DIET <sup>1</sup>	DIGESTA <sup>2</sup>	DIET <sup>1</sup>	DIGESTA <sup>2</sup>
PARTICLE DISTRIBUTION (%)						
>2.00 mm	3.6	2.2	18.6	6.4	8.5	4.3
1.00 to 2.00 mm	34.1	10.7	61.2	29.0	51.9	23.2
.50 to 1.00 mm	30.2	23.5	13.0	19.4	22.6	25.4
.25 to .50 mm	18.6	28.9	5.5	17.4	11.1	24.1
.125 to .25 mm	8.0	20.9	1.1	12.0	3.6	11.8
<.125 mm	5.4	14.1	0.9	16.0	2.1	11.4

<sup>1</sup> Determined after the diets were pelleted.

<sup>2</sup> Average from 3 ileal collections from 3 pigs chosen at random.

on ileal amino acid availabilities were not realized prior to the start of this experiment and the effect of dietary particle size distribution was reexamined in more detail in study 4b.

A certain degree of fractionation of the ingested dietary components will take place during stomach emptying. This in turn will determine the uniformity of flow and composition of digesta through the end of the ileum. The amount of fractionation of digesta by the stomach will be influenced by the particle size distribution of the diet. In these experiments, particle size distribution was largest for wheat and only minor differences in the amino acid composition of ileal digesta from the individual 8-hour collections were found (Table 21). Therefore, it would be safe to assume that the individual 8-hour collections of ileal digesta from corn and barley were also of a similar composition.

True ileal amino acid availabilities  
from the cereal grains based on  
data from the maintenance diets.

As was explained previously (page 76 ), the metabolic ileal and faecal amino acid levels in study 2 were originally determined by aid of the M-7 and M-14 diets. These diets contained 4% casein as the only protein source.

True faecal amino acid availabilities from casein for the pig were found to be very close to 100% (Eggum, 1973). The



availabilities of the essential amino acids, with the exception of ILE of which the availability was 95.4%, ranged from 97.9 to 99.9%. Consequently, the amino acids in faeces collected from pigs fed casein should be indicative of the metabolic faecal amino acid losses.

Two maintenance diets were formulated, namely with 7 and 14% alphafloc respectively. Dietary fiber level has been shown to affect the metabolic faecal nitrogen excretion in pigs and rats (Fehm, 1954; Meyer, 1956; Mitchell, 1954; Whiting and Bezeau, 1957 a,b). Therefore, it can be expected that dietary fiber level will also affect the metabolic faecal excretion of the individual amino acids.

Ileal and faecal samples from M-7 fed to the cannulated pigs after the cereal test diet sequence were lost and the effect of body weight on the metabolic ileal and faecal amino acid levels could not be assessed. The effect of weight on the metabolic faecal nitrogen and amino acid levels, when these were expressed as grams excreted per 100 grams of dry matter intake, was found to be very small for the non-cannulated pigs in study 2b (Table 26). In addition, only small differences due to weight of rats on the metabolic faecal amino acid excretion were found in study 1 (Table 18).

On the assumption that there was no effect of weight on the metabolic amino acid and nitrogen levels for the cannulated pigs, raising the dietary alphafloc level from 7 to

14% increased the metabolic ileal and faecal amino acid and nitrogen levels (Table 44). The metabolic faecal nitrogen level increased from approximately .13 to .17 grams per 100 grams of dry matter intake. This increase in faecal nitrogen excretion with increasing fiber levels compares favourably with data obtained by Whiting and Bezeau (1957 a, b) and Behm (1954).

For both M-7 and M-14, there was a consistent net disappearance of ARG, THR, GLU, GLY PRO and SER in the large intestine. The net disappearance of PRO, which was present in very abundant amounts in ileal digesta, was quite substantial (Table 44).

Bovine salivary mucin contains many repeating units of SER-GLY-THR-PRO peptides. The amino acids from these peptides make up half the amino acids in mucin. Glutamic acid and ASP were most prominent among the other amino acids present in mucin (Horowitz, 1967). The amino acids mentioned were also present in large amounts in ileal digesta from M-7 and M-14. The amino acids found in ileal digesta from the maintenance diets may therefore be mainly derived from mucus that is secreted by the small intestine and that is not digested and reabsorbed prior to the end of the ileum.

For both M-7 and M-14, there were small but consistent net increases of LEU, LYS, MET, PHE and TYR from the end

TABLE 44. Levels of amino acids, total nitrogen and dry matter in ileal digesta and faeces from the cannulated pigs fed the maintenance diets.

LEVEL OF ALPHAFLC (%)	7 <sup>1</sup>			14 <sup>2</sup>		
	ILEUM	FAECES	A <sup>3</sup>	ILEUM	FAECES	B <sup>3</sup>
AMINO ACIDS <sup>4</sup>						
<u>Essential</u>						
ARG	.033	.024	-.009	.046	.030	-.016
HIS	.012	.010	-.002	.018	.014	-.004
ILE	.033	.030	-.003	.036	.039	+.003
LEU	.039	.043	+.004	.044	.057	+.013
LYS <sup>5</sup>	.034	.040	+.006	.042	.049	+.007
MET	.009	.014	+.005	.011	.017	+.006
PHE	.021	.027	+.006	.023	.034	+.011
THR	.042	.030	-.008	.045	.040	-.005
VAL	.036	.034	-.002	.042	.044	+.002
<u>Non-Essential</u>						
ALA	.040	.041	+.001	.056	.054	-.002
ASP	.064	.064	.000	.073	.083	+.010
GLU	.120	.086	-.034	.147	.111	-.036
GLY	.097	.033	-.064	.148	.042	-.106
PRO	.314	.033	-.281	.576	.058	-.518
SER	.065	.034	-.031	.078	.045	-.033
TYR	.017	.023	+.006	.018	.025	+.007
NITROGEN	.201	.129	-.072	.306	.167	-.139
DRY MATTER	13.25	8.65	-4.60	23.34	13.42	-9.92

<sup>1</sup> M-7 was fed prior to the cereal test diet sequence, results from pig number 1 were not included.

<sup>2</sup> M-14 was fed following the cereal test diet sequence, results from pig number 1 were not included.

<sup>3</sup> Columns A and B indicate the differences between ileal and faecal levels.

<sup>4</sup> Expressed as grams per 100 grams dry matter intake.

<sup>5</sup> Determined by acid hydrolysis.

of the ileum to the faeces. These increases may have been due to de novo microbial synthesis of these amino acids from nitrogen that was derived from microbial deamination of the major amino acids present in mucin.

The levels of PRO in ileal digesta from M-7 and M-14 were .314 and .576 grams per 100 grams of dry matter intake respectively (Table 44). The PRO levels in ileal digesta from corn, wheat and barley were .222, .191 and .232 respectively (Table 42). True ileal availabilities of PRO exceeding 100% would be obtained for all cereal grains if one simply used the levels of metabolic ileal PRO obtained from either M-7 or M-14. Of course, the maximum true availability of any amino acid can only be 100%.

According to Whiting and Ezeau (1957 a, b) metabolic nitrogen levels corresponding to particular test diets should be determined with protein-free diets that contain similar levels of fiber as the test diets. Therefore, it can be expected that the same would apply for the determination of metabolic ileal and faecal amino acid levels. The crude fiber level of the maintenance diets were 4.95% for M-7 and 9.62% for M-14 and the ileal PRO levels were .314 and .576 grams respectively (Table 44). The crude fiber levels of corn, wheat and barley were 2.62, 2.56 and 5.37% respectively (Table 19). Corrections based on equal crude fiber intake

per 100 grams of dry matter intake, of the cereal and maintenance diets gave corresponding metabolic ileal PRO levels of .190, .180 and .340 grams for corn, wheat and barley respectively. Thus, true ileal PRO availabilities of 97.3, 99.9 and 108.9% were obtained for corn, wheat and barley respectively. The calculation carried out assumed a linear relationship between crude fiber intake and ileal PRO levels.

According to Sauer et al. (1974) metabolic amino acid levels corresponding to particular test diets should be determined with protein-free diets that contain the same level of undigestible dry matter as the test diets from which one determines the true amino acid availabilities. The amounts of dry matter passing through the end of the ileum, per 100 grams of dry matter intake, were 13.25, 23.34, 19.80, 26.70 and 33.80 grams for M-7, M-14, corn, wheat and barley respectively (Table 42 and 44). Similar type of corrections (as for equal crude fiber intake) for the determination of metabolic ileal PRO levels would result in true PRO availabilities exceeding 100% for all cereal grains tested.

The interpretation of data obtained from the maintenance and cereal test diets in the previous section were based on results that were expressed as grams per 100 grams of dry matter intake. The average daily dry matter intake by the pigs fed M-7 and M-14 was 1486 (87.8% of

allowable intake) and 935 grams (48.9% of allowable intake) respectively (Appendix, Table 9). The average daily dry matter intake of the cereal diets by the pigs was equal to the allowable daily intake in all cases and was 1855, 1849 and 1857 for corn, wheat and barley respectively (Appendix, Table 3, 4 and 5). Calculations based on equal total crude fiber intake for the cereal and maintenance diets (in the same manner as they were carried out for crude fiber intake per 100 grams of dry matter intake) will result in true PRO availabilities of 96.7, 100.4 and 107.1% for corn, wheat and barley respectively.

From the previous discussion one might believe that PRO of casein was of relatively low availability at the end of the ileum. If this was the case for PRO, what then about the true ileal availabilities of the other amino acids from casein?

Taking into account the possibility that casein was perhaps not completely digested and absorbed by the end of the ileum, complete protein-free diets were fed to the cannulated pigs as part of study 4a. Therefore, the data obtained for metabolic ileal and faecal amino acid levels from feeding the protein-free diets in study 4a will be used for the estimations of true ileal amino acid availabilities from corn, wheat and barley.

True ileal amino acid availabilities  
from the cereal grains  
based on data from the protein-free diets.

Three different protein-free diets containing 5, 10 and 15% alphafloc respectively were fed to the pigs in study 4a. The average daily dry matter intake were 1551 g. for PF-5; 1357 g. for PF-10 and 1109 g. for PF-15 (Appendix, Table 23, 24 and 25).

Continuous problems were experienced during the feeding of the PF-15 diets. Blockage occurred frequently. Often the cannulae became disconnected, resulting in large losses of digesta. Three of the 6 pigs went completely off feed after the first or second day that they were offered the PF-15 diets. Therefore, suitable statistical analyses for the determination of the effect of body weight and level of dietary fiber on metabolic ileal and faecal amino acid levels could not be performed. Previous results obtained from pigs fed the maintenance diets (Table 26) and from rats fed the protein-free diets (Table 18) showed that there was virtually no effect of weight on the metabolic faecal amino acid levels when these were expressed as grams per 100 grams of dry matter intake. Although data were available only from pigs numbered 6 and 9, the results obtained seemed to indicate that weight had no major effect on the metabolic ileal amino acid levels (Appendix, Table 24 and 25).

In general, increasing the fiber levels of the protein-free diets increased the metabolic ileal and faecal amino acid and nitrogen levels. The increment in the levels of ileal and faecal amino acids levelled off as the alphafloc content of the protein-free diets was raised from 10 to 15% (Table 45). Due to the limited amount of results available from diet PF-15, it is not possible to tell if the increase in metabolic ileal and faecal amino acid levels with increasing fiber intake is of a linear or a curvilinear relationship. Theoretically, it is not unreasonable to postulate a limit to metabolic losses by the animal and assume a curvilinear relationship.

The relationships of the ileal and faecal amino acid and nitrogen levels between M-7 and M-14 were basically similar to those observed between PF-5 and PF-10 (Table 44 and 45). For both PF-5 and PF-10 (and also for PF-15), there was a consistent net disappearance of ARG, THR, GLY, PRO and SER in the large intestine. Consistent net increases were found for ILE, LEU, LYS, MET, PHE and ASP.

It seemed that as the level of alphafloc was increased from 5 to 10%, particular amino acids decreased more (for example GLY and PRO) and particular amino acids increased more (for example ILE, LEU, LYS and PHE). In other words, more microbial fermentation seemed to take place in the large intestine with PF-10 than with PF-5.



TABLE 45. Amino acid, total nitrogen and dry matter appearance (+) or disappearance (-)<sup>1</sup> in the large intestine in pigs fed the protein-free diets.

LEVEL OF ALPHAFLC (%)	5			10			15			14.5 <sup>3</sup>		
LOCATION	ILEUM <sup>2</sup>	FAECES <sup>2</sup>	A	ILEUM <sup>2</sup>	FAECES <sup>2</sup>	B	ILEUM <sup>2</sup>	FAECES <sup>2</sup>	C	ILEUM <sup>2</sup>	FAECES <sup>2</sup>	D
AMINO ACIDS												
<u>Essential</u>												
ARG	.049	.022	-.027	.058	.029	-.029	.061	.033	-.028	.031	.137	+.106
HIS	.014	.010	-.004	.017	.014	-.003	.022	.015	-.007	.003	.006	+.003
ILE	.021	.027	+.006	.029	.039	+.010	.031	.045	+.024	.026	.051	+.025
LEU	.039	.042	+.003	.051	.059	+.008	.057	.067	+.010	.041	.089	+.048
LYS	.027	.036	+.009	.036	.048	+.012	.039	.054	+.015	.036	.092	+.056
MET	.006	.012	+.006	.008	.017	+.009	--	--	--	.003	.020	+.017
PHE	.023	.026	+.003	.030	.037	+.007	.033	.040	+.007	.022	.055	+.033
THR	.039	.030	-.009	.051	.041	-.010	.059	.045	-.014	.042	.072	+.030
VAL	.031	.033	+.002	.043	.049	+.006	.044	.053	+.009	.038	.059	+.021
<u>Non-Essential</u>												
ALA	.041	.036	-.005	.056	.053	-.003	.058	.059	+.001	.048	.082	+.034
ASP	.056	.061	+.005	.078	.084	+.006	.081	.096	+.015	.058	.130	+.072
CYS	.013	.010	-.003	.016	.013	-.003	--	--	--	.005	.025	+.020
GLU	.071	.068	-.003	.094	.096	+.002	.100	.108	+.008	.080	.162	+.082
GLY	.139	.031	-.108	.171	.043	-.128	.169	.047	-.122	.082	.078	-.004
PRO	.474	.026	-.448	.577	.046	-.531	.585	.038	-.547	.331	.034	-.297
SER	.038	.026	-.012	.049	.035	-.014	.053	.037	-.016	.018	.050	+.032
TYR	.013	.014	+.001	.015	.016	+.001	.016	.018	+.002	.040	.071	+.031
NITROGEN	.205	.101	-.104	.256	.139	-.117	.271	.161	-.110	.427	.360	-.067
DRY MATTER	11.67	6.48	-5.19	17.69	10.53	-7.16	26.32	14.78	-11.54	19.20	13.40	-5.80

<sup>1</sup> Expressed as grams per 100 gram dry matter intake, shown in columns A, B, C and D.

<sup>2</sup> Columns indicate ileal or faecal levels.

<sup>3</sup> Converted results from Holmes *et al.*, (1974).

Holmes et al. (1974) fed protein-free diets containing 14.5% alphafloc to 6 cannulated pigs that weighed approximately 45 kg. Their results and those obtained in these studies were quite different (Table 45). With the exception of PRO and GLY, Holmes et al. (1974) found a net increase of all amino acids between the end of the ileum and the anus. These increases were very substantial for ARG especially, LEU, LYS, ASP and GLU. The amino acid composition (grams per 16 grams of nitrogen) of metabolic faecal protein from pigs determined at different laboratories is shown in Table 46. Although there were differences in the composition for some amino acids (PHE, SER and TYR in particular), generally speaking the results obtained in these studies, those of Dammers (1964), Carlson and Bayley (1970) and Eggum (1971) compare reasonably well. The metabolic faecal amino acid composition obtained by Holmes et al. (1974) differed markedly from those obtained in these studies and from the other workers, especially with regard to the content of ARG and HIS. Insufficient dietary equilibration may have taken place by the time Holmes et al. (1974) collected faecal material. The latter may have contributed to the relatively large increases that were found for some amino acids between the end of the ileum and the anus in their studies (Table 45).

The level of fiber inclusion in the protein-free diets in these studies did not seem to have a major effect

TABLE 46. Amino acid composition<sup>1</sup> of metabolic protein determined at different laboratories.

SOURCE	SAUER (1976) <sup>2</sup>			DAMMERS (1964)	CARLSON and BAYLEY (1970)	EGGUM (1971)	HOLMES et al (1974)
	PF-5	PF-10	PF-15				
AMINO ACIDS							
<u>Essential</u>							
ARG	3.49	3.34	3.28	3.20	--	4.19	6.09
HIS	1.58	1.62	1.49	1.40	--	2.09	0.27
ILE	4.28	4.49	4.47	4.00	4.20	3.97	2.27
LEU	6.65	6.79	6.66	5.60	6.40	6.07	3.96
LYS	5.70	5.53	5.37	4.90	4.30	5.65	4.09
MET	1.90	1.96	--	2.10	--	2.03	0.89
PHE	4.12	4.26	3.98	3.60	3.80	5.09	2.44
THR	4.75	4.72	4.47	4.80	5.10	4.38	3.20
VAL	5.23	5.64	5.27	5.00	4.90	4.82	2.62
<u>Non-Essential</u>							
ALA	5.70	6.10	5.86	5.30	5.00	5.64	3.64
ASP	9.66	9.67	9.54	9.00	8.30	8.34	5.78
CYS	1.58	1.50	--	1.50	--	1.61	1.11
GLU	10.77	11.05	10.73	9.40	8.00	10.66	7.20
GLY	4.91	4.95	4.67	4.20	4.30	4.48	3.47
PRO	4.12	5.29	3.78	--	2.20	--	1.51
SER	4.12	4.03	3.68	3.80	4.70	7.01	2.22
TYR	2.22	1.84	1.79	1.80	3.00	3.44	3.16

<sup>1</sup> Expressed as grams per 16 grams of nitrogen.

<sup>2</sup> Determined for protein-free diets with different levels of alphafloc inclusion.

on the composition of metabolic faecal protein (Table 45).

Proline in particular and GLY were present in abundant amounts in ileal digesta from the protein-free diets (Table 45). These observations would eliminate the possibility that PRO from casein was of low availability by the end of the ileum. The level of PRO in ileal digesta from PF-5 was even higher than that obtained from M-7 (Table 44 and 45). The levels of PRO obtained from either PF-10, PF-15 or M-14 were of the same order.

Apparent ileal amino acid availabilities and "true" ileal availabilities that were calculated by using different corrections for metabolic ileal amino acid levels are shown in Table 47, 48 and 49 for corn, wheat and barley respectively.

Metabolic ileal amino acid corrections for obtaining true availabilities were carried out as follows: 1) On the basis of equal crude fiber intake of the cereal and protein-free diets and 2) On the basis of equal amounts of undigestible dry matter passing through the end of the ileum from the cereal and protein-free diets. For example, as the level of crude fiber intake increased from 3.86% for PF-5 to 6.80% for PF-10, the level of metabolic ileal LYS increased from .027 to .036 grams per 100 grams of dry matter intake (Table 32 and 45). Then, by extrapolation of the line connecting these 2 points one can estimate the metabolic ileal LYS levels at

TABLE 47. Apparent ileal amino acid availabilities from corn and the estimated true availabilities as calculated by the use of different type of corrections.

TYPE OF CORRECTION	APPARENT					TRUE			
	NONE		CRUDE FIBER INTAKE			DRY MATTER PASSAGE		PROLINE	
	A (%) <sup>1</sup>	B <sup>2</sup>	C (%) <sup>3</sup>	D <sup>4</sup>	E (%) <sup>5</sup>	D <sup>4</sup>	E (%) <sup>5</sup>	D <sup>4</sup>	E (%) <sup>5</sup>
AMINO ACIDS									
<u>Essential</u>									
ARG	.445	.056	87.4	.046	97.8	.058	100.4	.028	93.7
HIS	.254	.030	88.2	.013	93.3	.017	94.9	.007	90.9
ILE	.354	.045	87.3	.018	92.4	.029	95.5	.003	88.1
LEU	1.307	.099	92.4	.035	95.1	.051	96.3	.012	93.3
LYS <sup>6</sup>	.338	.061	82.0	.024	89.1	.035	92.3	.009	84.6
MET	.250	.020	91.9	.005	94.0	--	--	.002	92.8
PHE	.523	.049	90.6	.020	94.5	.030	96.4	.007	92.0
THR	.364	.077	78.8	.035	88.5	.051	92.9	.012	82.1
VAL	.456	.069	84.9	.027	90.8	.043	94.3	.003	85.5
<u>Non-Essential</u>									
ALA	.760	.088	88.4	.036	93.2	.056	95.8	.008	89.5
ASP	.723	.116	84.0	.049	90.7	.078	94.7	.004	84.5
CYS	.200	.036	82.1	.012	88.0	--	--	.009	86.5
GLU	2.085	.166	92.0	.063	95.1	.094	96.5	.020	93.1
GLY	.399	.129	67.7	.129	100.0	.171	110.5	.074	86.2
PRO	.843	.222	73.7	.438	125.6	.577	142.1	.222	100.0
SER	.505	.077	84.8	.034	93.5	.049	94.5	.013	87.3
TYR	.395	.043	89.1	.012	92.2	.015	92.9	.009	91.4
NITROGEN	1.995	.348	82.6	.188	92.0	.256	95.4	.090	87.1

<sup>1</sup> Column A indicates the dietary amino acid content. <sup>2</sup> Column B indicates the apparent ileal amino acid levels, expressed as grams per 100 gram dry matter intake.

<sup>3</sup> Column C indicates the apparent ileal amino acid availabilities.

<sup>4</sup> Columns D indicate the respective corrected metabolic ileal amino acid levels, expressed as grams per 100 grams dry matter intake.

<sup>5</sup> Columns E indicate the respective true ileal amino acid availability estimates.

<sup>6</sup> Determined by the oxidation method.

TABLE 48. Apparent ileal amino acid availabilities from wheat and the estimated true availabilities as calculated by the use of different type of corrections.

TYPE OF CORRECTION	APPARENT					TRUE			
	NONE			CRUDE FIBER INTAKE		DRY MATTER PASSAGE		PROLINE	
	A(%) <sup>1</sup>	B <sup>2</sup>	C(%) <sup>3</sup>	D <sup>4</sup>	E(%) <sup>5</sup>	D <sup>4</sup>	E(%) <sup>5</sup>	D <sup>4</sup>	E(%) <sup>5</sup>
AMINO ACIDS (%)									
<u>Essential</u>									
ARG	.575	.082	85.7	.045	93.6	.061	96.3	.026	90.3
HIS	.294	.032	89.1	.013	93.5	.022	95.6	.006	91.2
ILE	.444	.065	85.4	.018	89.4	.031	92.3	.001	85.6
LEU	.970	.127	86.9	.034	90.4	.057	92.8	.009	87.8
LYS	.362	.088	75.7	.024	82.3	.039	86.5	.006	77.4
MET <sup>6</sup>	.241	.032	86.7	.005	88.8	--	--	.001	87.1
PHE	.671	.075	88.8	.020	91.8	.033	93.7	.005	89.6
THR	.386	.091	76.4	.034	85.2	.059	91.7	.009	78.8
VAL	.545	.094	82.8	.026	87.5	.044	90.8	.000	82.8
<u>Non-Essential</u>									
ALA	.488	.127	74.0	.035	81.1	.058	85.9	.004	74.8
ASP <sup>6</sup>	.677	.167	75.3	.047	82.3	.081	87.3	.000	75.3
CYS <sup>6</sup>	.281	.042	85.1	.012	89.3	--	--	.008	87.9
GLU	4.440	.324	92.7	.062	94.1	.100	95.0	.016	93.1
GLY	.566	.149	73.7	.126	95.9	.169	103.5	.066	85.3
PRO	1.445	.191	86.8	.432	116.7	.585	127.3	.191	100.0
SER	.633	.101	84.0	.033	89.3	.053	92.4	.010	85.6
TYR	.407	.057	86.0	.012	88.9	.016	89.9	.008	88.0
NITROGEN	2.607	.445	82.9	.184	90.0	.271	93.3	.076	85.8

1, 2, 3, 4, 5, 6 Refer to Table 47 for explanation.

TABLE 49. Apparent ileal amino acid availabilities from barley and the estimated true availabilities as calculated by the use of different type of corrections.

TYPE OF CORRECTION	APPARENT					TRUE			
	NONE		CRUDE FIBER INTAKE			DRY MATTER PASSAGE		PROLINE	
	A (%) <sup>1</sup>	B <sup>2</sup>	C (%) <sup>3</sup>	D <sup>4</sup>	E (%) <sup>5</sup>	D <sup>4</sup>	E (%) <sup>5</sup>	D <sup>4</sup>	E (%) <sup>5</sup>
AMINO ACIDS									
<u>Essential</u>									
ARG	.569	.106	81.4	.053	90.7	.061	92.1	.029	86.5
HIS	.229	.045	80.3	.015	86.9	.022	90.0	.007	83.4
ILE	.390	.082	79.0	.024	85.1	.031	86.9	.004	80.5
LEU	.855	.158	81.5	.044	86.7	.057	88.2	.013	83.0
LYS <sup>6</sup>	.415	.111	73.3	.030	80.5	.039	82.7	.009	75.4
MET	.193	.038	80.3	.007	83.9	--	--	.002	81.3
PHE	.601	.107	82.2	.026	86.5	.033	87.4	.007	83.4
THR	.381	.110	71.1	.044	82.7	.059	86.6	.013	74.5
VAL	.544	.120	77.9	.036	84.6	.044	86.0	.004	78.7
<u>Non-Essential</u>									
ALA	.482	.146	69.7	.047	79.5	.058	81.7	.009	71.6
ASP	.694	.200	71.2	.070	81.3	.081	82.9	.006	72.0
CYS <sup>6</sup>	.219	.049	77.6	.014	84.0	--	--	.009	81.7
GLU	2.927	.393	86.6	.080	89.3	.100	90.0	.022	87.3
GLY	.485	.168	65.4	.152	96.7	.169	100.2	.076	81.0
PRO	1.215	.232	80.9	.516	123.4	.585	129.1	.232	100.0
SER	.501	.119	76.2	.042	84.6	.053	86.8	.014	79.0
TYR	.364	.074	79.2	.014	83.5	.016	84.1	.009	82.1
NITROGEN	2.222	.558	74.9	.225	85.0	.271	87.1	.094	79.1

1, 2, 3, 4, 5, 6 Refer to Table 47 for explanation.

other levels of crude fiber i.e. at the crude fiber levels of the cereal grains. The crude fiber levels of corn, wheat and barley were 2.62, 2.56 and 5.37% respectively (Table 19). In the same order, these crude fiber levels would give corresponding metabolic ileal LYS levels of 0.24, 0.24 and 0.30 grams per 100 grams of dry matter intake (Table 47, 48 and 49). The apparent ileal amino acid levels were then corrected for the corresponding metabolic ileal amino acid levels and true ileal amino acid availabilities were obtained.

For the estimation of true ileal availabilities of amino acids based on "equal dry matter passage", the calculations were based on the levels of indigestible dry matter passing through the end of the ileum for the test and protein-free diets. The type of corrections that were carried out resulted in true availabilities of PRO that exceeded 100% for all cereal grains. In addition, true availabilities of GLY exceeded 100% in some instances (Table 47, 48 and 49).

It should be kept in mind that metabolic ileal amino acid levels corresponding to the cereal test diets were extrapolated linearly from data obtained from diets PF-5 and PF-10. Metabolic ileal data were obtained only from 3 pigs from diet PF-15, and these may have been biased because of the problems that were encountered during the feeding of this diet. However,



the interpretation of results obtained would not change, essentially, even if data from diet PF-15 were included for the extrapolation of metabolic ileal amino acid levels from the protein-free diets. Similarly, interpretation of results would not change if calculations based on total crude fiber intake or total "dry matter passage" were carried out.

The last columns in Tables 47, 48 and 49 (Column C under the heading of Proline) show the estimated true ileal amino acid availabilities from the cereal grains when the true PRO availability was set at 100%. As the ileal levels of PRO increase with increasing dietary fiber level (from PF-5 to PF-10), the ileal levels of all the other amino acids also increase, though not always in the same proportion (Table 45). If the true availability of PRO from corn is set at 100% for example, then the apparent ileal PRO level in corn digesta would be equal to the metabolic ileal PRO level for corn, namely .222 grams per 100 grams of dry matter intake (Table 47). At this particular metabolic ileal PRO level, corresponding metabolic ileal levels of the other amino acids can be obtained by extrapolation from amino acid levels that corresponded to .474 grams of PRO that was obtained by feeding the PF-5 diet and to .577 grams of PRO per 100 grams of dry matter intake that was obtained by feeding the PF-10 diet.

(Table 45). These extrapolations resulted in very low levels of metabolic ileal amino acids and gave true ileal availability estimates that were very close to the corresponding apparent availability estimates for most amino acids (Table 47, 48 and 49).

It is reasonable to assume that the true ileal availabilities of PRO from cereal grains will be less than 100%. Therefore, even the calculations based on "100% true PRO availability from cereal grains" will overestimate the metabolic ileal amino acid levels associated with the feeding of the cereal grains.

Regardless of the corrections used, the estimated true ileal LYS availabilities from corn, wheat and barley were still relatively low in relation to the other amino acids. Corrections based on equal crude fiber levels resulted in estimated true ileal LYS availabilities of 89.1, 82.3 and 80.5% for corn, wheat and barley respectively. In that same order, they were 92.3, 86.5 and 82.7% when based on equal "dry matter passage".

However, the type of metabolic corrections performed can not be valid since true ileal PRO and GLY availabilities were obtained that exceeded 100%. Due to the extensive disappearance of these amino acids in the large intestine (presumably by microbial deamination), true faecal availabilities of these 2 amino acids did not exceed 100%.

The calculations and postulations made are subject to several possible errors: 1) The variability of the ileal PRO levels between pigs fed the protein-free diets (Appendix, Table 23, 24 and 25), 2) Although consistent results were obtained with regard to data from diets PF-5 and PF-10, these could not be subjected to statistical analysis and 3) Estimated true availabilities were based on linear responses from data obtained from the PF-5 and PF-10 diets.

In conclusion, there is the possibility that the use of protein-free diets for estimation of metabolic ileal amino acid levels (and therefore also for metabolic faecal amino acid levels) may result in overestimation of the actual levels.

Problems associated with the use of  
protein-free diets for the estimation  
of true ileal and faecal amino acid availabilities.

The following discussion will deal with some possible explanations and postulations as to why protein-free diets can overestimate the metabolic ileal and faecal amino acid losses as well as nitrogen losses.

According to Nasset (1965) ingested protein is mixed with several times its mass of endogenous protein in the small intestine so that wide fluctuations in the amino acid mixture available for absorption will not occur. The relatively constant amino acid mixture produced will contribute

to maximum efficiency in the utilization of amino acids by the animal. Of course, this proposed homeostatic mechanism is of a short term nature and will fail if a protein deficiency (or a specific amino acid deficiency) is imposed on the animal for longer periods of time.

Nasset (1965) estimated that the total daily amount of endogenous protein secreted into the lumen of the small intestine of a 70 kg. man to be 141-354 grams (Table 50). The estimate calculated by Nasset is much higher than that calculated by Fauconneau and Michel (1970), which was only 67 grams (Table 50). The latter authors' did not include protein from intestinal juice as part of the endogenous protein derived from digestive secretions. Not including estimates from intestinal juice in Nasset's calculations, the amount of protein secreted from digestive juices into the lumen will be 24-63 grams. The minimum estimate obtained, 24 grams, compares reasonably well with that obtained by Fauconneau and Michel (1970), which was 17 grams (Table 50). The estimates from both sources for protein derived from mucosal shedding were not that different, namely 50 and 77-91 grams (Table 50).

The manner in which Nasset (1965) arrived at the relatively large contribution of endogenous protein from intestinal juice to the total endogenous protein from the

TABLE 50. Estimation of the daily endogeneous protein secretion into the lumen of the small intestine of a 70 kg. man.

REFERENCE	NASSET (1965)		FAUCONNEAU and MICHEL (1970)	
ITEMS	VOLUME <sup>1</sup>	PROTEIN		PROTEIN
	liters (range)	% (range)	grams (range)	grams
PROTEIN FROM DIGESTIVE SECRETIONS				
Saliva	1.0- 1.5	0.2-0.8	2-12	3
Gastric Juice	2.5- 3.5	0.3-0.5	8-18	5
Pancreatic Juice	1.2- 1.5	1.0-2.0	12-30	8
Intestinal Juice	1.0- 2.0	0.4-1.0	40-200	-
Bile	0.5	0.4-0.6	2-3	1
Subtotal (A) <sup>2</sup>	15.2-27.0		64-263	17
Subtotal (B) <sup>3</sup>	5.2- 7.0		24-63	17
PROTEIN FROM MUCOSAL SHEDDING			77-91	50
TOTAL PROTEIN (A) <sup>2</sup>			141-354	67
TOTAL PROTEIN (B) <sup>3</sup>			101-154	67
MUCOSAL PROTEIN % OF TOTAL (E) <sup>3</sup>			50-90	75

<sup>1</sup> Daily volume of the digestive secretions.

<sup>2</sup> Additions include data from intestinal juice.

<sup>3</sup> Additions do not include data from intestinal juice.

digestive secretions is not clear. Nasset (1965) stated as follows: "The volumes of secretion were derived from many sources and except for intestinal juice, they are generally available and accepted. The large volume for intestinal juice was computed from the fact that the pylorus admits 7 liters of gastric contents to the duodenum in 24 hr. and that gastric contents are diluted three to five times in the duodenum and upper jejunum (Eörgstrom et al., 1957)." Subsequently, Nasset estimated the total volume of intestinal juice secreted daily into the lumen to be 10 to 20 liters, with variations in protein content from 0.4 to 1.0% (Table 50).

In interpreting Nasset's data, first of all it is unclear how he arrived at 7 liters of gastric contents to be emptied daily into the duodenum. Of these 7 liters, a maximum of only 3.5 to 5 liters can be accounted for by saliva and gastric juice (Table 50). Secondly, Nasset infers from work by Borgström et al. (1957) that gastric contents are diluted three to five times in the duodenum and upper jejunum. Borgström et al. (1957) fed 500 ml. liquid test diets to 6 human subjects. Samples of digesta were taken by intubation. The test meal during its passage through the duodenum was diluted to a volume of 1.5 to 2.5 liters. Polyethylenglycol, which is not absorbed in the intestine,

was used as a reference substance to measure the dilution. However, the dilutions that were found can easily be explained by digestive secretions of saliva, gastric juice, pancreatic juice and bile. The total daily secretion of these digestive juices may range from 5.2 to 7 liters (Table 50). Therefore, the dilutions measured by Borgström et al. (1957) are not necessarily due to the secretion of intestinal juice. Even if there is a secretion of intestinal juice into the lumen, this secretion may perhaps just simply represent movement of water from the epithelial cells into the lumen in response to an increase in osmotic pressure. After pancreatic juice has been added to the intestinal contents, protein and carbohydrate digestion, in so far as it relates to the formation of significant amounts of digestive endproducts (such as free amino acids and glucose) becomes of importance. The formation of these free digestive endproducts will increase the osmotic pressure in the lumen and naturally water will move from the epithelial cells into the lumen. Notwithstanding, some free amino acids from the epithelial cells may initially move along with the water into the lumen by a "solvent drag" type of mechanism.

From the previous discussion, it follows that Nasset may have overestimated the endogenous protein secretions into the lumen of the small intestine. The results obtained by

Zébrowska (1973 a, b) did not indicate a significant dilution of feed nitrogen by the endogenous nitrogen secreted in the proximal parts of the alimentary tract as has been suggested by Nasset. Zébrowska (1973 a, b) fitted duodenal re-entrant cannulae to 60 kg. pigs. With the exception of GLY, the amino acid composition of digesta obtained at the end of the duodenum and that of the diet fed were very similar. Glycine is found in relatively high amounts in glycocholic acid of the bile and its salts.

Not taking into account the doubtful levels of endogenous protein secretions from intestinal juice, protein derived from mucosal shedding will make up the major proportion of the total endogenous protein, namely 75% using data from Fauconneau and Michel and 50 to 90% (average is 70%) using data from Nasset (Table 50).

The endogenous protein derived from shedded cells may be subdivided into: 1) protein or amino acids loosely contained in these cells. The protein will be largely made up of cytoplasmic enzymes that hydrolyse di- and tripeptides and disaccharides, 2) protein and/or amino acids tightly associated with inner and outer membrane structures, and 3) mucus protein associated with the outer membrane part of the cells. The last 2 types of protein are of a lower digestibility than the cytoplasmic protein and will therefore make up the



major portion of indigestible endogenous protein that was found at the end of the ileum. Peptido-glycans (part of mucus protein) are resistant to trypsin and pepsin (Allison, 1970). The relative digestibilities of whole cells of bacteria and cell contents indicated that nitrogen in wall structures was undigestible (Tannenbaum, 1968).

Is it possible, then, that the protein-free diets that were used in these studies increased the shedding of epithelial cells above the level normally taking place during the feeding of the cereal test diets? Feeding protein-deficient diets to rats for short periods of time resulted in the reduction of the number of cells of the small intestine. Cell size was not affected (Durand et al., 1966; Munro and Goldberg, 1964; Vandermeers et al., 1966). The small intestine of the rat lost 25% of its nitrogen after 8 days of feeding a protein-free diet (Ju and Nasset, 1959). The loss of nitrogen may possibly be due largely to a decrease in the number of epithelial cells of the small intestine.

Part of the cells of the small intestine may actually be a subunit of the so-called "labile protein storage reservoir" of the animal. Increased cellular losses of these cells, in response to the feeding of protein-free or protein-deficient diets, with subsequent digestion of protein components and absorption of amino acids derived thereof for use at other

locations in the body, may have taken place. As was explained previously, a part of the endogenous protein derived from increased cellular slough-off may escape normal digestion (i.e. by digestive enzymes) prior to the end of the ileum. This would result in higher metabolic ileal amino acid levels than those that would be obtained during the feeding of balanced diets. The higher metabolic ileal amino acid levels in turn may give rise to higher metabolic faecal levels.

Protein-free diets are usually employed to determine the metabolic faecal amino acid and nitrogen losses. In most experiments in which protein-free diets are used, faecal collections are carried out after 5 to 10 days i.e. before feed consumption becomes too low. The determination of metabolic faecal amino acids and nitrogen during this time may well have been the measurement of the undigestible protein components derived from the increased loss of intestinal cells. Due to the extensive modification of metabolic ileal amino acid levels by the flora in the large intestine, this measurement will be more evident when determined at the end of the ileum than when determined by faecal collection.

The epithelial cells of the small intestine may have been shedded at an increased rate, initially during the feeding of protein-free diets. The amino acid needs of these cells could be directly dependent on the supply of amino acids produced during digestion. This supply would be very low during

the feeding of protein-free diets and thus result in increased shedding of these cells.

The reduction in cell number of the small intestine can be attributed to decreased rate of cell formation and/or increased rate of cell loss. The increased rate of cell loss would likely be the first response of the small intestine to protein deficiency i.e. a short term control mechanism. Decreased rate of cell formation and cell loss will probably become more prominent as protein deficiency continues for longer periods of time. The latter mechanism represents a long term control mechanism. Hopper et al. (1968) and Brown et al. (1963) observed slower cell renewal of the intestinal cells and reduced migration of cells from crypts to the tip of the villi during advanced stages of protein deficiency.

The previous interpretation of events taking place in the small intestine upon feeding of low protein or protein-free diets would be of use to the animal only if the reduction in cell number is not accompanied by reduced absorption of endogenous or exogenous amino acids. Release of the individual amino acids by the proteolytic enzymes of the digestive tract rather than absorption of amino acids from the small intestine of the pig was found to be rate limiting in the utilization of protein at the intestinal level (Zebrowska, 1973 a,b). Everted sacs of the small intestine from protein depleted animals were found to transport amino acids more rapidly into

the sac than from well nourished rats (Herskovic, 1969; Kirsch et al., 1968). Therefore, the increase in absorptive capacity of the reduced number of cells in the small intestine during protein deprivation may partially compensate for the reduction in absorption due to cellular loss. In addition, the intake of protein-free diets by the test animals is usually much lower than that of more balanced diets and less epithelial cells will be necessary for absorption of the digestive end-products.

Snook (1965) studied the effect of the feeding of protein-free diets on the content of proteolytic enzymes in the pancreas of the rat. Compared to casein, a protein-free diet depressed the pancreatic content of chymotrypsinogen and trypsinogen. In addition, digestive enzymes were found to be less rapidly inactivated (by autodigestion) in the small intestine when diets containing protein were fed (Snook and Meyer, 1964 a and b). Peptides of dietary origin seemed to be responsible for the decrease in autodigestion. Consequently, the feeding of protein-free diets will result in reduced enzyme secretion from the pancreas and in faster inactivation of enzymes secreted into the intestinal lumen. The lower enzyme activity, in turn, may result in a reduction in digestion of endogenous protein that is derived from shedded mucosal cells.

The work by Snook (1965) shows that enzyme secretions are reduced when protein-free diets are fed. Therefore, one

would expect underestimation of metabolic ileal and faecal amino acid levels when determined by the use of protein-free diets. This seems to be in contradiction to statements made earlier that protein-free diets overestimate metabolic ileal and faecal amino acid levels. However, one should keep in mind that protein derived from digestive secretions make up only a small proportion of the total endogeneous protein (Table 50). In addition, one may expect the digestibility of enzyme protein (by autodigestion) to be higher than that of protein from shedded mucosal cells. The enzymes secreted by the digestive tract are "simply" folded peptides, without undigestible protein components that are associated with mucus protein or cellular membrane material from shedded epithelial cells.

In addition to the level of fiber in the diet, the type of fiber was found to affect the metabolic faecal nitrogen excretion in pigs (Whiting and Bezeau, 1957b). As the dietary level of pure wood cellulose (Solka-Floc) was increased from 5 to 10 to 15%, the level of metabolic faecal nitrogen increased from .10 to .14 to .15 grams per 100 grams of dry matter intake. Raising the dietary level of methocel in protein-free diets from 7 to 14% increased the metabolic faecal nitrogen excretion from 0.06 to 0.07 grams. Protein-free diets containing 14 and 28% oat hulls resulted in metabolic faecal nitrogen excretions of .09 and .14 grams per 100 grams of dry matter intake

respectively. The actual crude fiber contents of solka-floc, methocel and oat hulls were approximately 75, 90 and 50% respectively. The differences in metabolic faecal nitrogen excretion between the three fiber sources were still very evident when these were corrected for equal crude fiber content.

Alphafloc was the source of fiber used in these studies for the determination of metabolic ileal and faecal amino acid levels. The composition of crude fiber (or that of acid and neutral-detergent fiber which more closely relate to the absolute amount of undigestible dietary dry matter content) of Alphafloc may be quite different from that of crude fiber of other sources, such as from cereal grains. The differences in composition would bring about different physical characteristics which would result in different physiological and/or nutritional responses by the test animals upon ingestion of the diets. Crude fiber is composed of cellulose, lignin and other polysaccharides in variable amounts depending on the source of crude fiber. There were large differences between the components of crude fiber in ability to retain water and to form gels. Cellulose was least able to retain water (Eastwood, 1973).

As was explained previously, the test diet will be diluted by digestive juices during its passage through the stomach, duodenum and upper jejunum. In addition, water will move

from the epithelial cells into the lumen. From the upper jejunum on to the end of the ileum (and to the anus), water will be removed by absorption. The degree to which water will be absorbed may depend on the type of undigested dietary components remaining in the end of the small intestine i.e. the types of undigestible components in relation to their water holding capacities. As the water holding capacity of the remaining digesta decreases, presumably the extent of physical abrasion of epithelial cells by the digesta will increase and vice versa. In addition (when the water holding capacity is low), the physical pressure of digesta on the intestinal wall will be increased and this would result in increased peristaltic activity. The latter, in turn, will cause more friction between the digesta and the epithelial cell wall, thereby increasing the physical abrasion of cells. The relationship between mucus secretion and peristaltic activity is not known. However, it is reasonable to assume that digesta that is difficult to move needs more mucus for lubrication purposes so that passage is eased.

Ileal digesta from the cereal diets seemed to be of a much more "liquid" nature than digesta from the protein-free and maintenance diets. Unfortunately, dry matter contents were not measured. The ileal digesta from the cereal grains flowed with ease into the collection bags during sampling. Ileal digesta from the protein-free and maintenance diets were

of a thick, viscous and sticky nature. The small intestine seemed to have difficulty in moving the ileal digesta from these diets into the collection bags. Flockage occurred at times during collection of ileal digesta from these diets and flow had to be assisted sometimes by gently flushing water into the ileal cannulae.

In summary, protein-free diets of a composition similar to the ones used in these studies, may conceivably produce changes in the nutritional and physiological mechanisms operating in the small intestine, compared with those taking place when natural diets such as cereal grains are fed. As a result, metabolic ileal amino acid and nitrogen levels and therefore also faecal levels may have been overestimated.

Overestimation of metabolic losses were postulated to be due to increased cellular losses of epithelial cells and/or increased mucus secretions. On the other hand, a decrease in the digestion and reabsorption of endogenous protein secreted into the lumen of the small intestine may be held responsible. The latter aspect is discussed in more detail, in relation to rate of passage, in the discussion part of study 4.

The use of protein-free diets has its importance in nutritional research. First of all, it may be of importance to identify the undigestible protein fractions from a feed as



such and to determine factors affecting the metabolic losses from the animal. Secondly, protein-free diets are used to determine the maintenance losses of nitrogen (urinary plus faecal plus cutaneous nitrogen losses). The latter is important for the assessment of protein needs for maintaining animals or adults (Munro, 1969) and for the differentiation between protein requirements for growth (and/or production) and those needed for maintenance. Depending on the type of protein-free diet used, maintenance losses of nitrogen derived from faeces may actually be overestimated.

Certainly, knowledge of metabolic ileal and faecal amino acid levels (and generally speaking those of other dietary ingredients) is of importance. However, to say the least, the use of protein-free diets for arriving at these estimates should be reevaluated. Future studies i.e. by aid of isotopes, may be warranted to study this topic.

On a practical basis, i.e. for formulation of dietary amino acid levels in relation to requirements for monogastric animals, it is not important to have separate knowledge of the amounts of undigested and/or unabsorbed dietary and metabolic amino acids. Both sources of amino acids (protein) are lost to the animal. The determination of apparent amino acid availabilities will be sufficient.

In the previous discussion most attention was paid to the estimation of true ileal amino acid availabilities.

True faecal availabilities were not discussed. Interpretation of calculated true faecal amino acid availabilities would be more misleading since an additional error comes into play, namely that certain amino acids are synthesized by the flora in the large intestine upon feeding of protein-free diets.

## STUDY 4.

The results obtained from study 4b will be discussed prior to those of study 4a.

The ileal nitrogen and dry matter digestibilities and the ileal availabilities of all amino acids were higher for finely ground than for cracked wheat (Table 51). For most amino acids, these increases were significant. The increased availabilities were due to 1) the absence of cracked or broken feed particles that would have escaped physical digestive breakdown in case cracked wheat was fed and 2) fine grinding may have resulted in an increased release of aleurone protein from the aleurone cells and more of this protein may have been available for digestion.

The first reason for increased availabilities of amino acids from finely ground over cracked wheat is shown in Table 36. The dietary particle size distribution of finely ground and cracked wheat is reflected in the particle size distribution of ileal digesta. Only 5% of the particles in ileal digesta from finely ground wheat were larger than 1.00 mm. However, approximately 25% of the particles in ileal digesta from cracked wheat were larger than 1.00 mm. (Table 36).

The largest percentage increases in ileal availabilities due to fine grinding were found for LYS, GLY and ALA and were 8.9, 7.6 and 7.3% respectively. The smallest increases were found for PHE, GLU and PRO and were 3.5, 1.2 and 0.7% respectively

TABLE 51. The apparent ileal and faecal amino acid availabilities from ground and cracked wheat in addition to the digestibilities of amino acids in the large intestine and the differences in percentage units between ileal and faecal amino acid availabilities

TYPE OF PRE-PROCESSING	GROUND				CRACKED				C <sup>3</sup>
LOCATION	ILEUM	FAECES	A <sup>1</sup>	B <sup>2</sup>	ILEUM	FAECES	A <sup>1</sup>	B <sup>2</sup>	
AMINO ACIDS (%)									
<u>Essential</u>									
ARG	92.0	94.4	30.0	2.2	86.5	93.9	54.8	7.4	5.5
HIS	91.1	94.4	37.1	3.3	85.7	93.9	57.3	8.2	5.4
ILE	90.2	91.2	10.2	1.0	85.6	89.9	29.9	4.3	4.2
LEU	91.5	92.9	16.5	1.4	87.6	92.1	36.3	4.5	3.9
LYS	81.5	85.3	20.5	3.8	72.6	83.8	40.9	11.2	8.9
PHE	92.9	93.5	8.5	0.6	89.4	92.9	33.0	3.5	3.5
THR	83.7	88.7	30.7	5.0	78.0	87.3	42.3	9.3	5.7
VAL	88.6	91.3	23.7	2.7	83.1	90.5	43.8	7.4	5.5
<u>Non-Essential</u>									
ALA	82.1	87.1	27.9	5.0	74.5	86.5	47.1	12.0	7.6
ASP	82.8	87.1	25.0	4.3	77.3	86.2	39.2	8.9	5.5
GLU	95.3	97.8	53.2	2.5	94.1	97.6	59.3	3.5	1.2
GLY	81.7	90.1	45.9	8.4	74.4	89.8	60.2	15.4	7.3
PRO	84.5	95.9	73.5	11.4	83.8	96.3	77.2	12.5	0.7
SER	88.2	93.3	43.2	5.1	84.7	90.3	36.6	5.6	3.5
TYR	89.8	90.7	8.8	0.9	85.3	90.6	36.1	5.3	4.5
NITROGEN (%)	88.4	93.4	51.7	5.0	84.7	92.3	49.7	7.6	3.7
DRY MATTER (%)	80.4	88.8	42.9	8.4	76.8	88.8	51.7	12.0	3.6

- 1 Digestibilities of amino acids, nitrogen and dry matter in the large intestine.  
 2 Differences in percentage units between ileal and faecal amino acid availabilities.  
 3 Differences between ileal amino acid availabilities from finely ground and cracked wheat.

(Table 51). Lysine, GLY and ALA are relatively high in aleurone protein. Phenylalanine, GLU and PRO are relatively low in aleurone protein (Table 2 and 33). Extrapolations as to PRO and GLY with regard to increased availability of aleurone protein for digestion due to grinding should be considered carefully since these 2 amino acids are found in relatively high amounts in metabolic ileal protein.

The faecal amino acid availabilities were only slightly and non-significantly higher for finely ground than for cracked wheat (Table 51). Therefore, it is very difficult and unreliable to measure differences due to "pre-processing" by simply evaluating faecal digestibility measurements. Additional measurements, such as urinary nitrogen losses or rate of gain, should be carried out concomitantly. The latter finding is in agreement with data that indicate that availability values determined by faecal analysis are generally higher than those determined by growth methods (de Muelenaere et al., 1967; Nesheim and Carpenter, 1967; Payne et al., 1968). The ileal and faecal availabilities of LYS from ground wheat differed only to a small extent (81.5 and 85.3%). In this case it would have made little difference if LYS availability had been determined by the growth or the faecal analysis method (Table 51).

The net disappearance of amino acids in the large intestine was higher for cracked than for finely ground wheat (Table 51). A higher level of undigested starch from cracked

wheat entering the large intestine may have allowed for a higher level of microbial fermentation and amino acid disappearance. On the other hand, simply more nutrients in general may have been available for microbial fermentation.

The results obtained from finely ground and cracked wheat of study 4b are compared with those of ground wheat from study 4a and with those of cracked wheat from study 2a (Table 52). Even though different varieties of wheat were tested, the ileal amino acid availabilities of cracked wheat from study 2a and 4b compare very well. The ileal availabilities of amino acids from ground wheat in study 4a were between those obtained from finely ground and cracked wheat from study 4b but were closer to those of finely ground wheat. The comparisons of results from studies 2a and 4 show the reproducibility of data as they relate to the type of "pre-processing" of wheat and to the methods for determining ileal amino acid availabilities. Moreover, the differences between the ileal amino acid availabilities from corn, wheat and barley in study 2a were not only due to the type of cereal grain tested but also to the manner of "pre-processing" to which each cereal grain was subjected.

The ileal amino acid availabilities decreased from flour to whole wheat to E+S+M (Table 53). The LYS availabilities, in particular, decreased from 84.2 to 79.5 to 66.4.

Lysine availabilities from flour, determined either by

TABLE 52. Comparisons of ileal and faecal amino acid availabilities from wheat from studies 2a and 4 as affected by the type of pre-processing.

LOCATION	ILEUM				FAECES			
TYPE OF PRE-PROCESSING	CRACKED		GROUND		CRACKED		GROUND	
STUDY	4b	2a	4b	4a	4b	2a	4b	4a
AMINO ACIDS (%)								
<u>Essential</u>								
ARG	86.5	85.8	92.0	87.1	93.9	92.7	94.4	94.6
HIS	85.7	89.1	91.1	88.4	93.9	94.9	94.4	93.9
ILE	85.6	85.3	90.2	89.1	89.9	89.4	91.2	91.6
LEU	87.6	86.9	91.5	89.9	92.1	91.5	92.9	93.2
LYS	72.6	75.7	81.5	79.5	83.8	80.7	85.3	86.1
PHE	89.4	88.8	92.9	91.5	92.9	92.5	93.5	94.3
THR	78.0	76.5	83.7	78.4	87.3	86.7	88.7	89.1
VAL	83.1	82.8	88.6	86.7	90.5	88.9	91.3	91.7
<u>Non-Essential</u>								
ALA	74.5	74.0	82.1	79.6	86.5	84.0	87.1	88.1
ASP	77.3	75.4	83.8	80.8	86.2	83.1	87.1	88.0
GLU	94.1	92.7	95.3	95.6	97.6	97.0	97.8	97.9
GLY	74.4	73.7	81.7	72.6	89.8	89.3	90.1	90.6
PRO	83.8	86.8	84.5	79.1	96.3	96.7	95.9	96.8
SER	84.7	84.1	88.2	86.3	90.3	92.5	93.3	94.3
TYR	85.3	85.9	89.8	89.2	90.6	89.5	90.7	92.9
NITROGEN (%)	84.7	82.9	88.4	85.2	92.3	91.2	93.4	93.3
DRY MATTER (%)	76.8	73.3	80.4	78.2	88.8	89.0	88.8	89.4

TABLE 53. Apparent ileal and faecal amino acid availabilities, in addition to nitrogen and dry matter digestibilities and the disappearance of these substances in the large intestine.

DIET	WHOLE WHEAT				FLOUR				E+S+M <sup>1</sup>				D+S+M (D)			
	ILEUM	FAECES	L.I. <sup>1</sup>	$\Delta^2$	ILEUM	FAECES	L.I. <sup>1</sup>	$\Delta^2$	ILEUM	FAECES	L.I. <sup>1</sup>	$\Delta^2$	ILEUM	FAECES	L.I. <sup>1</sup>	$\Delta^2$
AMINO ACIDS (%)																
<u>Essential</u>																
ARG	87.1	94.6	56.6	-7.5	90.7	95.6	52.7	-4.9	84.8	90.0	34.2	-5.2	71.5	90.2	65.6	-18.7
HIS	88.4	93.9	47.4	-5.5	93.9	96.6	44.3	-2.7	78.5	88.8	47.9	-10.3	73.6	88.0	54.5	-14.4
ILE	89.1	91.6	22.9	-2.5	93.9	94.7	13.1	-0.8	72.9	74.6	6.3	-1.7	69.6	75.5	19.4	-5.9
LEU	89.9	93.2	32.7	-3.3	94.6	95.5	16.7	-0.9	74.4	78.7	16.8	-4.3	71.7	80.7	30.4	-9.0
LYS <sub>3</sub>	79.5	86.1	32.2	-6.6	84.2	86.0	11.4	-1.8	66.4	75.5	27.1	-9.1	57.5	77.6	47.3	-20.1
MET	92.4	93.4	13.2	-1.0	93.7	93.4	105.8	+0.3	77.8	81.8	18.0	-4.0	76.5	73.2	114.0	+3.3
PHE	91.5	94.3	32.9	-2.8	95.5	96.3	17.8	-0.8	76.0	79.5	14.6	-3.5	73.9	81.5	29.1	-7.6
THR	78.4	89.1	49.5	-10.7	85.4	92.3	47.3	-6.9	54.0	71.3	37.7	-17.3	47.8	74.7	51.5	-26.9
VAL	86.7	91.7	37.6	-5.0	92.7	94.3	21.9	-1.6	71.3	76.0	15.2	-4.7	66.5	78.2	34.9	-11.7
<u>Non-Essential</u>																
ALA	79.6	88.1	41.7	-8.5	86.1	90.8	33.8	-4.7	70.2	75.5	17.8	-5.3	60.7	76.2	39.9	-15.5
ASP	80.8	88.0	37.5	-7.2	85.5	89.2	25.5	-3.7	69.8	75.6	19.2	-5.8	64.2	78.2	39.1	-14.0
GLU	95.6	97.9	52.3	-2.3	97.9	98.6	33.3	-0.7	85.8	89.9	28.9	-4.1	83.8	90.9	43.2	-7.1
GLY	72.6	90.6	65.7	-18.0	78.5	93.6	70.2	-15.1	57.3	78.3	49.2	-21.0	31.9	79.3	69.6	-47.4
PRO	79.1	96.8	84.7	-17.7	83.0	98.5	91.2	-15.5	70.3	89.5	64.6	-19.2	-47.5	78.4	85.4	-125.9
SER	86.3	94.3	58.4	-8.0	91.7	95.8	49.9	-4.1	72.2	83.1	39.2	-10.9	67.7	84.2	51.1	-16.5
TYR	89.2	92.9	34.3	-3.7	93.1	94.4	18.8	-1.3	71.1	78.8	26.6	-7.7	68.5	78.6	32.1	-10.1
NITROGEN (%)	85.2	93.3	54.7	-8.1	90.5	96.4	62.1	-5.9	69.8	80.9	36.8	-11.1	59.2	83.0	58.3	-23.8
DRY MATTER (%)	78.2	89.4	51.4	-11.2	90.2	95.0	49.0	-4.8	43.3	65.0	38.3	-21.7	69.2	83.9	47.7	-14.7

<sup>1</sup> Digestibilities of amino acids, nitrogen and dry matter in the large intestine.

<sup>2</sup> Differences between ileal and faecal estimates.

<sup>3</sup> Determined by acid hydrolysis.



ileal or faecal collection, were very similar. Calhoun et al (1960), using rats, found the LYS availabilities from flour to be 84.5% and 82.0% using weight and carcass nitrogen gain respectively as the method for measurement. Their estimates compare very well to those found in these experiments, irrespective of measurement by ileal or faecal collection.

The relatively low LYS availability of flour (Table 53) eliminates the notion that LYS availability from cereal grains is solely determined by the amount of LYS present or associated with aleurone cells. The relatively low availability of LYS from flour in relation to the availabilities of the other amino acids may possibly be due to the large number of lysyl-prolyl peptide linkages found in flour. These peptide linkages are completely resistant to trypsin digestion (page 14 and 15).

Generally, the disappearance of amino acids in the large intestine increased from flour to whole wheat to E+S+M (Table 53). In other words, the lower the availabilities of amino acids at the end of the ileum, the more the faecal availability estimates will deviate from the ileal availabilities. With the exception of THR, the faecal availabilities of the essential amino acids may still reflect the ileal availabilities on a qualitative basis (i.e. the order of least availability).

As was discussed previously, the ileal availabilities from whole wheat were dependent on the type of "pre-processing". Whole wheat was ground through a 2.00 mm screen in this study. If the

wheat had been cracked then larger differences in amino acid availabilities between those of whole wheat and flour may well have been observed. If the whole wheat had been finely ground (as in study 4b), the availabilities obtained might have approximated those of flour (Table 51 and 53). The ileal availabilities of flour are unlikely to be improved by grinding and therefore represent maximum values. Flour, as obtained, was already of a very powdery consistency. The E+S+M diet was not ground prior to pelleting. Consequently, there might be an improvement in amino acid availabilities from E+S+M over those obtained in this study (Table 53).

Whole wheat was milled to give 75% flour and 25% E+S+M. Therefore, the total amounts of the individual amino acids from flour ( $\times 0.75$ ) plus those of E+S+M ( $\times 0.25$ ) should be equal to the total amount of the individual amino acids contained in whole wheat ( $\times 1.0$ ). The recovery of the separate amino acids from flour plus E+S+M as a percentage of those in whole wheat ranged from 94.0 to 104.0% (Table 54).

The level of each amino acid found in ileal digesta derived from 100 grams of whole wheat should be equal to that derived from 75 grams of flour plus that from 25 grams of E+S+M. The latter would be the case if no associative or other unspecified effects occur.

The ileal levels of amino acids derived from 100 grams of whole wheat can be calculated from the individual amino acid

TABLE 54. The recovery<sup>1</sup> of amino acids from B+S+M and flour (column A), in addition to the ileal amino acid availabilities from B+S+M that were measured directly (column B) and those of B+S+M that were calculated by difference from cracked wheat and flour (column C) and from finely ground wheat and flour (column D).

COLUMN	A	B	C	D
AMINO ACIDS (%)				
<u>Essential</u>				
ARG	96.0	84.8	80.7	90.2
HIS	104.0	78.5	68.4	85.0
ILE	103.0	72.9	60.7	79.1
LEU	102.0	74.4	67.4	82.6
LYS	96.0	66.4	57.7	77.8
PHE	102.0	77.8	68.6	84.0
THR	98.0	54.0	71.7	90.7
VAL	100.0	71.3	60.6	79.0
<u>Non-Essential</u>				
ALA	102.0	70.2	56.2	75.4
ASP	95.0	69.8	65.3	79.0
GLU	102.0	85.8	79.3	86.2
GLY	97.0	57.3	68.2	87.8
PRO	102.0	70.3	88.0	92.3
SER	103.0	72.2	66.7	79.3
TYR	94.0	71.1	69.8	86.4

<sup>1</sup> The recovery of the individual amino acids from flour plus B+S+M as a percentage of those in whole wheat.

availabilities and the dietary levels. The ileal levels of amino acids derived from 75 grams of flour can be calculated in the same manner. The ileal excretion levels from 25 grams of B+S+M are then calculated by difference from those derived from 100 grams of whole wheat and 75 grams of flour. As the dietary level of each amino acid in B+S+M is known, the availability of each amino acid can be calculated. These calculations were carried out using ileal amino acid availabilities from finely ground and cracked wheat (Table 51) in order to arrive at maximum and minimum possible amino acid availability estimates respectively from B+S+M (Table 54).

The minimum possible essential ileal amino acid availabilities from B+S+M ranged from 57.7 for LYS to 80.7% for ARG. The maximum availabilities ranged from 77.8 for LYS to 90.7% for THR (Table 54). Therefore, the potential availabilities of amino acids from B+S+M may approximate those of whole wheat (Table 53).

With the exception of THR, GLY and PRO, the directly determined amino acid availabilities for B+S+M were found to be between the minimum and maximum calculated availabilities from B+S+M. Threonine, GLY and PRO are present in relative large amounts in metabolic protein, even at corrected levels (Table 47, 48 and 49). The directly determined availabilities of these three amino acids were all lower than the calculated estimates. Fiber level as such has been shown to affect the

metabolic ileal amino acid levels (Table 45). The level of crude fiber of E+S+M (8.49%) was much higher than that of whole wheat (2.35%) or flour (0.25%). The calculations for the availabilities of E+S+M were based on differences between ileal amino acid levels from whole wheat and flour and may have resulted in overestimation of ileal availabilities of THR, GLY and PRO.

The calculated availabilities of the other amino acids were in the range of those found directly. This latter would only occur if the dilution of undigested ileal dietary amino acids by the endogenous amino acids (with the exception of THR, GLY and PRO) was of a relatively small proportion.

In order to obtain more data on the availabilities of amino acids from E+S+M (since only 3 sets of results were available), the E+S+M diet was diluted with 60% cornstarch to give the E+S+M-D diet. The ileal availabilities of all amino acids from E+S+M-D were lower than those of E+S+M (Table 53). The availabilities of ARG, GLY and PRO in particular were much lower. These amino acids are also very prominent in metabolic ileal protein (Table 45).

The higher intake of the E+S+M-D diet (1649 g/day) than of the E+S+M diet (833 g/day) by the pigs could have been partially responsible for the lower ileal amino acid availabilities obtained from E+S+M-D. Dietary and metabolic protein will be retained for a longer period of time in the small intestine when

food intake is low than when food intake is high. Therefore, when intake is low, more time will be permitted for digestion and absorption of protein and other nutrients.

Differences in ileal amino acid availabilities and nitrogen digestibilities should be detected if there were relatively large variations in dietary intake by pigs fed the same diets. The dietary intake of the pigs fed B+S+M-D varied from a minimum of 1386 g/day for pig no. 1 to a maximum of 1848 for pig no. 4. The ileal nitrogen digestibilities were 62.9 and 48.9% for pig no. 1 and 4 respectively. Generally speaking, ileal amino acid availabilities were also higher for pig no. 1. The differences in ileal amino acid availabilities and nitrogen digestibilities between pig no. 1 and 4 were not reflected by data obtained from faecal analyses (Appendix, Table 22). The "evidence" presented may be somewhat circumstantial but can be substantiated by interpretation of comparative data from Easter (1972), Holmes et al., (1974), Zébrowska (1973 a, b) and those obtained from this thesis.

Holmes et al. (1974) fed a semi-purified diet containing 31% SBM to 45 kg pigs at a rate of 22 g per kg body weight. Zébrowska (1973 a, b) fed a semi-purified diet containing 35% SBM to 60 kg pigs, at a rate of 30 g per kg body weight per day. The average ileal amino acid availability obtained by Holmes et al. (1974) was 10.8 percentage units higher than that obtained by Zébrowska (1973). The differences in percentage units for the

individual amino acid availabilities ranged from 1.8 for PRO to 21.0% for HIS.

Easter (1973) fed corn to 55 kg pigs, at a rate of 45 g per kg body weight. In the present studies, corn was fed to the pigs at an approximate rate of 27 g per kg body weight. The average ileal amino acid availabilities obtained in the present studies were 15.6 percentage units higher than those obtained by Easter (1972). The differences in percentage units for the individual amino acid availabilities between the two studies ranged from 10.2 for TYR to 28.2% for GLY (Table 41). The average faecal amino acid availability obtained by Easter (1973) was only 4.1% lower than that obtained in these studies.

The ileal amino acid availabilities obtained for barley in the present studies were very similar to those obtained by Zebrowska (1973 a, b) (Table 41). Barley was fed to the pigs at almost the same rate in both studies, namely at 27 and 30 g per kg body weight in these studies and those by Zebrowska respectively.

From the data discussed it seems that dietary intake (in relation to body weight) may also be a factor in determining amino acid availabilities at the end of the ileum. Differences due to intake may not always be detected, or only to a lesser extent, when amino acid availabilities are determined by the faecal analysis method. The latter postulation would indicate that there is relatively more wastage of protein of

dietary and/or metabolic origin (caused by microbial fermentation) in the large intestine at higher than at lower food intakes. Therefore, in certain cases, the large intestine in addition to the liver may also contribute to the deamination of excess protein i.e. when protein intake exceeds the amount required. The question may be asked now, why does the animal not excrete the excess protein as such in the faeces? The by-products of protein metabolism in the large intestine, which are mainly short chain volatile fatty acids, may still be of use to the animal. These compounds may be used as precursors for glucose or for fat synthesis. In addition, microbial fermentation in the large intestine will make volatile fatty acids, produced from undigestible carbohydrate sources, available to the animal.

The ileal availabilities of amino acids which are relatively high in metabolic ileal protein (ARG, GLY and PRO) were especially low from E+S+M-D in relation to those obtained from E+S+M (Table 53). A change in the rate of passage would particularly change the availabilities of amino acids from protein that is already of a relatively low digestibility, such as protein from sloughed-off cells or from mucus.

On the other hand, cornstarch as such may have been responsible for the high levels of metabolic ileal protein found at the end of the ileum upon feeding of the E+S+M-D diet. As for the maintenance and protein-free diets, ileal PRO and



GLY levels were also found to be relatively high in ileal digesta from E+S+M-D (Table 53). It may or may not be coincidental but all these diets contained 60% cornstarch.

Casein was fed (24.3 mg. nitrogen per 100 g body weight) with and without different carbohydrates (167 mg per 100 g body weight) to rats (Buraczewski et al., 1971). The rate of stomach emptying of casein was decreased when corn dextrin, glucose, soluble starch, sucrose or lactose (in ascending order) were the carbohydrate sources. However, the rate of stomach emptying of casein was increased when cornstarch was the added carbohydrate source. The authors related the differences in rate of stomach emptying of casein to differences in solubility and rate of absorption of the carbohydrates. Possibly, the increased rate of stomach emptying of casein due to cornstarch inclusion (which is very soluble and rapidly absorbed) may have resulted also in an increase in rate of passage of digesta through the small intestine. As was mentioned previously, increased rate of passage of digesta through the small intestine may especially decrease the digestion and reabsorption of endogenous protein.

Differential losses of amino acids in the large intestine (due to microbial fermentation) caused by different types of carbohydrates may have some effect on the nutritional status of the animal if the diets fed were low in protein or deficient in a particular amino acid. The P.E.R.'s (protein efficiency

ratio's) were  $0.83^1$ ,  $0.90^1$  and  $0.93^1$  when diets containing 24% wheat gluten and either 60% cornstarch, dextrin or glucose respectively were fed to rats. In the same order, the P.E.R.'s were  $1.61^2$ ,  $1.58^2$  and  $1.54^2$  when the diets were supplemented with LYS (Chang, 1962).

Although there may be other explanations, the effect of cornstarch (in relation to possible increased rate of passage through the small intestine) may explain the lower P.E.R. found when cornstarch rather than dextrin or glucose was included in the non-LYS-supplemented wheat gluten diets.

It may have been coincidental but true ileal amino acid availabilities from E+S+M-D could be obtained in such a manner that true PRO and GLY availabilities obtained did not exceed 100%. The availabilities of these amino acids were close to those of the other amino acids (Table 55). Corrections for metabolic ileal amino acid levels were based on equal crude fiber levels per 100 grams of dry matter intake for the E+S+M-D and protein-free diets. The crude fiber level of E+S+M-D (3.88%) was very close to that of PF-5 (3.86%). Therefore metabolic ileal amino acid data from PF-5 were used. Generally speaking,

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<sup>1</sup>P.E.R. of the glucose containing diet was significantly higher than that of the cornstarch containing diet.

<sup>2</sup>P.E.R.'s were not significantly different.

TABLE 55. The apparent ileal amino acid availabilities<sup>1</sup> from B+S+M-D and B+S+M, in addition to the estimated true availabilities from B+S+M-D.

DIET	B+S+M-D		E+S+M
AVAILABILITIES	APPARENT	TRUE	APPARENT
AMINO ACIDS (%)			
<u>Essential</u>			
ARG	71.5 $\pm$ 6.7	82.4	84.8 $\pm$ 1.5
HIS	73.6 $\pm$ 3.1	80.9	78.5 $\pm$ 2.6
ILE	69.6 $\pm$ 1.6	78.2	72.9 $\pm$ 2.3
LEU	71.7 $\pm$ 1.7	80.3	74.4 $\pm$ 3.0
LYS	57.5 $\pm$ 3.9	69.1	66.4 $\pm$ 4.8
MET	76.5 $\pm$ 6.0	81.6	77.8 $\pm$ 3.8
PHE	73.9 $\pm$ 1.3	81.7	76.0 $\pm$ 2.5
THR	47.8 $\pm$ 3.9	67.2	53.9 $\pm$ 6.5
VAL	66.5 $\pm$ 2.6	75.5	71.3 $\pm$ 2.7
<u>Non-Essential</u>			
ALA	60.7 $\pm$ 5.9	72.3	70.2 $\pm$ 0.7
ASP	64.2 $\pm$ 3.3	75.4	69.8 $\pm$ 1.2
GLU	83.8 $\pm$ 1.6	88.7	85.8 $\pm$ 1.2
GLY	31.9 $\pm$ 21.0	69.2	57.3 $\pm$ 8.9
PRO	-47.5 $\pm$ 66.5	62.3	70.3 $\pm$ 11.0
SER	67.7 $\pm$ 2.8	80.5	72.2 $\pm$ 2.1
TYR	68.5 $\pm$ 2.6	77.2	71.1 $\pm$ 3.0

<sup>1</sup> Means and standard deviation.

the true ileal amino acid availabilities obtained from B+S+M-D were very close to the apparent availabilities from B+S+M fed as such. Indirectly, the latter indicates again that metabolic ileal amino acid levels of natural diets (such as B+S+M and cereal grains) may be overestimated by the use of protein-free diets.

The reason that "true" ileal amino acid availabilities from diet B+S+M-D could be obtained may have been due to the similarities between the B+S+M-D and protein-free diets with respect to certain nutrient(s). Both diets contained 60% cornstarch. The reason that no precise "true" ileal amino acid availabilities (or associated metabolic ileal amino acid levels) could be obtained from the cereal grains is probably due to the fact that there was much less similarity between the cereal and protein-free diets. Because of the differences between these diets, different responses with regard to the amounts of endogenous protein secreted and/or digested and reabsorbed in the small intestine were elicited.

Accurate determination of "true" ileal availabilities is best if the test and protein-free diets resemble each other. For example, in determining true amino acid availabilities of a protein source, the composition of semi-purified diet containing this protein source (i.e. at a level of 10%) can be formulated in identical manner (with the exception of the protein itself) to that of the protein-free diet. However,

in determining true amino acid availabilities of an energy-protein source (e.g. barley), the situation is different. A natural type diet will differ in a number of characteristics from a protein-free diet.

## STUDY 3

The apparent availabilities of amino acids from barley, corn and wheat (Table 40) and those of whole wheat, flour and B+S+M (Table 53) were found to be higher when they were determined by faecal analysis than when they were determined by ileal analysis. The differences between ileal and faecal availabilities were found to be dependent on the type of diet and on the "pre-processing" conditions these diets were subjected to prior to feeding. Differences between ileal and faecal estimates were more pronounced for some than for other amino acids.

The apparent faecal amino acid availabilities from sorghum grain and corn were higher than those obtained at the end of the ileum from pigs fitted with ileocaecal re-entrant cannulae (Easter, 1972). Generally speaking, apparent amino acid availabilities of SBM and RSM were higher when they were determined from faecal collection than when they were determined at the end of the ileum of pigs fitted with ileal re-entrant cannulae (Holmes et al., 1974). The average amino acid availabilities based on ileal samples from chicks that were sacrificed were lower than those determined by faecal analysis (Ivy et al., 1971; Filipot et al., 1971; Soares and Kifer, 1971). Caecectomy of chicks significantly reduced the protein digestibility of heat-damaged cod protein (Nesheim and Carpenter, 1967).

The amino acid availabilities determined by the faecal analysis method may overestimate the amounts of amino acids that

are actually absorbed by the animal. Bacterial enzymes in the large intestine bring about the hydrolysis of undigested protein through peptides of decreasing length to free amino acids. These free amino acids may be either absorbed as such by the animal or they may be fermented further by the micro-organisms to yield ammonia and carbon skeletons. The rate of deaminative fermentation varied from amino acid to amino acid (Michel, 1966). The extent to which amino acid absorption versus deamination takes place in the large intestine will determine which method is more valid i.e. the faecal or ileal analysis method.

These experiments were carried out to determine the extent to which LYS was absorbed as such in the large intestine. Lysine was either caecally infused as a part of the ISP protein complex (study 3a) or as free LYS monohydrochloride (study 3b). Lysine was selected since it is often the first limiting amino acid to pigs.

The B+2ISP diet is representative of a barley-soybean pig grower ration. Reducing the level of soybean meal by half in the grower ration will result in a decreased rate of gain by the pigs. The decreased rate of gain can be largely overcome by aid of LYS supplementation (Bell, 1964; Bowland, 1962; Braude et al., 1972; Neilson et al., 1963).

The levels of LYS that were found in the caecal contents after infusion of ISP or free LYS would be much higher than those found under normal conditions. Therefore, a situation was created

whereby there was ample opportunity for substantial LYS absorption in the hindgut with a resultant improvement in nitrogen retention. It was reasoned that if only a small response in nitrogen retention took place upon LYS infusion, then LYS absorption in the large intestine was of little importance under normal circumstances.

The amounts of nitrogen retained per day were 18.00, 15.80 and 15.91 g. for B+2ISP, B+ISP and B+ISP+C-ISP respectively (Table 29). In the same order these values represent 112.5<sup>1</sup>, 98.8 and 99.4 g. of protein deposited per day. On the assumption that the average protein content of the pig's body tissue is 16%, then the average daily gains would be 703, 617 and 622 g. for pigs fed B+2ISP, B+ISP and B+ISP+C-ISP respectively. Similar calculations carried out on data obtained from study 3b will give the following daily rates of gain: 693, 600 and 613 g. for B+ISP+LYS, B+ISP and B+ISP+C-LYS respectively. Excluding the calculated missing value, an average daily gain of 607 g. would be obtained for pigs fed B+ISP+C-LYS (Table 31).

The data obtained from study 3a and 3b for B+ISP compare very well (Table 29 and 31). The amount of nitrogen retained per day from B+2ISP was only slightly higher than that obtained from B+ISP+LYS (Table 29 and 31).

Nitrogen retentions and biological values were higher

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<sup>1</sup>Nitrogen retained per day times 6.25.



for B+ISP+LYS than for B+ISP (Table 31). Lysine was the limiting amino acid in B+ISP and its addition resulted in lower amounts of the other amino acids that had to be catabolized. Nitrogen retentions and biological values for B+2ISP were lower than those obtained from B+ISP (Table 29). Since only LYS was needed for supplementation (B+ISP+LYS and B+2ISP gave nearly similar responses with regard to the amount of nitrogen retained per day), the other amino acids contained in the 1 unit of ISP added had to be catabolized.

For efficient protein synthesis, all amino acids have to be simultaneously present at the site where protein synthesis takes place. It can be argued that caecally infused LYS may arrive at a different time at the site of protein synthesis than the amino acids from orally ingested protein. Possibly, this may occur if the animals were fed once daily and the infusion had been carried out 12 hrs. later. However, in these studies, the pigs were fed 3 times daily and the infusions were performed when the animals were offered their meals.

In study 3b, LYS was mixed with cornstarch to facilitate LYS infusion into the caecum. The caecally infused cornstarch may have stimulated bacterial activity, resulting in an increase in deamination of amino acids (including caecally infused LYS). In retrospect, LYS should perhaps have been mixed with a fiber source from which energy is less available to bacteria than is the case with cornstarch.

The results obtained in these studies show that caecally supplied LYS did not improve the nutritional status of the animal. The amounts of nitrogen retained per day were nearly the same for B+ISP, B+ISP+C-ISP and B+ISP+C-LYS.

However, a small amount of LYS absorption may have taken place in the large intestine of the pig. Slade et al. (1971) showed that there was LYS absorption from the large intestine of the horse. However, the authors' did not measure the magnitude of LYS absorption (page 26 ).

In summary, LYS availability determined by the faecal analysis method overestimates the actual amount of LYS that is available for utilization by the pig. There is no reason to assume that the other amino acids undergo a different fate from LYS (in relation to deamination and excretion of ammonia as urea in the urine) in the large intestine.

## STUDY 1

The amino acid composition of bran and endosperm from high and low protein wheat from study 1 and that of bran (E+S+H) and endosperm (flour) from study 4a are compared in Table 56. Aleurone protein contains relatively high levels of ARG and LYS and relatively low levels of GLU and PRO in relation to endosperm protein (Table 2). Therefore, the efficiency of separation of aleurone and endosperm protein can be measured by the degree of difference in the composition of these amino acids between the bran and endosperm fractions.

As shown in Table 56, a better separation of bran and endosperm protein was achieved by the conventional milling process (study 4a) than by pearling (study 1) of wheat. This was also reflected by the very low crude fiber content (0.25%) of the endosperm (flour) fraction obtained in study 4a.

The separation of aleurone protein from endosperm protein was much better for the low than for the high protein wheat (Table 56). In retrospect, abrasion should have been carried out for a longer period of time for the high protein wheat, namely for 36 instead of 20 seconds (Table 5). Therefore, a substantial amount of aleurone protein may have remained in the endosperm fraction, resulting in less distinct differences in the amino acid composition of ARG, GLY, GLU and PRO between bran and endosperm protein. Consequently, interpretation of amino acid availabilities from bran and endosperm from the high

TABLE 56. The amino acid composition<sup>1</sup> and crude fiber content of bran and endosperm from study 1 and study 4a.

NUMBER OF STUDY	STUDY 1				STUDY 4a.	
FRACTION OF CEREAL GRAIN	BRAN <sup>2</sup>	ENDO- <sup>2</sup> SPERM	BRAN <sup>3</sup>	ENDO- <sup>3</sup> SPERM	BRAN <sup>4</sup>	ENDO- <sup>4</sup> SPERM
AMINO ACIDS						
<u>Essential</u>						
ARG	4.86	3.81	5.19	3.81	6.11	3.24
HIS	1.83	1.93	1.85	1.95	2.46	2.01
ILE	2.94	3.40	2.84	3.44	3.18	3.74
LEU	6.44	7.21	6.32	7.44	5.99	6.86
LYS	2.35	2.11	3.27	2.33	3.43	1.74
PHE	4.44	5.10	3.91	5.12	3.87	5.23
THR	2.86	2.40	3.13	2.98	2.64	2.44
VAL	4.01	4.16	4.20	4.47	4.49	4.20
<u>Non-Essential</u>						
ALA	4.14	3.52	4.98	3.81	4.58	2.76
ASP	5.72	4.92	6.33	5.21	6.56	3.89
CLU	30.04	38.10	21.90	35.16	18.82	35.57
GLY	4.74	4.28	5.19	4.37	4.89	3.29
PRO	9.94	12.42	7.25	12.28	5.67	11.45
SER	4.48	4.92	4.34	4.84	3.84	3.99
TYR	2.94	3.22	2.56	2.98	1.95	2.05
CRUDE FIBER (%)	5.84	1.37	6.93	1.17	8.49	0.25

<sup>1</sup> Expressed as grams per 16 grams of nitrogen.

<sup>2</sup> Bran and endosperm derived from high protein wheat.

<sup>3</sup> Bran and endosperm derived from low protein wheat.

<sup>4</sup> Bran and endosperm indicate B+S+M and flour respectively for study 4a.

protein wheat variety in relation to amino acid availabilities from aleurone and endosperm protein should be considered carefully.

The apparent faecal amino acid availabilities obtained from pigs fed wheat in study 4a and those from pigs fed wheat in study 2 are compared with those obtained from the rats, fed high and low protein wheat, in this study (Table 57).

With the exception of the availabilities obtained from low protein wheat (11.2%) fed to rats, the availabilities compare reasonably well between rats and pigs especially those obtained from pigs from study 2 and those from rats fed high protein wheat (Table 57). The differences between faecal availabilities varied only from 0.3 for SER to 2.4 percentage units for ASP between these 2 studies. It may have been a coincidental observation but the same variety of wheat, namely Glenlea, was tested. In addition, the protein contents of both Glenlea wheat sources were relatively similar (Table 57).

The differences between faecal availabilities were somewhat larger when those from rats fed high protein wheat and those obtained from the pigs from study 4a were compared. The differences ranged from 0.3 for PHE to 7.2% for LYS (Table 57). The wheat sources in this comparison, although not too different in protein content, were of a different variety.

The faecal amino acid availabilities obtained from rats fed the low protein wheat were lower than those obtained

TABLE 57. Comparisons of apparent faecal amino acid availabilities, in addition to nitrogen and dry matter digestibilities, of wheat and barley fed to pigs and rats.

CEREAL GRAIN	WHEAT				BARLEY		
LEVEL OF PROTEIN (%)	15.4 <sup>1</sup>	16.3 <sup>2</sup>	18.3 <sup>3</sup>	11.2 <sup>3</sup>	13.9 <sup>2</sup>	11.4 <sup>3</sup>	8.5 <sup>3</sup>
ANIMAL SPECIES	PIGS	PIGS	RATS	RATS	PIGS	RATS	RATS
AMINO ACIDS (%)							
<u>Essential</u>							
ARG	94.6	92.4	91.2	87.8	88.4	82.3	74.7
HIS	93.9	94.5	93.7	90.8	89.9	83.6	78.0
ILE	91.6	89.2	90.4	85.3	81.3	79.0	70.6
LEU	93.2	91.4	92.7	89.4	85.4	82.5	77.3
LYS	86.1	80.3	78.9	73.4	75.5	67.7	60.1
PHE	94.3	92.6	94.0	90.8	87.1	86.0	78.6
THR	89.1	86.4	86.8	81.9	79.9	76.0	67.8
VAL	91.7	88.8	90.4	86.3	82.7	82.0	73.7
<u>Non-Essential</u>							
ALA	88.1	83.6	85.2	80.9	76.1	73.2	67.0
ASP	88.0	82.9	85.3	79.1	76.3	72.8	67.2
GLU	97.9	97.0	97.4	95.7	92.1	89.8	84.1
GLY	90.6	89.1	89.8	84.8	81.2	76.0	70.0
PRO	96.8	96.9	97.3	95.9	91.7	90.5	83.6
SER	94.3	92.3	92.6	89.4	85.6	82.8	74.2
TYR	92.9	82.5	91.8	85.6	83.8	82.8	74.7
NITROGEN (%)	93.3	91.2	89.7	84.4	84.0	78.7	72.8
DRY MATTER (%)	89.4	88.8	90.3	88.8	82.7	83.7	83.5

1 Data from study 4a.

2 Data from study 2: average of cannulated and non-cannulated pigs.

3 Data from study 1.

from the rats fed the high protein wheat and the differences ranged from 6.2 for TYR and ASP to 1.4% for PRO (Table 57).

For barley, the faecal availabilities for pigs from study 2 and those obtained from the rats fed the high protein barley do not compare too unfavourably. The differences ranged from 0.7 for VAL to 7.8% for LYS (Table 57). The barley source fed to the pigs was of the Herta variety while the high protein barley fed to the rats was of the Fergus variety. In the same order, the protein contents of these barley sources were 13.9 and 11.4% respectively.

The faecal amino acid availabilities from rats fed the low protein barley source (8.5% protein) were much lower than those obtained from pigs fed barley (13.9% protein) in study 2 (Table 57). The differences ranged from 8.0 for GLU to 15.4% for LYS. Even though these barley sources were of the same variety (Herta), they differed markedly with regard to their levels of protein. In contrast, the 2 sources of Glenlea wheat, that were fed to rats (18.3% protein) and pigs (16.3% protein) and resulted in very similar faecal availability estimates, were almost similar in protein content.

The faecal amino acid availabilities from rats fed the high protein barley were higher than those obtained from low protein barley (as for high and low protein wheat). The differences ranged from 5.2 for LEU to 8.6% for SER (Table 57).

Generally speaking, faecal amino acid availabilities

from cereal grains fed to rats and pigs compare reasonably well if cereal grains containing approximately similar protein content are fed to both species. Faecal availabilities may compare even better if in addition to equal protein content the same variety is tested.

Eggum (1973) compared faecal amino acid availabilities between rats and pigs that were fed the same sources of wheat and barley. The experiments on pigs and rats were carried out under similar experimental conditions. For wheat, faecal amino acid availabilities obtained from pigs were slightly higher than those from rats. The difference varied from 4.9 for LYS to 0.2% for THR and HIS. For barley, faecal amino acid availabilities obtained from rats were slightly higher than those obtained from pigs and varied from 6.3 for SER to 0.3% for GLY. Although Eggum (1973) found some differences between pigs and rats, these differences and those obtained from other cereal grains were of a small magnitude. In general, rats and pigs may be equally used for the determination of faecal amino acid availabilities from cereal grains.

Nitrogen digestibility was increased with higher protein content of different barley sources fed to rats (Eggum, 1969). Similar observations were made in the present studies for high and low protein barley or wheat fed to rats. This was postulated to be due to the decrease in the percentage of aleurone protein (as a % of total cereal protein) with increasing



protein content of a cereal grain (see page 12). However, as was shown in these studies (Table 15 and 16), the nitrogen digestibility (and amino acid availabilities) from endosperm from high protein barley (or wheat) was also higher than that obtained from endosperm protein from low protein barley (or wheat). Therefore, in addition to an increase in the relative amount of aleurone protein there seems to be an increase in the amount of undigestible protein from endosperm (or flour) from cereal grains as the protein content decreases (i.e. the overall protein digestibility is affected).

In comparing the data obtained from rats and pigs it should be kept in mind that there were differences in the physical treatment of the cereal grains prior to feeding. However, as was shown in study 4b, physical treatment of a cereal grain prior to feeding had only a minor effect on amino acid availabilities as they were measured by the faecal analysis method.

The methods used for determining faecal amino acid availabilities from rats and pigs were quite different. The amino acid availabilities obtained from the pig were determined by aid of the chromic oxide method (Crampton and Harris, 1969). The amino acid availabilities obtained from rats were determined by total faecal collection from feeding the test diets during a period of 4 consecutive days. Inclusion of ferric oxide in the basic diet (which was fed prior to and following the test

periods) permitted somewhat easier identification and collection of faeces resulting from the test diets. However, a gradual colour change is usually observed from red to that of the colour of the faeces from the test diets. To a certain extent, the amount of faeces collected will be dependent on the colour appraisal by the individual collecting the faeces. Therefore, this method for collecting faeces is somewhat subjective.

Due to the modifying action of the microflora of the large intestine, data obtained from studies 2, 3 and 4 indicate that apparent ileal amino acid availabilities are a more accurate measurement than apparent faecal availabilities. The differences between ileal and faecal amino acid availabilities varied from amino acid to amino acid. The differences were also found to be dietary dependent and were also affected by the physical treatment of the cereal grain prior to feeding. For the essential amino acids from wheat in study 2a, the differences between ileal and faecal amino acid availabilities were found to be the largest for THR, namely 10.2%, and the smallest for MET, namely 2.3% (Table 40). For study 4a, the differences varied from 10.7% for THR to 1.0% for MET (Table 53). The differences between ileal and faecal amino acid availabilities for wheat from study 2a and 4a were more or less the same for each amino acid. Therefore, the average differences were used for arriving at estimated ileal amino acid availabilities (by subtraction from the faecal availabilities) for the rats fed

the high and low protein wheat sources (Table 58).

For barley fed to pigs in study 2a, the differences between ileal and faecal essential amino acid availabilities varied from 11.5 for HIS to 0.5% for MET. These differences were used for arriving at estimated ileal amino acid availabilities for the rats fed the 2 barley sources (Table 40).

The changes from faecal to ileal amino acid availabilities for the rats tend to make LYS and THR about equally the least available essential amino acid (Table 58).

In general, the magnitude of the differences in percentage units of faecal amino acid availabilities between endosperm and bran decreased from those obtained from the wheat source fed to the pigs (study 4a) to those obtained from the low protein wheat source and to those obtained from the high protein wheat source fed to rats in this study (Table 59). In the same order, the average differences between faecal endosperm and bran amino acid availabilities were 13.9, 6.8 and 4.3 percentage units respectively. These differences may be partially explained by the less efficient separation of aleurone and endosperm protein from wheat fed to the rats (especially that of high protein wheat) than from wheat fed to the pigs (Table 56). In addition, the bran diets fed to the rats were ground through a 2.00 mm screen prior to preparation of crumbles. The bran fraction fed to pigs was not ground prior to being pelleted and fed. However, it should be kept in mind that

TABLE 58. Comparisons of the apparent ileal amino acid availabilities, in addition to nitrogen and dry matter digestibilities of wheat and barley fed to pigs and the estimated ileal values for rats.

CEREAL GRAIN	WHEAT				BARLEY		
LEVEL OF PROTEIN (%)	15.4 <sup>1</sup>	16.3 <sup>2</sup>	18.3 <sup>3</sup>	11.2 <sup>3</sup>	13.9 <sup>2</sup>	11.4 <sup>3</sup>	8.5 <sup>3</sup>
ANIMAL SPECIES	PIGS	PIGS	RATS	RATS	PIGS	RATS	RATS
AMINO ACIDS (%)							
<u>Essential</u>							
ARG	87.1	85.8	84.0	80.6	81.5	74.4	66.8
HIS	88.4	89.1	88.0	85.1	80.4	72.1	66.5
ILE	89.1	85.3	87.1	82.0	79.1	75.0	66.6
LEU	89.9	86.9	88.7	85.4	81.5	77.4	72.2
LYS	79.5	75.7	73.6	68.1	73.3	63.5	55.9
PHE	91.5	88.8	90.7	87.5	82.2	80.3	72.9
THR	78.4	76.5	76.3	71.4	71.2	65.8	57.6
VAL	86.7	82.8	84.3	80.7	78.0	75.7	67.4
<u>Non-Essential</u>							
ALA	79.6	74.0	75.9	71.6	69.7	65.5	59.3
ASP	80.8	75.4	77.8	71.6	71.2	66.1	60.5
CLU	95.6	92.7	94.1	92.4	86.6	83.7	78.0
GLY	72.6	73.7	73.0	68.0	71.2	64.6	58.6
PRO	79.1	86.8	83.5	82.1	80.9	79.1	72.2
SER	86.3	84.1	84.4	81.2	76.3	72.2	63.6
TYR	89.2	85.9	88.1	81.9	79.7	77.5	69.4
NITROGEN (%)	85.2	82.9	81.5	76.2	74.9	72.7	64.6
DRY MATTER (%)	78.2	73.3	76.8	75.3	66.2	66.4	66.2

<sup>1</sup> Data from study 4a.

<sup>2</sup> Data from study 2a.

<sup>3</sup> Estimated data from study 1.

TABLE 59. Comparison of faecal amino acid availabilities from bran and endosperm from wheat fed to pigs and rats.

ANIMAL SPECIES	PIGS			RATS			RATS		
CEREAL FRACTION	ENDO- <sup>1</sup> SPERM	BRAN <sup>1</sup>	A <sup>4</sup>	ENDO- <sup>2</sup> SPERM	BRAN <sup>2</sup>	B <sup>4</sup>	ENDO- <sup>3</sup> SPERM	BRAN <sup>3</sup>	C <sup>4</sup>
AMINO ACIDS (%)									
<u>Essential</u>									
ARG	95.6	90.0	5.6	92.9	92.4	0.5	88.7	87.8	0.9
HIS	96.6	88.0	7.8	94.6	93.1	1.5	91.9	87.9	4.0
ILE	94.7	74.6	20.1	92.8	87.0	5.8	88.1	77.7	10.4
LEU	95.5	78.7	16.8	94.3	89.8	4.5	91.0	83.0	8.0
LYS	86.0	75.5	10.5	82.2	76.9	5.3	72.7	71.4	2.0
PHE	96.3	79.5	16.8	95.4	91.7	3.7	92.2	84.8	7.4
THR	92.3	71.3	21.0	89.7	83.6	6.2	83.7	74.9	8.8
VAL	94.3	76.0	18.3	92.7	87.6	5.1	88.6	80.7	7.9
<u>Non-Essential</u>									
ALA	90.8	75.5	15.3	88.0	82.4	5.6	81.2	75.8	5.4
ASP	89.2	75.6	13.6	87.7	82.5	5.2	80.5	74.1	6.4
GLU	98.6	89.9	8.7	98.0	96.0	2.0	96.5	91.0	5.5
GLY	93.6	78.3	15.3	92.2	86.0	6.6	86.8	77.1	9.7
PRO	98.5	89.5	9.0	98.0	95.5	2.5	96.8	89.0	7.8
SER	95.8	83.1	12.7	94.4	89.1	5.3	90.7	82.0	8.7
TYR	94.4	78.8	15.6	93.3	88.4	4.9	87.4	78.1	9.3
AVERAGE (%)	94.2	80.3	13.9	92.4	88.1	4.3	87.8	81.0	6.8

<sup>1</sup> Endosperm and bran indicate flour and B+S+M respectively from study 4a.

<sup>2</sup> Endosperm and bran obtained from high protein wheat from study 1.

<sup>3</sup> Endosperm and bran obtained from low protein wheat from study 1.

<sup>4</sup> Columns A, B and C indicate the difference in percentage units between endosperm and bran.

differences in amino acid availabilities due to "pre-processing" may be partially eliminated by the microflora of the large intestine (as was shown in study 4b). Also, the method used for determining faecal amino acid availabilities in the rat studies was of a somewhat subjective nature. The degree of error that can be committed will depend on the total bulk of faeces that is collected. The latter depends on the daily dietary intake, the dry matter digestibility and the length of time a particular test diet is fed. Therefore, in the rat experiments one may expect a larger error for endosperm than when bran is fed for the endosperm fraction has a much higher dry matter digestibility than bran. Conceivably, faecal amino acid availabilities from endosperm fed to rats may have been underestimated.

As was pointed out in the discussion part of studies 2 and 4, the use of protein-free diets for calculating true amino acid availabilities from cereal grains may give misleading values especially if true availabilities are determined by the faecal analysis method. The determination of apparent faecal amino acid availabilities from cereal grains may have some value. With the exception of THR, apparent ileal and faecal availabilities of essential amino acids from cereal grains do not differ that much especially if the grains were finely ground prior to feeding. Consequently, true faecal amino acid availabilities from cereal grains fed to rats have not been determined.

## SUMMARY

The apparent ileal and faecal amino acid availabilities from corn, wheat and barley were determined with 6 growing Managra barrows.

Lysine was the least available indispensable amino acid when determined by the faecal analysis method, namely 83.0, 80.7 and 77.5% for corn, wheat and barley respectively. Lysine and THR were approximately equally the least available indispensable amino acids when determined at the end of the ileum. The ileal LYS availabilities were 82.0, 75.7 and 73.7% for corn, wheat and barley respectively. In the same order, they were 78.9, 76.5 and 71.2% for THR. The apparent ileal availabilities of MET were 91.9, 86.6 and 80.4% for corn, wheat and barley respectively and were essentially similar to their faecal availabilities.

The apparent availabilities of LYS especially, and also those of THR and MET, are of importance since these amino acids are present in limiting amounts in cereal grains. Generally speaking, the faecal analysis method is valid for determining the apparent availabilities of LYS and MET. However, the faecal analysis method overestimates the availability of THR due to its relatively large disappearance in the large intestine. Tryptophan is also present in limited amounts in cereal grains, especially in corn, and its ileal availability should be assessed in future research.

The average amino acid disappearance in the large intestine (as a percentage of amino acid intake) increased from corn to wheat to barley and was 2.5, 5.3 and 6.0% respectively for the indispensable amino acids and 6.3, 8.5 and 8.8% respectively for the dispensable amino acids. For the indispensable acids, ARG, HIS and THR disappeared to the greatest extent. For the dispensable amino acids, disappearance was most marked for GLY and PRO.

A separate study was carried out to determine if apparent faecal amino acid availabilities from cannulated pigs were representative of those that would be obtained from non-cannulated pigs. Essentially no differences were found between normal and cannulated pigs fed the same sources of corn, wheat and barley.

The differences that were found between ileal and faecal amino acid availabilities from corn, wheat and barley may have been partially confounded by the type of physical treatment each cereal grain was subjected to prior to feeding. Two diets, namely finely ground and cracked wheat, were fed each to 4 finishing pigs. The ileal availabilities of all amino acids were higher from finely ground than from cracked wheat. The differences in percentage units ranged from 8.9% for LYS to 0.7% for PRO. The ileal availabilities of amino acids, associated with or contained in relatively large amounts in aleurone cells (such as LYS and ALA), were increased the most upon fine



grinding. Those amino acids, associated or contained in relatively low amounts in aleurone cells (such as GLU and PHE), were increased the least upon fine grinding. Differences in amino acid availabilities due to physical treatment of wheat prior to feeding could hardly be detected by faecal analysis. More research as to the improvement of amino acid availabilities upon finer grinding (and other dietary components such as starch) may be warranted for such cereal grains as barley or oats. Barley and oats contain relatively more aleurone protein (as a % of total protein) than wheat or corn.

The ileal and faecal amino acid availabilities from whole wheat, flour and B+S+M were determined with 6 growing barrows. Generally, apparent ileal and faecal amino acid availabilities decreased from flour to whole wheat and to B+S+M. The average availabilities of the indispensable amino acids were 91.6, 87.0 and 72.9% when determined from ileal digesta and 93.9, 92.0 and 79.6% when determined on faeces for flour, whole wheat and B+S+M respectively. As was found in studies carried out on corn, wheat and barley, there was a greater disappearance of dispensable than of indispensable amino acids in the large intestine. Of the indispensable amino acids, ARG, HIS and THR especially, disappeared to the greatest extent. Of the dispensable amino acids GLY and PRO showed the largest disappearance. The average amino acid disappearance in the large intestine (as a % of amino acid intake) increased from flour, to whole wheat and to B+S+M and was 4.1, 7.0 and

8.4% respectively.

Lysine was the least available indispensable amino acid of flour and whole wheat when determined by the faecal analysis method and was approximately 86% for both diets. Threonine was the least available indispensable amino acid from B+S+M, namely 71.3% using the faecal analysis method.

Lysine and THR were about equal in being the least available from flour and whole wheat when determined on ileal digesta. The apparent availabilities were approximately 79 and 85% for whole wheat and flour respectively. Threonine was the least available indispensable amino acid from B+S+M when determined at the end of the ileum (54%).

The hypothesis that the low LYS availability of cereal grains is due to the fact that this amino acid is mainly deposited in the protein fraction of lowest digestibility (in albumin and globulin protein associated with/or contained in aleurone cells) is not completely valid. The apparent ileal LYS availability of flour (84.2%) was found to be only 4.9% higher than that of whole wheat (79.5%). The difference between ileal LYS availabilities from flour and whole wheat would be approximately 2.7% if the whole wheat had been finely ground. In addition, it was shown (indirectly by calculations) that the LYS availability of B+S+M may approximate those of whole wheat and flour upon fine grinding of diet B+S+M. The relatively low availability of LYS from flour may possibly be partly attributed

to the relative large numbers of lysylprolyl peptide linkages in flour which are resistant to cleavage by the digestive enzymes.

Amino acid availabilities determined at the end of the ileum will give a more accurate measurement of their actual availabilities to the animal than measurement by the faecal analysis method. Partial verification of this was obtained when LYS was infused into the caecum, as free LYS monohydrochloride or as part of the ISP-protein complex. Infusion of LYS did not result in an improvement in nitrogen retention (amount of nitrogen retained per day) over that obtained from the pigs when they received no LYS infusion.

Interpretations of faecal amino acid availabilities obtained from rats fed high and low protein wheat and barley sources and the bran and endosperm fractions derived from these cereal grains, should be interpreted carefully because of the following reasons: 1) separation of aleurone from endosperm protein by pearling was far from complete, especially that of high protein wheat, 2) the measurement of faecal amino acid availabilities, from endosperm fractions in particular, may have been biased because of subjective measurements (colour appraisal) and 3) the faecal analysis method does not accurately measure amino acid availabilities, particularly those of ARG, HIS and especially THR (indispensable amino acids).

Regardless of the previous criticisms, differences in

apparent faecal amino acid availabilities due to protein content and/or variety of wheat and barley were detected. For example, the apparent LYS availabilities were 78.9 and 73.4% for high and low protein wheat respectively. In the same order, they were 67.7 and 60.1% for high and low protein barley. As mentioned previously, ileal and faecal LYS availabilities would not differ that much in case the cereal grains had been ground relatively fine prior to feeding as was done in these studies. More research should be carried out to determine the ileal amino acid availabilities from different varieties of particular cereal grains with varying levels of protein.

Metabolic ileal and faecal amino acid levels were determined by aid of 3 protein-free diets containing 5, 10 and 15% Alphafloc respectively. As the level of Alphafloc increased from 5 to 10 to 15%, the average ileal level of the indispensable amino acids increased from .030 to .039 to .043 grams per 100 grams of dry matter intake. In the same order, the average level of the dispensable amino acids increased from .119 to .150 and to .151 grams per 100 grams of dry matter intake. The dispensable amino acids made up the major portion of the metabolic ileal amino acids, namely 75 to 80%. The latter could be important in minimizing the loss of indispensable amino acids, as a result of the processes of digestion (either by faecal loss or by deamination in the large intestine).

Of the indispensable amino acids, ARG, THR and LEU (in

descending order) were present in relatively high amounts in metabolic ileal protein. Methionine and HIS were present in relatively low amounts.

Of the dispensable amino acids, PRO and GLY (in descending order) were present in very high amounts in metabolic ileal protein. Proline and GLY made up approximately 55 and 16% respectively of the total amount of the dispensable amino acids present at the end of the ileum when feeding the protein-free diets. Cystine and TYR, which are actually semi-dispensable amino acids, were present in very low amounts in metabolic ileal protein.

There was a consistent net disappearance of ARG, THR, GLY and PRO between the end of the ileum and the anus of pigs fed the protein-free diets. Proline in particular and GLY disappeared to the largest extent (presumably due to microbial deamination). Consistent net increases were found for ILE, LEU, LYS, MET, PHE and ASP between the end of the ileum and the anus. This was probably due to de novo microbial synthesis of these amino acids.

Protein-free diets overestimate the levels of metabolic ileal amino acids that are normally associated with the feeding of natural type diets, such as cereal grains. This conclusion was arrived at by obtaining "true" ileal PRO availabilities (and in certain instances those of GLY and ARG) that exceeded 100%, regardless of the type of corrections for metabolic ileal amino

acid levels carried out. Interpretation of calculated "true" faecal amino acid availabilities would be even more misleading than "true" ileal amino acid availabilities since an additional error comes into play, namely that certain amino acids are synthesized by the flora in the large intestine upon feeding of protein-free diets.

Overestimation of metabolic losses due to the feeding of protein-free diets were postulated to be due to increased cellular losses of epithelial cells and/or increased mucus secretions. On the other hand, a decrease in the digestion and reabsorption of endogenous protein secreted into the lumen of the small intestine, as initiated by differences in the rate of passage of protein-free and natural type diets, may be responsible. The overestimation of metabolic ileal amino acid levels, as well as nitrogen (and therefore also of metabolic faecal amino acid and nitrogen levels) is probably caused by a combination of the factors discussed, in addition to other possible unidentified factors.

Protein-free diets are widely used in nutritional studies on man and animals i.e. for the assessment of protein quality and the determination of protein (amino acid) requirements. In light of the findings in these studies, the use of protein-free diets for arriving at metabolic amino acid losses should be reevaluated and more detailed research is warranted on this aspect.

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APPENDIX TABLE 1. Analysis of variance between treatments for study 1: Mean squares for apparent amino acid availabilities and apparent nitrogen and dry matter digestibilities.

Source of Variation	ROWS (Test Diets)	COLUMNS (Weight)	ROWS X COLUMNS	ERROR
Degrees of Freedom	9	1	9	80
(Mean Squares)				
AMINO ACIDS				
<u>Essential</u>				
ARG	326.33 <sup>xx</sup>	0.08	4.15	2.35
HIS	294.54 <sup>xx</sup>	25.81 <sup>xx</sup>	14.36 <sup>xx</sup>	2.27
ILE	451.28 <sup>xx</sup>	0.01	4.93	3.84
LEU	268.25 <sup>xx</sup>	13.32 <sup>x</sup>	3.53	3.25
LYS	390.80 <sup>xx</sup>	5.29	12.33	10.44
MET	424.72 <sup>xx</sup>	5.11	7.16	5.64
PHE	278.26 <sup>xx</sup>	0.01	1.61	1.60
THR	444.60 <sup>xx</sup>	0.41	6.62	5.32
VAL	315.69 <sup>xx</sup>	0.07	3.56	2.83
<u>Non-Essential</u>				
ALA	424.18 <sup>xx</sup>	35.16 <sup>xx</sup>	6.30	4.60
ASP	423.43 <sup>xx</sup>	15.29	7.70	6.83
GLU	212.50 <sup>xx</sup>	2.96 <sup>x</sup>	0.79	0.68
GLY	518.55 <sup>xx</sup>	5.62	3.68	3.05
PRO	223.73 <sup>xx</sup>	2.07	1.05	0.86
SER	394.68 <sup>xx</sup>	0.55	2.07	2.43
TYR	365.42 <sup>xx</sup>	1.49	3.14	3.45
NITROGEN	424.30 <sup>xx</sup>	0.38	6.73	4.40
DRY MATTER	778.89 <sup>xx</sup>	7.67 <sup>xx</sup>	1.05	0.85

x (P<0.05)

xx (P<0.01)

APPENDIX TABLE 2. Analysis of variance between treatments for study 1: Mean squares for metabolic faecal amino acid and nitrogen losses.

Source of Variation	ROWS (Test Diets)	COLUMNS (Weight)	ROWS X COLUMNS	ERROR
Degrees of Freedom	1	1	1	56
<hr/>				
AMINO ACIDS	(Mean Squares)			
<u>Essential</u>				
ARG	.00061056 <sup>xx</sup>	.00013202 <sup>xx</sup>	.00000602	.00077667
HIS	.00013054 <sup>xx</sup>	.00000499	.00000099	.00027965
ILE	.00163282 <sup>xx</sup>	.00006324	.00000232	.00086475
LEU	.00437077 <sup>xx</sup>	.00023800 <sup>x</sup>	.00000411	.00226328
LYS	.00252721 <sup>xx</sup>	.00002282	.00000273	.00146270
MET	.00035917 <sup>xx</sup>	.00012615 <sup>x</sup>	.00001561	.00027237
PHE	.00155449 <sup>xx</sup>	.00005152	.00001540	.00092374
THR	.00441184 <sup>xx</sup>	.00009805 <sup>x</sup>	.00000332	.00124160
VAL	.00267600 <sup>xx</sup>	.00012644 <sup>x</sup>	.00000007	.00146677
<u>Non-Essential</u>				
ALA	.00311760 <sup>xx</sup>	.00020944 <sup>x</sup>	.00002007	.00182109
ASP	.00775207 <sup>xx</sup>	.00042135 <sup>x</sup>	.00003082	.00452194
GLU	.01152875 <sup>xx</sup>	.00041870	.00002077	.00654177
GLY	.00241427 <sup>xx</sup>	.00000680	.00000680	.00152185
PRO	.00122402 <sup>xx</sup>	.00000187	.00000217	.00199625
SER	.00231633 <sup>xx</sup>	.00005339	.00000141	.00116564
TYR	.00114756 <sup>xx</sup>	.00002940	.00000004	.00089239
NITROGEN	.10634460 <sup>xx</sup>	.00285660	.01098907 <sup>x</sup>	.13080867

x (P<0.05)

xx (P<.001)

APPENDIX TABLE 3. The individual apparent ileal and faecal amino acid availabilities from corn in study 2a, in addition to nitrogen and dry matter digestibilities and average daily dry matter intake.

PERIOD	I				II				III			
PIG NUMBER	1		6		3		4		2		7	
LOCATION	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES
AMINO ACIDS (%)												
<u>Essential</u>												
ARG	89.8	91.1	88.2	92.5	89.6	93.2	89.6	93.3	85.9	91.6	81.5	91.5
HIS	89.1	93.2	89.7	92.6	89.3	95.0	88.9	96.1	87.6	92.6	85.0	92.1
ILE	89.4	87.6	87.3	86.6	90.2	90.1	88.6	90.2	86.4	86.8	83.0	87.4
LEU	94.0	93.5	92.4	93.3	94.4	95.1	93.4	94.8	91.8	93.5	88.8	92.3
LYS <sub>1</sub>	83.1	81.9	82.8	81.7	84.0	85.6	82.7	86.5	80.2	80.6	79.2	81.9
MET <sub>1</sub>	92.2	88.4	92.2	88.4	91.4	92.2	91.4	92.2	92.0	87.9	92.0	87.9
PHE	92.1	90.4	90.3	90.7	93.0	92.9	91.5	92.8	89.2	90.7	86.9	90.3
THR	81.1	85.3	80.0	85.4	82.5	88.5	80.1	88.3	77.0	85.4	72.4	84.8
VAL	87.5	87.3	84.4	87.1	88.1	89.5	86.3	90.0	83.9	87.5	79.3	87.6
<u>Non-Essential</u>												
ALA	90.2	89.6	89.2	90.7	91.1	92.6	89.4	92.3	88.1	90.1	82.8	89.5
ASP <sub>1</sub>	85.8	86.4	84.7	86.4	86.9	87.1	84.7	88.2	82.6	85.5	78.6	86.0
CYS <sub>1</sub>	84.9	89.3	84.9	89.3	78.4	91.8	78.4	91.8	83.1	89.6	83.1	89.6
GLU	93.2	93.4	92.3	94.5	94.0	95.0	92.4	95.1	91.3	93.9	87.8	92.8
GLY	73.3	85.2	73.7	85.3	77.2	87.8	69.2	87.8	72.7	85.4	61.1	85.6
PRO	79.2	93.1	89.6	92.3	84.4	94.2	75.6	95.2	84.3	94.0	69.3	91.4
SER	86.3	90.2	85.9	91.2	87.4	92.3	85.6	92.3	83.8	90.7	80.1	89.5
TYR	91.0	89.4	89.1	89.6	91.7	91.6	89.8	91.5	88.3	89.7	84.2	89.3
NITROGEN	83.2	88.3	83.9	88.5	84.2	91.2	82.2	91.8	82.8	88.7	78.4	87.8
DRY MATTER <sub>2</sub>	80.3	89.3	81.3	88.9	80.7	90.3	79.8	90.1	80.0	89.6	79.9	88.2
DRY MATTER <sub>3</sub>	81.8		77.9		82.4		81.4		79.7		77.1	
DRY MATTER INTAKE (g/day)	1765		1765		1900		1900		1900		1900	

<sup>1</sup> For the determination of MET and CYS availabilities, faeces from the 2 pigs, fed the same diet during each particular test period, were pooled.

<sup>2</sup> Dry matter digestibilities in this row were based on the chromic oxide method.

<sup>3</sup> Dry matter digestibilities in this row were based on total collection.

APPENDIX TABLE 4. The individual apparent ileal and faecal amino acid availabilities from barley in study 2a, in addition to nitrogen and dry matter digestibilities and average daily dry matter intake.

PERIOD	I				II				III			
PIG NUMBER	3		4		2		7		1		6	
LOCATION	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES
AMINO ACIDS (%)												
<u>Essential</u>												
ARG	79.0	87.5	83.9	90.7	84.4	89.2	80.9	88.4	81.6	90.1	79.1	90.5
HIS	78.8	91.7	81.9	91.3	83.7	92.3	77.8	88.0	82.0	90.7	78.0	92.2
ILE	77.0	81.8	81.0	83.1	83.1	83.1	75.7	81.4	77.0	84.4	80.6	84.5
LEU	78.6	85.6	83.0	87.0	85.6	87.0	79.4	85.1	80.8	87.0	81.5	88.0
LYS <sub>1</sub>	71.2	76.7	73.6	78.3	77.9	78.7	69.8	74.0	73.9	77.9	73.1	79.6
NET <sub>1</sub>	80.7	80.0	80.7	80.0	80.5	78.5	80.5	78.5	80.0	81.3	80.0	81.3
PHE	80.1	87.3	83.7	87.9	85.2	88.2	80.3	86.9	82.6	88.5	82.0	88.6
THR	66.6	79.6	72.8	81.8	76.6	81.8	68.8	79.7	70.6	82.4	71.7	82.9
VAL	75.3	83.1	79.3	84.9	82.1	84.3	76.3	82.8	77.3	84.9	77.5	85.6
<u>Non-Essential</u>												
ALA	65.9	76.7	71.1	77.0	75.0	77.6	65.5	75.3	71.3	78.6	69.4	79.0
ASP <sub>1</sub>	68.2	75.3	71.9	78.9	75.6	78.0	67.8	76.6	73.1	78.7	70.4	79.6
CYS <sub>1</sub>	81.5	88.6	81.5	88.6	77.3	89.0	77.3	89.0	74.1	88.6	74.1	88.6
GLU	85.5	92.3	88.0	92.7	88.9	93.2	84.8	91.7	87.7	93.0	85.4	93.4
GLY	59.9	81.8	64.6	82.4	74.0	82.7	57.4	81.4	68.4	83.4	67.3	83.9
PRO	77.2	92.0	83.7	91.7	83.9	93.1	74.9	91.0	83.1	92.7	82.8	93.2
SER	72.4	85.5	78.7	87.1	81.1	87.5	72.3	85.5	77.6	87.6	75.6	88.0
TYR	77.4	84.3	80.5	84.1	84.2	85.0	77.6	83.6	78.4	86.6	80.2	86.1
NITROGEN <sub>2</sub>	74.6	85.7	77.8	85.8	77.5	86.0	70.6	84.1	75.7	86.1	73.1	87.5
DRY MATTER <sub>2</sub>	62.6	82.7	70.4	83.9	71.3	83.9	66.1	83.9	62.6	83.6	64.0	83.7
DRY MATTER <sub>3</sub>	65.0		71.9		69.3		64.8		66.1		65.6	
DRY MATTER INTAKE (g/day)	1766		1766		1902		1902		1902		1902	

1, 2, 3 The same as in Appendix, Table 3 .



APPENDIX TABLE 5. The individual apparent ileal and faecal amino acid availabilities from wheat in study 2a, in addition to nitrogen and dry matter digestibilities and average daily dry matter intake.

PERIOD	I				II				III			
PIC NUMBER	2		7		1		6		3		4	
LOCATION	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES
AMINO ACIDS (%)												
<u>Essential</u>												
ARG	84.7	92.9	84.0	91.2	85.4	91.2	86.3	93.6	88.2	93.6	86.2	93.4
HIS	87.6	94.6	86.9	93.4	89.3	93.8	87.5	96.0	92.3	95.3	91.0	96.2
ILE	86.0	88.5	81.9	89.1	84.6	86.9	85.4	90.5	88.2	91.2	85.7	89.9
LEU	87.2	91.3	84.1	90.8	86.7	89.4	86.9	92.6	89.4	92.8	86.9	91.9
LYS	75.6	79.3	70.4	77.4	76.1	78.2	75.3	82.4	79.9	82.7	76.6	84.0
MET <sup>1</sup>	84.7	87.4	84.7	87.4	86.4	87.9	86.4	87.9	88.8	91.5	88.8	91.5
PHE	88.8	92.2	86.7	92.3	88.3	90.8	89.1	93.6	91.6	94.0	89.0	93.0
THR	77.2	86.1	71.5	85.5	76.9	84.3	76.1	88.0	79.9	88.5	77.4	87.8
VAL	83.3	88.7	78.7	88.1	83.1	86.1	82.6	90.3	85.8	90.5	83.5	89.5
<u>Non-Essential</u>												
ALA	74.5	83.7	67.7	83.2	74.2	80.1	73.9	85.2	79.5	86.5	74.0	85.1
ASP	76.2	82.9	67.5	81.0	76.3	80.1	74.8	84.5	81.1	85.6	76.6	84.3
CYS <sup>1</sup>	82.6	93.2	82.6	93.2	85.6	93.6	85.6	93.6	87.5	94.4	87.5	94.4
GLU	92.8	96.7	91.3	96.9	92.6	96.4	93.0	97.4	94.2	97.5	92.4	97.3
GLY	75.7	88.7	62.2	89.0	76.3	87.1	71.3	90.3	79.7	90.6	77.1	89.7
PRO	84.9	95.5	86.6	96.9	83.6	96.3	90.6	96.9	92.9	97.4	82.4	97.1
SER	84.8	92.1	80.8	92.0	85.1	91.0	83.3	93.2	85.4	93.6	85.0	92.9
TYR	86.0	89.1	83.2	88.6	85.2	87.4	86.0	90.4	88.3	91.4	86.4	90.1
NITROGEN	83.7	90.6	77.8	89.9	83.8	90.4	82.2	92.5	85.9	92.1	84.3	91.5
DRY MATTER <sup>2</sup>	74.1	88.6	70.3	89.2	73.7	88.6	72.9	89.2	75.0	89.4	73.6	88.8
DRY MATTER <sup>3</sup>	73.9		72.7		--		--		73.6		72.3	
DRY MATTER	1759		1759		1894		1894		1894		1894	
INTAKE (g/day)												

1, 2, 3 The same as in Appendix, Table 3.

APPENDIX TABLE 6. Mean squares of the analyses of variance for the apparent ileal and faecal availabilities of the essential amino acids for study 2a.

AMINO ACIDS	ARG	HIS	ILE	LEU	LYS	PHE	THR	VAL
<u>Source of variation</u>								
Treatment (2) <sup>1</sup>	58.79 <sup>xx</sup>	135.04 <sup>xx</sup>	169.90 <sup>xx</sup>	247.07 <sup>xx</sup>	154.84 <sup>xx</sup>	132.72 <sup>xx</sup>	137.75 <sup>xx</sup>	242.84 <sup>xx</sup>
Periods (2)	3.28	0.92	2.31	1.91	9.46	2.28	7.35	4.05
Pigs (5)	6.80 <sup>x</sup>	9.96 <sup>x</sup>	9.41	6.59	18.05 <sup>x</sup>	4.92	14.24	9.36 <sup>x</sup>
Location (1)	382.20 <sup>xx</sup>	476.69 <sup>xx</sup>	75.11 <sup>xx</sup>	121.73 <sup>xx</sup>	106.73 <sup>xx</sup>	106.43 <sup>xx</sup>	773.77 <sup>xx</sup>	104.63 <sup>xx</sup>
Treatments x Location (2)	8.16 <sup>x</sup>	27.27 <sup>xx</sup>	11.45	13.13 <sup>x</sup>	13.00	18.28 <sup>xx</sup>	7.61	8.57
Periods x Location (2)	5.56	0.07	2.23	1.58	0.61	1.23	3.71	3.98
Pigs x Location (5)	8.05 <sup>x</sup>	3.36	3.67	1.67	3.56	1.50	4.61	4.45
Error (16)	2.13	2.65	3.48	2.38	3.78	1.91	5.28	3.11

<sup>1</sup> Numbers in parentheses indicate the degrees of freedom.

<sup>x</sup> (P<0.05)

<sup>xx</sup> (P<0.01)

APPENDIX TABLE 7. Mean squares for the analysis of variance for the apparent ileal and faecal availabilities of the non-essential amino acids, in addition to nitrogen and dry matter digestibilities for study 2a.

AMINO ACIDS	ALA	ASP	GLU	GLY	PRO	SER	TYR	N.	D.M.
<u>Source of variation</u>									
Treatment (2) <sup>1</sup>	805.02 <sup>xx</sup>	347.22 <sup>xx</sup>	84.51 <sup>xx</sup>	174.53 <sup>xx</sup>	100.73 <sup>xx</sup>	170.58 <sup>xx</sup>	170.09 <sup>xx</sup>	153.30 <sup>xx</sup>	285.10 <sup>xx</sup>
Periods (2)	5.17	7.29	1.38	13.45	0.20	2.03	2.87	0.95	2.20
Pigs (5)	17.64 <sup>x</sup>	17.39 <sup>x</sup>	3.82	41.36 <sup>xx</sup>	25.08	10.03 <sup>x</sup>	7.26	14.33 <sup>xx</sup>	3.60
Location (1)	400.00 <sup>xx</sup>	290.70 <sup>xx</sup>	162.99 <sup>xx</sup>	2288.03 <sup>xx</sup>	1167.36 <sup>xx</sup>	633.36 <sup>xx</sup>	101.00 <sup>xx</sup>	682.95 <sup>xx</sup>	1796.34 <sup>xx</sup>
Treatments x Location (2)	46.34 <sup>xx</sup>	20.51 <sup>x</sup>	11.32 <sup>xx</sup>	4.56	7.29	14.52 <sup>x</sup>	12.61 <sup>x</sup>	12.86 <sup>xx</sup>	54.46 <sup>xx</sup>
Periods x Location (2)	2.29	2.23	0.83	3.37	4.19	1.35	3.51	1.33	0.51
Pigs x Location (5)	8.11	9.23	1.41	35.30 <sup>x</sup>	18.04	4.53	2.25	4.45	2.66
Error (16)	4.87	4.68	1.38	8.51	13.42	3.06	2.78	1.60	5.75

<sup>1</sup> Numbers in parentheses indicate the degrees of freedom.

<sup>x</sup> (P<0.05)

<sup>xx</sup> (P<0.01)

APPENDIX TABLE 8. Mean squares of the analyses of variance for the apparent ileal and faecal availabilities of MET and CYS for study 2a.

AMINO ACIDS	MET	CYS
<u>Source of variation</u>		
Treatment (2) <sup>1</sup>	177.03 <sup>xx</sup>	59.58
Periods (2)	2.75	0.83
Pigs (2)	7.86	2.60
Location (1)	0.14	383.64 <sup>xx</sup>
Treatment x Location (2)	8.26	3.98
Periods x Location (2)	0.21	5.05
Pigs x Location (2)	1.62	0.16
Error (4)	2.31	12.42

<sup>1</sup> Numbers in parentheses indicate the degrees of freedom.

<sup>xx</sup> (P<0.01)

APPENDIX TABLE 9. The individual metabolic ileal amino acid and nitrogen levels<sup>1</sup>, in addition to dry matter digestibilities and daily dry matter intake for pigs fed the maintenance diets.

LEVEL OF ALPHAFLOC (%)	7						14				
PIG NUMBER	1	2	3	4	7	1	2	3	4	7	6
<b>AMINO ACIDS</b>											
<u>Essential</u>											
ARG	.057	.030	.041	.027	.035	.046	.038	.049	.049	.053	.041
HIS	.016	.010	.016	.009	.012	.016	.016	.016	.016	.026	.016
ILE	.037	.033	.031	.029	.033	.030	.039	.035	.032	.041	.034
LEU	.055	.040	.035	.034	.048	.046	.043	.043	.040	.047	.049
LYS	.044	.034	.033	.028	.039	.046	.042	.043	.040	.046	.041
MET <sup>2</sup>	.009	.009	.007	.008	.011	.011	.012	.011	.012	.010	.010
PHE	.030	.020	.018	.017	.027	.029	.023	.022	.021	.023	.025
THR	.054	.036	.042	.038	.052	.047	.042	.047	.043	.051	.043
VAL	.047	.036	.034	.032	.043	.041	.042	.041	.039	.046	.042
<u>Non-Essential</u>											
ALA	.061	.039	.042	.035	.045	.061	.047	.054	.051	.079	.051
ASP	.075	.065	.060	.059	.072	.076	.074	.074	.071	.070	.074
GLU	.139	.127	.122	.107	.124	.120	.148	.144	.135	.176	.131
GLY	.186	.085	.122	.090	.090	.129	.095	.142	.126	.281	.098
PRO	.464	.243	.494	.248	.270	.449	.303	.649	.471	1.206	.251
SER	.075	.064	.072	.059	.055	.062	.075	.081	.067	.100	.066
TYR	.024	.017	.015	.014	.021	.019	.017	.017	.016	.019	.019
NITROGEN	.271	.181	.224	.176	.222	.310	.259	.329	.290	.400	.251
DRY MATTER DIGESTIBILITY (%) <sup>3</sup>	85.8	87.0	86.4	86.9	86.7	77.1	76.5	76.5	76.5	76.8	77.3
DRY MATTER DIGESTIBILITY (%) <sup>4</sup>	91.2	85.6	88.7	90.8	86.3	90.0	83.2	86.6	85.9	73.7	87.4
DRY MATTER INTAKE	825	1621	1580	1159	1586	521	1041	723	891	1337	684

<sup>1</sup> Expressed as grams per 100 gram dry matter intake. <sup>2</sup> Determined by acid hydrolysis.

<sup>3</sup> Ileal dry matter digestibilities in this row were based on the chromic oxide method.

<sup>4</sup> Ileal dry matter digestibilities in this row were based on total collection.

APPENDIX TABLE 10. The individual metabolic faecal amino acid and nitrogen levels<sup>1</sup>, in addition to dry matter digestibilities for pigs fed the maintenance diets.

LEVEL OF ALPHAFLOC (%)							14				
PIG NUMBER	1	2	3	4	7	1	2	3	4	7	1
AMINO ACIDS											
<u>Essential</u>											
ARG	.049	.030	.022	.019	.024	.058	.022	.041	.031	.027	.029
HIS	.017	.012	.010	.008	.010	.020	.009	.018	.018	.014	.012
ILE	.048	.032	.031	.027	.028	.071	.030	.046	.038	.044	.036
LEU	.080	.055	.037	.037	.042	.125	.044	.072	.057	.056	.055
LYS <sup>2</sup>	.068	.046	.040	.038	.036	.095	.037	.059	.054	.047	.058
MET	.025	.018	.015	.011	.012	.034	.013	.020	.019	.018	.015
PHE	.047	.034	.025	.023	.026	.074	.028	.043	.035	.030	.034
THR	.050	.035	.030	.026	.030	.080	.033	.050	.041	.037	.040
VAL	.055	.039	.034	.029	.033	.085	.036	.054	.045	.042	.044
<u>Non-Essential</u>											
ALA	.066	.048	.041	.036	.040	.108	.044	.066	.056	.051	.054
ASP	.104	.073	.065	.056	.062	.153	.066	.101	.086	.079	.081
GLU	.130	.092	.093	.082	.077	.197	.090	.130	.116	.111	.106
GLY	.055	.042	.031	.028	.032	.087	.033	.051	.044	.041	.042
PRO	.060	.041	.034	.032	.026	.109	.057	.063	.053	.062	.055
SER	.049	.034	.038	.034	.028	.075	.039	.053	.045	.045	.044
TYR	.036	.026	.018	.016	.020	.054	.022	.031	.026	.023	.025
NITROGEN	.204	.150	.120	.111	.133	.323	.135	.198	.166	.169	.169
DRY MATTER DIGESTIBILITY (%)	90.1	90.0	92.0	92.2	91.2	85.8	87.3	86.5	86.4	85.9	86.8

<sup>1</sup> Expressed as grams per 100 gram dry matter intake.

<sup>2</sup> Determined by acid hydrolysis.

APPENDIX TABLE 11. Mean squares of analyses of variance for the apparent amino acid availabilities, nitrogen and dry matter digestibilities for study 2b.

Source of Variation	TREATMENT	PIGS	PERIODS	ERROR
Degrees of Freedom	2	5	2	8
AMINO ACIDS				
<u>Essential</u>				
ARG	38.20 <sup>xx</sup>	2.58	2.65	2.31
HIS	63.74 <sup>xx</sup>	3.74	3.64	1.19
ILE	158.92 <sup>xx</sup>	9.06	0.82	4.35
LEU	146.18 <sup>xx</sup>	4.71	0.18	2.20
LYS	115.58 <sup>xx</sup>	24.48	12.54	11.16
MET	248.65 <sup>xx</sup>	15.35	0.50	8.61
PHE	72.94 <sup>xx</sup>	4.22 <sup>xx</sup>	0.35	0.55
THR	119.14 <sup>xx</sup>	11.05	1.36	4.32
VAL	100.53 <sup>xx</sup>	10.81	0.75	3.29
<u>Non-Essential</u>				
ALA	357.13 <sup>xx</sup>	17.30 <sup>x</sup>	1.67	4.21
ASP	203.40 <sup>xx</sup>	16.74	3.31	16.53
GLU	47.18 <sup>xx</sup>	1.87	0.36	1.04
GLY	129.85 <sup>xx</sup>	12.00	2.97	4.63
PRO	51.94 <sup>xx</sup>	0.89	0.08	0.69
SER	103.40 <sup>xx</sup>	3.55	0.12	1.61
TYR	104.91 <sup>xx</sup>	6.01	0.51	2.40
NITROGEN	131.57 <sup>xx</sup>	4.47	2.63	1.83
DRY MATTER	98.43 <sup>xx</sup>	0.96	0.19	0.20

x (P<0.05)

xx (P<0.01)

APPENDIX TABLE 12.

The individual apparent faecal amino acid availabilities from barley in study 2b, in addition to nitrogen and dry matter digestibilities.

PERIOD	I		II		III	
PIG NUMBER	5	6	1	2	3	4
AMINO ACIDS (%)						
<u>Essential</u>						
ARG	88.0	87.0	87.7	86.8	87.5	87.1
HIS	89.8	85.9	87.8	87.5	88.6	87.1
ILE	80.6	75.2	79.8	79.8	80.3	81.1
LEU	85.3	80.9	84.2	84.5	84.7	84.9
LYS	78.7	68.9	72.6	72.8	75.2	72.5
MET	75.8	68.7	78.7	76.0	77.2	80.9
PHE	87.1	82.3	86.9	87.2	86.6	86.8
THR	79.2	74.1	78.4	79.8	78.9	79.2
VAL	82.8	75.2	82.0	82.2	81.8	82.4
<u>Non-Essential</u>						
ALA	76.8	69.3	75.7	76.2	75.6	75.2
ASP	76.4	68.5	75.5	76.6	75.7	75.6
GLU	91.9	89.0	92.0	91.7	91.9	92.6
GLY	81.4	72.6	81.2	81.9	80.7	81.0
PRO	91.4	89.5	91.5	92.2	90.8	91.4
SER	84.9	81.4	84.6	85.0	84.7	84.9
TYR	83.7	78.6	83.2	83.3	82.5	83.8
NITROGEN (%)	82.3	82.7	82.7	82.1	83.5	78.8
DRY MATTER (%)	82.0	81.0	82.5	82.1	81.3	81.6



APPENDIX TABLE 13.

The individual apparent faecal amino acid availabilities from corn in study 2b, in addition to nitrogen and dry matter digestibilities.

PERIOD	I		II		III	
PIG NUMBER	3	4	5	6	1	2
AMINO ACIDS (%)						
<u>Essential</u>						
ARG	93.3	89.7	91.1	91.2	89.9	91.7
HIS	94.5	91.2	93.4	92.9	91.1	91.2
ILE	91.0	85.2	88.2	87.9	88.2	86.0
LEU	94.9	92.0	93.6	93.6	94.7	92.6
LYS	85.8	78.8	83.1	82.8	82.0	78.4
MET	88.8	87.5	89.4	86.3	86.8	85.6
PHE	92.7	89.7	91.3	91.2	92.0	90.5
THR	88.7	83.4	86.6	85.4	87.6	83.7
VAL	89.0	86.6	88.4	87.3	88.6	86.2
<u>Non-Essential</u>						
ALA	92.0	88.7	91.1	89.6	90.5	89.3
ASP	87.7	84.2	87.5	86.3	86.1	83.9
GLU	95.3	92.2	94.2	93.9	94.6	92.3
GLY	88.2	84.0	87.0	86.0	86.4	84.2
PRO	94.1	93.1	93.2	92.9	94.9	92.1
SER	92.5	88.9	90.9	90.3	91.5	89.2
TYR	91.7	88.4	90.3	89.9	90.9	89.0
NITROGEN (%)	91.3	86.2	88.6	88.9	88.7	87.2
DRY MATTER (%)	90.1	88.8	89.6	89.3	88.7	87.6

APPENDIX TABLE 14.

The individual apparent faecal amino acid availabilities from wheat in study 2b, in addition to nitrogen and dry matter digestibilities.

PERIOD	I		II		III	
PIG NUMBER	1	2	3	4	5	6
AMINO ACIDS (%)						
<u>Essential</u>						
ARG	92.8	93.4	92.8	93.2	93.1	87.4
HIS	94.7	95.1	95.0	94.6	94.7	90.4
ILE	90.0	90.5	90.1	88.5	90.8	83.5
LEU	92.4	92.7	91.3	91.2	92.5	87.1
LYS	80.9	82.8	81.8	81.8	83.2	69.1
MET	88.5	90.7	88.0	85.0	89.6	82.2
PHE	93.4	93.7	92.5	92.4	93.6	89.6
THR	86.7	88.2	87.7	85.5	88.2	80.1
VAL	89.8	90.4	89.0	88.1	90.4	83.6
<u>Non-Essential</u>						
ALA	84.5	86.1	85.5	81.3	86.4	76.2
ASP	83.7	85.6	84.0	81.6	85.7	75.0
GLU	97.3	97.5	97.4	96.8	97.6	95.4
GLY	89.4	90.8	89.9	88.2	90.6	84.6
PRO	97.1	97.2	97.3	96.6	97.4	96.2
SER	92.8	93.3	92.3	91.5	93.3	89.3
TYR	90.7	91.0	90.1	88.9	90.7	85.3
NITROGEN (%)	91.3	92.3	91.8	90.8	92.5	88.0
DRY MATTER (%)	88.8	88.9	87.8	87.8	88.9	88.7

APPENDIX TABLE 15 Mean squares of the analyses of variance of the metabolic faecal amino acid and nitrogen excretion from study 2b.

Source of variation	ROWS (TEST DIETS)	COLUMNS (WEIGHT)	ROWS X COLUMNS	ERROR
Degrees of freedom	1	1	1	20
AMINO ACIDS				
<u>Essential</u>				
ARG	0.000504 <sup>xx</sup>	0.000150 <sup>xx</sup>	0.000004	0.000017
HIS	0.000077 <sup>xx</sup>	0.000001	0.000003	0.000003
ILE	0.000682 <sup>xx</sup>	0.000001	0.000013	0.000038
LEU	0.002072 <sup>xx</sup>	0.000176	0.000001	0.000117
LYS	0.001276 <sup>xx</sup>	0.000040	0.000002	0.000054
MET	0.000216 <sup>xx</sup>	0.000016	0.000008	0.000013
PHE	0.000672 <sup>xx</sup>	0.000155 <sup>x</sup>	0.000001	0.000030
THR	0.001053 <sup>xx</sup>	0.000084	0.000009	0.000062
VAL	0.001426 <sup>xx</sup>	0.000030	0.000007	0.000092
<u>Non-Essential</u>				
ALA	0.002016 <sup>xx</sup>	0.000130	0.000001	0.000098
ASP	0.004902 <sup>xx</sup>	0.000040	0.000026	0.000298
GLU	0.005310 <sup>xx</sup>	0.000051	0.000108	0.000354
GLY	0.001148 <sup>xx</sup>	0.000170	0.000006	0.000053
PRO	0.000715 <sup>xx</sup>	0.000045	0.000001	0.000038
SER	0.000522 <sup>xx</sup>	0.000032	0.000020	0.000053
TYR	0.000337 <sup>xx</sup>	0.000073	0.000001	0.000023
NITROGEN	0.017985 <sup>xx</sup>	0.000035	0.000001	0.000445

x (P<0.05)  
xx (P<0.01)

APPENDIX TABLE 16. The individual metabolic fecal amino acid and nitrogen excretion from pigs fed the 7% alphafloc maintenance diet.

TIME OF COLLECTION	BEFORE CEREAL TEST DIETS						AFTER CEREAL TEST DIETS					
PIG NUMBER	1	2	3	4	5	6	1	2	3	4	5	6
AMINO ACIDS												
<u>Essential</u>												
ARG	.028	.025	.016	.016	.021	.024	.016	.013	.021	.015	.017	.023
HIS	.011	.007	.007	.007	.007	.008	.008	.006	.008	.006	.007	.009
ILE	.036	.032	.021	.018	.026	.030	.025	.033	.029	.023	.025	.036
LEU	.056	.045	.026	.028	.041	.049	.030	.037	.042	.029	.032	.045
LYS	.040	.035	.025	.024	.033	.040	.028	.028	.036	.023	.026	.037
MET	.017	.014	.007	.008	.012	.013	.009	.012	.013	.011	.012	.017
PHE	.035	.027	.016	.018	.026	.029	.019	.020	.021	.017	.018	.025
THR	.041	.033	.019	.021	.026	.032	.023	.026	.031	.021	.023	.033
VAL	.042	.035	.022	.022	.032	.037	.027	.034	.033	.024	.026	.039
<u>Non-Essential</u>												
ALA	.052	.043	.027	.029	.040	.046	.033	.035	.042	.028	.030	.042
ASP	.084	.068	.043	.043	.062	.069	.051	.057	.064	.046	.049	.074
GLU	.100	.094	.064	.055	.075	.089	.078	.106	.078	.071	.074	.113
GLY	.040	.035	.022	.024	.032	.036	.024	.026	.033	.023	.024	.033
PRO	.041	.033	.025	.024	.027	.043	.026	.033	.026	.028	.028	.036
SER	.041	.040	.026	.022	.029	.035	.034	.046	.031	.029	.032	.046
TYR	.026	.022	.012	.013	.018	.020	.012	.014	.019	.013	.013	.020
NITROGEN	.143	.127	.102	.094	.113	.129	.109	.131	.126	.101	.110	.147

APPENDIX TABLE 17. The individual metabolic fecal amino acid and nitrogen excretion from pigs fed the 14% alphafloc maintenance diets.

TIME OF COLLECTION	BEFORE CEREAL DIETS						AFTER CEREAL DIETS					
PIG NUMBER	1	2	3	4	5	6	1	2	3	4	5	6
AMINO ACIDS												
<u>Essential</u>												
ARG	.029	.030	.033	.028	.032	.038	.020	.029	.027	.028	.021	.030
HIS	.013	.010	.010	.008	.008	.015	.009	.014	.011	.012	.011	.013
ILE	.038	.042	.044	.033	.034	.045	.032	.045	.035	.032	.033	.049
LEU	.056	.069	.062	.052	.055	.065	.043	.064	.050	.045	.041	.081
LYS	.047	.049	.050	.041	.039	.055	.036	.055	.039	.042	.037	.060
MET	.018	.018	.018	.013	.015	.018	.017	.026	.022	.015	.013	.024
PHE	.035	.039	.037	.033	.033	.037	.025	.035	.030	.026	.024	.044
THR	.040	.045	.052	.039	.040	.043	.031	.043	.037	.031	.028	.059
VAL	.047	.048	.052	.043	.042	.057	.035	.050	.039	.037	.034	.074
<u>Non-Essential</u>												
ALA	.059	.060	.070	.052	.049	.058	.043	.066	.050	.044	.040	.076
ASP	.088	.091	.094	.077	.079	.099	.074	.094	.084	.070	.064	.139
GLU	.109	.111	.134	.100	.095	.132	.096	.138	.098	.093	.104	.144
GLY	.047	.048	.055	.043	.040	.045	.032	.046	.038	.034	.031	.059
PRO	.040	.039	.041	.044	.043	.052	.037	.043	.039	.031	.040	.052
SER	.041	.042	.047	.039	.039	.052	.037	.053	.037	.037	.044	.055
TYR	.026	.026	.029	.023	.025	.028	.020	.026	.022	.020	.013	.034
NITROGEN	.165	.180	.182	.161	.145	.205	.156	.211	.166	.162	.149	.207

APPENDIX TABLE 18

Analyses of variance between treatments for parameters measured in study 3a.

Source of Variation	ROWS (PICS)	COLUMNS (PERIODS)	TREATMENTS	ERROR
Degrees of Freedom	2	2	2	2
<u>ITEMS</u>	(Mean squares)			
Nitrogen retained (g/day)	0.7647	1.3803	4.6244 <sup>x</sup>	0.2216
Nitrogen retained <sup>1</sup>	0.002734	0.001269	0.011537	0.000698
Nitrogen retention (%) <sup>2</sup>	4.4275	1.6374	62.5195 <sup>x</sup>	1.1611
Biological value (%) <sup>3</sup>	5.6459	8.4700	78.4245 <sup>x</sup>	1.6782

1, 2, 3 As shown in Table 28.

<sup>x</sup> ( $P < 0.05$ )

APPENDIX TABLE 19 The individual apparent ileal and faecal amino acid availabilities from whole wheat in study 4a, in addition to nitrogen and dry matter digestibilities and average daily dry matter intake.

DAY	12-18				34-40				56-62			
PIG NUMBER	5		7		1		2		3		4	
LOCATION	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES
AMINO ACIDS (%)												
<u>Essential</u>												
ARG	87.2	94.1	88.6	94.3	89.4	94.2	85.9	94.8	86.7	95.4	84.7	94.8
HIS	88.6	93.4	89.8	92.5	89.7	94.0	85.9	94.5	88.2	94.9	88.4	94.5
ILE	89.1	91.6	90.6	91.8	90.4	89.8	85.2	92.0	89.6	92.8	89.5	91.5
LEU	89.7	92.8	91.0	93.3	90.9	92.5	86.9	93.1	90.2	94.0	90.5	93.2
LYS	80.2	86.1	81.2	87.0	81.6	84.2	72.8	85.9	80.1	87.6	81.0	86.0
MET	93.6	93.7	93.4	94.2	92.9	90.1	88.4	94.0	94.4	95.4	91.9	93.1
PHE	90.9	93.5	91.6	94.1	92.2	93.6	89.7	94.6	92.2	95.3	92.5	94.5
THR	79.0	88.8	78.6	89.6	81.4	88.1	73.7	88.8	78.1	90.7	79.8	89.0
VAL	86.3	91.3	86.9	91.9	88.2	90.3	83.4	91.7	87.6	93.6	87.9	91.8
<u>Non-Essential</u>												
ALA	79.2	88.4	80.9	88.8	83.4	85.4	75.6	88.2	78.8	90.0	79.5	87.9
ASP	81.4	87.3	81.8	88.6	84.1	86.3	76.5	87.7	79.7	89.9	81.5	88.0
GLU	95.8	97.8	95.9	97.9	96.3	97.7	94.5	97.9	95.4	98.1	95.8	97.9
GLY	69.9	90.2	75.8	91.0	81.2	89.8	76.4	90.6	65.5	91.5	66.7	90.6
PRO	80.8	96.5	85.7	97.6	92.5	94.8	91.5	97.0	69.5	98.1	54.5	96.6
SER	85.7	93.7	86.9	94.0	88.4	93.3	83.8	95.6	85.9	95.3	87.0	94.1
TYR	87.9	92.4	89.2	93.2	89.7	91.2	86.6	92.7	90.9	94.8	91.0	92.9
NITROGEN (%)	84.7	93.0	86.6	93.6	88.0	92.6	84.1	93.1	84.1	94.5	83.6	93.4
DRY MATTER (%)	78.0	89.2	80.4	89.9	78.9	89.8	75.8	88.6	77.1	89.5	78.9	89.3
DRY MATTER INTAKE (g/day)	1367		1367		1640		1640		1721		1530	

APPENDIX TABLE 20. The individual apparent ileal and faecal amino acid availabilities from flour in study 4a, in addition to nitrogen and dry matter digestibilities and average daily dry matter intake.

DAY	12-18				34-40				56-62			
PIG NUMBER	1		2		3		4		5		7	
LOCATION	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES
AMINO ACIDS (%)												
<u>Essential</u>												
ARG	92.5	93.7	92.5	97.0	90.4	95.1	92.7	96.3	89.7	96.8	86.3	94.7
HIS	93.8	94.9	94.0	97.7	94.0	96.5	94.3	97.0	94.2	97.7	93.3	95.8
ILE	93.1	93.2	93.4	95.8	94.4	94.7	94.6	95.4	94.2	96.5	94.1	92.7
LEU	94.0	94.5	94.2	96.4	95.2	95.7	94.4	96.1	95.1	97.3	95.0	93.3
LYS	81.8	80.2	83.8	91.3	85.0	84.1	86.4	88.2	84.7	90.0	83.4	82.0
MET	92.6	89.5	93.1	96.1	94.5	90.9	93.0	95.4	95.9	95.4	93.0	93.2
PHE	95.7	95.6	95.8	97.0	93.4	96.4	95.7	96.7	96.5	97.7	96.0	94.7
THR	83.8	91.2	84.3	94.3	85.0	91.9	84.7	92.8	87.4	94.8	87.3	88.6
VAL	91.8	93.0	92.2	95.5	93.1	94.2	92.6	95.0	93.3	96.3	93.1	91.9
<u>Non-Essential</u>												
ALA	86.6	88.3	86.1	93.2	86.7	90.5	87.2	92.2	85.6	93.8	84.7	86.7
ASP	84.1	86.9	87.0	92.5	85.8	88.4	85.7	90.2	84.7	92.4	85.5	84.9
GLU	97.6	98.4	97.9	98.9	97.9	98.6	97.9	98.8	98.0	99.1	98.0	98.0
GLY	86.9	92.7	83.6	95.2	76.6	93.3	85.5	94.1	69.6	95.6	69.3	90.8
PRO	96.2	98.4	96.5	98.6	76.1	98.3	94.2	98.8	73.4	99.1	61.7	98.0
SER	91.4	94.7	91.4	97.0	91.3	95.7	91.8	96.2	92.3	97.3	92.0	94.0
TYR	92.6	93.5	92.8	96.2	93.5	93.1	92.7	94.3	94.3	96.3	92.8	93.3
NITROGEN (%)	91.7	94.6	91.8	96.8	89.6	95.2	92.5	96.2	89.6	96.1	87.6	94.6
DRY MATTER (%)	90.7	94.9	90.5	95.7	90.0	94.4	90.4	95.2	90.4	95.8	89.5	93.8
DRY MATTER INTAKE (g/day)	1353		1285		1542		1624		1550		1550	



APPENDIX TABLE 21. The individual apparent ileal and faecal amino acid availabilities from the B+S+M diet in study 4, in addition to nitrogen and dry matter digestibilities and average daily dry matter intake.

DAY	12-18		34-40		23-29	
PIG NUMBER	3		5		6	
LOCATION	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES
AMINO ACIDS (%)						
<u>Essential</u>						
ARG	86.5	92.4	84.4	88.8	83.5	88.7
HIS	81.3	91.7	78.2	87.4	76.1	87.5
ILE	73.1	76.9	75.1	73.4	70.5	73.6
LEU	75.3	81.3	76.9	76.9	71.1	77.9
LYS	72.0	80.3	63.9	72.6	63.4	73.5
MET	73.5	81.0	80.9	80.7	79.0	83.8
PHE	75.3	81.5	78.8	78.7	74.0	78.3
THR	53.9	75.3	60.5	69.5	47.5	69.1
VAL	72.9	80.5	73.3	76.6	68.4	71.1
<u>Non-Essential</u>						
ALA	69.9	78.6	71.0	75.7	69.8	72.1
ASP	68.9	78.9	71.2	74.2	69.4	73.8
GLU	86.4	90.9	86.6	89.7	84.5	89.2
GLY	67.1	80.0	49.6	77.2	55.3	77.9
PRO	74.5	91.4	57.8	86.9	78.6	90.3
SER	71.8	85.1	74.5	82.5	70.3	81.6
TYR	71.3	81.2	74.0	77.7	68.1	77.4
NITROGEN (%)	71.3	81.4	69.4	81.3	68.6	80.1
DRY MATTER (%)	43.6	66.1	45.8	65.6	40.5	63.3
DRY MATTER INTAKE (g/day)	659		737		1168	

APPENDIX TABLE 22. The individual apparent ileal and faecal amino acid availabilities from diet B+S+M-D from study 4a, in addition to nitrogen and dry matter digestibilities and average daily dry matter intake.

DAY	67-73						67-73					
PIG NUMBER	1		2		3		4		5		6	
LOCATION	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES
AMINO ACID (%)												
<u>Essential</u>												
ARG	75.7	89.7	77.1	88.7	70.2	89.8	58.8	91.3	74.3	91.4	73.1	90.2
HIS	71.8	89.5	74.3	85.6	72.6	86.7	68.9	88.7	76.8	89.3	77.0	88.3
ILE	67.9	79.0	70.2	68.0	70.3	76.5	69.3	77.9	67.9	77.3	72.0	74.2
LEU	71.3	81.7	71.6	75.2	73.0	81.8	69.5	82.9	70.7	82.8	74.2	80.0
LYS	53.6	76.2	58.8	72.8	60.3	77.4	53.1	80.5	56.1	81.4	62.9	77.6
MET	70.0	74.0	78.7	64.5	79.0	74.2	85.6	80.1	70.0	82.4	75.5	64.4
PHE	72.5	82.2	73.4	75.9	74.4	82.3	74.7	83.5	72.5	83.6	75.9	81.6
THR	47.0	73.8	46.6	68.9	46.9	75.7	42.1	77.3	50.3	77.6	53.8	74.7
VAL	64.5	80.9	68.7	72.9	66.0	79.6	63.1	80.5	66.4	79.5	70.2	75.8
<u>Non-Essential</u>												
ALA	59.9	79.3	65.6	70.5	60.9	78.1	49.9	78.6	61.7	79.0	66.3	71.4
ASP	63.3	77.7	66.2	73.7	65.1	79.2	58.5	80.5	63.9	80.3	68.4	77.8
GLU	84.8	90.7	83.8	88.6	84.5	91.5	81.0	92.2	83.1	92.1	85.5	90.1
GLY	50.7	80.6	46.6	75.5	25.6	80.4	-7.0	80.8	40.6	81.6	35.2	76.8
PRO	27.1	81.0	11.9	73.0	-93.5	78.5	-151.7	80.0	-39.4	79.5	-39.9	78.4
SER	69.1	83.2	69.1	80.4	66.1	85.0	63.5	86.1	67.0	86.3	71.5	84.4
TYR	66.5	76.8	71.7	72.8	70.1	78.2	69.6	83.6	64.5	82.0	68.7	78.4
NITROGEN (%)	62.9	81.6	66.8	81.3	55.7	84.4	48.9	84.1	58.5	84.2	62.4	82.2
DRY MATTER (%)	69.7	84.1	70.7	83.6	66.8	84.2	68.5	84.2	69.7	83.8	69.9	83.6
DRY MATTER INTAKE (g/day)	1386		1478		1663		1848		1756		1756	

APPENDIX TABLE 23. The individual metabolic ileal and faecal amino acid, nitrogen and dry matter levels from pigs fed the 5% alphafloc protein-free diets, in addition to average daily dry matter intake.

DAY	1-7				23-29				45-51			
PIG NUMBER	1		2		3		4		5		7	
LOCATION	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES
AMINO ACIDS <sup>1</sup>												
<u>Essential</u>												
ARG	.026	.023	.026	.022	.059	.019	.038	.024	.064	.020	.083	.021
HIS	.011	.012	.011	.011	.015	.010	.016	.012	.016	.010	.015	.009
ILE	.016	.030	.019	.030	.021	.025	.025	.033	.021	.024	.026	.022
LEU	.029	.047	.035	.045	.038	.039	.047	.049	.037	.038	.046	.036
LYS	.022	.040	.027	.039	.028	.034	.030	.040	.028	.033	.029	.033
MET	.005	.014	.005	.014	.005	.010	.005	.010	.007	.011	.007	.011
PHE	.018	.029	.020	.028	.022	.024	.028	.029	.023	.023	.026	.022
THR	.033	.033	.033	.032	.043	.029	.045	.035	.037	.026	.042	.026
VAL	.028	.037	.027	.035	.032	.032	.038	.038	.032	.029	.029	.027
<u>Non-Essential</u>												
ALA	.029	.040	.034	.039	.048	.034	.040	.040	.048	.029	.050	.032
ASP	.044	.066	.047	.064	.060	.056	.062	.069	.055	.058	.067	.054
CYS	.011	.009	.011	.009	.013	.011	.013	.011	.014	.011	.014	.011
GLU	.053	.078	.060	.072	.079	.063	.077	.077	.073	.061	.084	.058
GLY	.066	.037	.078	.031	.191	.030	.099	.033	.189	.028	.211	.026
PRO	.220	.029	.146	.024	.646	.032	.210	.031	.691	.021	.932	.020
SER	.030	.027	.032	.026	.044	.025	.038	.028	.039	.024	.045	.023
TYR	.011	.012	.013	.015	.012	.008	.017	.012	.012	.018	.015	.016
NITROGEN <sup>1</sup>	.123	.110	.130	.104	.252	.090	.161	.111	.253	.095	.313	.095
DRY MATTER <sup>1</sup>	10.26	6.98	11.42	6.56	11.85	6.69	11.13	6.21	12.93	6.34	12.40	6.08
DRY MATTER INTAKE (g/day)	1368		1368		1642		1642		1642		1642	

<sup>1</sup> Metabolic ileal and faecal amino acid levels were expressed as grams per 100 grams of dry matter intake.

APPENDIX TABLE 24. The individual metabolic ileal and faecal amino acid, nitrogen and dry matter levels from pigs fed the 10% alphafloc protein-free diet, in addition to average daily dry matter intake.

DAY	1-7				23-29				12-18		34-40		56-62	
PIG NUMBER	3		4		5		7				9			
LOCATION	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES
AMINO ACIDS <sup>1</sup>														
<u>Essential</u>														
ARG	.075	.026	.041	.035	.062	.027	.046	.028	.082	.030	.063	.026	.056	.034
HIS	.019	.013	.019	.017	.014	.014	.015	.013	.022	.014	.017	.012	.019	.017
ILE	.030	.035	.039	.048	.020	.039	.024	.032	.032	.041	.029	.040	.038	.046
LEU	.049	.053	.065	.071	.038	.059	.043	.050	.060	.060	.054	.055	.061	.071
LYS	.039	.044	.046	.058	.027	.050	.029	.039	.038	.045	.036	.042	.040	.056
MET	.007	.015	.012	.020	.007	.018	.006	.013	---	---	---	---	---	---
PHE	.030	.034	.039	.045	.024	.038	.024	.032	.034	.035	.036	.034	.036	.043
THR	.050	.035	.055	.050	.042	.042	.046	.034	.068	.043	.061	.037	.061	.050
VAL	.041	.043	.055	.059	.033	.050	.039	.041	.050	.046	.048	.045	.050	.056
<u>Non-Essential</u>														
ALA	.066	.046	.055	.063	.049	.057	.044	.043	.070	.054	.063	.056	.063	.066
ASP	.086	.075	.085	.102	.062	.087	.066	.068	.093	.087	.086	.078	.092	.101
CYS	.016	.011	.019	.016	.013	.013	.014	.012	---	---	---	---	---	---
GLU	.105	.085	.110	.116	.069	.100	.080	.078	.107	.099	.100	.083	.107	.115
GLY	.196	.038	.111	.051	.188	.045	.143	.037	.250	.042	.222	.040	.184	.052
PRO	.763	.038	.134	.049	.668	.050	.511	.040	.780	.042	.899	.056	.742	.059
SER	.052	.030	.048	.041	.043	.036	.044	.031	.062	.036	.059	.032	.059	.042
TYR	.015	.013	.019	.019	.011	.016	.012	.016	.018	.014	.021	.016	.017	.019
NITROGEN <sup>1</sup>	.310	.126	.203	.166	.250	.142	.208	.116	.334	.141	.300	.124	.287	.171
DRY MATTER <sup>1</sup>	18.14	10.32	17.15	11.24	17.54	10.20	16.51	10.22	19.26	10.20	19.05	10.87	19.05	10.87
DRY MATTER INTAKE (g/day)	1285		1102		1561		1561		1040		1416		1366	

<sup>1</sup> Metabolic ileal and faecal levels were expressed as grams per 100 grams of dry matter intake.

APPENDIX TABLE 25. The individual metabolic ileal and faecal amino acid, nitrogen and dry matter levels from pigs fed the 15% alphafloc protein-free diets, in addition to average daily dry matter intake.

DAY	1-7		45-51		12-18		34-40	
PIG NUMBER	5		3		6			
LOCATION	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES
<u>AMINO ACIDS<sup>1</sup></u>								
<u>Essential</u>								
ARG	.050	.032	.061	.038	.071	.026	.084	.033
HIS	.013	.016	.025	.014	.024	.013	.029	.017
ILE	.026	.043	.030	.049	.034	.047	.038	.045
LEU	.050	.063	.052	.072	.064	.063	.072	.072
LYS	.029	.052	.039	.060	.046	.045	.054	.056
MET	.010	.028	.014	.021	--	--	--	--
PHE	.029	.038	.028	.044	.042	.043	.041	.037
THR	.055	.041	.047	.051	.077	.047	.073	.044
VAL	.039	.049	.039	.059	.052	.057	.059	.050
<u>Non-Essential</u>								
ALA	.050	.054	.061	.068	.063	.063	.067	.055
ASP	.073	.088	.074	.107	.098	.092	.093	.099
CYS	.018	.019	.021	.015	--	--	--	--
GLU	.084	.099	.097	.120	.116	.111	.125	.105
GLY	.152	.043	.174	.052	.182	.053	.180	.045
PRO	.444	.035	.761	.042	.467	.042	.635	.038
SER	.042	.035	.055	.039	.063	.037	.061	.036
TYR	.013	.016	.014	.023	.021	.015	.023	.017
NITROGEN <sup>1</sup>	.233	.153	.284	.172	.272	.164	.306	.153
DRY MATTER <sup>1</sup>	25.34	15.27	26.74	13.64	25.59	15.57	28.14	15.27
DRY MATTER INTAKE (g/day)	730		1277		1107		1532	

<sup>1</sup> Metabolic ileal and faecal levels were expressed as grams per 100 grams of dry matter intake.

APPENDIX TABLE 26. Analyses of variance: Mean squares for apparent ileal and faecal amino acid availabilities, nitrogen and dry matter digestibilities from finely ground and cracked wheat for study 4b.

Source of variation	LOCATION	TREATMENT	TREATMENT X LOCATION	ERROR
Degrees of freedom	1	1	1	12
<u>AMINO ACIDS</u>				
<u>Essential</u>				
ARG	98.258 <sup>xx</sup>	37.424 <sup>xx</sup>	26.807 <sup>xx</sup>	1.512
HIS	141.372 <sup>xx</sup>	39.564 <sup>xx</sup>	28.998 <sup>xx</sup>	1.238
ILE	38.069 <sup>xx</sup>	45.428 <sup>xx</sup>	17.016 <sup>x</sup>	2.405
LEU	41.056 <sup>xx</sup>	28.436 <sup>xx</sup>	13.746 <sup>xx</sup>	1.217
LYS	265.038 <sup>xx</sup>	136.189 <sup>xx</sup>	73.103 <sup>x</sup>	13.571
MET	51.984 <sup>x</sup>	15.445	28.944	6.232
PHE	20.885 <sup>xx</sup>	20.566 <sup>xx</sup>	10.791 <sup>xx</sup>	0.722
THR	222.905 <sup>xx</sup>	60.140 <sup>xx</sup>	23.717 <sup>x</sup>	4.440
VAL	113.050 <sup>xx</sup>	47.990 <sup>xx</sup>	28.918 <sup>xx</sup>	2.474
<u>Non-Essential</u>				
ALA	332.789 <sup>xx</sup>	99.052 <sup>xx</sup>	69.597 <sup>x</sup>	9.175
ASP	209.960 <sup>xx</sup>	60.840 <sup>xx</sup>	29.921 <sup>x</sup>	4.372
GLU	40.132 <sup>xx</sup>	2.624	1.575	1.127
GLY	614.172 <sup>xx</sup>	73.831 <sup>xx</sup>	66.300 <sup>xx</sup>	3.834
PRO	569.538 <sup>xx</sup>	0.109	1.300	10.815
SER	117.777 <sup>xx</sup>	46.478 <sup>x</sup>	0.473	8.484
TYR	44.023 <sup>xx</sup>	25.553 <sup>xx</sup>	22.801 <sup>x</sup>	2.617
NITROGEN	194.393 <sup>xx</sup>	23.160 <sup>xx</sup>	15.269 <sup>xx</sup>	0.712
DRY MATTER	416.874 <sup>xx</sup>	12.942 <sup>xx</sup>	12.479 <sup>xx</sup>	0.638

<sup>x</sup> (P<0.05).

<sup>xx</sup> (P 0.01).

APPENDIX TABLE 27. The individual apparent ileal and faecal amino acid availabilities of ground wheat from study 4b, in addition to nitrogen and dry matter digestibilities and average daily dry matter intake.

DAY	78-84							
PIG NUMBER	1		3		5		6	
LOCATION	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES
AMINO ACIDS (%)								
<u>Essential</u>								
ARG	91.7	93.0	90.0	95.2	95.0	95.1	91.3	94.2
HIS	90.8	93.1	89.4	95.4	93.6	94.8	90.8	94.2
ILE	90.6	88.6	89.8	93.4	90.1	92.6	90.2	90.4
LEU	92.1	91.0	90.6	94.2	91.9	94.1	91.7	92.3
LYS	78.7	81.2	80.3	88.1	87.1	86.7	79.7	85.5
MET	87.3	86.4	87.1	90.5	90.4	90.1	87.2	88.6
PHE	93.3	91.8	92.1	94.6	93.2	94.6	93.0	93.1
THR	83.8	86.0	87.7	91.0	83.0	89.9	80.3	88.0
VAL	89.4	88.9	87.5	93.2	89.1	92.6	88.5	90.4
<u>Non-Essential</u>								
ALA	82.8	82.6	81.8	90.1	82.3	89.5	81.6	86.1
ASP	83.2	84.0	82.6	89.6	82.8	89.0	82.6	86.8
GLU	96.4	97.3	96.3	98.2	92.2	98.0	96.1	97.8
GLY	85.1	87.5	78.9	91.8	81.6	91.1	81.4	89.8
PRO	90.3	94.6	86.3	97.1	80.7	97.5	80.8	94.5
SER	89.6	91.5	87.0	94.2	88.0	94.3	88.3	93.3
TYR	90.2	88.1	88.0	92.0	91.5	92.4	89.5	90.4
NITROGEN (%)	89.1	92.0	87.4	94.3	88.4	94.2	88.8	93.4
DRY MATTER (%)	81.1	88.3	79.4	89.1	80.1	89.2	81.0	88.8
DRY MATTER INTAKE (g/day)	1830		1830		2045		2153	

APPENDIX TABLE 28. The individual apparent ileal and faecal amino acid availabilities of cracked wheat from study 4b, in addition to nitrogen and dry matter digestibilities and average daily dry matter intake.

DAY	78-84							
PIG NUMBER	2		4		7		9	
LOCATION	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES	ILEUM	FAECES
AMINO ACIDS (%)								
<u>Essential</u>								
ARG	86.8	93.9	85.5	93.8	86.9	94.4	86.6	93.6
HIS	84.2	93.5	85.5	94.4	87.5	94.4	85.8	93.4
ILE	81.8	89.3	85.1	90.2	89.0	90.8	86.5	89.4
LEU	85.0	91.8	87.7	92.5	89.7	92.5	87.8	91.5
LYS	64.0	82.7	72.1	85.5	77.9	84.8	76.7	82.1
MET	78.7	87.6	82.9	88.6	84.5	91.7	88.3	90.7
PHE	87.8	93.0	89.6	93.1	90.9	93.1	89.2	92.4
THR	78.8	86.1	76.7	88.9	80.6	88.1	76.0	86.1
VAL	79.3	90.6	83.1	90.6	85.4	91.0	84.4	89.7
<u>Non-Essential</u>								
ALA	66.6	85.3	73.2	86.5	80.6	87.4	77.9	85.9
ASP	71.7	85.8	77.1	87.1	81.6	87.1	78.7	84.6
GLU	93.3	97.5	94.2	97.9	95.3	97.7	93.6	97.3
GLY	74.5	89.9	70.0	90.4	78.8	90.0	74.5	89.1
PRO	87.4	95.1	77.8	96.3	86.2	97.2	83.8	96.7
SER	83.6	92.4	85.4	93.5	85.7	93.1	84.1	82.0
TYR	82.2	89.4	85.5	90.5	86.9	91.9	86.5	90.6
NITROGEN (%)	82.7	92.7	84.9	93.4	87.0	93.3	84.0	92.5
DRY MATTER (%)	76.7	88.8	76.4	89.2	78.6	88.5	75.6	88.6
DRY MATTER INTAKE (g/day)	2167		2059		1842		2167	