

THE UNIVERSITY OF MANITOBA  
AVAILABILITY OF AMINO ACIDS FROM  
BARLEY, WHEAT, TRITICALE AND SOYBEAN MEAL  
FOR GROWING PIGS

by  
WILLEM CORNELIS SAUER

A THESIS  
SUBMITTED TO THE FACULTY OF GRADUATE  
STUDIES IN PARTIAL FULFILLMENT OF THE  
REQUIREMENTS FOR THE DEGREE  
OF MASTER OF SCIENCE

DEPARTMENT OF ANIMAL SCIENCE

WINNIPEG, MANITOBA

January, 1972



## ABSTRACT

The biological availability of sixteen amino acids from barley, wheat, triticale and soybean meal to Managra barrows weighing 10kg or 30kg was determined.

For the 10kg pigs, the true nitrogen digestibility decreased from 94.7 to 92.0 to 90.9 and to 85.8% for soybean meal, triticale, wheat and for barley respectively, and from 91.6 to 89.6 to 88.1 and to 83.7% for the 30kg pigs. Generally, the true availability of each essential amino acid determined decreased from soybean meal to triticale to wheat and to barley.

The true availabilities of the essential amino acids of the cereal grains could be classified in four groups from low to high availability : 1) Lysine, which was least available ( $P < .05$ ), its availability being 86.3, 80.8 and 77.1% for the lighter pigs and 77.5, 67.3 and 65.0% for the heavier pigs in triticale, wheat and barley respectively, 2) Isoleucine, methionine, threonine and valine of which the availabilities varied between 90.3-92.4, 87.3-89.8 and 84.7-87.6% for the lighter pigs and between 86.8-87.7, 85.4-86.1 and 81.5-84.6% for the heavier pigs fed triticale, wheat and barley respectively, 3) Leucine, its availability being 93.7, 91.2 and 88.7% for the lighter pigs and 91.3, 89.8 and 87.6% for the heavier pigs fed triticale, wheat

and barley respectively, 4) Arginine, histidine and phenylalanine were the most available essential amino acids. The availabilities of these amino acids varied between 93.8-94.5, 92.4-94.7 and 90.1-93.2% for the lighter pigs and between 93.3-94.1, 91.6-93.1 and 89.5-90.1% for the heavier pigs fed triticales, wheat and barley respectively.

Methionine was the least available essential amino acid in soybean meal for the heavier pigs, namely 86.3%. However, its availability was not significantly different ( $P < .05$ ) from that of valine, threonine and lysine which were 90.9, 91.0 and 91.2% respectively. No significant differences ( $P < .05$ ) in amino acid availabilities from soybean meal were detected for the lighter pigs. The true availabilities varied between 94.0 and 98.4%.

For the non-essential amino acids, alanine was the least available and glutamic acid, proline and serine were the most available for all feeds tested.

# TABLE OF CONTENTS

	Page
ACKNOWLEDGEMENTS .....	1
LIST OF TABLES .....	2
LIST OF FIGURES .....	4
LIST OF APPENDICES .....	5
INTRODUCTION .....	7
REVIEW OF THE LITERATURE .....	9
The effect of dry matter intake and digestibility on the metabolic fecal nitrogen and amino acid excretion .....	9
The effect of source and level of protein on the metabolic fecal nitrogen and amino acid excretion .....	12
Dilution of dietary amino acids by endogenous amino acids in intestinal contents and feces .....	14
The influence of microflora on amino acid availability .....	19
Factors determining lysine availability and its determination in different foodstuffs by different methods .....	25
MATERIALS AND METHODS .....	31
Availability Study 1 .....	31
Availability Study 2 .....	35
Analytical methods .....	38
RESULTS .....	40
Study 1 .....	40
1. Apparent and true availability between test diets	
a) Apparent availability .....	42
b) True availability .....	45
2. Apparent and true availability within test diets	
a) Apparent availability .....	46
b) True availability .....	50

## RESULTS (continued)

Study 2 .....	50
1. Apparent and true availability between test diets	
a) Apparent availability .....	52
b) True availability .....	53
2. Apparent and true availability within test diets	
a) Apparent availability .....	54
b) True availability .....	57
DISCUSSION .....	58
Comparisons between the dietary and fecal amino acid levels when expressed as g amino acid/16 g nitrogen and its implications .....	58
Comparisons between the dietary amino acid availabilities when the dry matter intake or excretion of the protein-free diet is made equal to that of each test diet .....	70
SUMMARY .....	79
BIBLIOGRAPHY .....	82
APPENDICES .....	87

## ACKNOWLEDGEMENTS

The author is very grateful to Dr S.C. Stothers for his assistance, guidance and advice, as well as his patience throughout the course of this study.

Appreciation is also expressed to Mr Peter Mills for his technical assistance with amino acid analyses.

Furthermore, the author is indebted to his parents for providing personal financial assistance when necessary and to his wife for her encouragement and occasional assistance towards the completion of this thesis.

## LIST OF TABLES

Table	Page
1. Metabolic fecal nitrogen excretion as influenced by dry matter intake and digestibility and its relation to dietary fecal nitrogen in growing rats when these were fed a 10% protein diet of which the true nitrogen digestibility varied between 98 and 80%..	11
2. Formulation and composition of diets for amino acid availability studies.....	33
3. Mean and standard error of the dry matter and nitrogen intake, excretion and digestibility for the test and protein-free diets.....	37
4. Comparison of apparent and true availability between test diets in study 1 .....	43
5. Comparison of the mean apparent amino acid availabilities within the test diets in study 1 .....	47
6. Comparison of the mean true amino acid availabilities within the test diets in study 1 .....	48
7. Comparison of apparent and true availability between test diets in study 2 .....	51
8. Comparison of the mean apparent amino acid availabilities within the test diets in study 2 (means for eight pigs) .....	55
9. Comparison of the mean true amino acid availabilities within the test diets in study 2 (means for eight pigs) .....	56
10. Correction factors for equal dry matter excretion and consumption of the protein-free and test diet and the corrected nitrogen digestibilities and lysine availabilities .....	59

## Table

## Page

11. Average true amino acid availability (%)  
corrected for equal dry matter  
excretion and consumption of the  
protein-free and test diet in study 1... 60
12. Average true amino acid availability (%)  
corrected for equal dry matter  
excretion and consumption of the  
protein-free and test diet in study 2... 61
13. Comparison of the corrected metabolic  
fecal amino acid excretion (g) from  
the lighter (study 1) and heavier pigs  
(study 2) ..... 64
14. Comparison between the dietary amino acid  
levels (g/16 gN) and the apparent or  
true fecal amino acid levels for study 1. 71
15. Comparison between the dietary amino acid  
levels (g/16GN) and the apparent or true  
fecal amino acid levels for study 2 ..... 72



## LIST OF FIGURES

Figure		Page
1.	Metabolic cages for digestibility trials with swine	32
2.	Experimental designs for study 1 and 2	36

## LIST OF APPENDICES

Appendix Table	Page
1. Analysis of variance between treatments for study 1: Mean squares for apparent amino acid availabilities and apparent nitrogen and dry matter digestibility .....	87
2. Analysis of variance between treatments for study 1: Mean squares for true amino acid availabilities and true nitrogen digestibility .....	88
3. Analysis of variance between treatments for study 2: Mean squares for apparent amino acid availabilities and apparent nitrogen and dry matter digestibility .....	89
4. Analysis of variance between treatments for study 2: Mean squares for true amino acid availabilities and true nitrogen digestibility .....	90
5. Analysis of variance within treatments for study 1: Mean squares for true and apparent amino acid availabilities	91
6. Analysis of variance within treatments for study 2: Mean squares for true and apparent amino acid availabilities	92
7. Mean and standard error of the apparent availability of amino acids in study 1 as influenced by the type of diet .....	93
8. Mean and standard error of the true availability of amino acids in study 1 as influenced by the type of diet .....	94
9. Mean and standard error of the apparent availability of amino acids in study 2 as influenced by the type of diet .....	95
10. Mean and standard error of the true availability of amino acids in study 2 as influenced by the type of diet .....	96

Appendix  
Table

Page

11.	Mean and standard error of the amino acid excretion (g) by the pigs in study 1 as influenced by the type of diet.....	97
12.	Mean and standard error of the amino acid excretion (g) by the pigs in study 2 as influenced by the type of diet .....	98
13.	Triticale repeat experiment of study 1: Mean and standard error of the true and apparent amino acid availability and excretion of amino acids by the pigs (g) .....	99
14.	Mean apparent and true essential amino acid availabilities, nitrogen and dry matter digestibility between the test periods in study 2. ....	100
15.	Comparison between the dietary amino acid levels (g amino acid N/16 g N) and the apparent fecal amino acid levels in Study 1 .....	101
16.	Comparison between the dietary amino acid levels (g amino acid N/16 g N) and the apparent fecal amino acid levels in Study 2 .....	102

## INTRODUCTION

Since the development of the column chromatographic method for amino acid determinations it has become possible, on a more extensive basis, to consider not only protein as the unit in nutrition but rather the individual amino acid.

Amino acid levels determined after acid hydrolysis of foods predict their potential value. However, their kinetic value, i.e. their availability to the animal may be significantly less for some foodstuffs.

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 in so far as it is dependent on the digestibility of the protein, presence of peptide linkages that are resistant to the digestive enzymes, digestive enzyme-inhibiting substances and rate of release of the amino acids in the intestinal tract". Amino acid availability defined as such may be determined by measuring the amount of ingested amino acids excreted in the feces of test animals fed different foodstuffs according to the fecal analysis method as was developed by Kuiken and Lyman (1948).

Lysine is the indispensable amino acid that is most limiting in many foods of plant origin, especially in cereal grains, which represent the major source of

protein for most of the "underdeveloped" countries. Consequently, particular attention has been given to determination of lysine availability. It may very well be that lysine is limiting in these foods due to its low availability. In view of possible lysine supplementation to foods in which lysine is limiting it will be of utmost importance to obtain an accurate estimation of its availability in these foods.

In view of the extensive use of cereal grains in swine rations, the objective of this study was to determine in the pig the apparent and true amino acid availabilities from different cereal grains relative to those of soybean meal. In addition, similarities between the functions of the digestive tracts of pig and man enable us to extrapolate data obtained from experimental work with pigs and compile results which might be applied to man. Furthermore, amino acid availabilities were assessed for groups of pigs weighing 10 kg. and 30 kg. at the start of the experiment to determine if an age factor was involved in availability of amino acids.

## REVIEW OF THE LITERATURE

Amino acid availability may be determined by measuring the amount of ingested amino acids excreted in the feces of test animals fed different foodstuffs. At least two major problems are inherent in this method for determining amino acid availability of foodstuffs:

1) The estimation of the contribution of the endogenous amino acids to the amino acids derived from the test diet in the feces, and 2) the effect of the intestinal microflora, especially those of the large intestine, on amino acid availability. An attempt is made in this review to determine to what extent these problems affect amino acid availability as determined by the fecal analysis method according to Kuiken and Lyman (1948).

A special chapter is devoted to lysine availability per se since it is the most limiting essential amino acid in many foods of plant origin, especially in cereal grains.

The effect of dry matter intake and digestibility on the metabolic fecal nitrogen and amino acid excretion

It is conceivable that the amount of metabolic fecal nitrogen (N) excreted varies with the type of diet fed. This diet-metabolic fecal N interaction is to be kept in mind when determining the true N digestibility of a particular foodstuff. The level of dietary fiber, in particular, has been studied regarding its effect on loss of endogenous N.

Meyer (1956) carried out N balance studies involving 48 growing rats which were pair-fed a 0, 5, 15 or 30% cellulose diet. Cellulose was added at the expense of the basal ration, so that the ratios of sucrose and fat to protein, vitamins and minerals remained constant. The basal ration contained 86.8% sucrose and 4.0% defatted whole-egg protein. There was no influence of fiber intake on endogenous urinary N excretion. Each added increment of cellulose increased total fecal N excretion, although its concentration decreased in the feces. Fecal N concentration and either the cellulose intake or indigestible dry matter of the feces were inversely related but less variation was present when metabolic fecal N concentration was related to the indigestible dry matter by the regression equation  $Y = 3.1 + 115.5 \cdot \frac{1}{X}$  in which Y is the metabolic fecal N in milligrams per gram of fecal dry matter (fecal N concentration) and X is the percentage of indigestible dry matter in the diet.

The total metabolic fecal N excretion has been computed, according to the equation above, under conditions of varying dry matter intake (2 - 6 g) and dry matter digestibility (80 - 95%). The diet was assumed to contain 10% protein of which the true N digestibility varied from 80 to 98% and the fecal N derived from dietary N has been indicated for comparative purposes (Table 1). The calculations show that the contribution

TABLE 1. Metabolic fecal nitrogen excretion as influenced by dry matter intake, digestibility and its relation to dietary fecal nitrogen in growing rats when these were fed a 10% protein diet of which the true nitrogen digestibility varied between 98 and 80%

Dry matter intake (g)	X <sup>1</sup>	Y <sup>1</sup>	Total M.F.N. (mg)	Dietary fecal nitrogen (mg) at varying true nitrogen digestibility			
				98.0	95.0	90.0	80.0
2.0	5.0	26.1	2.6	4.0	10.0	20.0	40.0
	10.0	14.6	2.9	4.0	10.0	20.0	40.0
	20.0	8.9	3.6	4.0	10.0	20.0	40.0
4.0	5.0	26.1	5.2	8.0	20.0	40.0	80.0
	10.0	14.6	5.8	8.0	20.0	40.0	80.0
	20.0	8.9	7.1	8.0	20.0	40.0	80.0
6.0	5.0	26.1	7.9	12.0	30.0	60.0	120.0
	10.0	14.6	8.8	12.0	30.0	60.0	120.0
	20.0	8.9	10.7	12.0	30.0	60.0	120.0

<sup>1</sup>  $Y = 3.10 + 115.45 \cdot 1/X$  in which Y indicates the metabolic fecal nitrogen (M.F.N.) in milligrams per gram of fecal dry matter and X the percentage of indigestible dry matter in the diet (Meyer, 1956).



of the metabolic fecal N to the total amount of fecal N is relatively small when the true N digestibility of a protein is 90% or lower. Apparent and true amino acid availability should not differ very much in this case. However, it would be advisable to formulate a protein-free diet in such a manner that it contains an equal amount of indigestible dry matter as the diet for which one determines the true N digestibility and the true amino acid availabilities. This may be achieved by including a certain amount of fiber in the protein-free diet. It is especially important to take this precaution when the test protein is highly digestible. The test animals should also be pair-fed the test and protein-free diet in order to guarantee equal dry matter intake.

The effect of source and level of protein on the metabolic fecal nitrogen and amino acid excretion

Mitchell and Bert (1954) reviewed the effect of the source of protein on the output of metabolic fecal N and concluded that the inclusion of different protein sources in a diet did not appreciably increase or decrease its output.

Snook (1965) studied the effect of source and level of protein on the content of proteolytic enzymes in the pancreas. As compared to casein, a protein-free diet depressed and whole-egg protein elevated the pancreatic

content of chymotrypsinogen and trypsinogen. Both zymogen levels were increased in the pancreas when a diet containing 30% casein was fed to the rats as compared to a 15% casein diet.

Data compiled by Nasset (1965) show that a total of 64 to 263 g. of protein, derived from digestive secretions, are secreted daily in the adult man. However, protein derived from pancreatic juice makes up only 12% of this fraction. The main protein fraction seems to be derived from intestinal juice which amounts to about 75%. This fraction does not present much proteolytic activity and it seems therefore unlikely that a specific response as to source and level of dietary protein is of any physiological or nutritional importance as in the case of the pancreatic juice. According to estimations by Snook and Meyer (1964 a, 1964 b) the activities of trypsin and chymotrypsin may account for as much as 78% of the total proteolytic activity of the digesta.

In summary, although there is a specific response to source and level of protein, this response concerns only the proteolytic enzymes from the pancreas. The protein derived from the pancreatic juice makes up only a small fraction of the total digestive secretions and it would seem therefore unlikely that it results in an increase or decrease in metabolic fecal N.

Protein derived from intestinal mucosal cells and mucus, that is daily delivered to the lumen of the gut, has been estimated by Nasset (1965) to be 77 to 91 g. in man. This protein fraction might be called "slough-off" protein and the extent of "slough-off" would probably be dependant on the dry matter intake and digestibility of a particular diet (the main factor being the level and intake of fiber by the animal). Rate of passage of a certain diet might also influence the extent of "slough-off". This protein fraction would be expected to make up a large proportion of the metabolic fecal N.

Snook and Meyer (1964a, 1964b) found that the extent of autodigestion and reabsorption of the endogenous protein by the rat was reduced in presence of exogenous protein. Consequently, determination of metabolic fecal amino acids by feeding a protein-free diet would underestimate endogenous losses of amino acids when these estimations are used for evaluating the true amino acid availabilities of a particular protein or protein source. However, their work was carried out in vitro and quantitative effects remain to be determined.

Dilution of dietary amino acids by endogenous amino acids in intestinal contents and feces

Results obtained by Nasset and his co-workers (1955, 1961) have made many nutritionists and physiologists ask more questions about the determination of dietary

amino acid availability as studied by the fecal analysis method. Nasset concludes from his experimental work that the presence of endogenous N in the intestinal tract acts as a homeostatic device to prevent wide fluctuations in the amino acid mixture available for absorption and that the amino acid mixture produced during digestion remained relatively constant irrespective of the composition of the ingested protein.

Nasset (1955) fed 3 different types of test meal to dogs, which were killed 1.5 hours after feeding. The dogs were fasted 24 hours before feeding. The contents of the stomach, duodenum, jejunum and ileum were collected separately. The total mass of nitrogen in the lumen of the small intestine was about the same for egg albumen and zein but it was reduced by two-thirds after feeding a protein-free meal. Zein is virtually devoid of tryptophan and lysine but after ingestion of about 3 g. of zein, 15 mg. of tryptophan and 89 mg. of lysine were found in the small intestine. Amino acid molar ratios for jejunal contents were very similar after feeding egg albumin, zein and the protein-free meal.

Nasset and Ju (1961) fed radioactive casein to rats by stomach tube for one meal. The rats were sacrificed at 1, 2 and 4 hours after feeding and the gastric and intestinal contents were analyzed separately. At all times they found that exogenous N in the intestine was diluted 6 or 7-fold and that the

molar ratios of free amino acids were markedly different from those found in the ingested casein.

Results from experimental work carried out by Bergen and Purser (1968) were more or less in agreement with those of Nasset. Growing rats were fed either a 9% casein, protozoal or bacterial protein diet for 8 days. The animals were sacrificed 2 hours after feeding and the jejunal and ileal contents were collected. The bulk amino acid composition was found to be similar regardless of the protein source fed. The dilution ratio of exogenous and endogenous N for the 9% casein diet was found to be 1:1 and 1:10 in the jejunum and ileum respectively. As for the 9% bacterial protein diet, the ratios were 1:1 and 1:2 and for the protozoal diet 1:1 and 1:2 in jejunal and ileal contents respectively. A protein-free diet resulted in a lower total amount of amino acids in the gut but the relative amino acid ratios were similar to those obtained from the test diets.

Tombly and Meyer (1961) demonstrated differential digestion and absorption of exogenous and endogenous protein. Rats were fed 5 levels of whole egg protein namely at 0, 5, 10, 15 and 20% of the diet for 1 week. They were trained to eat in 1-hour periods, 12 hours apart and were sacrificed at 1, 2.5, 5 and 8 hours after feeding on the seventh day. Two peaks in nitrogen concentration occurred in the contents of the small

intestine, one at 1 hour and the other at 8 hours after feeding the different levels of whole egg protein. The 1-hour peak was not observed in the animals receiving a protein-free diet and was therefore considered to be due to the accumulation of dietary protein in the intestinal lumen. However, the 8-hour peak was observed in rats fed the protein-free diet and was attributed to secretion of endogenous protein in response to feeding. Denton and Elvehjem (1954) observed 2 peaks in the concentration curve for amino acids in the portal vein following the ingestion of amino acids by dogs; the first peak at 2.5 hours, the second one at 6 hours after feeding.

Crompton and Nesheim (1969) fed diets to ducks in which the protein source was either a combination of corn and soybean meal or of corn and corn gluten meal. The pattern of amino acids in segments more distal to the midpoint of the small intestine did not reflect the dietary amino acid pattern as well as that in upper portions suggesting that digestion of dietary proteins occurs before that of endogenous protein. The dilution of dietary protein by endogenous protein was insufficient to mask the amino acid pattern of ingested protein. In their discussion it was noted that Nasset usually fasted his animals for 24 hours and then obtained samples 1.5 hours after feeding and it could have been possible

that digestion and absorption had nearly been completed and that most of the remaining protein in the intestine was of endogenous origin. This would explain the high dilution of exogenous protein by endogenous protein in the experimental work by Nasset and his co-workers (1955, 1961).

Interpretation of the results obtained from the experimental work discussed would seem to indicate that the fecal amino acid composition from a highly digestible protein like casein should resemble that of a protein-free diet due to the relative high dilution of exogenous amino acids by endogenous amino acids as a result of the low level of indigestible protein. Findings by Carlson and Bayley (1970), who determined the amino acid availability of a 7, 14 and 21% casein diet in young pigs, seem to substantiate this interpretation. On the contrary, the fecal amino acid composition from a protein with a relatively low digestibility such as protein from cereal grains would more closely resemble that of the indigestible protein portion. The availability of amino acids from proteins in so far as it is dependant on the rate of digestive release and absorption may be determined by the changes of free amino acid concentrations of portal blood. As was discussed, 2 distinct peaks (the first one for exogenous amino acids, the second one for endogenous amino acids) were detected in the portal

blood of dogs that were fed a diet in which casein or an amino acid mixture made up the protein source. Simultaneous absorption of exogenous and endogenous amino acids would likely occur when a more slowly digestible protein was fed (for example, a feed of which the protein digestibility would be similar to that of the endogenous protein). One peak, both for exogenous and endogenous amino acids, would be observed in the portal blood and changes in the levels of free amino acids would not be indicative of the exogenous amino acids alone. Thus, changes in the levels of free amino acids in the blood plasma resulting from the ingestion of protein feeds, as a method for comparing the availabilities of amino acids in different feeds, would only be valid for highly digestible proteins.

#### The influence of microflora on amino acid availability

The use of germ-free laboratory animals provides a means for helping to determine the effect of the microflora on protein metabolism in the gut.

Harmon et al. (1968) studied the influence of the flora on metabolic fecal N excretion with selected levels of egg white protein up to 21.5%, fed as a liquid or dry diet. Metabolic fecal N excretion was much less in germ-free than in normal rats. Gordon and Westmann (1960) reported lower weights of the small intestine in germ-free than in normal rats. The rate of renewal of



the mucosa of the small intestine was reduced in the germ-free rat according to Abrams et al. (1963). Gordon and Bruckner-Kardoss (1961) showed that the surface area of the small intestine was about 30% less in germ-free than in normal rats. Thus, it seems logical that there is a lower output of metabolic fecal N in the germ-free rat because of a reduced renewal rate of intestinal mucosa and less intestinal mucosa because of a reduced intestinal surface area. However, Evrard et al. (1964) showed that trypsin and chymotrypsin, as well as other digestive enzymes, were elevated in the cecal contents and feces of the germ-free rat. As indicated previously, the digestive enzymes make up only a small fraction of the endogenous protein. In summary it seems that the flora, indirectly by their presence, cause a loss of endogenous protein.

Harmon et al. (1968) found that the fecal N excretion (metabolic plus exogenous) of germ-free rats was about twice that of the normal rats for a 21.5% protein liquid diet. The protein source was provided by hydrolyzed casein fortified with amino acids. Levenson and Tennant (1963) found twice as much fecal N in germ-free rats than in normal rats for 13, 20 and 40% dietary protein levels. The source of protein was not specified. Evrard et al. (1964) obtained similar results for a 20% casein diet. Luckey (1963) and Combe et al. (1965) found about 30% more fecal N in germ-free than in normal rats. The

results obtained, in general, indicate that there is a higher excretion of fecal N in germ-free than in normal rats when the dietary protein level is about 20%.

This seems to indicate that the flora are mainly active on indigestible dietary protein. Nesheim and Carpenter (1967) used caecectomy as a tool for assessing bacterial activity in chicks. In chicks, as with rats, most of any such bacterial activity would be expected to occur in the caeca. Freeze-dried cod muscle protein was found to have an apparent N digestibility of 90 and 89% in intact and caecectomized chicks respectively. Heat-damaged cod muscle protein resulted in a N digestibility of 77 and 68% for intact and caecectomized chicks respectively. Nesheim and Carpenter (1967) postulated that, as a result of microbial action on indigested protein entering the cecum, nitrogenous compounds of no nutritional value (mainly ammonia) might be produced and then absorbed. Significant amounts of heat-damaged cod muscle protein may remain in the cecum. Findings by Salter and Coates (1971) support the latter postulate. Proteins which reach the lower intestinal region may undergo reactions that differ from simple proteolysis and result in the absorption of nitrogenous compounds from the gut in forms other than the useful amino acids, as was discussed by Barnes and Kwong (1964). It may be hypothesized that significant quantities of slowly digestible proteins

may remain indigested in the cecum which may then be de-aminated by fermentation so that the values for N digestibility and individual amino acid availabilities can be misleadingly high. Lloyd et al. (1958) determined protein digestibility in whole and caeectomized pigs 8 to 28 weeks of age. The removal of the cecum resulted only in a slight decrease in protein digestibility. Cranwell (1968) criticizes their work because of the fact that the test diets were changed too rapidly. Besides, no account is taken of the microbial fermentation in the remaining part of the large intestine.

Evrard et al. (1964) found that urea accounted for at least 25% of the fecal N in germ-free rats whereas only traces of urea were found in the feces of normal rats. Lepkovsky et al. (1966) determined the distribution of protein and non-protein N in the cecum of germ-free and normal rats. Protein N accounted for 20% of total N in the germ-free rat whereas it accounted for 50% in the normal rat. Levenson and Tennant (1963) reported that 70% of the fecal N from germ-free and about 50% from normal rats was filterable. Increased solubility of the fecal N was also suggested by the work of Harmon et al. (1967) which showed that the percent of total fecal N represented by urea N was greater in germ-free than in conventional rats while the percentage of

chromatographically determined amino acids was higher in normal rats than in germ-free rats. In summary, although the experiments discussed were carried out under widely different experimental conditions (for example, different dietary levels and sources of protein and different methods for sterilization of the diets) they show that the anabolism of N from dietary and/or endogenous sources into bacterial protein may be of a significant proportion.

Michel (1961, 1966) isolated the microbial flora from the gastro-intestinal tract of the pig. In vitro, under conditions as closely as possible resembling the conditions in the intestinal contents, they exerted a considerable catabolic activity towards amino acids which they can deaminate and decarboxylate. Deamination occurring was principally that of amides and non-essential amino acids. Dicarboxylic amino acids decarboxylated most rapidly. Certain indispensable amino acids in particular lysine and methionine were only very slowly degraded. Maximum catabolic activity as to arginine took place in the cecum while the microbial flora capable of degrading aspartic acid and cysteine were mainly located in the ileum. Catabolic activity of the microflora varied considerably from pig to pig. As was shown by Larson and Hill (1960) beginning with the ileum important catabolic activity

can be detected and then from the cecum the catabolic activity will be limited by the small amount of free amino acids. Michel (1961) showed that the colon is the main source of ammonia production, which is derived from autolysis of bacterial protein. Most of the ammonia is absorbed.

In summary, the microbial flora may be active in utilizing indigestible dietary N in the lower end of the gut. It is thought to accomplish this by deamination of amino acids in the ileum and cecum and by incorporation of soluble N into microbial protein which partly autolyzes in the large intestine (mainly in the colon). Both events result in the production and absorption of ammonia and may lead to overestimation of amino acid availabilities as determined by the fecal analysis method. However, the availabilities of some amino acids might even be underestimated. Lysine is an essential amino acid of special importance since it is limiting in many plant proteins. Anderson et al. (1958) have shown that the level of lysine is relatively high in microbial protein. Lysine is not easily degraded by the flora relative to the other essential amino acids as shown in vitro by Michel (1961, 1966). Thus, lysine availability may be underestimated relative to the availabilities of the other amino acids when they are determined by the fecal analysis method.

Payne et al. (1968) suggest that measuring the quantities of amino acids passing the end of the ileum would provide a more meaningful comparison of amino acid availabilities than measurement of amino acids in the feces. Cho et al. (1971)<sup>1</sup> fitted re-entrant cannulae immediately proximal to the ileocecal junction of growing pigs, which were fed a soybean meal diet (17.8% protein) or a rapeseed meal diet (19.2% protein). Time of sampling of ileal digesta, after ingestion of the test diet, was found to influence its amino acid levels very extensively (preliminary findings). Consequently amino acid availability studies, whereby the animal is sacrificed at a certain time after ingestion of the test diet for collection of the digesta, may be a very critical procedure. Although the use of re-entrant cannulae is not very practical for determining the amino acid availabilities of a particular test meal, a definite indication of the extent of microbial modification may be obtained in quantitative terms by comparing the levels of amino acids in ileal digesta and feces.

Factors determining lysine availability and its determination in different foodstuffs by different methods

Lysine is the first limiting amino acid in many foods of plant origin, especially in cereal grains. The nutritive value of cereal protein is thought to be directly related to the proportion of lysine in the

---

<sup>1</sup> Personal communication

protein molecule which has free E-amino groups.

Carpenter (1960) developed a method based on the reactivity of the E-amino group of lysine with fluorodinitrobenzene (FDNB). The test materials are treated with acid and the resulting E-dinitrophenyl lysine (E-DNP-lysine) is taken as a measure of the original available lysine.

Stott and Smith (1966) suggest that "the values for FDNB-reactive lysine might be supposed to represent the maximum available lysine, as being a determination of the lysine not rendered unavailable by binding of the E-amino group". Any other factor lowering the availability of lysine (for example, the overall digestibility of the protein) could be expected to influence biological assays and give rise to a lower available lysine estimate than that given by the FDNB method. They carried out a microbiological assay using the protozoan *Tetrahymena pyriformis* W for measurement of available lysine. Their *Tetrahymena* estimates of available lysine for animal protein sources were similar to those obtained by the FDNB procedure for samples with higher available lysine content ( $>6$  g./16 g.N) but lower than FDNB estimates for samples with lower available lysine content ( $<6$  g./16 g.N). Boctor and Harper (1968) showed a high correlation between lysine availability estimates obtained by the FDNB and the rat-growth assay

method for egg albumen, beef protein and casein.

However, lysine availability values obtained by the FDNB method were higher than those obtained by the rat-growth assay method when these proteins were heat-treated. Ford (1964) found that the availability of lysine was low in a beef protein concentrate, whereas the FDNB method gave higher values. It seems that lysine availability values as determined by the FDNB method correlate well with those obtained by biological methods for proteins in which lysine is of high availability. This does not seem to be true for protein in which lysine is of low availability. In this latter category one might include heat-treated animal proteins and most plant proteins like those of the cereal grains. Besides, for foodstuffs rich in carbohydrate and with a low protein content (for example, cereals), the instability of E-DNP-lysine on acid hydrolysis is a serious problem as was shown by Carpenter and March (1961).

Bjarnason and Carpenter (1969) fed lactalbumen, of which the E-amino groups of the lysine units were propionylated, to young rats. A considerable quantity of E-N-propionyl-L-lysine, which is not utilized by the rat was found in the urine. Valle-Riestra and Barnes (1970) detected E-N-glucose-L-lysine and derivatives of this compound in the urine of rats that were fed egg white protein which was heat-damaged in the presence of glucose. Heating pure proteins (in the absence of



reducing sugars) is thought to give rise to the formation of cross-linkages between the E-amino groups of lysine and the carboxyl groups of the dicarboxylic amino acids or their amides as postulated by Bjarnason and Carpenter (1970). Bjarnason and Carpenter (1969) found no evidence of lysine compounds being absorbed and excreted in the urine of rats that were fed heat-treated bovine plasma albumen. Valle-Riestra and Barnes (1970) fed heat-damaged egg white protein to rats and detected only insignificant amounts of lysine and/or lysine derivatives in the urine (as radioactivity recovered in the urine from dietary  $^{14}\text{C}$ -L-lysine from egg white protein). In summary it seems that lysine, when cross-linked to other amino acids, is resistant to hydrolysis by the digestive enzymes. Lysine may be hydrolyzed by the digestive enzymes from the peptides and absorbed when its E-amino group is merely blocked by carbon atoms as was shown for glucose and propionate.

The FDNB procedure, as previously discussed, does not appear to be suitable for the estimation of lysine availability in heat-treated proteins. Heat treatment causes the formation of cross-linkages between lysine and other amino acids (the intensity of cross-linkage formation depending on the severity of heat treatment). In order to explain this phenomenon one might visualize situations in which the formation of

cross-linkages is to occur mainly on the outside of a protein molecule and that "free" peptide lysine may be trapped on the inside of this protein molecule. One would expect a low digestibility for this particular protein molecule. Trypsin, an important digestive enzyme, does not hydrolyze peptides at the carboxyl group of lysine units if the E-amino group of lysine is cross-linked as was shown by Hill (1965). The FDNB method, which involves acid hydrolysis would take into account the "trapped" lysine as available whereas biological methods for measuring lysine availability would not. The FDNB method might overestimate lysine availability for foodstuffs in which the lysine availability is naturally low (for example, in cereal grains) for the same reason as for the heat-treated proteins.

The availability of lysine is relatively low in cereal grains. Gupta et al. (1958) determined the lysine availability by the rat-growth assay method. Lysine availability was found to be 58 and 72% for corn and wheat flour respectively. Stott and Smith (1966) determined the lysine availability from wheat and barley by a microbiological assay using the protozoan *Tetrahymena pyriformis* W. Lysine availability determined as such varied extensively. It ranged from 32 to 59% in wheat and from 34 to 63% in barley. Calhoun et al. (1960) determined the true availability of

lysine in wheat and wheat gluten. Lysine availability in wheat gluten (99%) was found to be much higher than in wheat (75%) as determined by the fecal analysis method. This would indicate that lysine availability is much lower from the non-gluten protein fraction than from gluten in wheat. One would expect extensive cross-linking of lysine with other amino acids in the non-gluten protein fraction of wheat.

## MATERIALS AND METHODS

Two studies were carried out using Managra<sup>1</sup> pigs in order to determine the biological availability of amino acids by the fecal analysis method from pure barley, triticale, wheat and soybean meal (Table 2). A protein-free diet was employed to estimate the metabolic fecal amino acids (Table 2). Diets used were made into pellets of 0.3 cm. diameter.

Availability Study 1

The first part of this study was carried out in January, 1969. Four barrows from the same litter, weighing 7.3-8.8 kg., were individually assigned to wire cages that were circular in shape (Fig. 1), as described by Bell (1948). Placement of these cages on a metal table with a round hole directly under the centre of the cage permitted separation of urine and feces.

The pigs received a basic diet during the three day adjustment period (Table 2). They were fed at 8 a.m., 12.30 p.m. and 5 p.m. for a 30-60 minute period. The experiment was conducted using a 4 x 4 Latin Square design in order to allow testing of the four different test diets

---

<sup>1</sup> The breed composition of Managra consists of 45% Swedish Landrace, 20% Wessex Saddleback, 15% Welsh and 20% Minnesota No. 1 - Berkshire - Yorkshire - Tamworth.

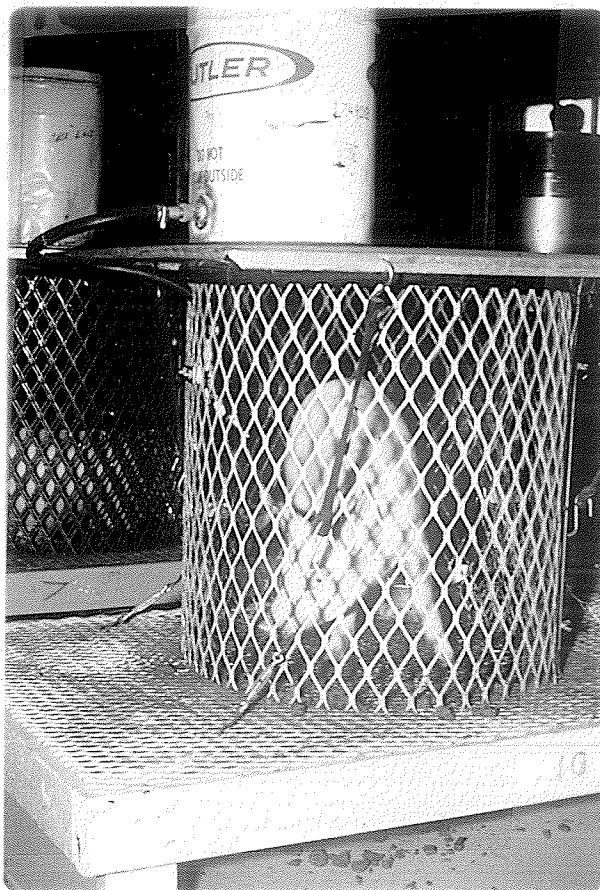


Fig. 1. Metabolic cages for digestibility trials with swine

TABLE 2. Formulation and composition of diets for amino acid availability studies

DIETS	Basic <sup>1</sup>	Soybean meal	Triticale	Wheat	Barley	Protein-free
Ingredients(%):						
Soybean meal	18.0	34.0				
Triticale			100.0			
Wheat	68.0			100.0		
Barley					100.0	
Fish meal	5.0					
Alfalfa	2.0					
Dextrose						
(sucrose)		49.5				75.0
Alphacel		13.2				20.0
Vegetable oil	3.0	3.2				5.0
Defluorinated						
rock phosphate	1.5					
Vitamin premix <sup>2</sup>	1.0					
Mineral premix <sup>3</sup>	0.5					
Ferric oxide	1.0					
Chemical analysis						
(%):						
Nitrogen <sup>4</sup>						
Study 1	2.98	2.46	2.47	2.47	1.81	
Study 1: Triti- cale repeat experiment			2.56			
Study 2	3.00	2.50	2.47	2.18	1.93	

<sup>1</sup> A "modified basic diet", containing 66% of the basic diet and 34% of the protein-free diet, was employed in study 1.

<sup>2</sup> Contains per kg diet: vitamin A (7150IU), vitamin D<sub>3</sub> (818ICU), vitamin E (5.5IU), vitamin B<sub>12</sub> (11 ug), vitamin B<sub>5</sub> premix (5.5 mg riboflavin, 11.0 mg pantothenic acid, 16.5 mg nicotinic acid, 275 mg choline chloride), DL methionine (0.5g), menadione (1.1mg), Santoquin (1.1mg) and penicillin-streptomycin (26.4 mg).

<sup>3</sup> Contains per kg diet: manganese oxide (81.4mg Mn), zinc oxide (11mg Zn), ferrous sulfate (35.2 mg Fe), copper sulfate (6.6mg Cu) and iodized salt (4.7g).

<sup>4</sup> Expressed as a percentage of dry matter

by the four pigs at four different periods. The test diets were fed for one day during each period. The pigs were permitted to consume up to 600 g. of test diet each day and it was attempted to have them consume 200 g. at each meal. They were allowed to drink up to 200 milliliters of water before and after each meal.

The basic diet gave rise to soft feces with a relatively high incidence of diarrhea, initially noted during the adjustment period. Consequently, the basic diet was diluted with the protein-free diet so that the protein content was reduced from 18.8 to 12.4%. This "modified basic diet", which was fed a day before and a day after the test diet, produced drier feces and a lower incidence of diarrhea.

One percent of ferric oxide was included in the original basic diet in order to permit easier identification and collection of feces resulting from the test and protein-free diets.

The protein-free diet was fed for one day following the feeding of the "modified basic diet" for two days, subsequent to the first test period. The pigs were fasted the noon and evening meal prior to the day they were fed the protein-free diet because of its relative unpalatability. The "modified basic diet" was fed again for one day after the protein-free diet and the

rest of the test diet - "modified basic diet" sequence was carried out (Fig. 2).

The experiment described was repeated in February, 1969. Four different barrows of the same litter and of similar weights were used. The protein-free diet, however, was fed immediately after the three day adjustment period.

In both studies it was observed that the consumption of triticale was relatively low compared to that of the other test diets (Table 3). Therefore, a separate availability study was carried out on triticale in which four litter-mate barrows were used, weighing 9.3-13.2 kg. This study was carried out in July, 1970. Triticale devoid of ergot was fed and its consumption increased to a level similar to that for the other test diets (Table 3).

#### Availability Study 2

This study was carried out in December, 1969. Eight barrows from four litters, two from each litter, were subjected to similar experimental conditions as described for study 1. The barrows ranged in weight from 28.6 - 34.1 kg. at the beginning of the experiment following the four day adjustment period.



Fig. 2 Experimental designs for study 1 and 2

		Day	1	2	3	4	5	6	7	8	9	10	11	12	13	14		
STUDY 1	Pig No.1	B.D <sup>1</sup>	B <sup>1</sup>	B.D <sub>M</sub> <sup>1</sup>	P-F <sup>1</sup>	B.D <sub>M</sub>	W <sup>1</sup>	B.D <sub>M</sub>	T <sup>1</sup>	B.D <sub>M</sub>	S <sup>1</sup>	B.D <sub>M</sub>						
	Pig No.2	B.D	S	B.D <sub>M</sub>	P-F	B.D <sub>M</sub>	B	B.D <sub>M</sub>	W	B.D <sub>M</sub>	T	B.D <sub>M</sub>						
	Pig No.3	B.D	T	B.D <sub>M</sub>	P-F	B.D <sub>M</sub>	S	B.D <sub>M</sub>	B	B.D <sub>M</sub>	W	B.D <sub>M</sub>						
	Pig No.4	B.D	W	B.D <sub>M</sub>	P-F	B.D <sub>M</sub>	T	B.D <sub>M</sub>	S	B.D <sub>M</sub>	B	B.D <sub>M</sub>						
		Day	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
STUDY 2	Pig No.1	B.D	P-F	B.D	B	B.D	S	B.D	W	B.D	T	B.D						
	Pig No.2																	
	Pig No.3	B.D	P-F	B.D	T	B.D	B	B.D	S	B.D	W	B.D						
	Pig No.4																	
	Pig No.5	B.D	P-F	B.D	W	B.D	T	B.D	B	B.D	S	B.D						
	Pig No.6																	
	Pig No.7	B.D	P-F	B.D	S	B.D	W	B.D	T	B.D	B	B.D						
	Pig No.8																	

<sup>1</sup> Basal diet (B.D); Modified basal diet (B.D<sub>M</sub>); Protein-free (P-F); Barley (B); Wheat (W); Triticale (T); Soybean meal (S).

TABLE 3. Mean and standard error of the dry matter and nitrogen intake, excretion (g/day) and digestibility for the test and protein-free diets.

DIETS	Soybean meal	Triticale	Wheat	Barley	Protein-free
<b>Study 1</b>					
Number of pigs	8	6	8	8	7
Dry matter intake (g)	387.4±30.5	254.6±23.8	444.6±30.2	468.2±22.5	379.9±40.8
Dry matter excretion (g)	60.6±7.0	35.4±2.4	58.4±6.0	68.3±4.9	52.0±5.5
Dry matter digestibility (%)	84.7±1.0	85.9±0.6	86.8±1.5	85.5±0.6	86.3±0.6
Nitrogen intake (g)	9.5±0.7	6.3±0.6	11.0±0.7	8.5±0.4	
Nitrogen excretion (g)	1.4±0.2	1.1±0.1	1.9±0.2	2.1±0.2	0.8±0.1
True nitrogen digestibility (%)	94.7±2.4	96.0±1.4	90.9±2.1	85.8±1.4	
Apparent nitrogen digestibility (%)	85.5±1.1	82.5±1.0	82.9±1.7	75.8±1.0	
<b>Triticale Repeat Study</b>					
Number of pigs		3			3
Dry matter intake (g)		497.6±5.1			338.8±16.5
Dry matter excretion (g)		61.6±1.5			60.5±4.4
Dry matter digestibility (%)		87.6±0.1			82.0±1.7
Nitrogen intake (g)		12.7±0.1			
Nitrogen excretion (g)		1.9±0.1			0.9±0.1
True nitrogen digestibility (%)		92.0±0.1			
Apparent nitrogen digestibility (%)		85.1±0.3			
<b>Study 2</b>					
Number of pigs	8	8	8	8	7
Dry matter intake (g)	1340.2±28.3	1240.3±87.5	1364.2±74.5	1417.6±36.9	864.4±5.2
Dry matter excretion (g)	142.6±16.6	133.8±17.2	150.6±18.5	174.1±18.8	106.9±7.4
Dry matter digestibility (%)	89.4±0.8	89.3±1.3	89.3±0.9	88.1±1.2	87.6±0.7
Nitrogen intake (g)	33.5±3.2	30.6±2.2	29.7±1.6	27.4±0.7	
Nitrogen excretion (g)	4.0±0.5	4.2±0.5	4.6±0.6	5.5±0.6	0.9±0.1
True nitrogen digestibility (%)	91.6±1.4	89.6±1.7	88.1±1.5	83.7±1.6	
Apparent nitrogen digestibility (%)	88.5±0.9	86.4±1.6	85.0±1.2	80.4±1.5	

The experiment was conducted using a simple cross-over design in which two barrows of the same litter were fed the four test diets at four different periods (Fig.2).

The basic diet did not seem to cause problems in stool formation for the heavier barrows during the adjustment period and it was fed as such in between the days the test and protein-free diets were fed.

Different sources of barley, triticale, wheat and soybean meal were used than in study 1 which resulted in somewhat different levels of nitrogen and amino acids (Table 2, 4 and 7).

The protein-free diet was fed for two days instead of one day as in study 1, after a four day adjustment period.

The pigs were allowed to consume up to 1800 g. of test diet each day over three meals whereby it was attempted to have them consume 600 g. at each meal.

#### Analytical Methods

The feces from the test and protein-free diets were collected and frozen. They were dried at 70°C. to constant weight and ground in a Wiley mill. Amino acid analysis was carried out according to the method of Bragg et al (1966). Modifications included a hydrolysis period of 15 hours and reconstitution to a volume of 100 milliliters (ml.) with a sodium citrate buffer at

p.H. 2.2. One half ml. of each sample was analyzed on a model 116 Beckman amino acid analyzer. Dietary and fecal N content were determined by the macro-Kjeldahl method (Horowitz, 1965). Duplicate analyses were carried out for N and amino acids on the diets while a single analysis was performed on fecal material.

Statistical methods used were analysis of variance, Duncan's multiple range test, determination of missing values and arcsin transformation according to Steel and Torrie (1960).

## RESULTS

The amino acid availabilities from barley, wheat, triticale and soybean meal were determined. The availabilities of tryptophan and cysteine could not be determined since tryptophan is destroyed and cysteine is partially destroyed during acid hydrolysis.

The first study was carried out on Managra barrows that weighed around 10 kg., while the second study was carried out on barrows that weighed around 30 kg. at the start of the experiment.

True amino acid availability from the test diets was calculated according to the equation : % amino acid availability =  $\left[ \text{amino acid intake} - (\text{fecal amino acids} - \text{metabolic fecal amino acids}) \right] \times 100 / \text{amino acid intake}$ . Metabolic fecal amino acids were not considered in the calculation of apparent amino acid availability.

### Study 1

#### 1. Apparent and true availability between test diets

Data from both experiments, which involved different pigs and which were carried out at different times in the form of a Latin Square design, were combined in one statistical analysis. One fecal collection was missing from each Latin Square due to the fact that a low dietary consumption of triticale made it impossible to separate the feces of it from the red coloured feces of

the "modified basic diet". Determination of missing values and their subsequent use in determining the F values in the analysis of variance contribute a bias towards the mean square for treatment. However, it was not necessary to remove this bias in the computations for arriving at the real F value since the F value obtained usually exceeded the 5% level of significance by a wide margin.

Metabolic fecal amino acid excretion was very similar for each of the four pigs in the first experiment (Latin Square 1) of study 1 as shown by the small standard errors (Appendix, Table 11). Therefore, in order to calculate the true amino acid availabilities, individual metabolic fecal amino acid excretions were taken into account for each of the four pigs. However, metabolic fecal amino acid excretion varied extensively between the pigs in the second experiment (Latin Square 2) of study 1 due to one of the pigs having consumed only about half the amount of protein-free diet (180 g.) as compared to the other three pigs. Fecal separation problems were encountered for this particular pig as well. As a result, the average metabolic fecal amino acid excretion of the other three pigs was used in calculating the true amino acid availabilities of the test diets (Appendix, Table 11).

One notices a low consumption of triticales relative to that of the other test diets and a low ratio of N from triticales feces and from the feces derived from the protein-free diet (Table 3). This obviously caused the high true availabilities for the amino acids from triticales (Table 4). There was also a problem in separating the triticales feces from the feces derived from the "modified basic diet". For this reason, true and apparent amino acid availabilities from the triticales repeat experiment were entered in Table 4. Most attention will be paid to these triticales amino acid availability estimates during the presentation of the results and their discussion.

The amino acid availability estimates of the triticales repeat experiment were from three pigs only instead of four. Data from one animal were ignored due to problems in fecal separation because of diarrhea.

a) Apparent availability

The availabilities of six of the ten essential amino acids from soybean meal were significantly higher ( $P < .05$ ) than those from triticales, wheat and barley.

Specifically these amino acids were arginine, isoleucine, leucine, lysine, threonine and valine (Table 4).

Especially remarkable were the large differences in lysine availability between the four test diets. Lysine availability was 82.5, 66.6, 61.6 and 57.6% for soybean meal, triticales (original study), wheat and barley

TABLE 4. Comparison of Apparent and True Availability between test diets in study 1

DIETS	Apparent Availability										True Availability									
	Soybean meal		Triticale		Triticale repeat		Wheat		Harley		Soybean meal		Triticale		Triticale repeat		Wheat		Harley	
	I <sup>2</sup>	II <sup>3</sup>	I	II	I	II	I	II	I	II	I	II	I	II	I	II	I	II	I	II
<b>AMINO ACIDS</b>																				
<b>Essential</b>																				
Arginine	1.11	93.2 <sup>A</sup>	.82	88.6 <sup>B</sup>	.73	88.2	.70	86.8 <sup>B</sup>	.52	82.5 <sup>C</sup>	1.11	98.4 <sup>A</sup>	.82	98.5 <sup>A</sup>	.73	94.5	.70	94.1 <sup>B</sup>	.52	91.4 <sup>B</sup>
Histidine	.38	91.6 <sup>A</sup>	.37	89.3 <sup>AB</sup>	.29	87.0	.32	87.6 <sup>BC</sup>	.21	83.1 <sup>C</sup>	.38	97.9 <sup>A</sup>	.37	99.1 <sup>A</sup>	.29	93.8	.32	94.7 <sup>B</sup>	.21	93.2 <sup>B</sup>
Isoleucine	.64	86.3 <sup>A</sup>	.49	78.4 <sup>B</sup>	.52	83.2	.49	78.7 <sup>B</sup>	.39	72.1 <sup>C</sup>	.68	95.2 <sup>A</sup>	.49	95.8 <sup>A</sup>	.52	91.5	.49	89.2 <sup>B</sup>	.39	84.7 <sup>B</sup>
Leucine	1.26	87.5 <sup>A</sup>	1.04	82.5 <sup>B</sup>	1.08	86.0	1.01	82.4 <sup>B</sup>	.86	79.1 <sup>B</sup>	1.26	95.7 <sup>A</sup>	1.04	96.6 <sup>A</sup>	1.08	93.7	1.01	91.2 <sup>B</sup>	.86	88.7 <sup>B</sup>
Lysine	.78	82.5 <sup>A</sup>	.49	66.6 <sup>B</sup>	.49	72.9	.44	61.6 <sup>B</sup>	.39	57.6 <sup>B</sup>	.78	94.9 <sup>A</sup>	.49	94.4 <sup>A</sup>	.49	86.3	.44	80.8 <sup>B</sup>	.39	77.1 <sup>B</sup>
Methionine	.20	78.5 <sup>A</sup>	.23	78.0 <sup>A</sup>	.18	78.5	.22	77.8 <sup>A</sup>	.19	74.5 <sup>A</sup>	.20	94.5 <sup>AB</sup>	.23	97.0 <sup>A</sup>	.18	90.3	.22	89.8 <sup>B</sup>	.19	87.3 <sup>B</sup>
Phenylalanine	.77	88.4 <sup>A</sup>	.74	85.6 <sup>AB</sup>	.74	88.1	.74	86.4 <sup>A</sup>	.61	82.5 <sup>B</sup>	.77	95.8 <sup>A</sup>	.74	96.5 <sup>A</sup>	.74	94.5	.74	92.4 <sup>B</sup>	.61	90.1 <sup>B</sup>
Threonine	.66	86.0 <sup>A</sup>	.38	71.1 <sup>B</sup>	.47	80.9	.43	75.0 <sup>B</sup>	.38	72.4 <sup>B</sup>	.66	95.4 <sup>A</sup>	.38	94.8 <sup>A</sup>	.47	90.9	.43	87.3 <sup>B</sup>	.38	85.5 <sup>B</sup>
Valine	.70	84.3 <sup>A</sup>	.61	79.1 <sup>B</sup>	.67	84.1	.58	77.8 <sup>B</sup>	.58	77.2 <sup>B</sup>	.70	94.5 <sup>A</sup>	.61	95.6 <sup>A</sup>	.67	92.4	.58	89.0 <sup>B</sup>	.58	87.6 <sup>B</sup>
<b>Non-Essential</b>																				
Alanine	.72	81.9 <sup>A</sup>	.59	72.9 <sup>B</sup>	.64	77.7	.56	70.6 <sup>B</sup>	.49	66.1 <sup>B</sup>	.72	94.1 <sup>A</sup>	.59	94.4 <sup>A</sup>	.64	88.4	.56	84.1 <sup>B</sup>	.49	80.6 <sup>B</sup>
Aspartic Acid	1.83	89.0 <sup>A</sup>	.79	69.2 <sup>B</sup>	1.00	79.5	.83	70.6 <sup>B</sup>	.82	71.8 <sup>B</sup>	1.83	96.6 <sup>A</sup>	.79	94.5 <sup>A</sup>	1.00	91.0	.83	84.6 <sup>B</sup>	.82	85.4 <sup>B</sup>
Glutamic Acid	3.02	92.5 <sup>A</sup>	4.04	92.9 <sup>A</sup>	4.37	94.7	4.71	94.1 <sup>A</sup>	2.68	88.4 <sup>B</sup>	3.02	97.8 <sup>AB</sup>	4.04	98.7 <sup>A</sup>	4.37	97.6	4.71	97.0 <sup>B</sup>	2.68	93.3 <sup>C</sup>
Glycine	.67	83.7 <sup>A</sup>	.62	78.5 <sup>AB</sup>	.65	84.2	.63	79.1 <sup>A</sup>	.48	73.2 <sup>B</sup>	.67	94.0 <sup>A</sup>	.62	94.5 <sup>A</sup>	.65	92.1	.63	88.3 <sup>B</sup>	.48	84.3 <sup>B</sup>
Proline	.79	90.8 <sup>B</sup>	1.60	94.1 <sup>A</sup>	1.55	94.7	1.53	93.6 <sup>A</sup>	1.31	90.3 <sup>B</sup>	.79	96.8 <sup>AB</sup>	1.60	98.2 <sup>A</sup>	1.55	97.1	1.53	96.3 <sup>B</sup>	1.31	93.2 <sup>C</sup>
Serine	.87	90.3 <sup>A</sup>	.63	83.2 <sup>BC</sup>	.71	87.5	.70	86.0 <sup>AB</sup>	.48	80.4 <sup>C</sup>	.87	96.9 <sup>A</sup>	.63	96.8 <sup>A</sup>	.71	94.2	.70	92.8 <sup>B</sup>	.48	89.9 <sup>B</sup>
Tyrosine	.52	85.6 <sup>A</sup>	.41	78.8 <sup>B</sup>	.41	83.2	.41	80.1 <sup>B</sup>	.35	76.6 <sup>B</sup>	.52	95.0 <sup>A</sup>	.41	95.1 <sup>A</sup>	.41	92.9	.41	90.3 <sup>B</sup>	.35	87.6 <sup>B</sup>
AVERAGE <sup>4</sup>	87.3		81.6		84.9		81.3		77.2		96.0		96.6		92.8		90.6		87.8	
NITROGEN	2.46	85.5 <sup>A</sup>	2.47	82.0 <sup>A</sup>	2.56	85.1	2.47	82.9 <sup>A</sup>	1.81	75.8 <sup>B</sup>	2.46	94.7 <sup>AB</sup>	2.47	95.7 <sup>A</sup>	2.56	92.0	2.47	90.9 <sup>BC</sup>	1.81	85.8 <sup>C</sup>
DRY MATTER	84.7 <sup>A</sup>		85.5 <sup>A</sup>		87.6		86.8 <sup>A</sup>		85.5											

<sup>1</sup> Comparison by Duncan's multiple range test at the 5% level of significance<sup>2</sup> I indicates dietary amino acid level (% of dry matter)<sup>3</sup> II indicates the mean amino acid availability (%), calculated on a dry matter basis<sup>4</sup> Simple average of the amino acid availabilities



respectively. The availability of histidine from soybean meal was higher than from triticale but not significantly. Neither was there a significant difference between the phenylalanine availabilities from soybean meal, triticale (original study) and wheat. Methionine availability remained essentially the same regardless of the type of diet fed.

The relative relationships between the apparent amino acid availabilities of the cereal grains were comparable for study 1 and 2 only when the availabilities of the original triticale experiment of study 1 were substituted by those of the triticale repeat experiment of study 1 (Table 4 and 7). Therefore, reference is made to the amino acid availabilities from the triticale repeat experiment for assessing the differences between the essential amino acid availabilities of the cereal grains in study 1. The availabilities of all the essential amino acids that were determined decreased from triticale to wheat to barley. The average amino acid availability decreased from 84.9 to 81.3 to 77.2% for triticale, wheat and barley respectively (Table 4).

There were only small differences between the apparent N digestibilities and the average apparent amino acid availabilities for each diet tested. The N

digestibilities from soybean meal, triticales (original study) and wheat were significantly higher than the N digestibility from barley while there were no significant differences among the dry matter digestibilities of the test diets (Table 4).

The amino acid availabilities from the triticales repeat experiment were generally higher than those from the original triticales study, concomitantly with a higher N and dry matter digestibility (Table 4).

b) True availability

The difference in the percentage of true and apparent average amino acid availability for the triticales repeat study was 7.9 which corresponded well with the differences for the other test diets, while the difference obtained in the original study on triticales was 15% (Table 4). Overestimation of the true amino acid availabilities for the original triticales study were caused by the low intake of triticales relative to that of the protein-free diet (Table 4). Therefore, for comparison of the true amino acid availabilities between the test diets, reference will be made only to those of the triticales repeat experiment.

The true availabilities, like the apparent availabilities, of the amino acids from soybean meal were higher than those from the cereal grains (Table 4).

The average true amino acid availability was 8.7, 7.9, 9.3 and 10.6% higher than the average apparent availability from soybean meal, triticale, wheat and barley respectively. The increase was highest for those amino acids that had a low apparent availability and the lowest for those that had a high apparent availability. For example, the true methionine availability in soybean meal was 16% higher than the apparent availability while the true arginine availability was only 5.3% higher.

The true N digestibility decreased from soybean meal to triticale to wheat to barley as did the average true amino acid availability. The average true amino acid availability was more or less the same as the N digestibility for each specific test diet (Table 4).

## 2. Apparent and true availability within test diets .

Combined data are shown in Table 5 and 6. Again, amino acid availability studies from the triticale repeat experiment have been included in the tables.

Statistical analyses on data of the original triticale study have been carried out on only six fecal collections from triticale due to fecal separation problems in two instances.

### a) Apparent availability .

Methionine, though not significantly different from lysine, was the least available essential

TABLE 5. Comparison of the mean apparent amino acid availabilities<sup>1</sup> within the test diets in study 1

DIETS	Amino Acids <sup>2</sup>															
	Met.	Ala.	Lys.	Gly.	Val.	Tyr.	Thr.	Ile.	Leu.	Phe.	Asp.	Ser.	Pro.	His.	Glu.	Arg.
Soybean meal (8 pigs)	78.5	81.9	82.5	83.7	84.3	85.6	86.0	86.3	87.5	88.4	89.0	90.3	90.8	91.6	92.5	93.2
Triticale (6 pigs)	66.1	70.0	71.9	73.5	78.5	78.8	78.8	79.1	79.4	82.8	83.8	86.1	88.5	89.0	93.1	94.2
Triticale repeat (3 pigs)	72.9	77.7	78.5	79.5	80.9	83.2	83.2	84.1	84.2	86.0	87.0	87.5	88.1	88.2	94.7	94.7
Wheat (8 pigs)	61.6	70.6	70.6	75.0	77.8	77.8	78.7	79.1	80.1	82.4	86.0	86.4	86.8	87.6	93.6	94.1
Harley (8 pigs)	57.6	66.1	71.8	72.1	72.4	73.2	74.5	76.6	77.2	79.1	80.4	82.5	82.5	83.1	88.4	90.3

<sup>1</sup> Means underscored by the same line are not significantly different (determined by Duncan's multiple range test at the 5% level of significance)

<sup>2</sup> Amino acid abbreviations according to Mahler and Cordes (1966)

TABLE 6. Comparison of the mean true amino acid availabilities<sup>1</sup> within the test diets in study 1

DIETS	Amino Acids <sup>2</sup>															
	Gly.	Ala.	Val.	Met.	Lys.	Tyr.	Ile.	Thr.	Leu.	Phe.	Asp.	Pro.	Ser.	Glu.	His.	Arg.
Soybean meal (8 pigs)	94.0	94.1	94.5	94.5	94.9	95.0	95.2	95.4	95.7	95.8	96.6	96.8	96.9	97.8	97.9	98.4
Triticale (6 pigs)	Lys. 94.2	Ala. 94.6	Asp. 94.6	Gly. 94.7	Thr. 95.2	Tyr. 95.3	Val. 95.6	Ile. 96.0	Leu. 96.6	Phe. 96.6	Ser. 96.9	Met. 97.8	Arg. 98.5	Pro. 98.5	His. 98.6	Glu. 98.9
Triticale repeat (3 pigs)	Lys. 86.3	Ala. 88.4	Met. 90.3	Thr. 90.9	Asp. 91.0	Ile. 91.5	Gly. 92.1	Val. 92.4	Tyr. 92.9	Leu. 93.7	His. 93.8	Ser. 94.2	Arg. 94.5	Phe. 94.5	Pro. 97.1	Glu. 97.6
Wheat (8 pigs)	Lys. 80.8	Ala. 84.1	Asp. 84.6	Thr. 87.3	Gly. 88.3	Val. 89.0	Ile. 89.2	Met. 89.8	Tyr. 90.3	Leu. 91.2	Phe. 92.4	Ser. 92.8	Arg. 94.1	His. 94.7	Pro. 96.3	Glu. 97.0
Barley (8 pigs)	Lys. 77.1	Ala. 80.6	Gly. 84.3	Ile. 84.7	Asp. 85.4	Thr. 85.5	Met. 87.3	Val. 87.6	Tyr. 87.6	Leu. 88.7	Ser. 89.9	Phe. 90.1	Arg. 91.4	His. 93.2	Pro. 93.2	Glu. 93.3

<sup>1</sup> Means underscored by the same line are not significantly different (determined by Duncan's multiple range test at the 5% level of significance)

<sup>2</sup> Amino acid abbreviations according to Mahler and Cordes (1966)

amino acid in soybean meal. Arginine and histidine were the most available essential amino acids in soybean meal, which was also the case for the cereal grains (Table 5).

For the cereal grains, lysine was the least available essential amino acid ( $P < .05$ ) with availabilities of 66.1, 61.6 and 57.6% for triticale, wheat and barley respectively. Lysine was also least available in the triticale repeat experiment, namely 72.9%.

Generally, the availabilities of the essential amino acids of the cereal grains can be classified in four groups from low to high availability: 1) Lysine, which is least available, 2) Isoleucine, methionine, threonine and valine which are second least available, 3) Leucine, 4) Arginine, histidine and phenylalanine which are the most available essential amino acids.

For the non-essential amino acids, alanine and glycine were least available in soybean meal. Alanine and aspartic acid were least available in wheat, triticale and barley. Glutamic acid and proline were the most available of the non-essential amino acids of the four test diets.

The sequence of amino acid availability obtained from the triticale repeat experiment did not compare too unfavourably to that obtained from the original triticale experiment in that lysine was least available and arginine and phenylalanine were the most available of the essential

amino acids in both experiments.

b) True availability

The margin between the percentages in availability of the amino acids was much smaller for true than that for apparent availability as was illustrated for soybean meal where the difference between the least and most available amino acid was only 4.8% (Table 6).

The sequence of true availability for the essential amino acids from wheat and barley was identical to that for apparent availability and was almost identical regarding the non-essential amino acids. This was also true for the triticale repeat experiment.

Study 2

1. Apparent and true availability between test diets

The comparisons of true and apparent amino acid availabilities between the test diets for the heavier pigs are shown in Table 7.

One animal did not consume the protein-free diet at all. The metabolic fecal amino acid excretion varied extensively for the other seven pigs as shown by the large standard errors (Appendix, Table 12). Therefore it was decided to use the average metabolic fecal amino acid excretion for calculating the true amino acid availabilities.

TABLE 7. Comparison of Apparent and True Availability between test diets in study 2

DIETS	Apparent Availability								True Availability							
	Soybean meal		Triticale		Wheat		Barley		Soybean meal		Triticale		Wheat		Barley	
	I <sup>2</sup>	II <sup>3</sup>	I	II	I	II	I	II	I	II	I	II	I	II	I	II
AMINO ACIDS																
Essential																
Arginine	1.06	94.7 <sup>A</sup>	.75	92.2 <sup>B</sup>	.64	91.2 <sup>B</sup>	.58	88.1 <sup>C</sup>	1.06	96.0 <sup>A</sup>	.75	94.1 <sup>AB</sup>	.64	93.1 <sup>B</sup>	.58	90.1 <sup>C</sup>
Histidine	.35	93.1 <sup>A</sup>	.30	91.6 <sup>A</sup>	.24	90.8 <sup>A</sup>	.21	87.3 <sup>B</sup>	.35	94.6 <sup>A</sup>	.30	93.3 <sup>A</sup>	.24	92.7 <sup>AB</sup>	.21	89.5 <sup>B</sup>
Isoleucine	.68	89.2 <sup>A</sup>	.50	84.0 <sup>B</sup>	.43	82.0 <sup>BC</sup>	.41	79.1 <sup>C</sup>	.68	92.1 <sup>A</sup>	.50	87.7 <sup>B</sup>	.43	86.1 <sup>B</sup>	.41	83.1 <sup>B</sup>
Leucine	1.27	90.9 <sup>A</sup>	1.10	88.6 <sup>AB</sup>	.94	86.9 <sup>BC</sup>	.90	84.9 <sup>C</sup>	1.27	93.2 <sup>A</sup>	1.10	91.3 <sup>AB</sup>	.94	89.8 <sup>BC</sup>	.90	87.6 <sup>C</sup>
Lysine	.93	88.6 <sup>A</sup>	.45	72.5 <sup>B</sup>	.35	61.3 <sup>C</sup>	.38	60.1 <sup>C</sup>	.93	91.2 <sup>A</sup>	.45	77.5 <sup>B</sup>	.35	67.3 <sup>BC</sup>	.38	65.0 <sup>C</sup>
Methionine	.19	81.7 <sup>A</sup>	.22	83.5 <sup>A</sup>	.20	81.7 <sup>A</sup>	.18	77.4 <sup>A</sup>	.19	86.3 <sup>A</sup>	.22	86.8 <sup>A</sup>	.20	85.7 <sup>A</sup>	.18	81.5 <sup>A</sup>
Phenylalanine	.81	91.7 <sup>A</sup>	.78	91.1 <sup>A</sup>	.91	89.3 <sup>AB</sup>	.65	87.2 <sup>B</sup>	.81	94.2 <sup>A</sup>	.78	93.3 <sup>A</sup>	.91	91.6 <sup>AB</sup>	.65	89.5 <sup>B</sup>
Threonine	.63	88.0 <sup>A</sup>	.48	83.7 <sup>AB</sup>	.40	81.1 <sup>B</sup>	.42	78.7 <sup>B</sup>	.63	91.0 <sup>A</sup>	.48	87.7 <sup>AB</sup>	.40	85.4 <sup>BC</sup>	.42	82.5 <sup>C</sup>
Valine	.71	87.2 <sup>A</sup>	.62	82.9 <sup>B</sup>	.57	81.9 <sup>B</sup>	.57	80.9 <sup>B</sup>	.71	90.9 <sup>A</sup>	.62	86.8 <sup>B</sup>	.57	85.9 <sup>B</sup>	.57	84.6 <sup>B</sup>
Non-Essential																
Alanine	.72	83.3 <sup>A</sup>	.65	77.8 <sup>AB</sup>	.55	71.6 <sup>B</sup>	.56	71.1 <sup>B</sup>	.72	89.2 <sup>A</sup>	.65	82.8 <sup>B</sup>	.55	77.1 <sup>B</sup>	.56	76.1 <sup>B</sup>
Aspartic Acid	2.01	91.4 <sup>A</sup>	1.00	80.5 <sup>B</sup>	.85	80.2 <sup>B</sup>	.83	76.6 <sup>B</sup>	2.01	93.9 <sup>A</sup>	1.00	84.9 <sup>B</sup>	.85	86.8 <sup>B</sup>	.83	81.1 <sup>B</sup>
Glutamic Acid	3.14	93.9 <sup>A</sup>	4.50	95.0 <sup>A</sup>	4.17	94.8 <sup>A</sup>	3.00	91.5 <sup>B</sup>	3.14	95.6 <sup>A</sup>	4.50	96.2 <sup>A</sup>	4.17	96.0 <sup>A</sup>	3.00	92.9 <sup>B</sup>
Glycine	.70	87.5 <sup>A</sup>	.67	85.2 <sup>A</sup>	.60	83.8 <sup>A</sup>	.54	81.2 <sup>A</sup>	.70	91.0 <sup>A</sup>	.67	88.7 <sup>A</sup>	.60	87.5 <sup>A</sup>	.54	85.0 <sup>A</sup>
Proline	.82	92.5 <sup>B</sup>	1.53	95.2 <sup>A</sup>	1.35	94.9 <sup>A</sup>	1.19	92.6 <sup>B</sup>	.82	94.8 <sup>BC</sup>	1.53	96.4 <sup>A</sup>	1.35	96.2 <sup>AB</sup>	1.19	93.9 <sup>C</sup>
Serine	.87	92.3 <sup>A</sup>	.74	90.7 <sup>AB</sup>	.67	89.1 <sup>B</sup>	.57	85.6 <sup>C</sup>	.87	94.6 <sup>A</sup>	.74	93.1 <sup>AB</sup>	.67	91.6 <sup>B</sup>	.57	88.5 <sup>C</sup>
Tyrosine	.51	90.4 <sup>A</sup>	.42	86.5 <sup>B</sup>	.36	84.0 <sup>BC</sup>	.35	81.4 <sup>C</sup>	.51	93.1 <sup>A</sup>	.42	89.7 <sup>B</sup>	.36	87.5 <sup>BC</sup>	.35	85.0 <sup>C</sup>
AVERAGE <sup>4</sup>	90.0		86.9		84.8		82.1		92.8		89.9		88.2		85.3	
NITROGEN	2.50	88.5 <sup>A</sup>	2.47	86.4 <sup>AB</sup>	2.18	85.0 <sup>B</sup>	1.93	80.4 <sup>C</sup>	2.50	91.6 <sup>A</sup>	2.47	89.6 <sup>AB</sup>	2.18	88.1 <sup>B</sup>	1.93	83.7 <sup>C</sup>
DRY MATTER	89.4 <sup>A</sup>		89.3 <sup>A</sup>		89.3 <sup>A</sup>		88.1 <sup>A</sup>									

<sup>1</sup> Comparison by Duncan's multiple range test at the 5% level of significance

<sup>2</sup> I indicates dietary amino acid level (% of dry matter)

<sup>3</sup> II indicates the mean amino acid availability (%), calculated on a dry matter basis

<sup>4</sup> Simple average of the amino acid availabilities



a) Apparent availability

But for methionine, the availabilities of all essential amino acids studied in soybean meal were higher than those from the cereal grains. However, this was only true for arginine, isoleucine, lysine and valine on a significant ( $P < 0.05$ ) basis. There were no differences in availability for histidine and phenylalanine between soybean meal, triticale and wheat. Neither were there significant differences ( $P < 0.05$ ) for leucine and threonine between soybean meal and triticale. As in study 1, methionine availability did not seem to depend on the diets used (Table 7).

Numerous differences in amino acid availabilities between the cereal grains were noted. From all the essential amino acids studied, only the availability of lysine was significantly higher ( $P < 0.05$ ) in triticale than in wheat. But for threonine and valine, the availabilities of the essential amino acids in triticale were significantly higher ( $P < 0.05$ ) than those from barley. The availabilities of arginine and histidine in wheat were significantly higher ( $P < 0.05$ ) than those from barley.

The availabilities of the non-essential amino acids in soybean meal were higher than those from the cereal grains, excepting glutamic acid and proline whose availabilities were highest in triticale and wheat.

Nitrogen digestibility followed the average amino acid availability very closely for each of the test diets as in study 1. There were no significant differences in dry matter digestibilities between the test diets (Table 7).

Although the individual availability estimates of the amino acids in this study differed to some extent from those in study 1 in each of the test diets, the relative relationship of the amino acids between the test diets was much the same (Table 4 and 7).

b) True availability

The average true amino acid availability was 2.8, 3.0, 3.4 and 3.2% higher than the average apparent amino acid availability for soybean meal, triticale, wheat and barley respectively and they closely followed the true N digestibility (as in study 1), which was about 3% higher than the apparent N digestibility (Table 7).

The differences between the average apparent and true amino acid availabilities were much smaller in this study than in study 1. Relatively, there was a higher ratio of N excreted in the feces of the test diets to that in the feces of the protein-free diet in this study (Table 3).

Generally, similar significant trends were noted for the true and apparent amino acid availabilities from the four test diets.

2. Apparent and true availability within test diets

The comparisons of true and apparent amino acid availability within the test diets are shown in table 8 and 9.

a) Apparent availability

Lysine was significantly ( $P < 0.05$ ) the least available essential amino acid in triticale, wheat and barley. Methionine was the least available essential amino acid in soybean meal (Table 8).

A remarkable similarity in the sequence for amino acid availability was noted for the cereal grains. The sequence was the same except for isoleucine, methionine, threonine and valine but regarding their availabilities these amino acids were not significantly different from each other with each cereal grain.

The availabilities of the essential amino acids in the cereal grains studied can be classified in the same groups as described for study 1.

Alanine was the least available non-essential amino acid, glutamic acid and proline were the most available in all diets tested. Aspartic acid was the second least available non-essential amino acid in the cereal grains. On the contrary, aspartic acid had a relatively high availability in soybean meal.

TABLE 8. Comparison of the mean apparent amino acid availabilities<sup>1</sup> within the test diets in study 2 (means for eight pigs)

DIETS	Amino Acids <sup>2</sup>															
	Met.	Ala.	Val.	Gly.	Thr.	Lys.	Ile.	Tyr.	Leu.	Asp.	Phe.	Ser.	Pro.	His.	Glu.	Arg.
Soybean meal	81.7	83.3	87.2	87.5	88.0	88.6	89.2	90.4	90.9	91.4	91.7	92.3	92.5	93.1	93.9	94.7
Triticale	Lys. 72.5	Ala. 77.8	Asp. 80.5	Val. 82.9	Met. 83.5	Thr. 83.7	Ile. 84.0	Gly. 85.2	Tyr. 86.5	Leu. 88.6	Ser. 90.7	Phe. 91.1	His. 91.6	Arg. 92.2	Glu. 95.0	Pro. 95.2
Wheat	Lys. 61.3	Ala. 71.6	Asp. 80.2	Thr. 81.1	Met. 81.7	Val. 81.9	Ile. 82.0	Gly. 83.8	Tyr. 84.0	Leu. 86.9	Ser. 89.1	Phe. 89.3	His. 90.8	Arg. 91.2	Glu. 94.8	Pro. 94.9
Barley	Lys. 60.1	Ala. 71.1	Asp. 76.6	Met. 77.4	Thr. 78.7	Ile. 79.1	Val. 80.9	Gly. 81.2	Tyr. 81.4	Leu. 84.9	Ser. 85.6	Phe. 87.2	His. 87.3	Arg. 88.1	Glu. 91.5	Pro. 92.6

<sup>1</sup> Means underscored by the same line are not significantly different (determined by Duncan's multiple range test at the 5% level of significance)

<sup>2</sup> Amino acid abbreviations according to Mahler and Cordes (1966)

TABLE 9. Comparison of the mean true amino acid availabilities<sup>1</sup> within the test diets in study 2 (means for eight pigs)

	Met. <sup>2</sup>	Ala.	Val.	Gly.	Thr.	Lys.	Ile.	Tyr.	Leu.	Asp.	Phe.	Ser.	His.	Pro.	Glu.	Arg.
Soybean meal	86.3	89.2	90.9	91.0	91.0	91.2	92.1	93.1	93.2	93.9	94.2	94.6	94.6	94.8	95.6	96.0
<hr/>																
	Lys.	Ala.	Asp.	Val.	Met.	Thr.	Ile.	Gly.	Tyr.	Leu.	Ser.	Phe.	His.	Arg.	Glu.	Pro.
Triticale	77.5	82.8	84.9	86.8	86.8	87.7	87.7	88.7	89.7	91.3	93.1	93.3	93.3	94.1	96.2	96.4
<hr/>																
	Lys.	Ala.	Thr.	Met.	Val.	Ile.	Asp.	Gly.	Tyr.	Leu.	Ser.	Phe.	His.	Arg.	Glu.	Pro.
Wheat	67.3	77.1	85.4	85.7	85.9	86.1	86.8	87.5	87.5	89.8	91.6	91.6	92.7	93.1	96.0	96.2
<hr/>																
	Lys.	Ala.	Asp.	Met.	Thr.	Ile.	Val.	Gly.	Tyr.	Leu.	Ser.	Phe.	His.	Arg.	Glu.	Pro.
Barley	65.0	76.1	81.1	81.5	82.5	83.1	84.6	85.0	85.0	87.6	88.5	89.5	89.5	90.1	92.9	93.9

<sup>1</sup> Means underscored by the same line are not significantly different (determined by Duncan's multiple range test at the 5% level of significance)

<sup>2</sup> Amino acid abbreviations according to Mahler and Cordes (1966)

b) True availability

The sequence for the amino acids in true availability was nearly identical to that for apparent availability for all diets studied (Table 9).

## DISCUSSION

Comparisons between the dietary amino acid availabilities when the dry matter intake or excretion of the protein-free diet is made equal to that of each test diet

As was pointed out in the literature review, it is important to have the same amount of indigestible material in the protein-free and test diet in order to determine the true N digestibility and the true amino acid availabilities of a protein (or protein source). Besides, the test animals should consume the same amount of both diets.

Cellulose was included at a level of 13.2 and 20% in the soybean meal and protein-free diet respectively in order to obtain about equal dry matter digestibilities for the test and protein-free diets (Table 2). Although the dry matter digestibilities were nearly similar, the amount of dry matter excreted differed widely due to the difference in dietary consumption of the test and protein-free diets (Table 3). Therefore, calculations for true amino acid availability estimates were carried out in which the amount of each metabolic fecal amino acid, as determined by feeding the protein-free diet, was corrected for equal consumption and dry matter excretion of the protein-free diet and each test diet. The correction factors are given in Table 10, while the corrected true amino acid

TABLE 10. Correction factors for equal dry matter excretion and consumption of the protein-free and test diet and the corrected nitrogen digestibilities and lysine availabilities

Items	Dry matter intake (g)	C.F. 1	Dry matter excretion (g)	C.F. 2	True Nitrogen Digestibility (%)			True Lysine Availability (%)		
					A <sup>3</sup>	B <sup>4</sup>	C <sup>5</sup>	A <sup>3</sup>	B <sup>4</sup>	C <sup>5</sup>
Study 1										
Soybean meal	387.4	1.02	60.6	1.17	93.7	93.9	95.2	93.4	93.8	95.6
Triticale	254.6	0.67	35.4	0.68	95.2	91.1	91.1	93.6	84.8	85.1
Wheat	444.6	1.17	58.4	1.12	90.0	91.3	90.9	79.6	82.8	81.9
Barley	468.6	1.23	68.3	1.31	84.7	86.8	87.6	76.5	81.1	82.7
Protein-free	380.0		52.0							
Triticale repeat										
Triticale	497.6	1.47	61.6	1.00	92.1	95.3	92.1	86.5	93.0	86.5
Protein-free	338.8		60.5							
Study 2										
Soybean meal	1340.2	1.54	142.6	1.33	90.7	92.2	91.6	90.1	91.3	90.9
Triticale	1240.3	1.43	133.8	1.25	89.2	90.5	90.0	76.3	78.3	77.6
Wheat	1364.2	1.57	150.6	1.41	87.5	89.3	88.8	66.5	69.6	68.8
Barley	1417.6	1.63	174.1	1.63	83.2	85.3	85.3	64.2	67.2	67.2
Protein-free	864.4		106.9							

- 1 C.F.<sub>1</sub> indicates correction factor 1, which is the ratio of dry matter intake of the test and protein-free diet
- 2 C.F.<sub>2</sub> indicates correction factor 2, which is the ratio of dry matter excretion from the test and protein-free diet
- 3 Uncorrected average true N digestibility or lysine availability
- 4 Average true N digestibility or lysine availability corrected for C.F.<sub>1</sub>
- 5 Average true N digestibility or lysine availability corrected for C.F.<sub>2</sub>



TABLE 11. Average true amino acid availability (%) corrected for equal dry matter excretion and consumption of the protein-free and test diet in study 1

DIETS	Soybean meal(8) <sup>1</sup>				Triticale (6)				Wheat(8)				Harley (8)				Triticale Repeat(3)			
	A <sup>2</sup>	B <sup>3</sup>	C <sup>4</sup>		A <sup>2</sup>	B <sup>3</sup>	C <sup>4</sup>		A <sup>2</sup>	B <sup>3</sup>	C <sup>4</sup>		A <sup>2</sup>	B <sup>3</sup>	C <sup>4</sup>		A <sup>2</sup>	B <sup>3</sup>	C <sup>4</sup>	
Amino Acids																				
Essential																				
Arginine	97.9	98.6	97.9		96.7	95.0	94.9		92.6	93.4	93.7		81.0	93.3	92.6		94.2	94.2	97.5	
Histidine	95.9	97.4	96.5		98.9	96.0	96.0		93.0	94.2	94.4		90.8	93.9	93.2		93.4	93.4	97.2	
Isoleucine	93.2	94.6	93.5		95.2	90.3	90.1		87.2	88.4	88.8		83.6	86.9	86.0		91.5	91.5	95.4	
Leucine	94.9	96.1	94.9		98.1	93.4	93.2		90.9	91.8	92.2		84.6	87.5	86.7		93.7	93.8	97.2	
Lysine	93.4	95.6	93.8		93.6	85.1	84.8		79.6	81.9	82.8		76.5	82.7	81.1		86.5	86.5	93.0	
Methionine	91.0	93.7	91.5		100.0	93.9	93.7		87.8	89.2	89.8		87.6	91.0	90.0		91.1	91.1	96.7	
Phenylalanine	96.3	97.7	96.5		97.9	94.3	94.2		93.0	94.0	94.3		89.2	91.8	91.2		94.6	94.6	97.6	
Threonine	93.8	95.2	94.1		90.7	84.0	83.8		84.8	86.4	86.9		83.7	87.2	86.3		90.6	90.6	95.3	
Valine	93.7	95.5	94.0		96.1	90.8	90.6		87.2	88.6	89.1		85.3	88.4	87.6		92.5	92.5	96.4	
Non-Essential																				
Alanine	93.2	94.9	93.2		94.0	87.0	86.8		82.7	84.2	84.9		79.6	83.5	82.4		88.4	88.4	93.4	
Aspartic Acid	95.8	97.0	95.9		95.0	87.0	86.7		83.7	85.5	86.2		84.4	88.4	87.4		90.4	90.4	95.4	
Glutamic Acid	97.1	97.9	97.2		98.6	96.9	96.8		96.6	97.0	97.1		92.8	94.2	93.8		97.5	97.5	98.9	
Glycine	91.9	93.7	92.3		93.0	88.2	88.0		91.1	88.5	88.9		83.1	86.4	85.5		92.0	92.0	95.7	
Proline	95.8	96.7	95.8		98.5	97.0	97.0		96.2	96.4	96.6		93.0	93.8	93.5		97.1	97.1	98.3	
Serine	95.8	96.8	96.0		94.4	90.4	90.3		91.3	92.0	92.3		88.0	90.2	89.6		94.3	94.3	97.5	
Tyrosine	93.5	94.4	93.3		93.3	88.2	88.1		89.6	90.5	91.0		85.4	87.9	87.2		93.1	93.1	97.5	
Nitrogen	93.7	95.2	93.9		95.2	91.1	91.1		90.0	90.9	91.3		84.7	87.6	86.8		92.1	92.1	95.3	

<sup>1</sup> Numbers in parentheses indicate the number of pigs from which the average true availabilities were determined.

<sup>2</sup> Uncorrected average true availability

<sup>3</sup> Average true availability corrected for equal dry matter excretion of the protein-free and test diet

<sup>4</sup> Average true availability corrected for equal consumption of the protein-free and test diet

TABLE 12. Average true amino acid availability (%) corrected for equal dry matter excretion and consumption of the protein-free and test diet in study 2

DIETS	Soybean meal (8) <sup>1</sup>			Triticale (8)			Wheat (8)			Barley (4)		
	A <sup>2</sup>	B <sup>3</sup>	C <sup>4</sup>	A <sup>2</sup>	B <sup>3</sup>	C <sup>4</sup>	A <sup>2</sup>	B <sup>3</sup>	C <sup>4</sup>	A <sup>2</sup>	B <sup>3</sup>	C <sup>4</sup>
<b>Amino Acids</b>												
<u>Essential</u>												
Arginine	95.5	95.8	96.1	93.5	94.0	94.3	92.6	93.4	93.6	89.8	91.0	91.0
Histidine	93.8	94.2	94.5	93.0	93.5	93.8	92.0	92.7	93.0	88.9	90.3	90.3
Isoleucine	91.3	92.1	92.6	86.0	86.9	87.4	84.0	85.5	86.2	82.3	84.7	84.7
Leucine	92.5	93.1	93.5	90.8	91.4	91.9	89.4	90.5	90.9	87.3	88.9	88.9
Lysine	90.1	90.9	91.3	76.3	77.6	78.3	66.5	68.8	69.6	64.2	67.2	67.2
Methionine	83.9	85.1	85.9	86.1	87.2	87.5	85.7	87.2	87.9	81.2	83.5	83.5
Phenylalanine	93.7	94.4	94.8	92.9	93.4	93.8	93.5	94.2	94.4	89.0	90.4	90.4
Threonine	90.0	90.9	91.5	86.7	87.7	88.2	84.8	86.4	87.2	82.0	84.4	84.4
Valine	89.4	90.3	91.0	85.8	86.7	87.4	85.0	86.4	87.0	83.4	85.6	85.6
<u>Non-Essential</u>												
Alanine	86.3	87.6	88.4	82.3	83.4	84.2	76.4	78.4	79.2	75.4	78.3	78.3
Aspartic Acid	93.2	93.7	94.0	83.9	85.6	86.2	83.6	85.3	86.1	80.4	83.1	83.1
Glutamic Acid	95.0	95.5	95.8	96.0	96.2	96.4	95.7	96.2	96.3	92.6	93.5	93.5
Glycine	89.9	90.8	91.5	88.2	89.0	89.7	86.7	88.0	88.5	84.3	86.6	86.6
Proline	94.1	94.7	95.1	96.2	96.5	96.7	95.8	96.3	96.4	93.5	94.3	94.3
Serine	93.8	94.4	94.8	92.7	93.4	93.7	91.0	92.0	92.5	87.7	89.5	89.5
Tyrosine	92.4	93.1	93.7	88.7	89.4	90.0	86.6	88.0	88.4	83.5	85.5	85.5
Nitrogen	90.7	91.6	92.2	89.2	90.0	90.5	87.5	88.8	89.3	83.2	85.3	85.3

<sup>1</sup> Numbers in parentheses indicate the number of pigs from which the average true availabilities were determined.

<sup>2</sup> Uncorrected average true availability

<sup>3</sup> Average true availability corrected for equal dry matter excretion of the protein-free and test diet

<sup>4</sup> Average true availability corrected for equal consumption of the protein-free and test diet.

availabilities are given in Table 11 for study 1 and in Table 12 for study 2. Average dietary consumption and fecal excretion of amino acids for each of the test diets and the average metabolic fecal amino acid excretion were used in order to minimize computations (Appendix, Tables 11, 12 and 13).

Corrections of metabolic fecal amino acid levels by factors larger than 1 increased true amino acid availability estimates, those smaller than 1 decreased these estimates. The least available amino acids decreased or increased the most. Both types of corrections resulted in nearly similar true N digestibility estimates except for the triticale repeat experiment where they differed by 3.2% (Table 10). Presumably too much indigestible fecal dry matter was collected in this case from the protein-free diet (page 69). The dry matter digestibility of the protein-free diet from the triticale repeat experiment was about 5% lower than those obtained from study 1 and 2 (Table 3) and correction for equal intake of the protein-free diet and triticale increased the metabolic fecal N excretion once more so that it resulted in a true N digestibility estimate that was even higher than those from soybean meal (Table 10). Therefore, the true N digestibility and true amino acid availability estimates obtained by correcting for equal dry matter excretion will be cited from the triticale

repeat experiment when compared to those obtained from triticale in study 2.

The corrected true N digestibility estimates<sup>1</sup> between studies 1 and 2 for the same diets were closer than the original estimates, i.e. 1.7, 1.6, 2.0 and 1.5% for soybean meal, triticale, wheat and barley respectively (Tables 10, 11 and 12). However, the corrections failed to make the true availability estimates of some amino acids, in particular those of lysine from the cereal grains, more comparable (Tables 10, 11 and 12).

The corrected metabolic fecal N and amino acid levels are given in Table 13 for each of the diets. About 50% more N was excreted by the pigs from study 2, which were about 20 kg heavier than the pigs from study 1 during the course of the experiments. Whiting and Bezeau (1957) reported that for each unit increase in body weight (kg) to the 0.3 power, there was a 0.12 g decrease in metabolic fecal N excretion per 100 g dry matter intake in diets containing between 5 and 18% protein (by extrapolation of the regression line of fecal N on N intake). However, by direct measurements using low protein diets (0.3%N), they found a 0.06 g decrease in metabolic fecal N excretion per 100 g dry matter intake.

---

<sup>1</sup> Compared on a basis of equal dry matter consumption of the protein-free and test diet in case of soybean meal, wheat and barley. Triticale in study 1 from the triticale repeat experiment (corrected for equal dry matter excretion) and triticale from study 2 (corrected for equal intake) were compared.

TABLE 13. Comparison of the corrected metabolic fecal amino acid excretion (g) from the lighter (study 1) and heavier pigs (study 2)

DIETS	Soybean meal		Triticale <sup>1</sup>		Wheat		Barley	
EXPERIMENT	1(7) <sup>2</sup>	2(7)	1(3)	2(7)	1(7)	2(7)	1(7)	2(7)
Amino Acids <sup>3</sup>								
<u>Essential</u>								
Arginine	0.21	0.25	0.23	0.23	0.24	0.25	0.25	0.26
Histidine	0.09	0.09	0.10	0.09	0.10	0.09	0.10	0.10
Isoleucine	0.21	0.34	0.22	0.31	0.24	0.35	0.25	0.36
Leucine	0.38	0.52	0.41	0.49	0.44	0.53	0.46	0.55
Lysine	0.36	0.40	0.33	0.37	0.41	0.41	0.44	0.42
Methionine	0.11	0.15	0.11	0.14	0.13	0.16	0.13	0.16
Phenylalanine	0.23	0.32	0.24	0.30	0.26	0.33	0.28	0.34
Threonine	0.21	0.34	0.23	0.31	0.24	0.35	0.25	0.36
Valine	0.28	0.43	0.28	0.40	0.32	0.44	0.34	0.46
Subtotal	2.08	2.84	2.15	2.64	2.38	2.91	2.50	3.01
<u>Non-Essential</u>								
Alanine	0.32	0.57	0.34	0.53	0.37	0.58	0.39	0.60
Aspartic Acid	0.51	0.77	0.54	0.71	0.58	0.79	0.61	0.82
Glutamic Acid	0.59	0.94	0.62	0.87	0.68	0.96	0.71	0.99
Glycine	0.25	0.42	0.25	0.39	0.28	0.42	0.30	0.44
Proline	0.18	0.32	0.19	0.30	0.21	0.33	0.22	0.34
Serine	0.18	0.34	0.24	0.31	0.21	0.35	0.22	0.36
Tyrosine	0.15	0.25	0.20	0.23	0.17	0.25	0.18	0.26
Subtotal	2.18	3.61	2.38	3.34	2.50	3.68	2.63	3.81
Total Amino Acids	4.26	6.45	4.53	5.98	4.88	6.59	5.13	6.82
Nitrogen	0.82	1.39	0.91	1.28	0.94	1.41	0.98	1.47

<sup>1</sup> Comparison of metabolic fecal amino acid excretion from the triticale repeat experiment in study 1 with that of study 2

<sup>2</sup> Numbers in parentheses indicate the number of pigs from which the average metabolic fecal amino acid excretion was determined.

<sup>3</sup> Amino acid excretion expressed as grams per day

The lighter pigs from study 1 excreted 0.211 g of metabolic fecal N per 100 g dry matter intake as compared to 0.104 g for the heavier pigs from study 2 (Table 3). A change in weight from 10 to 35 kg constitutes one unit increase in body weight to the 0.3 power, which corresponds approximately to the weight difference between the pigs from study 1 and 2. Thus, the difference obtained in metabolic fecal N excretion per 100 g dry matter intake between the pigs from study 1 and 2 compared well with the one obtained by Whiting and Bezeau (1957) by extrapolation but was higher than the one that they obtained directly by feeding a low protein diet.

The corrected metabolic fecal lysine excretion did not increase in the same proportion as total N from the lighter to the heavier pigs (Table 13). This may explain to a certain extent the lower corrected true lysine availabilities, in particular those of wheat and barley, of the test diets in study 2. The corrected true lysine availabilities were 82.8 and 81.1% in study 1 and 69.6 and 67.2% in study 2 for wheat and barley respectively (Table 10 and 12). Assuming that lysine would increase proportionally to total N from the younger to the older barrows, the recorrected metabolic fecal lysine excretion would be 0.61 g<sup>1</sup> from wheat. The resulting recorrected

---

<sup>1</sup>  $1.41/0.94 \times 0.41 = 0.61$  (Table 13)

true lysine availability from wheat would then be 73.0%<sup>1</sup> which differs now by only 9.8% (instead of 13.2%) from that of study 1 (Table 10 and 13). Several reasons may be postulated as to why the metabolic fecal lysine excretion did not increase in the same proportion as total N from the younger to the older pigs. First of all, the protein-free diet was fed for 2 days in study 2 as compared to 1 day in study 1. The pigs in study 2 might have adapted to the feeding of the protein-free diet by the second day by losing less metabolic fecal lysine. This adaptation (if it occurred) seemed to be true for the essential amino acids as a whole, because the increase in total N excretion was mainly due to increased losses of the non-essential amino acids (Table 13). Secondly, the intestinal flora change with age as was shown by Michel (1961). This might have been another reason for the difference in metabolic fecal amino acid composition between that of study 1 and study 2 (Table 14 and 15).

Whiting and Bezeau (1957) reported that, as the pigs increased in weight from 15 to 50 kg, the apparent digestibility of a protein increased. This was due to

---

<sup>1</sup> The average fecal lysine excretion from wheat was 1.90 g (Appendix, Table 12); the recorrected metabolic fecal lysine excretion was 0.61 g (Table 13); the average dietary lysine consumption from wheat was 4.77 g (Table 2 and 7).

the fact that the metabolic fecal N excretion per 100 g dry matter consumed, decreased in proportion to the 0.3 power of body weight. The apparent N digestibility was 3.0, 1.3<sup>1</sup>, 2.1 and 4.6% higher in study 2 than in study 1 for soybean meal, triticale, wheat and barley respectively (Table 4 and 7) and confirms the observations by Whiting and Bezeau. However, as was shown by Whiting and Bezeau (1957), the true protein digestibility (at dietary levels of 5 to 22% from wheat, skimmilk powder and fishmeal) did not change significantly as the pigs increased in weight from 15 to 60 kg. The true N digestibilities, corrected for equal intake of the test and protein-free diet, were slightly higher in study 1 than in study 2. For example, the true N digestibility of wheat was 2% higher in study 1 (Table 10). The recorrected true lysine availability from wheat in study 2 would be 76%<sup>2</sup>, if the true N digestibility of wheat in study 2 is made equal to that

---

<sup>1</sup> Between triticale from the triticale repeat experiment in study 1 and triticale in study 2.

<sup>2</sup> The average fecal lysine excretion from wheat was 1.90 g (Appendix, Table 12); the corrected metabolic fecal lysine excretion was 0.41 g (Table 13); the recorrected N excretion would be  $100 - 91.3 = 8.7$  g instead of  $100 - 89.3 = 10.7$  g per 100 g of dietary N in study 2 (Table 10) and the recorrected dietary lysine loss would then be  $\frac{8.7}{10.7} (1.90 - 0.41) = 1.15$  g; the average dietary lysine intake from wheat was 4.77 g (Table 2 and 7).



of wheat in study 1, which differs now by only 6.8% from the corrected true lysine availability of wheat in study 1 (Table 10). Thus, the relatively large differences in true lysine availabilities from the cereal grains between study 1 and 2 may be attributed partly to the relatively smaller differences in true N digestibilities between both studies for each of the cereal grains. Corrections on a basis of equivalent N digestibility would make the true availabilities of the least available amino acids, since these increase or decrease the most, more comparable between both studies for the cereal grains.

Significant differences ( $P < 0.05$ ) were found for dry matter digestibilities between the test periods in study 2. Consequently, significant differences ( $P < 0.05$ ) were found for N digestibility and amino acid availabilities (Appendix, Tables 3, 4 and 14). However, no significant differences ( $P < 0.05$ ) were found between the test diets regarding their dry matter digestibilities and they can thus not be held responsible for the differences in N digestibility and amino acid availabilities that were found between the test diets.

The dry matter digestibility decreased from 92.1% in test period I to 86.5% in test period IV. Inclusion of ferric oxide in the basic diet permits easier identification and collection of feces resulting from the test and protein-free diets. However, a gradual colour

change is usually observed from red to that of the colour of the feces from the test diets. Therefore, N digestibility and amino acid availabilities from a test diet will also be dependent on the colour appraisal by the individual collecting the feces. The author has to admit to have erred by being inconsistent in colour appraisal over the 4 test periods in separating the red coloured feces of the basic diet from that of the feces of the test diets in study 2. The individual<sup>1</sup>, who collected and separated feces in study 1, did not commit this error.

The so-called "colour appraisal factor" is an error which is inherent in this type of experimental work. This error might be minimized by feeding the test diets for longer periods instead of only 1 day. It was not possible to feed the test diets for more than 1 day in study 2. The pigs, which weighed around 30 kg at the start of the experiment, would have been too big for the cages by the end of the experiment. It would have been possible to feed the test diets for 2 days in the first study which involved pigs that weighed around 10 kg at the start of the experiment.

In future availability research, it would be advisable to use a different basic diet, for example, a basic diet containing casein as the protein source. The

---

<sup>1</sup> Dr P.M. Giovannetti

amino acids from casein are highly available as was shown by Carlson and Bayley (1970). Contamination of unavailable amino acids in the feces from the test diets by the unavailable amino acids in the feces of the basic diet would then be minimized.

Comparisons between the dietary and fecal amino acid levels when expressed as g amino acid/16 g nitrogen and its implications

The fecal levels (g/16 g N) of arginine, histidine, phenylalanine, glutamic acid, proline and serine, corrected or not corrected for metabolic fecal contributions, were lower than the dietary levels in both study 1 and study 2 (Table 14 and 15). The availabilities of these amino acids were higher than the total N digestibility and were usually the most available amino acids of the four test diets (Table 4 and 7). These observations may indicate: 1) the indigestible N residues contain low levels of these amino acids relative to the digested and absorbed N fractions from these plant protein sources and/or, 2) these amino acids are preferentially degraded by the microflora of the alimentary tract. Carlson and Bayley (1970) fed piglets a 14% protein diet using casein, an animal protein source. They found lower fecal than dietary levels (g/16 g N) of phenylalanine, glutamic acid, proline and serine. The levels of arginine and histidine were not determined in their studies. It would seem that the

TABLE 14. Comparison between the dietary amino acid levels ( $\mu\text{g}/16\text{gN}$ ) and the apparent or true fecal amino acid levels for study 1

DIETS	Soybean meal		Triticale		Wheat		Barley		Protein-free		Triticale Repeat		Protein-free			
	Feces(8) <sup>1</sup>		Feces(6)		Feces(8)		Feces(8)		Feces(7)		Feces(3)		Feces(3)			
	Diet	Apparent <sup>2</sup> True	Diet	Apparent <sup>2</sup> True <sup>3</sup>	Diet	Apparent True	Diet	Apparent <sup>2</sup> True	Diet	Apparent True	Diet	Apparent True	Diet	Apparent True		
Amino Acids																
Essential																
Arginine	7.2	3.4±0.1	2.4	5.3	3.5±0.1	4.5	3.7±0.1	3.4	4.6	3.3±0.2	2.7	4.0±0.3	4.6	3.7±0.0	3.1	4.3±0.1
Histidine	2.5	1.6±0.1	1.6	2.4	1.4±0.1	2.1	1.5±0.1	1.5	1.9	1.3±0.1	1.1	1.6±0.1	1.8	1.6±0.0	1.4	1.9±0.1
Isoleucine	4.5	4.3±0.2	4.8	3.2	3.8±0.2	3.2	4.0±0.1	4.1	3.5	3.8±0.1	3.7	3.9±0.1	3.3	3.7±0.2	3.4	4.0±0.1
Leucine	8.2	7.2±0.2	6.7	6.7	6.3±0.2	6.6	6.7±0.2	6.0	7.6	7.6±0.1	7.6	7.3±0.3	6.7	6.4±0.2	5.3	7.7±0.3
Lysine	5.1	6.3±0.3	5.3	3.2	6.2±0.2	2.8	6.3±0.3	5.8	3.5	6.0±0.2	5.3	6.9±0.3	3.1	5.6±0.1	5.1	6.0±0.3
Methionine	1.3	2.0±0.1	1.9	1.5	1.6±0.1	1.5	1.9±0.1	1.8	1.7	1.7±0.1	1.4	2.1±0.1	1.2	1.8±0.1	1.3	2.0±0.1
Phenylalanine	5.0	3.8±0.2	2.9	4.8	3.8±0.2	4.8	3.8±0.1	3.4	5.4	4.0±0.1	3.8	4.4±0.4	4.6	3.7±0.2	3.1	4.4±0.1
Threonine	4.3	4.1±0.1	4.3	2.5	4.3±0.2	2.8	4.1±0.2	4.2	3.4	3.7±0.2	3.6	3.9±0.3	2.9	3.6±0.1	3.3	4.3±0.2
Valine	4.6	5.0±0.2	4.5	4.0	4.8±0.2	3.7	5.0±0.1	4.8	5.1	5.1±0.2	4.9	5.3±0.1	4.3	4.6±0.2	3.9	5.4±0.2
Non-Essential																
Alanine	4.7	5.9±0.2	5.1	3.8	6.0±0.1	3.6	6.3±0.2	6.3	4.3	6.0±0.1	5.8	6.2±0.2	4.0	6.1±0.1	5.8	6.3±0.1
Aspartic Acid	11.9	9.1±0.3	8.0	5.1	8.7±0.3	5.4	9.2±0.5	8.7	7.2	8.4±0.2	7.4	9.7±0.5	6.3	8.6±0.1	7.0	10.0±0.2
Glutamic Acid	19.7	10.5±0.5	9.1	26.2	10.4±0.3	30.5	10.8±0.3	30.3	23.7	11.3±0.3	11.2	11.2±0.5	27.3	9.5±0.1	8.3	11.6±0.5
Glycine	4.4	5.1±0.2	5.6	4.0	5.1±0.2	4.1	5.0±0.1	5.1	4.3	4.7±0.1	4.7	4.7±0.2	4.1	4.4±0.1	4.1	4.7±0.1
Proline	5.1	3.5±0.1	3.5	10.4	3.5±0.3	9.9	3.7±0.1	3.8	11.6	4.7±0.3	5.3	3.5±0.1	9.7	3.5±0.1	3.4	3.5±0.1
Serine	5.7	3.6±0.1	3.7	4.1	4.0±0.2	4.5	3.8±0.2	3.9	4.3	3.5±0.2	3.3	3.5±0.3	4.4	3.6±0.1	3.1	4.5±0.2
Tyrosine	3.4	3.2±0.1	3.5	2.7	3.2±0.3	2.7	2.8±0.1	2.8	3.1	3.0±0.1	3.0	2.8±0.3	2.6	2.8±0.3	2.3	3.6±0.1
Total Amino Acids	97.6	78.6	72.9	89.9	76.6	92.7	78.6	75.9	95.2	78.1	74.8	81.0	90.9	73.2	63.9	84.2

<sup>1</sup> Numbers in parentheses represent the number of pigs from which the average fecal amino acid composition was determined.<sup>2</sup> Standard error for apparent fecal amino acid levels is indicated.<sup>3</sup> Irrational values were obtained for the average true fecal amino acid levels from the original triticale experiment and were omitted.

TABLE 15. Comparison between the dietary amino acid levels ( $\mu$ /16gN) and the apparent or true fecal amino acid levels for study 2

DIETS	Soybean meal		Triticale		Wheat		Barley		Protein-free				
	Diet		Diet		Diet		Diet		Diet				
	Feces(8) <sup>1</sup>	True	Feces(8)	True	Feces(8)	True	Feces(8)	True	Feces(7)	True			
Amino Acids													
Essential													
Arginine	6.8	3.2±0.2	3.3	4.9	2.9±0.2	2.9	4.7	2.8±0.2	2.8	4.8	2.9±0.1	2.9	2.8±0.1
Histidine	2.3	1.4±0.1	1.5	1.9	1.2±0.1	1.3	1.8	1.1±0.1	1.1	1.7	1.1±0.1	1.2	1.1±0.1
Isoleucine	4.4	4.0±0.1	4.1	3.2	3.8±0.1	4.2	3.2	4.0±0.3	4.1	3.4	3.7±0.1	3.6	4.0±0.1
Leucine	8.2	6.5±0.1	6.6	7.1	6.1±0.2	6.1	6.8	5.9±0.2	5.9	7.5	5.7±0.2	5.6	6.3±0.1
Lysine	6.0	6.0±0.3	6.4	2.9	6.0±0.4	6.4	2.6	6.5±0.3	6.9	3.2	6.4±0.3	6.7	4.8±0.1
Methionine	1.3	2.0±0.1	2.1	1.5	1.8±0.1	1.8	1.5	1.7±0.1	1.7	1.5	1.7±0.1	1.7	1.8±0.2
Phenylalanine	5.2	3.5±0.1	3.5	5.1	3.4±0.1	3.4	6.7	3.5±0.1	3.5	5.4	3.5±0.1	3.5	3.8±0.1
Threonine	4.0	4.3±0.1	4.3	3.1	3.8±0.2	3.8	2.9	3.7±0.2	3.6	3.5	3.7±0.1	3.7	3.9±0.0
Valine	4.5	5.2±0.2	5.2	4.0	5.2±0.1	5.3	4.2	5.1±0.2	5.1	4.7	4.7±0.2	4.7	5.3±0.2
Non-Essential													
Alanine	4.6	6.7±0.2	6.8	4.2	6.9±0.1	6.9	4.0	7.4±0.5	7.7	4.6	6.7±0.1	6.8	6.8±0.1
Aspartic Acid	12.8	9.5±0.2	9.7	6.5	9.2±0.4	9.3	6.2	8.4±0.4	8.2	6.8	8.2±0.2	8.0	9.2±0.2
Glutamic Acid	20.1	10.8±0.3	10.8	29.1	10.9±0.2	11.0	30.6	10.6±0.3	10.5	24.9	10.9±0.4	10.9	11.0±0.3
Glycine	4.5	4.9±0.1	4.9	4.3	4.8±0.1	4.8	4.4	4.7±0.2	4.7	4.5	4.3±0.1	4.2	4.9±0.1
Proline	5.3	3.5±0.2	3.4	9.9	3.5±0.1	3.5	9.9	3.5±0.2	3.4	9.9	3.8±0.2	3.8	3.9±0.2
Serine	5.6	3.8±0.2	3.7	4.8	3.4±0.2	3.3	4.9	3.6±0.1	3.5	4.7	3.5±0.1	3.5	4.0±0.2
Tyrosine	3.2	2.7±0.1	2.7	2.7	2.8±0.1	2.9	2.7	2.8±0.1	2.9	2.9	2.8±0.1	2.9	3.0±0.3
Total Amino Acids	98.8	78.0	79.0	95.2	75.7	76.9	97.1	75.3	75.6	94.0	73.6	73.7	76.6

<sup>1</sup> Numbers in parentheses represent the number of pigs from which the average fecal amino acid composition was determined.<sup>2</sup> Standard error for apparent fecal amino acid levels is indicated.

relatively high availability of phenylalanine, glutamic acid, proline and serine (and probably also of arginine and histidine) is not dependent on the type of protein, i.e. whether animal protein or different plant protein sources are dealt with. It is likely that the microflora degrade these amino acids preferentially.

Michel (1961) carried out in vitro experiments to determine the relative susceptibility of free amino acids to degradation by the cecal flora of the pig. According to his findings one might divide amino acids into 3 groups from relatively high to medium to low susceptibility to microbial degradation : 1) arginine, histidine, aspartic acid, glutamic acid and serine were easily degraded, 2) threonine, isoleucine, leucine and alanine were intermediate in susceptibility to degradation, with respect to group 1 and group 3, 3) lysine, methionine, phenylalanine, valine, proline and tyrosine were relatively insusceptible to degradation by the microflora. Michel's observations might explain the high availabilities of arginine, histidine, glutamic acid and serine (as was found in both studies) but not those of phenylalanine and proline which, in vitro, were found to be relatively insusceptible to microbial degradation. Neither do his observations explain the relatively low availability of aspartic acid from the cereal grains and its relatively high availability from

soybean meal (Tables 5, 6, 8 and 9).

Combe and Pion (1966) determined the amino acid composition (g/16 g N) of the cecal contents of germ-free and normal rats that were fed a 22% casein diet. The levels of lysine and alanine were markedly higher in the cecal contents from the normal rat than in the cecal contents from the germ-free rat. Their findings could perhaps explain to a certain extent the low availability of alanine, resulting from microbial synthesis, from all four diets tested (Tables 5, 6, 8 and 9).

As noted previously, lysine is the indispensable amino acid that is most limiting in many foods of plant origin especially in cereal grains. Therefore, knowledge of its availability is of primary importance. Lysine was found to be less digestible (either true or apparent digestibility) than total N in triticale, wheat and barley. Its digestibility (or availability) was very similar to that of total N in soybean meal (Tables 4 and 7). Surprisingly, the fecal lysine levels (g/16 g N) from soybean meal, triticale, wheat and barley varied only slightly in spite of the large variation in their dietary levels (Table 14 and 15). The latter variation was mainly due to the higher dietary lysine level in soybean meal relative to those in the cereal grains. Lysine, derived from bacterial synthesis, might cause its dietary availability to be underestimated.

Also, its relatively low susceptibility to bacterial degradation could result in its availability to be underestimated relative to those of the other amino acids. Assuming that the true lysine availability (or digestibility) is similar to that of total N for the diets tested, about 0.1<sup>1</sup>, 0.7<sup>1</sup>, 1.0<sup>1</sup> and 1.0<sup>1</sup> g of fecal lysine would be derived from bacterial synthesis for soybean meal, triticale, wheat and barley respectively in study 2 (Table 15; Appendix, Table 12). The amount of bacterial lysine synthesized is likely to depend to a certain extent on the type of diet fed but it seems unlikely that 7<sup>2</sup> to 10<sup>2</sup> times as much bacterial lysine will be synthesized when the animals are fed the cereal grains as when they are fed soybean meal. Therefore, irrespective of bacterial synthesis of lysine, the availability of lysine from the protein fraction of the cereal grains has to be lower than that of soybean meal.

In general, the dietary levels (g/16 g N) of the different amino acids from soybean meal and the cereal grains differed widely (Table 14 and 15). However, the fecal levels (g/16 g N) were more or less similar. These findings seem to indicate that the indigestible N

$$^1 \left\{ \frac{\text{True fecal lysine level (g/16gN)} - \text{Dietary lysine level (G/16gN)}}{\text{True fecal lysine level (g/16gN)}} \right\} \times \left\{ \text{total amount of fecal lysine from dietary source (g)} \right\}$$

$$^2 \quad 0.7/0.1 = 7 ; 1.0/0.1 = 10.$$



fractions from soybean meal, triticale, wheat and barley differ only in total quantity and not in composition (Table 14 and 15). Perhaps one might divide the dietary N of plant protein into 2 components as to their amino acid availability and composition: 1) fraction A, from which the amino acids are readily available and from which the amino acid composition is specific for each type of plant protein, 2) fraction B, from which the amino acids are not readily available and from which the amino acid composition is more or less similar for the types of plant proteins that were tested. This fraction B would increase from soybean meal to triticale to wheat and to barley as a percentage of the total dietary N fraction.

One of the reasons, apart from other factors, that could cause differences in amino acid availabilities between different kinds of protein is likely to be the levels of basic and aromatic amino acids in these proteins. Trypsin catalyzes the hydrolysis of peptide bonds whose carbonyl function is donated by a basic amino acid such as arginine or lysine. Chymotrypsin prefers to catalyze the hydrolysis of peptide linkages involving the aromatic amino acids, particularly phenylalanine and tyrosine. Therefore, in all likelihood, the higher the levels (g/16g N) of basic and aromatic amino acids in a protein the easier (and more complete) the digestion of that protein. The peptide bond at the lysine position is

resistant to hydrolysis by trypsin in case lysine is cross-linked to other amino acids (page 29). From these considerations it may be seen that the degree of digestion of a certain type of protein may depend on its level of available lysine. Consequently, the level of unavailable lysine, as to peptide digestion by trypsin, would in turn decrease the amino acid availabilities of other amino acids because of a less complete digestion of the protein. Cross-linked lysine might well be one of the factors that resulted in the indigestibility of protein fraction B (page 76).

Methionine, the limiting amino acid in soybean protein, also seems to be its least available essential amino acid (Tables 5, 6, 8 and 9). This was confirmed by Cho and Bayley (1970). The true fecal level (g/16 g N) was markedly higher than the dietary level of soybean meal, i.e. by 46% (1.9/1.3) in study 1 and by 60% (2.1/1.3) in study 2 (Table 14 and 15). In general, the true fecal methionine levels were only slightly higher than the dietary methionine levels regarding the cereal grains (Table 14 and 15). The question as to why methionine is less available in soybean meal relative to the other amino acids remains to be answered.

The total amino acid levels (g/16 g N) for diets and feces represent the recovery of amino acid N (as a percentage of total N determined by the Kjeldahl

procedure) on the assumption that each amino acid contains 16% N (Table 14 and 15). However, the percentage of N varies from 32.16% for arginine to 7.73% for tyrosine. Nevertheless, total amino acid levels from the diets and their feces have been calculated when expressed as g amino acid N/16 g N (Appendix, Table 15 and 16). Soybean meal was found to contain more N from amino acid N than the cereal grains. Approximately 83 and 75% of the total N was accounted for by amino acid N in soybean meal and the cereal grains respectively. The recovery of fecal amino acid N was about the same for the feces from soybean meal and the cereal grains and ranged in between 61.7 and 66.1% in study 1 and 61.4 and 64.9% in study 2. The lower fecal than dietary amino acid N recovery might have been due to : 1) the formation of bacterial by-products from amino acids in the gut which are not recovered by amino acid analysis and/or, 2) the non-recoverable dietary N compounds, in case these are absorbed to a lesser extent than the recoverable dietary N compounds, would be present at more concentrated levels in the feces than in the diet. Inclusion of ammonia levels would raise the recovery by about 5 to 10% in the diets and their feces (Appendix, Table 15 and 16).

## SUMMARY

Managra barrows weighing 10 kg or 30 kg were used to determine the biological availability of amino acids from barley, wheat, triticales and soybean meal.

The true nitrogen digestibility and in general the true availability of each essential amino acid determined decreased from soybean meal to triticales to wheat and to barley.

The true availabilities of the essential amino acids from the cereal grains could be divided into four groups from low to high availability: 1) Lysine which was least available ( $P < .05$ ), 2) Isoleucine, methionine, threonine and valine, 3) Leucine, 4) Arginine, histidine and phenylalanine which were most available.

For the heavier pigs, methionine was the least true available essential amino acid from soybean meal though its availability was not significantly different ( $P < .05$ ) from that of valine and lysine. No significant differences ( $P < .05$ ) between the true availabilities of the amino acids from soybean meal were detected for the lighter pigs.

The true nitrogen digestibilities and true amino acid availabilities were lower for the heavier pigs than for the lighter ones for each similar diet tested.

This was especially the case for lysine from the cereal grains. Corrections for metabolic fecal nitrogen and amino acid excretion, by equalizing the intake or dry matter excretion of the protein-free diet to that of each test diet, resulted in more comparable true nitrogen digestibilities and true amino acid availabilities. However, the corrections failed to make the true availability estimates, in particular those of lysine from the cereal grains, more comparable. This was due to the fact that the corrected metabolic fecal lysine excretion did not increase in the same proportion as total nitrogen from the lighter to the heavier pigs. Besides, the relatively large differences in true lysine availability (and other low available amino acids) from the cereal grains between the lighter and heavier pigs might also be attributed partly to the relatively smaller differences that were obtained between the true nitrogen digestibilities for each of the cereal grains.

The relatively high availability of phenylalanine, glutamic acid, proline and serine (and probably also of arginine and histidine) from all diets tested might be due to bacterial degradation in the gut.

The low availability of lysine from the cereal grains, though its availability may be underestimated to a certain extent due to bacterial synthesis, seems to be related to the presence of cross-linked lysine in these feeds.

Perhaps one might divide the dietary nitrogen of plant proteins into two components as to their amino acid availability and composition: 1) Fraction A, from which the amino acids are readily available and from which the amino acid composition is specific for each type of plant protein, 2) Fraction B, from which the amino acids are not readily available and from which the amino acid composition is more or less similar for the types of plant proteins that were tested. This fraction B would seem to increase from soybean meal to triticales to wheat and to barley as a percentage of the total dietary nitrogen fraction.

## BIBLIOGRAPHY

- Abrams, G.D., H. Brauer and H. Sprinz. 1963.  
Influence of normal microbial flora on mucosal morphology and cellular renewal in the ileum. A comparison of germ-free and conventional mice. Lab. Invest. 12: 355-359.
- Anderson, R.F., R.A. Rhodes, G.E.N. Nelson, M.C. Shekleton, A. Barreto, Jr. and M. Arnold. 1958.  
Lysine, methionine and tryptophan content of microorganisms. I. Bacteria. J. Bact. 76: 131-135.
- Barnes, R.H. and E. Kwong. 1964.  
Methionine absorption and utilization from soybean protein and the effect of soybean trypsin inhibitor - A study of amino acid availability. In The Role of the gastrointestinal Tract in Protein Metabolism, pp.41-54. H.N. Munro, ed. Academic Press, New York.
- Bell, J.M. 1948.  
An adjustable cylindrical cage for use in metabolism studies with young pigs. J. Nutr. 35: 365-369.
- Bergen, W.G. and D.B. Purser. 1968.  
Effect of feeding different protein sources on plasma and gut amino acids in the growing rat. J. Nutr. 95: 333-340.
- Bjarnason, J. and K.J. Carpenter. 1969.  
Mechanism of heat damage in proteins. 1. Models with acylated lysine units. Brit. J. Nutr. 23: 859-868.
- Bjarnason, J. and K.J. Carpenter. 1970.  
Mechanism of heat damage in proteins. 2. Chemical changes in pure proteins. Brit. J. Nutr. 24: 313-329.
- Boctor, A.M. and A.E. Harper. 1968.  
Measurement of available lysine in heated and unheated foodstuffs by chemical and biological methods. J. Nutr. 94: 289-296.
- Calhoun, W.K., F.N. Hepburn and W.B. Bradley. 1960.  
The availability of lysine in wheat, flour, bread and gluten. J. Nutr. 70: 337-347.
- Carlson, K.H. and H.S. Bayley. 1970.  
Nitrogen and amino acids in the feces of young pigs receiving a protein-free diet and diets containing graded levels of soybean oil meal or casein. J. Nutr. 100: 1353-1362.

- Carpenter, K.J. 1960.  
The estimation of available lysine in animal-protein foods. *Biochem. J.* 77: 604-610.
- Carpenter, K.J. and B.E. March. 1961.  
The availability of lysine in groundnut biscuits used in the treatment of kwashiorkor. 2. *Brit. J. Nutr.* 15: 403-409.
- Cho, C.Y. and H.S. Bayley. 1970.  
Evaluations of rapeseed and soybean meals as protein sources for swine: Apparent digestibilities of amino acids. *Canad. J. Anim. Sci.* 50: 521-528.
- Combe, E., E. Penot, H. Charlier and E. Sacquet. 1965.  
Métabolisme du rat "germ-free".  
*Ann. Biol. Anim. Biochim. Biophys.* 5: 183-206.
- Combe, E. and R. Pion. 1966.  
Note sur la composition en acides aminés du contenu de caecum de rats axéniques et de rats témoins.  
*Ann. Biol. Anim. Biochim. Biophys.* 6: 255-259.
- Cranwell, P.D. 1968.  
Microbial fermentation in the alimentary tract of the pig. *Nutr. Abstr. Rev.* 38: 721-730.
- Crompton, D.W.T. and M.C. Nesheim. 1969.  
Amino acid patterns during digestion in the small intestine of ducks. *J. Nutr.* 99: 43-50.
- de Muelenaere, H.J.H., M.L. Chen and A.E. Harper. 1967.  
Assessment of factors influencing estimation of availability of threonine, isoleucine and valine in cereal products. *J. Agr. Food Chem.* 15: 318-323.
- Denton, A.E. and C.A. Elvehjem. 1954.  
Amino acid concentration in the portal vein after ingestion of amino acids. *J. Biol. Chem.* 206: 455-460.
- Evrard, E. P.P. Hoet, H. Eyssen, H. Charlier and E. Sacquet. 1964.  
Faecal lipids in germ-free and conventional rats.  
*Brit. J. Exp. Pathol.* 45: 409-414.
- Ford, J.E. 1964.  
A microbiological method for assessing the nutritional value of proteins. 3. Further studies on the measurement of available amino acids.  
*Brit. J. Nutr.* 18: 449-460.



- Giovanetti, P.M., S.C. Stothers and R.J. Parker. 1970.  
Coprophagy prevention and availability of amino acids in wheat for the growing rat.  
Canad. J. Anim. Sci. 50: 269-277.
- Gordon, H.A. and B.S. Westmann. 1960.  
Morphological studies on the germ-free albino rats.  
Anat. Rec. 137: 65-67.
- Gordon, H.A. and E. Bruckner-Kardoss. 1961.  
Effect of normal microbial flora on various tissue elements of the small intestine.  
Acta Anat. 44: 210-219.
- Gupta, J.D., A.M. Dakroury, A.E. Harper and C.A. Elvehjem. 1958.  
Biological availability of lysine.  
J. Nutr. 64: 259-270.
- Harmon, B.G., D.E. Becker and A.H. Jensen. 1967.  
Effect of microbial flora on nitrogen excretion products (Abstract). J. Anim. Sci. 26: 907.
- Harmon, B.G., D.E. Becker, A.H. Jensen and D.H. Baker. 1968.  
Influence of microbiota on metabolic fecal nitrogen in rats. J. Nutr. 96: 391-396.
- Hill, R.L. 1965.  
Hydrolysis of proteins. Adv. Protein Chem. 20: 37-107.
- Horowitz, W. ed. 1965.  
Official Methods of Analysis of the Association of Agricultural Chemists. 10th ed. A.O.A.C., Washington, D.C.
- Kuiken, K.A. and C.M. Lyman. 1948.  
Availability of amino acids in some foods.  
J. Nutr. 36: 359-368.
- Larson, N.L. and F.G. Hill. 1960.  
Amine formation and metabolic activity of microorganisms in the ileum of young swine fed chlortetracycline.
- Lepkovsky, S., F. Furuta, K. Ozone and T. Koike. 1966.  
The proteases, amylase and lipase of the pancreas and intestinal contents of germ-free and conventional rats. Brit. J. Nutr. 20: 257-261.

- Levenson, S.M. and B. Tennant. 1963.  
Contributions of intestinal microflora to the  
nutrition of the host animal. Some metabolic  
and nutritional studies with germ-free animals.  
Fed. Proc. 22: 109-122.
- Lloyd, L.E., D.G. Dale and E.W. Crampton. 1958.  
The role of the caecum in nutrient utilization  
by the pig. J. Anim. Sci. 17: 684-691.
- Luckey, T.D. 1963.  
Germfree Life and Gnotobiology.  
Academic Press Inc., New York.
- Mahler, H.R. and E.H. Cordes. 1966.  
Basic Biological Chemistry.  
Harper & Row, Publishers, New York.
- Meyer, J.H. 1956.  
Influence of dietary fiber on metabolic and  
endogenous nitrogen excretion. J. Nutr. 58: 407-413.
- Michel, M.C. 1961.  
Activité métabolique de la flora totale isolée de  
l'intestin de porc.  
Ann. Biol. Anim. Biochim. Biophys. 1: 16-27.
- Michel, M.C. 1966.  
Métabolisme de la flora intestinal du porc  
degradation des formes L and D des acides aminés.  
Ann. Biol. Anim. Biochim. Biophys. 6: 33-46.
- Mitchell, H.H. and M.H. Bert. 1954.  
The determination of metabolic fecal nitrogen.  
J. Nutr. 52: 483-497.
- Nasset, E.S., P. Schwartz and H.V. Weiss. 1955.  
The digestion of proteins in vivo.  
J. Nutr. 56: 83-94.
- Nasset, E.S. and J.S. Ju. 1961.  
Mixture of endogenous and exogenous protein in the  
alimentary tract. J. Nutr. 74: 461-465.
- Nasset, E.S. 1965.  
Role of the digestive system in protein metabolism.  
Fed. Proc. 24: 953-958.
- Nesheim, M.C. and K.J. Carpenter. 1967.  
The digestion of heat-damaged protein.  
Brit. J. Nutr. 21: 399-411.

- Payne, W.L., G.F. Combs, R.R. Kifer and D.G. Snyder. 1968.  
Investigation of protein quality: ileal recovery of  
amino acids. Fed. Proc. 27: 1199-1203.
- Salter, D.N. and M.E. Coates. 1971.  
The influence of the microflora of the alimentary  
tract on protein digestion in the chick.  
Brit. J. Nutr. 26: 55-69.
- Snook, J.T. and J.H. Meyer. 1964a.  
Response of digestive enzymes to dietary protein.  
J. Nutr. 82: 409-414.
- Snook, J.T. and J.H. Meyer. 1964b.  
Effect of diet and digestive processes on  
proteolytic enzymes. J. Nutr. 83: 94-102.
- Snook, J.T. 1965.  
Effect of diet on intestinal proteolysis.  
Fed. Proc. 24: 941-945.
- Steel, R.G.D. and J.H. Torrie. 1960.  
Principles and procedures of statistics.  
McGraw-Hill Book Company, New York.
- Stott, J.A. and H. Smith. 1966.  
Microbiological assay of protein quality with  
Tetrahymena pyriformis W. 4. Measurement of available  
lysine, methionine, arginine and histidine.  
Brit. J. Nutr. 20: 663-673.
- Twombly, J. and J.H. Meyer. 1961.  
Endogenous nitrogen secretions into the digestive  
tract. J. Nutr. 74: 453-460.
- Valle-Riestra, J. and R.H. Barnes. 1970.  
Digestion of heat-damaged egg albumen by the rat.  
J. Nutr. 100: 873-882.
- Whiting, F. and L.M. Bezeau. 1957.  
The metabolic fecal nitrogen excretion of the pig  
as influenced by the amount of fibre in the ration  
and by body weight. Canad. J. Anim. Sci. 37: 95-105.

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

Item Source of Variation Degrees of freedom	Treatments 3	Squares 1	Pigs/Square 6	Periods/Square 6	Error 13
	(Mean squares)				
<u>Essential</u>					
Arginine	127.8 <sup>xx</sup>	0.1	20.2 <sup>x</sup>	10.5	6.1
Histidine	78.1 <sup>xx</sup>	0.1	23.4 <sup>x</sup>	13.5	6.0
Isoleucine	139.7 <sup>xx</sup>	8.5	27.1	22.1	11.6
Leucine	58.2 <sup>xx</sup>	4.3	21.4	15.4	7.9
Lysine	356.7 <sup>xx</sup>	1.6	57.5	48.2	42.8
Methionine	12.0	0.8	47.7	25.0	16.6
Phenylalanine	31.1 <sup>x</sup>	5.2	16.4	14.1	6.3
Threonine	184.3 <sup>xx</sup>	15.8	23.7	20.3	15.7
Valine	45.9 <sup>x</sup>	3.5	25.8	16.9	10.5
<u>Non-Essential</u>					
Alanine	153.3 <sup>xx</sup>	5.8	35.2	25.5	14.9
Aspartic Acid	368.0 <sup>xx</sup>	13.5	25.1	24.5	16.8
Glutamic Acid	50.7 <sup>xx</sup>	1.5	7.0	4.6	3.6
Glycine	73.2 <sup>xx</sup>	3.8	29.1	19.4	11.4
Proline	33.5 <sup>xx</sup>	1.0	7.9	7.4	3.3
Serine	98.2 <sup>xx</sup>	5.8	13.1	12.9	7.3
Tyrosine	65.4 <sup>xx</sup>	2.6	31.9	21.5	11.3
Nitrogen	72.0 <sup>xx</sup>	1.1	23.2	11.6	9.0
Dry Matter	3.9	0.6	16.9 <sup>x</sup>	9.4	5.4

<sup>x</sup> ( $P < 0.05$ )  
<sup>xx</sup> ( $P < 0.01$ )

APPENDIX Table 2. Analysis of variance between treatments for study 1: Mean squares for true amino acid availabilities and true nitrogen digestibility

Item Source of variation Degrees of freedom	Treatments 3	Squares 1	Pigs/Square 6	Periods/Square 6	Error 13
	(Mean squares)				
<u>Essential</u>					
Arginine	196.2 <sup>xx</sup>	0.2	72.0 <sup>xx</sup>	30.7	14.5
Histidine	142.9 <sup>xx</sup>	1.7	87.2 <sup>xx</sup>	43.2 <sup>x</sup>	13.3
Isoleucine	230.2 <sup>xx</sup>	1.9	52.3	39.2	19.6
Leucine	142.7 <sup>xx</sup>	13.1	45.5 <sup>x</sup>	30.3	13.7
Lysine	527.4 <sup>xx</sup>	12.3	80.9	90.3	48.5
Methionine	193.6 <sup>x</sup>	0.6	188.0 <sup>x</sup>	50.7	44.0
Phenylalanine	101.2 <sup>xx</sup>	14.6	27.9 <sup>x</sup>	31.4 <sup>x</sup>	9.6
Threonine	214.0 <sup>xx</sup>	55.7	40.8	56.3	22.8
Valine	140.4 <sup>xx</sup>	13.0	45.2	40.5	23.1
<u>Non-Essential</u>					
Alanine	326.8 <sup>xx</sup>	31.6	57.4	49.7	23.2
Aspartic Acid	314.6 <sup>xx</sup>	63.0	56.8	62.9	26.1
Glutamic Acid	104.6 <sup>xx</sup>	27.0	11.9	16.0	6.6
Glycine	174.2 <sup>xx</sup>	27.6	47.5	36.2	16.4
Proline	77.1 <sup>xx</sup>	11.4	14.3	17.1	6.6
Serine	134.2 <sup>xx</sup>	62.4 <sup>x</sup>	27.3	35.1	12.3
Tyrosine	122.1 <sup>x</sup>	2.2	65.3 <sup>x</sup>	44.8	22.3
Nitrogen	168.6 <sup>xx</sup>	28.5	48.3	26.8	17.8

<sup>x</sup> ( $P < 0.05$ )  
<sup>xx</sup> ( $P < 0.01$ )

APPENDIX Table 3. Analysis of variance between treatments for study 2: Mean squares for apparent amino acid availabilities and apparent nitrogen and dry matter digestibility

Item Source of variation Degrees of freedom	Treatments	Periods	Pigs	Error
	3	3	7	18
	(Mean squares)			
<u>Essential</u>				
Arginine	64.1 <sup>xx</sup>	17.9	3.2	5.8
Histidine	45.3 <sup>xx</sup>	14.1	8.0	8.6
Isoleucine	90.8 <sup>xx</sup>	58.5 <sup>xx</sup>	7.2	10.1
Leucine	40.1 <sup>xx</sup>	39.5 <sup>xx</sup>	3.3	7.5
Lysine	653.1 <sup>xx</sup>	135.5 <sup>x</sup>	20.2	38.3
Methionine	28.3	74.7 <sup>xx</sup>	11.7	12.0
Phenylalanine	29.8 <sup>xx</sup>	31.7 <sup>xx</sup>	4.3	5.6
Threonine	75.5 <sup>xx</sup>	46.9 <sup>x</sup>	4.6	12.1
Valine	46.2 <sup>xx</sup>	67.6 <sup>xx</sup>	12.2	7.7
<u>Non-Essential</u>				
Alanine	124.0 <sup>xx</sup>	146.2 <sup>xx</sup>	12.1	19.3
Aspartic Acid	213.4 <sup>xx</sup>	67.7 <sup>x</sup>	12.1	19.4
Glutamic Acid	27.7 <sup>xx</sup>	23.7 <sup>xx</sup>	2.1	4.1
Glycine	34.7	65.9 <sup>xx</sup>	4.5	11.1
Proline	25.1 <sup>xx</sup>	18.5 <sup>xx</sup>	3.1	3.4
Serine	55.3 <sup>xx</sup>	31.5 <sup>xx</sup>	0.6	5.7
Tyrosine	79.1 <sup>xx</sup>	47.4 <sup>xx</sup>	3.9	8.2
Nitrogen	61.1 <sup>xx</sup>	59.0 <sup>xx</sup>	4.4	9.5
Dry Matter	2.6	39.4 <sup>xx</sup>	3.2	6.7

<sup>x</sup> (P<0.05)  
<sup>xx</sup> (P<0.01)

APPENDIX Table 4. Analysis of variance between treatments for study 2: Mean squares for true amino acid availabilities and true nitrogen digestibility

Item Source of variation Degrees of freedom	Treatment  3	Periods  3 (Mean squares)	Pigs  7	Error  18
<u>Essential</u>				
Arginine	63.4 <sup>xx</sup>	33.7 <sup>x</sup>	3.7	7.0
Histidine	43.7 <sup>x</sup>	25.3	8.3	10.5
Isoleucine	90.2 <sup>xx</sup>	105.8 <sup>xx</sup>	11.5	11.9
Leucine	42.9 <sup>x</sup>	70.8 <sup>xx</sup>	6.0	8.8
Lysine	599.0 <sup>xx</sup>	192.9 <sup>x</sup>	22.6	40.0
Methionine	29.4	116.8 <sup>xx</sup>	17.9	15.4
Phenylalanine	37.3 <sup>xx</sup>	58.6 <sup>xx</sup>	6.7	6.6
Threonine	75.1 <sup>xx</sup>	87.8 <sup>xx</sup>	7.0	13.7
Valine	45.1 <sup>x</sup>	121.7 <sup>xx</sup>	14.3	12.3
<u>Non-Essential</u>				
Alanine	171.1 <sup>xx</sup>	222.9 <sup>xx</sup>	24.5	19.6
Aspartic Acid	189.2 <sup>xx</sup>	85.1	26.4	28.1
Glutamic Acid	28.4 <sup>xx</sup>	43.5 <sup>xx</sup>	4.5	5.0
Glycine	39.5	118.9 <sup>xx</sup>	8.8	13.2
Proline	20.3 <sup>x</sup>	38.6 <sup>xx</sup>	6.2	4.5
Serine	59.1 <sup>xx</sup>	61.6 <sup>xx</sup>	10.3	3.5
Tyrosine	85.1 <sup>xx</sup>	84.5 <sup>xx</sup>	5.9	8.9
Nitrogen	65.6 <sup>xx</sup>	101.6 <sup>xx</sup>	4.9	12.0

<sup>x</sup> ( $P < 0.05$ )  
<sup>xx</sup> ( $P < 0.01$ )

APPENDIX Table 5. Analysis of variance within treatments for study 1: Mean squares for true and apparent amino acid availabilities

Item Source of variation	Between amino acids	Within amino acids (error)
Degrees of freedom	15	112
<u>True availability</u> (Mean squares)		
Soybean meal	37.5	37.4
Triticale <sup>1</sup>	43.3 <sup>x</sup>	23.2
Triticale repeat <sup>2</sup>	32.4 <sup>xx</sup>	1.2
Wheat	158.0 <sup>xx</sup>	47.5
Barley	122.5 <sup>xx</sup>	24.8
<u>Apparent Availability</u>		
Soybean meal	102.8 <sup>xx</sup>	8.0
Triticale <sup>1</sup>	231.8 <sup>xx</sup>	8.1
Triticale repeat <sup>2</sup>	72.0 <sup>xx</sup>	0.4
Wheat	336.2 <sup>xx</sup>	30.8
Barley	254.7 <sup>xx</sup>	10.2

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

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

<sup>1</sup> Degrees of freedom of error is 80

<sup>2</sup> Degrees of freedom of error is 32



APPENDIX Table 6. Analysis of variance within treatments for study 2: Mean squares for true and apparent amino acid availabilities

Item Source of variation	Between amino acids	Within amino acids (error)
Degrees of freedom	15	112
<u>True Availability</u>		
Soybean meal	60.5 <sup>xx</sup>	15.1
Triticale	183.1 <sup>xx</sup>	18.8
Wheat	282.3 <sup>xx</sup>	27.4
Barley	232.7 <sup>xx</sup>	23.6
<u>Apparent Availability</u>		
Soybean meal	89.8 <sup>xx</sup>	5.8
Triticale	228.0 <sup>xx</sup>	16.0
Wheat	344.0 <sup>xx</sup>	19.3
Barley	274.9 <sup>xx</sup>	18.7

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

APPENDIX Table 7. Mean and standard error of the apparent availability of amino acids in study 1 as influenced by the type of diet

Diets	Soybean meal(8) <sup>1</sup>	Triticale(6)	Wheat(8)	Barley(8)
Amino acids				
<u>Essential</u>				
Arginine	93.2 <sup>±</sup> 0.6	88.6 <sup>±</sup> 1.0	86.8 <sup>±</sup> 1.5	82.5 <sup>±</sup> 1.1
Histidine	91.6 <sup>±</sup> 0.7	89.3 <sup>±</sup> 0.8	87.6 <sup>±</sup> 1.5	83.1 <sup>±</sup> 1.3
Isoleucine	86.3 <sup>±</sup> 1.0	78.4 <sup>±</sup> 1.1	78.7 <sup>±</sup> 2.0	72.1 <sup>±</sup> 1.3
Leucine	87.5 <sup>±</sup> 0.9	82.5 <sup>±</sup> 1.0	82.4 <sup>±</sup> 1.8	79.1 <sup>±</sup> 0.9
Lysine	82.5 <sup>±</sup> 1.0	66.6 <sup>±</sup> 1.8	61.6 <sup>±</sup> 3.5	57.6 <sup>±</sup> 1.3
Methionine	78.5 <sup>±</sup> 1.7	78.0 <sup>±</sup> 1.2	77.8 <sup>±</sup> 2.3	74.5 <sup>±</sup> 1.4
Phenylalanine	88.4 <sup>±</sup> 0.9	85.6 <sup>±</sup> 0.9	86.4 <sup>±</sup> 1.5	82.5 <sup>±</sup> 0.8
Threonine	86.0 <sup>±</sup> 1.0	71.1 <sup>±</sup> 1.7	75.0 <sup>±</sup> 1.8	72.4 <sup>±</sup> 1.3
Valine	84.3 <sup>±</sup> 1.0	79.1 <sup>±</sup> 1.0	77.8 <sup>±</sup> 2.0	77.2 <sup>±</sup> 1.0
<u>Non-Essential</u>				
Alanine	81.9 <sup>±</sup> 1.1	72.9 <sup>±</sup> 1.2	70.6 <sup>±</sup> 2.3	66.1 <sup>±</sup> 1.3
Aspartic Acid	89.0 <sup>±</sup> 0.8	69.2 <sup>±</sup> 1.8	70.6 <sup>±</sup> 2.2	71.8 <sup>±</sup> 1.3
Glutamic Acid	92.5 <sup>±</sup> 0.7	92.9 <sup>±</sup> 0.6	94.1 <sup>±</sup> 0.8	88.4 <sup>±</sup> 0.6
Glycine	83.7 <sup>±</sup> 1.2	78.5 <sup>±</sup> 1.1	79.1 <sup>±</sup> 2.0	73.2 <sup>±</sup> 1.1
Proline	90.8 <sup>±</sup> 0.8	94.1 <sup>±</sup> 0.5	93.6 <sup>±</sup> 1.1	90.3 <sup>±</sup> 0.3
Serine	90.3 <sup>±</sup> 0.7	83.2 <sup>±</sup> 1.0	86.0 <sup>±</sup> 1.4	80.4 <sup>±</sup> 1.1
Tyrosine	85.6 <sup>±</sup> 1.2	78.8 <sup>±</sup> 1.1	80.1 <sup>±</sup> 2.1	76.6 <sup>±</sup> 1.1

<sup>1</sup> Numbers in parentheses indicate the number of determinations for which the mean and standard error were determined.

APPENDIX Table 8. Mean and standard error of the true availability of amino acids in study 1 as influenced by the type of diet

Diets	Soybean meal(8) <sup>1</sup>	Triticale(6)	Wheat(8)	Barley(8)
Amino acids				
<u>Essential</u>				
Arginine	98.4 $\pm$ 1.4	98.5 $\pm$ 2.3	94.1 $\pm$ 2.5	91.4 $\pm$ 1.8
Histidine	97.9 $\pm$ 1.5	99.1 $\pm$ 2.1	94.7 $\pm$ 2.4	93.2 $\pm$ 2.4
Isoleucine	95.2 $\pm$ 2.2	95.8 $\pm$ 0.9	89.2 $\pm$ 2.4	84.7 $\pm$ 1.7
Leucine	95.7 $\pm$ 2.0	96.6 $\pm$ 0.9	91.2 $\pm$ 2.2	88.7 $\pm$ 1.5
Lysine	94.9 $\pm$ 2.2	94.4 $\pm$ 3.2	80.8 $\pm$ 3.8	77.1 $\pm$ 1.9
Methionine	94.5 $\pm$ 4.0	97.0 $\pm$ 2.9	89.8 $\pm$ 2.9	87.3 $\pm$ 2.4
Phenylalanine	95.8 $\pm$ 1.9	96.5 $\pm$ 0.9	92.4 $\pm$ 1.7	90.1 $\pm$ 1.3
Threonine	95.4 $\pm$ 2.1	94.8 $\pm$ 2.8	87.3 $\pm$ 2.2	85.5 $\pm$ 1.9
Valine	94.5 $\pm$ 2.2	95.6 $\pm$ 1.1	89.0 $\pm$ 2.6	87.6 $\pm$ 1.6
<u>Non-Essential</u>				
Alanine	94.1 $\pm$ 2.3	94.4 $\pm$ 1.8	84.1 $\pm$ 2.6	80.6 $\pm$ 1.9
Aspartic Acid	96.6 $\pm$ 1.8	94.5 $\pm$ 3.2	84.6 $\pm$ 2.8	85.4 $\pm$ 2.0
Glutamic Acid	97.8 $\pm$ 1.5	98.7 $\pm$ 1.4	97.0 $\pm$ 1.1	93.3 $\pm$ 1.0
Glycine	94.0 $\pm$ 2.4	94.5 $\pm$ 0.9	88.3 $\pm$ 2.3	84.3 $\pm$ 1.5
Proline	96.8 $\pm$ 1.7	98.2 $\pm$ 0.4	96.3 $\pm$ 1.3	93.2 $\pm$ 0.5
Serine	96.9 $\pm$ 1.7	96.8 $\pm$ 2.2	92.8 $\pm$ 1.8	89.9 $\pm$ 1.7
Tyrosine	95.0 $\pm$ 2.3	95.1 $\pm$ 1.1	90.3 $\pm$ 2.7	87.6 $\pm$ 1.7

<sup>1</sup> Numbers in parentheses indicate the number of determinations for which the mean and standard error were determined.

APPENDIX Table 9. Mean and standard<sup>1</sup> error of the apparent availability of amino acids in study 2 as influenced by the type of diet

Diets	Soybean meal	Triticale	Wheat	Barley
Amino acids				
<u>Essential</u>				
Arginine	94.7±0.7	92.2±0.7	91.2±0.9	88.1±1.2
Histidine	93.1±0.7	91.6±1.1	90.8±1.3	87.3±1.1
Isoleucine	89.2±0.9	84.0±1.4	82.0±1.4	79.1±1.5
Leucine	90.9±0.8	88.6±1.1	86.9±1.2	84.9±1.3
Lysine	88.6±1.0	72.5±2.2	61.3±3.1	60.1±2.6
Methionine	81.7±1.1	83.5±1.1	81.7±1.8	77.4±1.9
Phenylalanine	91.7±0.7	91.1±0.9	89.3±1.2	87.2±1.1
Threonine	88.0±0.9	83.7±1.4	81.1±1.3	78.7±1.6
Valine	87.2±1.0	82.9±1.6	81.9±1.5	90.9±1.5
<u>Non-Essential</u>				
Alanine	83.3±1.2	77.8±2.0	71.6±2.4	71.1±2.0
Aspartic Acid	91.4±0.7	80.5±2.4	80.2±1.6	76.6±1.7
Glutamic Acid	93.9±0.6	95.0±1.0	94.8±0.7	91.5±1.0
Glycine	87.5±0.9	85.2±1.5	83.8±1.5	81.2±1.5
Proline	92.5±0.7	95.2±0.8	94.9±0.8	92.6±0.8
Serine	92.3±0.6	90.7±0.9	89.1±0.9	85.6±1.2
Tyrosine	90.4±0.8	86.5±1.1	84.0±1.4	81.4±1.3

<sup>1</sup> Mean and standard error of 8 determinations

APPENDIX Table 10. Mean and standard error<sup>1</sup> of the true availability of amino acids in study 2 as influenced by the type of diet

Diets	Soybean meal	Triticale	Wheat	Barley
Amino acids				
<u>Essential</u>				
Arginine	96.0±1.0	94.1±0.8	93.1±1.1	90.1±1.3
Histidine	94.6±0.7	93.3±1.2	92.7±1.5	89.5±1.2
Isoleucine	92.1±1.5	87.7±1.6	86.1±1.8	83.1±1.7
Leucine	93.2±1.2	91.3±1.2	89.8±1.5	87.6±1.5
Lysine	91.2±1.4	77.5±2.4	67.3±3.2	65.0±2.7
Methionine	86.3±1.6	86.8±1.3	85.7±2.1	81.5±2.2
Phenylalanine	94.2±1.2	93.3±0.9	91.6±1.4	89.5±1.3
Threonine	91.0±1.3	87.7±1.5	85.4±1.6	82.5±1.8
Valine	90.9±1.7	86.8±1.7	85.9±1.8	84.6±1.7
<u>Non-Essential</u>				
Alanine	89.2±2.1	82.8±2.1	77.1±2.8	76.1±2.2
Aspartic acid	93.9±1.3	84.9±2.5	86.8±2.3	81.1±1.9
Glutamic acid	95.6±1.1	96.2±1.1	96.0±0.9	92.9±1.1
Glycine	91.0±1.5	88.7±1.6	87.5±1.8	85.0±1.8
Proline	94.8±1.2	96.4±0.9	96.2±1.0	93.9±1.0
Serine	94.6±1.1	93.1±1.0	91.6±1.2	88.5±1.4
Tyrosine	93.1±1.3	89.7±1.2	87.5±1.6	85.0±1.5

<sup>1</sup> Mean and standard error of 8 determinations

APPENDIX Table 11. Mean and standard error of the amino acid excretion (g/day) for study 1. as influenced by the type of diet

Diets	Soybean meal	Triticale <sup>1</sup>	Wheat	Barley	Protein-free	
					Latin Square 1 <sup>2</sup>	Latin Square 2 <sup>3</sup>
Amino acids						
<u>Essential</u>						
Arginine	0.30±0.03	0.24±0.02	0.42±0.05	0.43±0.04	0.20±0.02	0.21 0.04
Histidine	0.13±0.01	0.10±0.01	0.18±0.02	0.17±0.02	0.09±0.01	0.09 0.01
Isoleucine	0.37±0.04	0.26±0.01	0.46±0.05	0.50±0.04	0.22±0.01	0.21 0.03
Leucine	0.62±0.07	0.44±0.02	0.78±0.09	0.84±0.06	0.38±0.03	0.35 0.06
Lysine	0.54±0.06	0.41±0.03	0.73±0.09	0.78±0.05	0.34±0.02	0.34 0.04
Methionine	0.18±0.02	0.12±0.01	0.21±0.03	0.23±0.02	0.10±0.01	0.11 0.02
Phenylalanine	0.35±0.04	0.26±0.01	0.44±0.05	0.50±0.03	0.21±0.03	0.19 0.03
Threonine	0.37±0.04	0.27±0.01	0.47±0.04	0.49±0.04	0.24±0.02	0.20 0.04
Valine	0.43±0.05	0.31±0.02	0.57±0.06	0.62±0.05	0.26±0.02	0.24 0.04
<u>Non-Essential</u>						
Alanine	0.51±0.05	0.39±0.03	0.72±0.07	0.78±0.06	0.33±0.02	0.30 0.05
Aspartic Acid	0.80±0.08	0.59±0.04	1.06±0.10	1.08±0.08	0.53±0.06	0.44 0.08
Glutamic Acid	0.89±0.09	0.69±0.04	1.22±0.10	1.46±0.10	0.61±0.06	0.53 0.09
Glycine	0.43±0.05	0.33±0.01	0.58±0.06	0.61±0.04	0.26±0.01	0.23 0.04
Proline	0.29±0.03	0.23±0.01	0.43±0.05	0.60±0.04	0.18±0.01	0.16 0.03
Serine	0.33±0.03	0.26±0.01	0.43±0.04	0.44±0.03	0.22±0.02	0.18 0.03
Tyrosine	0.30±0.04	0.21±0.01	0.36±0.04	0.39±0.03	0.17±0.01	0.17 0.03

<sup>1</sup> Mean and standard error of 6 determinations

<sup>2</sup> Mean and standard error of 4 determinations

<sup>3</sup> Mean and standard error of 3 determinations

APPENDIX Table 12. Mean and standard error of the amino acid excretion (g) by the pigs in study 2 as influenced by the type of diet

Diets	Soybean meal	Triticale	Wheat	Barley	Protein-free <sup>1</sup>
Amino acids <sup>2</sup>					
<u>Essential</u>					
Arginine	0.78±0.11	0.72±0.07	0.80±0.10	1.01±0.11	0.16±0.02
Histidine	0.32±0.03	0.32±0.04	0.32±0.05	0.39±0.04	0.06±0.01
Isoleucine	1.03±0.14	1.01±0.12	1.10±0.15	1.24±0.13	0.22±0.02
Leucine	1.61±0.21	1.57±0.18	1.74±0.24	1.98±0.22	0.34±0.03
Lysine	1.49±0.22	1.57±0.24	1.90±0.30	2.19±0.26	0.26±0.03
Methionine	0.48±0.06	0.46±0.06	0.52±0.08	0.60±0.08	0.10±0.02
Phenylalanine	0.92±0.12	0.87±0.09	1.02±0.13	1.21±0.12	0.21±0.03
Threonine	1.05±0.13	0.97±0.10	1.07±0.13	1.30±0.14	0.22±0.02
Valine	1.27±0.17	1.33±0.17	1.47±0.20	1.57±0.16	0.28±0.03
<u>Non-essential</u>					
Alanine	1.68±0.23	1.80±0.23	2.23±0.35	2.34±0.26	0.37±0.04
Aspartic Acid	2.36±0.31	2.43±0.37	2.43±0.33	2.81±0.29	0.50±0.05
Glutamic Acid	2.69±0.35	2.84±0.40	3.09±0.41	3.73±0.43	0.61±0.07
Glycine	1.21±0.16	1.24±0.15	1.39±0.20	1.48±0.16	0.27±0.03
Proline	0.86±0.11	0.91±0.11	0.99±0.14	1.28±0.13	0.21±0.02
Serine	0.92±0.11	0.86±0.08	1.03±0.13	1.20±0.13	0.22±0.03
Tyrosine	0.66±0.09	0.71±0.08	0.82±0.12	0.94±0.10	0.16±0.02

<sup>1</sup> Mean and standard error of 7 determinations

<sup>2</sup> Amino acid excretion expressed as grams per day

APPENDIX Table 13. Triticale repeat experiment of study 1:  
Mean and standard error<sup>1</sup> of the true and apparent  
amino acid availability and excretion of amino acids  
by the pigs (g)

Amino acids	Availability		Fecal Excretion <sup>2</sup>	
	Apparent	True	Triticale	Protein-free
<u>Essential</u>				
Arginine	88.2±0.4	94.5±0.3	0.43±0.02	0.23±0.02
Histidine	87.0±0.4	93.8±1.0	0.19±0.00	0.10±0.01
Isoleucine	83.2±0.2	91.5±0.6	0.44±0.01	0.22±0.02
Leucine	86.0±0.2	93.7±0.8	0.75±0.02	0.41±0.04
Lysine	72.9±0.3	86.3±1.0	0.66±0.01	0.33±0.04
Methionine	78.5±0.6	90.3±1.4	0.19±0.01	0.11±0.01
Phenylalanine	88.1±0.3	94.5±0.4	0.44±0.02	0.24±0.02
Threonine	80.9±0.3	90.9±0.2	0.45±0.01	0.23±0.01
Valine	84.1±0.4	92.4±0.2	0.53±0.02	0.28±0.02
<u>Non-essential</u>				
Alanine	77.7±0.3	88.4±0.3	0.71±0.02	0.34±0.03
Aspartic Acid	79.5±0.2	91.0±0.7	1.02±0.02	0.54±0.05
Glutamic acid	94.7±0.2	97.6±0.4	1.16±0.04	0.62±0.07
Glycine	84.2±0.3	92.1±0.3	0.51±0.02	0.25±0.02
Proline	94.7±0.1	97.1±0.3	0.41±0.01	0.19±0.02
Serine	87.5±0.3	94.2±0.3	0.44±0.02	0.24±0.02
Tyrosine	83.2±0.7	92.9±0.7	0.34±0.02	0.20±0.01

<sup>1</sup> Mean and standard error of 3 determinations

<sup>2</sup> Amino acid excretion expressed as grams per day



APPENDIX Table 14. Mean<sup>1</sup> apparent and true essential amino acid availabilities, nitrogen and dry matter digestibility between the test periods in study 2.

TEST PERIODS	I	II	III	IV
Apparent availability				
<u>Amino acids</u>				
Arginine	93.6	91.3	91.0	90.7
Histidine	92.1	91.6	89.5	89.6
Isoleucine	87.8	85.0	81.2	80.4
Leucine	91.0	88.5	86.2	85.5
Lysine	79.2	73.6	65.8	66.5
Methionine	85.4	82.8	76.9	78.1
Phenylalanine	92.0	91.1	88.4	87.6
Threonine	87.0	83.6	80.9	80.1
Valine	87.8	84.2	80.9	79.5
Nitrogen	89.4	85.5	83.6	81.7
Dry matter	92.1	89.1	88.1	86.5
True availability				
<u>Amino acids</u>				
Arginine	95.9	93.0	92.4	92.3
Histidine	94.3	93.5	91.1	91.4
Isoleucine	92.4	88.5	84.2	83.7
Leucine	94.2	91.2	88.4	87.9
Lysine	84.9	78.1	69.9	70.9
Methionine	90.4	86.8	80.4	81.9
Phenylalanine	95.1	93.1	90.2	89.8
Threonine	91.7	87.4	84.0	83.6
Valine	92.6	88.0	84.0	82.9
Nitrogen	93.3	88.6	86.2	84.6

<sup>1</sup> Mean of 8 determinations

APPENDIX Table 15. Comparison between the dietary amino acid levels (g amino acid N/16 g N) and the apparent fecal amino acid levels in Study 1.

Diets	Soybean meal		Triticale		Wheat		Barley		Protein-free		Triticale repeat		Protein-free	
	Diet	Feces(8) <sup>1</sup>	Diet	Feces(6)	Diet	Feces(8)	Diet	Feces(8)	Feces(7)	Diet	Feces(3)	Feces(3)	Feces(3)	Feces(3)
<u>Essential</u>														
Arginine	2.32	1.10±0.04	1.71	1.14±0.03	1.46	1.18±0.02	1.48	1.06±0.05	1.28±0.08	1.47	1.18±0.01	1.37±0.03		
Histidine	0.67	0.42±0.02	0.65	0.37±0.01	0.56	0.40±0.02	0.50	0.35±0.03	0.44±0.03	0.49	0.42±0.01	0.50±0.03		
Isoleucine	0.48	0.46±0.02	0.34	0.40±0.02	0.34	0.43±0.01	0.37	0.41±0.01	0.42±0.01	0.35	0.39±0.02	0.43±0.01		
Leucine	0.88	0.77±0.02	0.72	0.67±0.02	0.70	0.71±0.02	0.81	0.69±0.01	0.78±0.03	0.72	0.68±0.02	0.82±0.03		
Lysine	0.97	1.20±0.06	0.61	1.19±0.04	0.54	1.21±0.05	0.66	1.14±0.03	1.32±0.06	0.59	1.07±0.02	1.16±0.06		
Methionine	0.12	0.19±0.01	0.14	0.15±0.01	0.14	0.18±0.01	0.16	0.16±0.01	0.20±0.01	0.11	0.17±0.01	0.19±0.01		
Phenylalanine	0.42	0.32±0.02	0.41	0.32±0.02	0.41	0.32±0.01	0.46	0.34±0.01	0.37±0.03	0.39	0.31±0.02	0.37±0.01		
Threonine	0.51	0.48±0.01	0.29	0.50±0.02	0.33	0.48±0.02	0.40	0.44±0.02	0.46±0.03	0.34	0.42±0.01	0.51±0.02		
Valine	0.55	0.58±0.02	0.47	0.55±0.02	0.45	0.58±0.01	0.61	0.59±0.02	0.61±0.01	0.50	0.53±0.02	0.63±0.02		
<u>Non-Essential</u>														
Alanine	0.74	0.92±0.03	0.60	0.94±0.01	0.57	0.99±0.03	0.68	0.95±0.02	0.97±0.03	0.63	0.96±0.02	0.99±0.02		
Aspartic Acid	1.26	0.96±0.03	0.54	0.92±0.03	0.56	0.97±0.05	0.76	0.88±0.02	1.02±0.05	0.66	0.90±0.01	1.05±0.02		
Glutamic Acid	1.87	1.00±0.05	2.49	0.99±0.03	2.90	1.03±0.03	2.25	1.08±0.03	1.07±0.05	2.60	0.90±0.01	1.10±0.05		
Glycine	0.81	0.95±0.03	0.75	0.95±0.04	0.76	0.93±0.01	0.80	0.88±0.02	0.87±0.03	0.76	0.82±0.01	0.88±0.02		
Proline	0.62	0.43±0.02	1.26	0.43±0.04	1.21	0.45±0.01	1.41	0.57±0.03	0.42±0.01	1.18	0.42±0.01	0.43±0.01		
Serine	0.75	0.48±0.01	0.54	0.43±0.03	0.60	0.50±0.02	0.57	0.46±0.02	0.46±0.04	0.59	0.48±0.01	0.60±0.02		
Tyrosine	0.26	0.25±0.01	0.21	0.25±0.02	0.21	0.22±0.01	0.24	0.23±0.01	0.22±0.02	0.20	0.22±0.02	0.28±0.01		
Total Amino Acid N	13.23	10.51±0.27	11.74	10.30±0.21	11.74	10.58±0.17	12.16	10.23±0.25	10.90±0.38	11.58	9.87±0.06	11.31±0.30		
Amino Acid N (%)	82.7	65.7	73.4	64.4	73.4	66.1	76.0	63.9	68.2	72.4	61.7	70.7		

<sup>1</sup> Numbers in parentheses indicate the number of determinations from which the mean and standard errors were determined.

APPENDIX Table 16. Comparison between the dietary amino acid levels (g amino acid N/16 g N) and the apparent fecal amino acid levels in Study 2.

Diets	Soybean meal		Triticale		Wheat		Barley		Protein-free	
	Diet	Feces(8) <sup>1</sup>	Diet	Feces(8)	Diet	Feces(8)	Diet	Feces(8)	Diet	Feces(7)
<b>Amino acids</b>										
<u>Essential</u>										
Arginine	2.18	1.03±0.07	1.56	0.93±0.06	1.51	0.91±0.06	1.55	0.94±0.04		0.91±0.03
Histidine	0.61	0.38±0.03	0.52	0.33±0.03	0.48	0.30±0.03	0.47	0.31±0.02		0.29±0.03
Isoleucine	0.47	0.43±0.01	0.34	0.41±0.01	0.34	0.43±0.03	0.36	0.39±0.01		0.43±0.00
Leucine	0.87	0.69±0.01	0.76	0.65±0.02	0.73	0.63±0.02	0.80	0.61±0.02		0.67±0.01
Lysine	1.14	1.14±0.05	0.56	1.15±0.08	0.49	1.24±0.06	0.61	1.22±0.05		0.92±0.02
Methionine	0.12	0.19±0.01	0.14	0.17±0.01	0.14	0.16±0.01	0.14	0.16±0.01		0.17±0.02
Phenylalanine	0.44	0.30±0.01	0.43	0.29±0.01	0.57	0.30±0.01	0.46	0.30±0.01		0.32±0.01
Threonine	0.47	0.50±0.01	0.36	0.45±0.02	0.34	0.43±0.02	0.41	0.44±0.01		0.46±0.00
Valine	0.54	0.60±0.02	0.48	0.61±0.01	0.50	0.59±0.02	0.56	0.55±0.02		0.62±0.02
<u>Non-Essential</u>										
Alanine	0.72	1.06±0.03	0.66	1.08±0.02	0.63	1.17±0.07	0.73	1.06±0.02		1.07±0.02
Aspartic Acid	1.35	1.00±0.02	0.68	0.97±0.04	0.65	0.88±0.04	0.72	0.86±0.02		0.97±0.02
Glutamic Acid	1.91	1.03±0.03	2.77	1.04±0.02	2.91	1.01±0.03	2.37	1.04±0.04		1.05±0.03
Glycine	0.84	0.91±0.02	0.81	0.89±0.02	0.82	0.88±0.04	0.84	0.80±0.02		0.92±0.01
Proline	0.64	0.42±0.02	1.21	0.43±0.01	1.20	0.42±0.02	1.20	0.46±0.02		0.47±0.02
Serine	0.74	0.50±0.02	0.64	0.45±0.02	0.65	0.48±0.01	0.63	0.47±0.01		0.53±0.02
Tyrosine	0.25	0.21±0.01	0.21	0.22±0.01	0.21	0.22±0.01	0.22	0.22±0.01		0.23±0.01
Total Amino Acid N	13.29	10.38±0.18	12.13	10.05±0.23	12.17	10.04±0.27	12.07	9.83±0.19		10.03±0.18
Amino Acid N (%)	83.1	64.9	75.8	62.8	76.1	62.8	75.4	61.4		62.7

<sup>1</sup> Numbers in parentheses indicate the number of determinations from which the mean and standard errors were determined.