The effect of protein quality and fibre level in the diet and microbial activity in the digestive tract on protein utilization and energy digestibility in rats

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1. Two nitrogen-balance experiments were performed with growing rats to test the effect of dietary fibre level, protein quality and antibiotic inclusion on microbial activity, N excretion patterns and energy digestibility. Each experiment involved eight dietary treatments in a $2 \times 2 \times 2$ factorial design, with five rats per treatment. The eight treatments resulted from a combination of two protein treatments, two fibre treatments and two antibiotic treatments. In Expt 1 the protein was provided as barley, or barley plus 2 g L-lysine hydrochloride/kg dry matter (DM) (at 15 g N/kg DM) and in Expt 2 as soya-bean meal or soya-bean meal plus 2 g DL-methionine/kg DM (at 15 g N/kg DM). In both experiments the basal diet was provided with or without additional fibre as 100 g barley husk/kg DM and with or without antibiotic as 7 g Nebacitin/kg DM.

2. With both barley and soya-bean meal, true protein digestibility (TD) was improved with the addition of amino acids. Only with the soya-bean meal diets was TD increased with Nebacitin treatment, with the effect of Nebacitin and methionine being additive. Barley husk slightly reduced the TD of soya-bean meal.

3. The effect of treatments on biological value (BV) was considerable. Lysine increased BV of the barley diet from 0·741 to 0·815 whereas Nebacitin reduced BV from 0·799 to 0·757. Methionine increased the BV of soya-bean meal from 0·754 to 0·911 while BV was reduced by Nebacitin from 0·843 to 0·821 and by barley husk from 0·845 to 0·820.

4. Net protein utilization (NPU) was markedly improved by the addition of amino acids and reduced by the addition of Nebacitin. Barley husk reduced NPU with soya-bean meal diets only.

5. Amino acid addition had no effect on energy digestibility of either diet, but Nebacitin reduced this value by approximately 5% on the barley diets, with a smaller reduction on the soya-bean meal diets. Fibre reduced energy digestibility of both diets.

6. On both diets, blood urea (BU) decreased with addition of amino acids. Nebacitin increased BU with the barley diets, but decreased BU with the soya-bean meal diets. With the soya-bean meal diets, the decrease with a combination of barley husk and methionine was particularly marked.

7. Microbial activity, as measured by caecal ATP activity, was strongly reduced by antibiotic addition with both protein sources. On the soya-bean meal diets ATP activity was increased by the addition of methionine.

8. The results demonstrated a difference in response to the addition of fibre and the first limiting amino acid to barley and soya-bean meal, as measured by several indicators of protein and energy utilization.

The effect of fibre on dietary energy and nitrogen utilization in the monogastric animal has formed the basis of many studies. The contribution of fibre to the energy needs of the pig and rat have been debated. Although fermentation of fibre occurs in the hind-gut (Nyman

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& Asp, 1982) to produce organic acids (Friend *et al.* 1962; Clemens *et al.* 1975), the quantification of the energy absorbed in the form of these acids (Farrell & Johnson 1972; Kass *et al.* 1980; Eggum & Chwalibog, 1983) has been difficult because of errors associated with currently used methods (Argenzio, 1982). Some results showing dietary propionic acid to cause reduced growth rates have been confounded by a concomitant reduction in feed intake (Hoque & Elliot, 1964; Thacker & Bowland, 1980). Partridge (1982) has presented results indicating that the net energy value of fibre for pigs and rats may be close to zero; however, the reasons for such an apparently poor utilization of fibre do not appear to have been clearly identified.

The effect of fibre on N balance and N excretory patterns is influenced by many factors, including its chemical composition and degradability (Bach Knudsen *et al.* 1982). Hind-gut fermentation has been shown to be increased by the presence of starch either undigested, or infused into the caecum (Mason *et al.* 1976; Zebrowska *et al.* 1980) resulting in a change in the excretory pattern of N (Beames & Eggum, 1981) presumably as a result of the flow of N into the lumen of the gut from the blood (Rerat *et al.* 1979*a*) as indicated by a reduction in blood urea-N. The effect of fibre on the apparent digestibility of N depends on the nature of the dietary fibre, digestibility of dietary carbohydrate and the digestibility and level of dietary protein (Beames & Eggum, 1981). Furthermore, fibre reduces the transit time significantly (Raczynski *et al.* 1982), and thus leaves less time for microbial fermentation. Responses to dietary variations largely result from changes in the extent of fermentation and the demands of the microbial population causing this fermentation. The effect of fermentation has been studied with the use of germ-free animals (Salter & Coates, 1971) and antibiotics, in particular Nebacitin, both with rats (Eggum *et al.* 1979; Sauer *et al.* 1980; Bach Knudsen *et al.* 1982) and pigs (Mason *et al.* 1976, 1982).

The present experiment was designed to study the effect of protein quality and natural fibre on N and energy utilization in rats, and to investigate the role of bacterial fermentation by the use of the antibiotic Nebacitin. Correction of faecal values for the N contribution from Nebacitin was performed as described by Mason *et al.* (1982). When Nebacitin was subjected to both 3- and 4-enzyme in vitro digestibility (Pedersen & Eggum, 1981) no digestion was observed.

EXPERIMENTAL

Diets

In each of two experiments there were eight dietary treatments in a $2 \times 2 \times 2$ factorial design, with five rats per treatment. Both experiments were of a similar design, differing only in the source of dietary protein. In Expt 1 protein was provided by barley and in Expt 2 by soya-bean meal. The eight treatments in each experiment resulted from a combination of two protein treatments, two fibre treatments and two antibiotic treatments. Each diet, which contained 15 g N/kg dry matter (DM), was based on a N-free mixture containing the following amounts/kg air-dry matter (89·2 g sucrose, 52·0 g cellulose, 52·0 g peanut oil, and 806·8 g autoclaved potato starch) plus minerals and vitamins (Eggum, 1973).

The basal protein was provided with or without supplemental amino acids (2 g L-lysine hydrochloride/kg DM in the barley diets and 2 g DL-methionine/kg DM in the soya-bean meal diets), with or without 7 g Nebacitin (neomycin sulphate–bacitracin; 1:2, w/w)/kg DM and with or without barley husk (100 g/kg DM). Husk was included at the expense of the N-free portion of the basal diet. Formulations are given in Tables 1 and 2.

Animals and feeding

The experimental procedure has been described by Eggum (1973). Groups of five Wistar male rats weighing approximately 70 g were used, with preliminary periods of 4 d and balance periods of 5 d.

Table 1. *Expt 1. Formulation of barley-based diets (g dry matter (DM) from each component)*

Nitrogen and fibre source...	Barley		Barley + lysine		Barley + hulls		Barley + hulls + lysine	
Antibiotic...	−	+	−	+	−	+	−	+
Barley	768·17	768·17	768·17	768·17	768·17	768·17	768·17	768·17
Lysine*	—	—	2·00	2·00	—	—	2·00	2·00
Barley hulls	—	—	—	—	100·00	100·00	100·00	100·00
Mineral mixture†	40·00	40·00	40·00	40·00	40·00	40·00	40·00	40·00
Vitamin mixture†	16·00	16·00	16·00	16·00	16·00	16·00	16·00	16·00
N-free mixture‡	175·83	175·83	175·83	175·83	75·83	75·83	75·83	75·83
Nebacitin	—	7·00	—	7·00	—	7·00	—	7·00

+, With antibiotic; −, without antibiotic.
* As L-lysine hydrochloride.
† Composition according to Eggum (1973).
‡ For details, see p. 306.

Table 2. *Expt 2. Formulation of soya-bean meal based diets (g dry matter (DM) from each component)*

Nitrogen and fibre source...	Soya-bean meal		Soya-bean meal + methionine		Soya-bean meal + barley hulls		Soya-bean meal + barley hulls + methionine	
Antibiotic...	−	+	−	+	−	+	−	+
Soya-bean meal	181·45	181·45	181·45	181·45	181·45	181·45	181·45	181·45
Methionine*	—	—	2·00	2·00	—	—	2·00	2·00
Barley hulls	—	—	—	—	100·00	100·00	100·00	100·00
Mineral mixture†	40·00	40·00	40·00	40·00	40·00	40·00	40·00	40·00
Vitamin mixture †	16·00	16·00	16·00	16·00	16·00	16·00	16·00	16·00
N-free mixture‡	762·55	762·55	762·55	662·55	762·55	662·55	662·55	662·55
Nebacitin	—	7·00	—	7·00	—	7·00	—	7·00

+, With antibiotic; −, without antibiotic.
* As DL-methionine.
† Composition according to Eggum (1973).
‡ For details, see p. 306.

True digestibility (TD), biological value (BV), net protein utilization (NPU) and digestibility of energy (DE) were measured. At the end of the experiment, when all rats were killed, samples of whole blood were taken from the vena cava for urea determination and samples of caecal contents for measurement of ATP concentration.

Chemical analyses

Dry matter, N, ash and N-free extract were determined according to standard methods (Association of Official Analytical Chemists, 1975). Fat was measured after acid-hydrolysis followed by diethyl ether extraction (Stoldt, 1957).

Amino acid analyses were carried out according to Mason *et al.* (1980). Levels of ATP in caecal contents were measured by a modified method of Wolstrup & Jensen (1976) as described by Bach Knudsen *et al.* (1982).

Table 3. *Proximate analysis and amino acid content of barley, soya-bean meal and barley husk*

	Barley	Soya-bean meal	Barley husk
Dry matter (DM) (g/kg)	871	890	921
Composition (g/kg DM)			
Crude protein (nitrogen × 6·25)	126·5	553·2	31·3
Diethyl ether extract after acid-hydrolysis	23·5	10·0	21·0
Neutral-detergent fibre	164	157*	772
Acid-detergent fibre	69		394
Ash	29·7	63·9	56·1
N-free extract	772·2	340·2	516·5
Amino acids (g/kg crude protein)			
Alanine	40·1	42·2	55·5
Arginine	47·1	71·1	44·4
Aspartic acid	53·7	107·7	73·1
Cystine	21·1	15·1	15·9
Glutamic acid	259·9	178·7	129·6
Glycine	37·9	41·6	46·9
Histidine	20·7	33·8	16·6
Isoleucine	39·0	45·1	31·8
Leucine	70·8	74·9	59·0
Lysine	32·9	59·9	45·9
Methionine	17·8	14·9	17·8
Phenylalanine	51·8	52·2	34·8
Proline	116·6	49·6	59·5
Serine	42·2	49·6	42·3
Threonine	33·1	37·2	37·8
Tryptophan	10·9	10·8	11·7
Tyrosine	30·1	30·0	20·1
Valine	50·9	50·2	45·2

* Value for soya-bean meal supplement from McQueen & Nicholson (1979).

Statistical analyses

All results were subjected to analysis of variance as outlined in the University of British Columbia programme BMD:10 V (Bjerring *et al.* 1975).

Differences between means were tested at $P < 0{\cdot}05$, using Student Newman-Keuls' multiple-range test included in the previously-mentioned programme.

RESULTS

Chemical composition

The proximate analysis of dietary components, and the amino acid composition of the barley, soya-bean meal and barley husk are given in Table 3. As can be seen, the values for barley and soya-bean meal correspond to values normally found in these food sources. The husk of barley contained high levels of neutral-detergent fibre (772 g/kg DM) and high concentrations in the protein of the amino acids lysine, threonine and tryptophan, when compared to the whole grain.

TD

Expt 1. With the barley diets, the addition of lysine slightly increased TD from 0·869 to 0·877, whereas the addition of either Nebacitin or fibre resulted in a decrease of a similar small magnitude (Table 4). None of the interactions was significant.

Table 4. *The effect of lysine, Nebacitin and fibre on protein utilization, energy digestibility, blood urea and microbial activity in the hind-gut of rats given a diet with barley as the only protein source*

Measurement	Main effects								
	Lysine			Nebacitin			Fibre		
	0	+	SEM	0	+	SEM	0	+	SEM
True digestibility (TD)	0·869[a]	0·877[b]	0·002	0·879[a]	0·867[b]	0·002	0·878[a]	0·869[b]	0·002
Biological value (BV)	0·741[a]	0·815[b]	0·003	0·799[a]	0·757[b]	0·003	0·774[a]	0·781[a]	0·003
Net protein utilization (NPU)	0·644[a]	0·715[b]	0·003	0·703[a]	0·656[b]	0·003	0·680[a]	0·679[a]	0·003
Energy digestibility (DE)	0·757[a]	0·755[a]	0·002	0·782[a]	0·731[b]	0·002	0·795[a]	0·717[b]	0·002
Blood urea (mg/l)	184[a]	123[b]	2·0	142[a]	166[b]	2·0	153	154	2·0
ATP (μg/g wet caecal contents)	27·8[a]	31·1[a]	2·1	50·6[a]	8·3[b]	2·1	28·4[a]	30·5[a]	2·1

	First-order interactions														
	Lysine					Lysine					Nebacitin				
	0	0	+	+		0	0	+	+		0	0	+	+	
	Nebacitin					Fibre					Fibre				
	0	+	0	+	SEM	0	+	0	+	SEM	0	+	0	+	SEM
TD	0·873	0·866	0·866	0·869	0·003	0·873	0·866	0·883	0·872	0·003	0·883	0·876	0·873	0·862	0·003
BV	0·761	0·720	0·836	0·793	0·004	0·736	0·745	0·812	0·817	0·004	0·800[a]	0·798[a]	0·749[c]	0·765[b]	0·004
NPU	0·665	0·623	0·741	0·689	0·004	0·643	0·645	0·717	0·713	0·004	0·706	0·699	0·654	0·659	0·004
DE	0·783	0·732	0·781	0·729	0·003	0·796	0·719	0·795	0·715	0·003	0·820	0·744	0·771	0·690	0·003
Blood urea (mg/l)	166	203	118	128	2·8	184	185	122	123	2·8	130[c]	154[b]	176[a]	155[b]	2·8
ATP (μg/g wet caecal contents)	47·1	8·4	54·0	8·2	3·0	24·9	30·7	31·8	30·4	3·0	49·6	51·6	7·1	9·5	3·0

[a, b, c] Within comparison groups, mean values with unlike superscript letters were significantly ($P < 0·05$) different according to Student Newman-Keuls' test. Where no superscripts are provided, interactions were not significant.

Lysine, 2 g L-lysine hydrochloride/kg dry matter (DM); Nebacitin, 7 g Nebacitin (neomycine sulphate–bacitracin; 1:2, w/w)/kg DM; fibre, 100 g barley husk/kg DM; 0, without; +, with.

Expt 2. In contrast to the responses in Expt 1, the addition of amino acid, in this case methionine, to the basal soya-bean protein improved TD only when the diet contained no Nebacitin. The interaction between methionine and fibre indicated a positive response to methionine addition when no fibre was added to the diet, but a negative response, albeit small, when there was an addition of fibre.

BV

Expt 1. Lysine addition to the barley diet improved BV from 0·741 to 0·815. This response was not modified by the addition of Nebacitin or of fibre. The effect of Nebacitin was to decrease BV markedly, with an average reduction for all diets from 0·799 to 0·757. Fibre affected BV only when the diets contained Nebacitin, where addition of fibre increased BV from 0·749 to 0·765.

Expt. 2. The addition of methionine to the soya-bean meal diet increased BV from 0·754 to 0·911. As in Expt 1, this response was not modified by the addition of either Nebacitin or fibre. Both Nebacitin and fibre reduced BV. The reduction with Nebacitin was greater with diets containing no added methionine and no added fibre. Likewise, the reduction in BV as a result of fibre addition was greater in diets containing no added methionine or Nebacitin.

NPU

Expt 1. Lysine addition improved the NPU of barley protein from 0·644 to 0·715, whereas Nebacitin reduced NPU from 0·703 to 0·656. Fibre addition had no effect. Also, none of the interactions were significant.

Expt 2. In common with Expt 1, amino acid addition caused a marked improvement in NPU. In contrast, however, were the effects of Nebacitin and fibre. Fibre addition markedly reduced NPU from a mean of 0·793 to a mean of 0·747. The response to Nebacitin varied, depending on the fibre level; with no additional fibre, there was a decrease in NPU from 0·801 to 0·785 whereas, in the presence of added fibre, NPU rose from 0·740 to 0·754.

DE

Expt 1. DE was not affected by supplementation with lysine, but was markedly reduced as a result of Nebacitin addition (from 0·782 to 0·731) and fibre addition (from 0·795 to 0·717). No interactions were significant.

Expt 2. As in Expt 1, DE was not affected by amino acid supplementation, but was reduced by the addition of Nebacitin (from 0·839 to 0·811) and fibre (from 0·867 to 0·783). No interactions were significant.

Blood urea concentration (BU)

Expt 1. Addition of lysine decreased BU from 184 to 123 mg/l. The addition of Nebacitin to the diet increased BU, but only in those diets which did not contain any additional fibre.

Expt. 2. On the soya-bean meal diets, BU was decreased when additional methionine was provided. A reduction occurred also as a result of Nebacitin addition, although this effect was not evident in diets containing additional methionine. Fibre addition caused a reduction in BU, with this reduction being more marked in diets containing additional methionine (from 121 to 46 mg/l) than in diets not supplemented with methionine (from 187 to 178 mg/l).

Caecal ATP concentration

Expt 1. The only factor to affect caecal ATP concentration was antibiotic supplementation, with the mean concentration reduced five to seven times on diets with Nebacitin, compared to diets with no Nebacitin.

Expt. 2. The results with the soya-bean meal diets were similar to those obtained with

Table 5. *The effect of methionine, Nebacitin and fibre on protein utilization, energy digestibility, blood urea and microbial activity in the hind-gut of rats given a diet with soya-bean meal as the only protein source*

Measurement	Main effects								
	Methionine			Nebacitin			Fibre		
	0	+	SEM	0	+	SEM	0	+	SEM
True digestibility (TD)	0·923[a]	0·926[a]	0·002	0·913[a]	0·937[b]	0·002	0·938[a]	0·912[b]	0·002
Biological value (BV)	0·754[b]	0·911[a]	0·002	0·843[a]	0·821[b]	0·002	0·845[a]	0·820[b]	0·002
Net protein utilization (NPU)	0·696[b]	0·844[a]	0·003	0·771[a]	0·769[a]	0·003	0·793[a]	0·747[b]	0·003
Energy digestibility (DE)	0·829[a]	0·821[a]	0·002	0·839[a]	0·811[b]	0·002	0·867[a]	0·783[b]	0·002
Blood urea (mg/l)	182[a]	84[b]	1·6	147[a]	119[b]	1·6	154[a]	112[b]	1·6
ATP (μg/g wet caecal contents)	6·6[a]	14·5[b]	1·3	18·4[a]	2·8[b]	1·3	10·9[a]	10·2[a]	1·3

Measurement	First-order interactions														
	Methionine					Methionine					Nebacitin				
	0	0	+	+		0	0	+	+		0	0	+	+	
	Nebacitin					Fibre					Fibre				
	0	+	0	+	SEM	0	+	0	+	SEM	0	+	0	+	SEM
TD	0·908[c]	0·939[a]	0·917[b]	0·935[a]	0·003	0·930[b]	0·917[c]	0·946[a]	0·906[d]	0·003	0·928	0·898	0·948	0·925	0·003
BV	0·770[c]	0·737[d]	0·917[a]	0·905[b]	0·003	0·773[c]	0·734[d]	0·917[a]	0·905[b]	0·003	0·863	0·824[bc]	0·827[b]	0·815[c]	0·003
NPU	0·699	0·692	0·842	0·846	0·004	0·718	0·673	0·868	0·820	0·004	0·801[a]	0·740[d]	0·785[b]	0·754[c]	0·004
DE	0·841	0·817	0·837	0·805	0·003	0·870	0·789	0·865	0·777	0·003	0·883	0·795	0·851	0·771	0·003
Blood urea (mg/l)	208[a]	157[b]	86[c]	81[c]	2·2	187[a]	178[b]	121[c]	46d	2·2	165	129	143	95	2·2
ATP (μg/g wet caecal contents)	10·7[b]	2·5[a]	26·0[c]	3·0[a]	1·8	7·1	6·2	14·7	14·3	1·8	19·4	17·4	2·4	3·1	1·8

a,b,c,d, Within comparison groups, mean values with unlike superscript letters were significantly ($P < 0·05$) different according to Student Newman-Keuls' test. Where no superscripts are provided, interactions were not significant.

Methionine, 2 g DL-methionine/kg dry matter (DM); Nebacitin, 7 g Nebacitin (neomycin sulphate–bacitracin; 1:2, w/w)/kg DM; fibre, 100 g barley husk/kg DM; 0, without; +, with.

the barley diets insofar as the reduction in ATP concentration associated with antibiotic inclusion was concerned. The addition of methionine increased ATP concentration, but this effect was not evident when the diet also contained Nebacitin.

DISCUSSION

In both experiments TD of the basal diets was slightly elevated by amino acid supplementation, although significantly only with lysine supplementation. This is in agreement with the results of Tao *et al.* (1971), Gruhn (1976) and Ostrowski (1975) who have shown that lysine supplementation of lysine-deficient diets can improve protein digestibility. This slight elevation in TD could have been due to a lower urea diffusion into the gut as a result of a reduction of BU levels associated with the amino acid supplementation. The simultaneous elevation of BV supports this hypothesis.

The large increase in ATP activity as a result of methionine addition to the soya-bean meal diets seems strange. Complete absorption of purified amino acids could be justifiably assumed (although not easily proven (Rerat *et al.* 1979*b*)), particularly if, as was the case in this experiment, they are intimately mixed with the diet and consumed over an extended portion of each day (Williams & Dunkin, 1980). Consequently, undigested DL-methionine should not have been a significant source of N for the stimulation of hind-gut microbial activity. Neither would an increase in endogenous N be expected with DL-methionine supplementation, nor an increased urea diffusion into the gut (Beames & Eggum, 1981), particularly in view of the fact that TD was not affected. It is thus hypothesized that the stimulation of hind-gut activity by DL-methionine was by some mechanism independent of N concentration. This is supported by the fact that hind-gut microbial activity was not increased by supplementation of the barley with lysine. The stimulation may have been associated with sulphur. S can pass from the serosal to the mucosal side of the gut as inorganic sulphate (Dziewiatkowski, 1970) is a component of the bile acid, taurocholic acid, and has been shown to stimulate microbial growth in the rumen (Moir, 1970).

In each experiment the addition of antibiotic caused a dramatic reduction in gut microbial activity, as also was shown by Eggum *et al.* (1979, 1982*b*) with a concomitant reduction in energy digestibility. The effect was most pronounced with the barley basal diets which had a lower DE value than the soya-bean meal basal diets. The same experience was recorded also by Eggum *et al.* (1982*b*) in experiments with both rats and pigs given diets with different fibre content. The lower values for DE when Nebacitin was added could be explained by a reduction in the production and absorption of organic acids (Eggum *et al.* 1982*a*) although the contribution to NE by such fermentation is still being debated (Partridge, 1982; Eggum & Chwalibog, 1983). By the same reasoning, one would have expected an improvement in DE to result from the increased hind-gut ATP activity on the methionine supplemented soya-bean meal diets. We cannot suggest why this response did not occur. With the barley diets, antibiotic supplementation increased BU concentration, but the effect was restricted to those diets containing no barley husks. In contrast, antibiotic addition to the soya-bean meal diets increased BU concentration, with the effect not apparent on diets containing methionine, where values were already low. An increase in BU associated with a reduction in BV, but virtually no change in TD, as occurred on the barley diets, was expected, but the reason for the reduction in BV as a result of antibiotic feeding is not so clear. It had been postulated by Eggum *et al.* (1979) that such a reduction could be an indication of an absorption of microbially-synthesized essential amino acids in the hind-gut of animals with normal gut flora, although experiments on the infusion of amino acids into the caecum of the pig have been unable to demonstrate any significant improvement in N balance as a result of this infusion (Zebrowska & Buraczewski, 1977; Sauer *et al.* 1977; Hodgdon *et al.* 1977). In both experiments Nebacitin had a lower negative

effect on BV when barley husk was included. As fibre decreases transit time (Raczynski *et al.* 1982) its addition would allow less time for microbial growth and production of the limiting amino acid in the hind-gut.

Hind-gut fermentation could influence BU in one of several ways. With a high N level in the biomass, but a low level of available carbohydrate, the ammonia produced from deamination could be in excess of that required for bacterial synthesis, with absorption facilitated by the buffering action of the alkaline secretions of the glands of the large intestine.

BU normally is inversely correlated with BV (Eggum & Christensen, 1974). This was also demonstrated in the present experiment, where addition of lysine to barley and methionine to soya-bean meal reduced BU markedly, which effect was not reduced by antibiotic or fibre additions. Both fibre and starch, when present in the hind-gut, increase the movement of endogenous N into the hind-gut. In work with pigs, Sauer *et al.* (1977, 1980) have shown cellulose and barley straw to cause an increase, while Mason *et al.* (1976) showed raw potato starch to have a similar effect. However, the effect of these nutrients on BU is not similar. Fibre, when provided in the form of cellulose with brown beans (*Phaseolus vulgaris*) has been shown to increase BU in rats, but to have no effect where the cellulose is provided with casein (Beames & Eggum, 1981) while, in pigs, the fibre has been shown to have no effect (Gargallo & Zimmerman, 1980). If all the nitrogenous end products of fermentation are amines and ammonia, this N would appear to be of little or no nutritional value, with any N in excess of fermentative requirements being absorbed, to cause a large rise in BU (Zebrowska, 1982). However, results in the present study indicate that hind-gut microbially-synthesized amino acids might be absorbed and thus improve dietary protein quality.

Some of the apparent inconsistencies encountered in the results of the present experiments would indicate that the relationships which exist between bacterial activity in the hind-gut, the source and quality of dietary protein and the level and source of carbohydrates may be considerably affected by variations in the form of each nutrient.

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