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Apparent ileal digestibility of amino acids in wet wheat protein and soya bean meal for growing pigs

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A study was conducted on four castrated male pigs to determine the apparent ileal digestibility (AID) of crude protein (CP) and amino acids in wheat protein (WP), a wet by-product of the starch and gluten industry, and in soya bean meal (SBM). The pigs were fitted with a steered ileo-caecal valve cannula at a liveweight of 35 kg. They were assigned to two semi-purified wheat starch based diets, with either WP or SBM as a sole protein source, and fed according to a three-period reversal design. The diets were formulated to contain 140 g CP/kg DM, 11.3 MJ net energy/kg DM and similar amounts of lysine, methionine and threonine.

The CP content and the lysine and threonine contents in CP were lower in WP than in SBM. Nearly half of the DM in WP was starch, and the crude fibre content of the product was very low. The apparent ileal and total tract digestibilities of CP were very similar in both the WP and SBM diets. The AID of methionine was higher in WP (88.2%) than in SBM (84.6%) diets ($P<0.01$) but no differences were found in the AID of the other amino acids. The results show that the AID values of CP and amino acids in WP in growing pigs are comparable to those in SBM.

Key words: cannulation; proteinous by-products; chemical composition

Introduction

The grain starch industry provides several protein-rich by-products useful for pig feeding, such as wheat or maize gluten feed (Knabe et al. 1989, Smits et al. 1992), and barley or oat protein (Näsi 1989, Näsi and Aimonen 1992). In the vicinity to starch plants they can easily be used in undehydrated form on farms with wet feeding systems. Although the protein content and the apparent ileal digestibility (AID) of protein and amino acids in these by-products is usually much higher than that in the grain (Knabe et al. 1989, Buraczewska et al. 1996), the low lysine content limits their use as a sole supplementary protein source for pigs. Fortified with lysine, however, grain protein can replace as much as two thirds of soya bean meal (SBM) in barley-based pig diets (Näsi 1989).

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Wheat protein (WP) is a wet by-product of the grain starch industry of the Raisio Group's Grain Starch Division (Raisio, Finland). During processing, the outer parts of the wheat kernel are first removed as bran and middlings by dry milling. The large A-starch granules of the endosperm are separated from a water-flour mixture by fractionation and the gluten proteins by a maturation process. The soluble proteins of the residual are coagulated by cooking at 104°C for a few minutes. Finally, after cooling, WP is separated from the residual starch, which mainly consists of small B-type granules. The remaining starch is hydrolysed and, after evaporation, formed into wheat syrup.

WP is a low-fibrous feedstuff, containing about 15% dry matter (DM) and 22% crude protein in DM. Data on the apparent total tract digestibility (ATTD) of protein and amino acids in a dried by-product from a roughly similar wheat starch process have been published by Smits et al. (1992). However, it is accepted that ileal digestibility is the most reliable method for determining the amino acid digestibility of feedstuffs for pigs (Sauer and Ozimek 1986, Moughan 1993), but no information on AID values in the wet by-products of wheat starch industry has so far been published.

The objective of the present trial was, first, to evaluate the AID and ATTD of crude protein and the AID of amino acids in WP for growing pigs and, second, to compare the digestibility of the nutrients in WP with those in SBM.

Material and methods

Animals and experimental procedure

Four castrated Finnish Yorkshire male pigs, at an average initial liveweight of 35 kg, were surgically fitted with steered ileo-caecal valve (SICV) cannulae under halothane anaesthesia. A schematic view of the cannulation and details of the surgical operation were presented by Mroz et al. (1996). The pigs were individually housed in metabolic pens (143 x 123 cm, slatted plastic floor), at a room temperature of +23°C during the recovery period and at +18°C during the trial. The pigs weighed 44.4 (SE 3.2) kg at the beginning and 91 (SE 1.8) kg at the end of the trial. The pigs were randomly assigned to two semi-purified wheat starch-based diets containing either WP or normal solvent-extracted SBM as the sole protein source. The trial consisted of three experimental periods, and the pigs were transferred from one diet to the other after each period. The duration of the period was 16 days, including 7 days of adaptation, 5 days of faeces collection and finally, on days 13 and 16, ileal digesta collection. The faeces were collected quantitatively into a plastic bag attached to the pig with a Velcro support system (Van Kleef et al. 1994), that was glued to the skin around the anus. The faeces were stored frozen at –20°C. Ileal digesta was collected for 12 hours on each of 2 days, between 6.00 and 18.00 h, into a plastic bag connected to the barrel of the cannula. The bags were replaced with new ones at least hourly and the digesta was quick-frozen at –30°C. Cumulative feed samples of both protein sources and the semi-purified parts of the diets were collected during the experiment.

Feeds and feeding

Before the operation and during the 3-week recovery period, the pigs were fed a standard barley-soya bean meal diet. During recovery, the feed allowance was gradually increased to the experimental level. Two experimental diets were formulated to be isonitrogenous and isoenergetic and to contain equal amounts of lysine, threonine and methionine (Table 1). The WP batch used was frozen in plastic containers and defrosted for a few days in a cold store before feeding.

Chromium (Cr), ytterbium (Yb) and cobalt (Co) were used as indigestible markers in the trial. Chromium-mordanted straw (70 g/kg Cr) was prepared as described by Udén et al. (1980), and 3 g of the ground straw was mixed with each

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Table 1. Composition and calculated analyses of the experimental diets based on wet wheat protein or soya bean meal (g/kg DM).

	WP diet	SBM diet
Wheat protein	516	–
Soya bean meal	–	278
Wheat starch	339	574
Cellulose	55	40
Sugar	50	50
Vegetable oil	–	20.7
Dicalcium monophosphate	21.4	23.2
Calcium carbonate	7.3	5.6
Mineral+vitamin premix[1]	5.0	5.0
NaCl	3.0	3.0
L lysine HCl	2.7	–
DL methionine	–	0.6
L threonine	0.8	–
Calculated analyses		
Crude protein	141	141
Crude fibre	46	47
Lysine	8.5	8.5
Methionine	2.7	2.5
Threonine	5.5	5.4
Phosphorus	6.0	6.0
Calcium	7.5	7.5
Energy value, NE, MJ/kg DM[2]	11.27	11.26

WP=wheat protein; SBM=soya bean meal

1) Mineral+vitamin-premix provided the following vitamins and trace minerals per kilogram dry matter of diet: 2500 IU vitamin A, 500 IU vitamin D, 31.2 mg vitamin E, 1.5 mg thiamine, 3.0 mg riboflavin, 1.25 mg pyridoxine, 10 µg vitamin B_{12}, 50 mg choline, 7.5 mg pantothenic acid, 6.2 mg nicotinic acid, 1.1 mg folic acid, 48 mg Fe, 110 mg Zn, 19 mg Cu, 15.5 mg Mn, 0.34 mg I and 0.11 mg Se and barley as a carrier.

2) According to Tuori et al. (1996).

meal. Ytterbium acetate ($Yb(CH_3COO)_3$) and lithium-Co-EDTA complex (LiCoEDTA) were dissolved in water in a ratio of 1:2.5:40, and 20 ml of the solution was first mixed with the liquid part of the diets (WP or water), providing 200 mg of Yb and Co for each meal. The dry and liquid feed ingredients were mixed directly prior to feeding. The water to feed ratio in the SBM diet was 2:1. The pigs were fed twice daily (at 6.00 and 18.00), at a feeding level of 90 g DM/kg liveweight$^{0.75}$/day. Water was freely available from low-pressure drinking nipples.

Analytical procedure

The faeces were defrosted before pooling, but the digesta was pooled in a frozen state. All the samples were freeze-dried and ground to pass a 1-mm sieve. Their proximate composition was analysed by the methods of AOAC (1984). Ether extract was determined after 4 N HCl hydrolysis. The amino acid composition of WP, SBM and ileal digesta was assayed on an LKB Alfa plus II amino acid analyser after hydrolysis for 23 h in 6 N HCl. Methionine and cystine were determined after oxidation with performic acid before acid hydrolysis, whereas tryptophan was determined as barium hydroxide hydrolyzate. The starch in WP and SBM was measured as glucose after hydrolysis with thermamyl and amyloglucosidase and the sugar composition with HPLC after ethanol extraction and Sep-Pak C18 purification. The lactic acid in WP was determined with a colorimetric method of Barker and Summerson (1941). The calcium in WP and SBM and the concentrations of Cr, Yb and Co in the chromium-mordanted straw, ($Yb(CH_3COO)_3$) – LiCoEDTA solution, faeces and digesta were analysed by atomic absorption spectroscopy. The phosphorus in WP and SBM was assayed by photometry after ammonium vanadate pretreatment. All analyses were made in duplicate, except those of the marker concentrations, which were determined with a single assay.

The AID values were calculated from the marker ratio of the feeds and digesta and the ATTD values from the total collection of faeces. The digestibility coefficients calculated using Cr as the marker are presented here, since they showed the smallest variation (standard error of the mean). The results of a comparison of the different markers will be later published by Valaja et al..

The digestibility data were subjected to a least-squares analysis of variance (Snedecor and Cochran 1989) according to the following model:

$$Y_{ijk}=\mu+A_i+P_j+D_k+e_{ijk},$$

where μ = the overall mean, A_i = the effect of animal i, P_j = the effect of period j, D_k = the effect of diet k and e_{ijk}= the residual term.

Table 2. Analysed chemical compositions of wet wheat protein, soya bean meal and diets based on wet wheat protein or soya bean meal.

	WP	SBM	WP diet[1]	SBM diet[1]
Dry matter, g/kg	153	906	250	910
Ash, g/kg DM	25	63	43	50
Crude protein, g/kg DM	240	478	135	144
Ether extract, g/kg DM	38	46	22	37
Crude fibre, g/kg DM	20	85	51	58
Nitrogen free extract, g/kg DM	677	328	749	711
Starch, g/kg DM	490	10		
Total sugars, g/kg DM	86	18		
Lactic acid, g/kg DM	18	ND		
Phosphorus, g/kg DM	4.5	7.5		
Calcium	1.3	3.3		
Amino acids[2]	g/16 g N		g/kg DM	
Indispensable				
Arginine	6.5	7.6	8.5	10.5
Histidine	2.6	2.8	3.4	3.8
Isoleucine	3.8	4.7	4.9	6.5
Leucine	7.6	7.9	9.9	10.8
Lysine	4.5	6.5	8.1	9.0
Methionine	2.0	1.5	2.6	2.7
Phenylalanine	4.4	5.3	5.8	7.2
Threonine	3.5	4.2	5.5	5.8
Tryptophan	1.5	1.4	2.1	1.9
Valine	5.7	5.1	7.4	7.0
Dispensable				
Alanine	5.2	4.5	6.7	6.2
Aspartic acid	7.3	11.8	9.6	16.2
Cystine	2.5	1.5	3.3	2.0
Glutamic acid	18.8	17.5	24.7	24.3
Glycine	4.7	4.5	6.1	6.3
Proline	7.5	5.3	9.9	7.4
Serine	4.7	5.4	6.1	7.4
Tyrosine	3.6	3.8	4.7	5.2

WP=wheat protein; SBM=soya bean meal; ND=not determined.

[1]Analysed chemical composition of the diets: sum of nutrients content in WP or in SBM and in the mixture of the other ingredients.

[2]Amino acid composition in WP and SBM: n=2.

Results and discussion

Chemical composition

As compared with SBM, WP protein was relatively poor in lysine and threonine but rich in sulphur-containing amino acids (Table 2). The contents of indispensable amino acids, except phenylalanine, were somewhat higher (0.3–2.0% units) in WP protein than in whole wheat (Just et al. 1983, Tuori et al. 1996). By contrast, the protein in WP contained 4.2% units less glutamic acid and 2.8% units less proline than did CP in wheat grains, the gluten storage proteins having been separated from wheat endosperm after the

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starch process. Up to 90% of CP in wheat is gluten, rich in both glutamic acid and proline (36% and 13% of total amino acids), but poor in lysine and threonine (Lookhart 1991). The essential to non-essential amino acids ratio in WP, 44/56, was slightly higher than that in wheat grains, 38/62 (Just et al. 1983, Tuori et al. 1996), which may also reflect a reduction in the gluten content in WP.

The content of lysine in WP (4.5 g/16 g N) was higher than that found by Smits et al. (1992) in a protein fraction (wheat gluten feed) derived from the wheat starch and gluten industry (3 g/16 g N), which may be caused by the different efficiency of the gluten process. Grain protein from the barley starch industry (Näsi and Aimonen 1992, Buraczewska et al. 1996) had a lower lysine content than the protein in WP, whereas oat protein (Näsi and Aimonen 1992) had a similar content. The CP content of these products ranged, however, from 158 (wheat gluten feed) to 408 g/kg DM (oat protein), resulting in large differences in the total content of lysine and other amino acids.

The crude fat of the grain was enriched in WP, as it was in the protein fractions mentioned above. Separation of the residual starch and coagulated proteins seemed to be rather incomplete, as nearly half of the DM in WP was starch. The sugar fraction consisted of maltose (47 g/kg DM), glucose (37 g/kg DM) and small amounts of fructose and saccharose (2.5 and 0.6 g/kg DM). Comparable starch and sugar contents were found in barley protein by Näsi and Aimonen (1992). WP also contained a small amount of lactic acid and the average pH of the product was 5.6.

The phosphorus content of WP, 4.5 g/kg DM, was slightly higher than that of whole wheat, 3.8–4.0 g/kg DM (Just et al. 1983). Remarkably high phosphorus contents (10–13 g/kg DM) were found in wheat gluten feed by Smits et al. (1992), even though the phosphorus-rich aleurone was not present in the product. Barley protein also had a higher phosphorus content (7 g/kg DM) than did WP (Buraczewska et al. 1996). The calcium content of these protein fractions was, however, rather similar (1–1.3 g/kg DM).

The experimental diets (Table 2) contained nearly similar amounts of threonine and methionine but the analysed lysine content of the WP diet was lower than that of the SBM diet. The formulation of the WP diet was based on the analyses a sample removed from the process before the experiment, which had a higher lysine content than the sample collected during the trial.

Nutrient digestibility

One pig, which recovered slowly from the operation, was excluded from the first experimental period but taken back to the trial for the last two periods. At the beginning of the second period, the cannula of another pig slipped out of the gut, and the pig was removed from the trial. Therefore, the final number of observations was five in WP and four in SBM diets. The pigs ate the experimental diets readily and their average daily weight gain was 1017 (SE 21) g in the course of the trial. Necropsy revealed that, as in the case of pigs with simple T or re-entrant cannulae, some adherences existed around of the terminal ileum of the pigs, but this had no impact on the digesta passage and quantitative collection.

The AID coefficients of CP and individual amino acids in WP and SBM are presented in Table 3. Their calculation was based on the assumption that the supplemented synthetic amino acids were completely absorbed in the ileum (Leibholz et al. 1986). The AID of methionine was higher in WP than in SBM (3.6% units; $P<0.01$), but there was no difference in the AID of the other amino acids between these protein sources ($P>0.1$).

The AID values of the essential amino acids, except methionine, in SBM were somewhat higher than those reported by Sauer and Ozimek (1986), Furuya and Kaji (1989), Knabe et al. (1989), De Lange et al. (1990) and Marty et al. (1994), but lower than those presented by ITCF-Eurolysine (1993), which were determined with ileo-rectal anastomised pigs. The variation noted between individual experiments may have been caused by the quantity and quality of die-

Table 3. Apparent ileal digestibility (%) of amino acids in pigs fed wet wheat protein or soya bean meal diets.

	WP diet	SBM diet	SEM	Statistical significance
Amino acids, indispensable				
Arginine	91.1	92.5	0.72	NS
Histidine	88.6	89.4	1.18	NS
Isoleucine	85.6	86.8	0.76	NS
Leucine	88.4	87.1	0.95	NS
Lysine	83.2	87.6	1.37	NS
Methionine	88.2	84.6	0.26	**
Phenylalanine	88.3	87.7	0.91	NS
Threonine	80.2	82.3	2.00	NS
Tryptophan	86.1	84.4	1.37	NS
Valine	87.0	84.8	1.11	NS
Total indispensable amino acids	87.1	87.4	1.02	NS
Amino acids, dispensable				
Alanine	84.8	82.2	1.32	NS
Aspartic acid	81.6	87.2	2.10	NS
Cystine	86.3	85.1	1.93	NS
Glutamic acid	91.1	91.0	0.78	NS
Glycine	80.4	79.6	1.45	NS
Proline	90.6	87.4	1.30	NS
Serine	85.5	86.7	1.65	NS
Tyrosine	87.9	85.9	0.87	NS
Total indispensable amino acids	87.3	87.2	1.27	NS

WP=wet wheat protein; SBM=soya bean meal; SEM=standard error of the mean.
Statistical significance: NS = non-significant ($P>0.1$), ** = $P<0.01$.
SEM has been shown for WP diets. To obtain the corresponding values for SBM diets, the values for WP diets should be multiplied by 0.827.

tary fibre, processing conditions and residual levels of trypsin inhibitors in SBM (Sauer and Ozimek 1986, Schulze et al. 1994).

Threonine was the least digestible indispensable amino acid and glycine the least digestible dispensable amino acid in both WP and SBM. The consistently low AID obtained for these amino acids is partly due to their relatively high concentrations in endogenous secretions (Sauer et al. 1977, Low 1979). Small intestinal secretion is very rich in threonine, and glycine accounts for more than 90% of all amino acids secreted in porcine bile juice (Souffrant 1991). According to Low (1980), threonine has the shortest potential absorption time, because it appears last in the small intestine, after enzymic hydrolysis.

The AID values of the indispensable amino acids, except phenylalanine, were higher in WP than in whole wheat (Sauer and Ozimek 1986, De Lange et al. 1990). The high AID of amino acids can partly be explained by the very low content of crude fibre in WP. Sauer et al. (1977) showed that the AID of lysine and threonine in wheat milling fractions was related to the fibre content and declined from wheat flour to whole wheat to wheat offal (a mixture of bran, shorts and middlings).

The high AID of the amino acids in WP indicates that heat treatment at +104°C during the coagulation step was unlikely to have caused extensive damage to the proteins. Moreover, the AID of CP and amino acids in WP was closely similar to that in wet barley protein (Buracze-

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Table 4. Apparent ileal and total tract digestibilities of dietary proximate components in pigs fed wet wheat protein or soya bean meal diets.

	Apparent ileal digestibility, %				Total tract digestibility, %			
	WP diet	SBM diet	SEM	Significance	WP diet	SBM diet	SEM	Significance
Dry matter	83.3	83.3	2.12	NS	91.1	92.4	0.64	NS
Organic matter	85.0	85.5	2.07	NS	92.6	94.2	0.65	NS
Ash	46.3	40.9	3.46	NS	57.2	58.4	0.49	NS
Crude protein	82.5	82.9	1.18	NS	89.2	89.6	1.17	NS
Ether extract	77.8	81.3	2.59	NS	78.0	79.7	0.09	**
Crude fibre	1.7	12.6	15.0	NS	41.0	60.9	9.47	NS
Nitrogen free extract	91.9	92.8	1.33	NS	97.3	98.8	0.21	*

WP=wet wheat protein; SBM= soya been meal; SEM=standard error of the mean.
Statistical significance: NS= non-significant (P>0.1), * = P<0.05, ** = P<0.01.
SEM has been shown for WP diets. To obtain the corresponding values for SBM diets, the values for WP diets should be multiplied by 0.827.

wska et al. 1996), processed at much lower temperatures (Linko et al. 1989). Additional heat damage to the proteins can be avoided by using protein feedstuffs of the starch industry in a wet form. Linko et al. (1989) showed that direct hot-air drying of barley protein was detrimental to protein quality.

The lactic acid present in WP may have a favourable effect on protein digestion and absorption. Acidification of grower pigs' diets with 2% propionic acid has promoted the AID of several indispensable amino acids and decreased the concentration of amines in the caecal digesta, indicating a suppressive effect on microbial activity (Mosenthin et al. 1992).

The AID and the ATTD of the proximate components in both WP and SBM diets were high (Table 4), as they typically are in low-fibrous semi-purified diets. The differences in the AID of the proximate components between the diets were insignificant (P>0.1). The ATTD values of ether extract (P<0.01) and nitrogen-free extract (P<0.05) were higher the SBM diet than in the WP diet. For nitrogen-free extract, the difference is obviously caused by the greater proportion of fully digestible starch in the SBM diet than in the WP diet and, for ether extract, by the addition of vegetable oil to the SBM diet. Just et al. (1980) showed that the apparent digestibility of fat is improved by increasing the concentration of crude fat in the diet.

The ATTD of CP in the SBM batch used was comparable with that reported by Knabe et al. (1989), but clearly higher than that found in normal and processed SBMs in the study of Näsi (1991). The ATTD of protein in WP was similar to that in wet barley protein in the study of Buraczewska et al. (1996), but higher than that found in dried wheat gluten feed (Smits et al. 1992).

Nearly 90% of CP in both protein sources was digested in the ileum, and disappearance of CP from the large intestine of pigs fed WP and SBM diets was rather low, 6.9% and 6.7% units, as expressed by CP intake. The protein absorbed from the large intestine is lost from the protein synthesis of the pig and excreted in the urine (Zebrowska 1973).

In conclusion, our results show that the protein content of WP from the wheat starch and gluten industry was lower than that of SBM, and the protein in WP contained less lysine and threonine than did that of SBM. Starch was the main component in the DM of WP, and the crude fibre content of the product was very low. The apparent ileal digestibilities of protein and amino acids in WP were as high as in SBM and the digestibility of methionine was even higher

(3.6 %-units). Although the number of observations in the study was small, the digestibility values noted for WP and SBM agreed well with the data presented earlier for similar by-products from the starch or oil industry.

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SELOSTUS

Kuivaamattoman vehnäproteiinin ja soijarouheen aminohappojen ohutsuolisulavuus sioilla

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Kokeessa määritettiin vehnätärkkelys- ja gluteeniteollisuuden (Raision Yhtymä) sivutuotteena syntyvän kuivaamattoman vehnäproteiinin ja vertailurehuna soijarouheen valkuaisen ja aminohappojen ohutsuolisulavuus neljällä leikkosialla. Sioille asennettiin 35 kilon painoisina umpisuoleen SICV-kanyyli, jonka kautta voitiin kerätä ohutsuolen ruokasulaa. Siat ruokittiin vehnätärkkelyspohjaisella puolipuhdistetulla rehulla, jonka ainoana valkuaisen lähteenä oli joko vehnäproteiini tai soijarouhe. Rehuissa oli 140 g raakavalkuaista ja 11,3 MJ nettoenergiaa kuiva-ainekilossa ja niiden lysiini-, metioniini- ja treoniinipitoisuus oli samanlainen. Kokeessa oli kolme jaksoa, ja jakson vaihtuessa siat siirrettiin ruokinnalta toiselle.

Tutkitun vehnäproteiinierän valkuaispitoisuus oli pienempi kuin soijarouheen (240 g/kg ka vs. 478 g kg/ka), ja sen valkuainen sisälsi vähemmän lysiiniä (4,5 vs. 6,5 g/16 g N) ja treoniinia (3,5 vs. 4,2 g/ 16 g N) kuin soijarouheen. Noin puolet vehnäproteiinin kuiva-aineesta oli tärkkelystä, ja raakakuitupitoisuus oli pieni (20 g/kg ka). Vehnäproteiinin valkuaisen ohutsuolisulavuus (82,5 vs. 82,9 %) ja kokonaissulavuus (89,4 vs. 89,6 %) olivat lähes samat kuin soijarouheen. Vehnäproteiinin aminohappojen ohutsuolisulavuus oli hyvä, metioniinin ohutsuolisulavuus (88,2 %) jopa parempi kuin soijarouheen (84,6 %). Kokeen tulosten perusteella vehnäproteiinin valkuaisen ja aminohappojen ohutsuolisulavuus on hyvin verrattavissa soijarouheen sulavuusarvoihin.