

# The enzymatic activity of the small intestine mucosa of young pigs as affected by high fibre diets fed after weaning

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## ABSTRACT

Two groups of gilts were fed from 10 to 25 kg BW on a standard low fibre (LF) diet, and two groups on a LF diet supplemented with 10 or 20% grass meal (MF and HF diet, respectively). The fibre content was 37.9, 53.5 and 76.4 g/kg in the LF, MF and HF diets, respectively. At 25 kg BW the pigs from one group fed on LF, and groups on MF and HF diets, were assigned to the LF diet, while a second group fed on LF was transferred to the HF diet. After 14 days the pigs were slaughtered and samples of mucosa from the proximal, middle and distal part of the small intestine were taken and analysed for the activity of sucrase, aminopeptidase A and N, and dipeptidyl peptidase IV. The activity of peptidases was higher in pigs fed MF and HF diets from 10 to 25 kg and the HF diet 14 days before slaughter than in animals continuously fed on the standard LF diet, while the activity of sucrase was higher in pigs fed on the MF diet from 10 to 25 kg than continuously on the LF diet.

KEY WORDS: piglets, fibre, enzyme activity

## INTRODUCTION

Development of digestive-tract enzymatic functions in piglets around weaning is well known (Hedemann et al., 2003; Hedemann and Jensen, 2004). However, no information is available on the effects of feeding a high fibre diet after weaning on enzymatic function of the small intestine mucosa in pigs fed during the following period on a standard low fibre diet.

The objective of the study was to determine the effect of temporary fibre supplementation of diets fed to piglets after weaning, on the activity of sucrase and peptidases in the mucosa of the small intestine after a period of feeding a standard diet.

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## MATERIAL AND METHODS

The diets differing in fibre content were formulated by mixing a standard low fibre diet (LF) with 10 or 20% of grass meal (diet MF and HF, respectively). The crude fibre contents in diets LF, MF and HF were 37.9, 53.5 and 76.4 g/kg, respectively. The experiment was carried out with four litters of five gilts allotted after weaning to four groups, and comprised two periods. From 10 to 25 kg BW the piglets were fed on diets differing in fibre contents, i.e. on LF (group 1 and 4), MF (group 2) or HF diets (group 3), and then were transferred for 14 days to the LF (group 1, 2 and 3) or to HF (group 4) diet (Table 1). After 14 days the pigs were slaughtered, the small intestine removed, separated from mesenteric fat, and divided into three equal sections: proximal, middle and distal. From each section a sample of mucosa was taken, frozen in liquid nitrogen and stored at 70°C until analysis. The activities of sucrase (Dahlquist, 1964), aminopeptidases A and N (Maroux et al., 1973) and dipepidyl peptidase (Nagatsu et al., 1976) were determined in mucosa homogenates.

Analysis of variance was performed using Statgraphics version 6.0 Plus software.

Table 1. Enzyme activity of the small intestine mucosa (mmol/g mucosa) as affected by previous fibre diet

Group	Diets fed in the period		Segments	Sucrase	Aminopeptidase		Dipepidyl peptidase IV
	10-25 kg BW 14 days				A	N	
1	LF	LF	Proximal	3.54	2.42	6.23	4.70
			Middle	4.66	5.54	9.96	6.13
			Distal	3.98	9.01	11.66	8.48
2	MF	LF	Proximal	3.59	6.38	15.44	7.97
			Middle	6.96	11.05	13.20	12.72
			Distal	7.35	15.49	17.61	17.47
3	HF	LF	Proximal	3.98	4.36	7.91	6.00
			Middle	6.02	8.88	14.77	11.49
			Distal	6.46	12.93	18.76	16.90
4	LF	HF	Proximal	2.93	3.74	7.06	6.47
			Middle	6.78	10.67	17.93	11.22
			Distal	6.09	9.35	10.28	12.52
		SEM		0.97	0.95	1.02	1.08
Statistical significance	Group			*	**	**	**
		Segment		NS	**	**	**
Interaction				NS	NS	NS	NS

\* P<0.05, \*\* P<0.01, NS - not significant

## RESULTS AND DISCUSSION

Sucrase activity was the lowest in pigs fed the standard low fibre diet in both periods (group 1, mean from three segments 4.06 mmol/g mucosa) and the highest in pigs fed on the MF diet followed by LF (group 2, mean 5.97 mmol/g;  $P < 0.05$ ; Table 1). Increasing the fibre content and feeding the high fibre diet in the period preceding slaughter (group 3 and 4, respectively) did not significantly affect sucrase activity. The activity of this enzyme was not significantly related to the segment of the intestine, but a tendency was observed towards lower activity in the proximal part than in the middle and distal ones.

Proteolytic enzyme activity was affected both by the segment of intestine and the diet. The activity of all aminopeptidases was greater in the middle and distal segments than in the proximal part. It was also higher in pigs from groups 2 and 3 fed on both diets with a higher fibre content from 10 to 25 kg body weight, and in group 4 fed on the high-fibre diet 14 days before slaughter. The increase of aminopeptidase activities was found in all segments.

The results indicate that feeding a diet with increased fibre for 14 days increases the enzymatic activity of intestinal mucosa. This may be considered a compensatory response of the digestive tract to the inhibitory effect of fibre on digestion of nutrients (Jørgensen et al., 1996). The considerably higher enzymatic activity of mucosa found in pigs after a 14-day period of feeding a low fibre diet, following feeding diets with a higher fibre content, points to a persistent effect of fibre on the digestive tract of young pigs.

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## STRESZCZENIE

**Aktywność enzymatyczna śluzówki jelita cienkiego prosiąt żywionych po odsadzeniu dietami z dużą zawartością włókna**

Dwie grupy loszek odsadzonych przy masie ciała 10 kg żywiono dietą standardową o niskiej zawartości włókna (LF), a dwie grupy dietą LF z dodatkiem 10 lub 20% mączki z traw (odpowiednio dieta MF i HF). Zawartość włókna w diecie LF, MF i HF wynosiła odpowiednio 37,9; 53,5 i 76,4 g/kg. Po osiągnięciu 25 kg m.c. loszki z jednej grupy LF oraz z grup MF i HF żywiono dietą LF, zaś zwierzętom z drugiej grupy LF podawano dietę HF. Po 14 dniach świnie ubito, pobrano śluzówkę z trzech odcinków jelita cienkiego i oznaczono aktywność sacharazy, aminopeptydazy A i N oraz dipepidyl peptydazy IV. Aktywność peptydaz była wyższa u świń żywionych dietami MF i HF od m.c. 10 do 25 kg oraz dietą HF przez 14 dni przed ubojem, niż u zwierząt żywionych przez cały czas doświadczania dietą LF. Aktywność sacharazy była wyższa u świń żywionych dietą MF od 10 do 25 kg m.c. niż otrzymujących przez cały czas dietę LF.

# The effect of vitamin E on the quality of meat of pigs fed maize grain silage

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## ABSTRACT

In an experiment carried out on 48 fattening pigs the effects of adding vitamin E to maize grain silage fed during the whole fattening period from 60 to 111 kg BW on meat quality and storage stability were studied. Pigs originated from PL × LWP sows mated to a D × Pi boar were fed restrictively according to body weight. All animals received the same daily amounts of maize grain silage and soyabean meal, but the diet for group II was supplemented with vitamin E. Vitamin E supplementation slightly improved meat quality traits and significantly ( $P \leq 0.05$ ) improved meat shelf-life during frozen storage.

KEY WORDS: maize grain silage, vitamin E, pork quality

## INTRODUCTION

Over the last two years the use of farm-grown feeds in pig fattening has attracted growing attention for economic reasons. Maize grain, characterized by a high energy concentration, nutrient digestibility, lack of antinutritive factors, and satisfactory yields is recommended, particularly in the form of grain silage. The expected cost reduction can be obtained especially when this silage is fed up to the end of the fattening period. The high content of unsaturated fatty acids in maize grain creates problems with meat quality and stability during storage. This kind of diet influences the meat fatty acid profile and stimulates lipid oxidation processes (D'Arrigo et al., 2002), which generate undesirable organoleptic characteristics such as odour, taste and colour as well as decrease the shelf-life of meat shortly after slaughter. Meat and fat quality can be protected from these oxidation products either by limiting this kind of diet before slaughter (Sommer, 1994; Burgstaller, 1995) or by supplementing it with higher doses of the biological antioxidant, vitamin E (Jensen et al., 1998).

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The aim of this experiment was to investigate the effectiveness of adding vitamin E to maize grain silage fed to pigs during the entire fattening period on meat quality and stability.

## MATERIAL AND METHODS

The experiment was carried out on 48 pigs originated from Polish Landrace × Large White Polish sows mated to a Duroc × Pietrain boar, and divided into two restrictively fed groups according to body weight from approximately 60 to 111 kg. For the whole fattening period, group I received crushed maize grain silage in amounts of 2.2-2.8 kg daily and, as the protein component, soyabean meal (0.6 to 0.5 kg/d). For pigs in group II the same diet was supplemented with 600 mg of vitamin E per kg of soyabean meal. Daily feed rations for pigs in both groups were supplemented with vitamins and minerals. All animals were kept in individual, straw-bedded pens. At the end of the experiment all of the pigs were slaughtered, right sides of carcasses were evaluated and samples of *longissimus dorsi* muscle, obtained from the area of the last thoracic and first lumbar vertebra, were collected for basic chemical analysis (AOAC, 1990), TBA-RS content (Pikul et al., 1989) and fatty acid profile (gas chromatograph Varian 3400). Backfat samples were taken from the same place for peroxide value estimation (AOAC, 1990). TBA-RS, peroxide value and meat colour (colourimeter Minolta CR-310) were measured after 2 weeks and 4 months of storage at -25°C. All data were subjected to analysis of variance (Statistica, 1997).

## RESULTS

The results presented in Table 1 indicate that maize silage supplemented with vitamin E had no effect on pigs' carcass quality. All measured parameters were slightly better in pigs receiving vitamin E, but the differences were not statistically significant.

Table 1. Results of carcass quality evaluation

Item	Experimental groups		SEM
	I - maize grain silage	II - maize grain silage with vitamin E	
Body weight at slaughter, kg	111	111	0.282
Meat content in primal cuts, kg	26.08	26.25	0.477
Meat in proper ham, %	75.18	75.38	0.778
Loin eye area, cm <sup>2</sup>	59.02	59.39	1.754
Carcass meatiness, %	58.73	60.07	0.873
Backfat thickness of 5 measurements, cm	2.42	2.37	0.066

Some of the meat quality and stability parameters assessed in the present experiment are given in Table 2. The chemical composition of *M. longissimus dorsi*

was similar in both groups. The vitamin E-supplemented maize silage increased the polyunsaturated fatty acid content in meat by 1.9%, but the differences were not significant. A tendency towards improved (by 1.7 to 3.33%) meat redness and yellowness, measured after 2 weeks and 4 months of storage, was observed in pigs receiving vitamin E. Supplementation of the maize silage with vitamin E significantly ( $P \leq 0.05$ ) decreased the peroxide value and TBA-RS content in meat after 4 months of frozen storage by approximately 36 and 18%, respectively in comparison with the unsupplemented group.

Table 2. Some indices of *M. longissimus dorsi* meat quality and stability

Item	Experimental groups		SEM
	I - maize grain silage	II – maize grain silage with vitamin E	
Crude protein content, %	23.08	23.40	0.129
Crude fat content, %	1.97	1.78	0.144
PUFA, %	21.64	22.05	1.075
Meat colour on Hunter scale after 2 weeks			
lightness	44.58	45.40	0.590
redness	12.86	13.08	0.120
yellowness	2.40	2.32	0.103
Meat colour on Hunter scale after 4 months			
lightness	42.94	44.63	0.561
redness	12.29	12.69	0.121
yellowness	3.62	3.52	0.106
Peroxide value after 2 weeks, meq/kg	0.473	0.401	0.056
Peroxide value after 4 months, meq/kg	0.959 <sup>b</sup>	0.614 <sup>a</sup>	0.075
TBA-RS after 2 weeks, mg/kg	0.457	0.454	0.014
TBA-RS after 4 months, mg/kg	0.625 <sup>b</sup>	0.510 <sup>a</sup>	0.028

<sup>a,b</sup>-  $P \leq 0.05$

## DISCUSSION

The main hypothesis examined in the present experiment was the possibility of improving the quality and storage stability of meat from pigs fed high amounts of maize grain silage to the end of the fattening period. The obtained results indicate a tendency towards improved carcass and meat quality in pigs fed maize grain silage with added vitamin E, but the differences were not significant. Supplementation prevented the meat from yellowing and increased meat redness after frozen storage, which is always desired by consumers (Jensen et al., 1998). Meat colour stabilization agrees with the findings obtained by the present author in a previous experiment (Hanczakowska, 2004) as well as in other studies (Monahan et al., 1992). The antioxidative role of vitamin E was confirmed by a

slight upward tendency in the PUFA content of meat. The iodometric peroxide value is used as the main indicator of the initial phases of unsaturated fatty acid oxidation, while a typical indicator of the secondary products of lipid oxidation is 2-thiobarbituric acid-reactive substances (TBA-RS). Both indices demonstrate that vitamin E is 56 and 66% effective in inhibiting lipid oxidation processes at the initial and final phase, respectively. Similar vitamin E activity was also noted by D'Arrigo et al. (2002).

## CONCLUSIONS

The quality and shelf-life of meat obtained from pigs receiving ensiled maize grain for the whole fattening period can be improved by supplementing their diet with the antioxidant, vitamin E.

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## STRESZCZENIE

### **Wpływ dodatku witaminy E na jakość mięsa świń żywionych kiszonym ziarnem kukurydzy**

W doświadczeniu przeprowadzonym na 48 tucznikach określano wpływ dodatku witaminy E do dawek dla świń żywionych kiszonym ziarnem kukurydzy przez cały okres tuczu od 60 do 111 kg m.c. na jakość i trwałość mięsa. Tuczniaki pochodzące od loch (wbp  $\times$  pbz) pokrytych knurem (D  $\times$  Pi) żywiono ograniczonymi dawkami paszy odpowiednio do masy ciała. Wszystkie zwierzęta otrzymywały taką samą dzienną dawkę kiszonego ziarna kukurydzy i śruty sojowej, ale dla grupy II zastosowano dodatek witaminy E. Dodatek witaminy E do dawek pokarmowych polepszył nieco badane cechy jakości mięsa oraz istotnie ( $P \leq 0,05$ ) poprawił trwałość mięsa w czasie mrożenia.

# The effect of organic feeding on carcass and meat quality of fattening pigs

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## ABSTRACT

The effect of organic feeding on carcass and meat quality was investigated in 160 fattening pigs. All animals received a feed mixture composed of either standard (group I-M1) or organically grown cereals, leguminous seeds and rapeseed cake (experimental groups - M). The pigs additionally received organically cultivated whole-plant maize silage (SM) in groups II and III, and grass silage (SG) in groups IV and V. In groups III and V the animals also received a herb mixture (H). Feeding animals with SM or SG and the herb supplement had an apparent positive effect on carcass quality, meat tenderness and taste. The backfat of the experimental animals contained more PUFA.

KEY WORDS: organic feed, pig, carcass, meat quality

## INTRODUCTION

Increasing consumer demands in terms of animal product quality are associated with concern about farm animal welfare and feeding. This has resulted in growing numbers of organic farms (Foster and Lumpkin, 1999). Their goal is to produce higher quality animal products that meet consumer expectations. Organically fed animals must receive roughage feeds, which affect not only the fattening results but also carcass dressing and meat quality (Sandrum et al., 2000). This type of feeding also influences the fatty acid composition of meat fat (Högberg et al., 2003) and its sensory qualities (Danielsen et al., 2000).

The purpose of this experiment was to evaluate the effect of organic feeding on the carcass dressing percentage, meat quality and fatty acid composition of backfat in comparison with traditional feeding.

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## MATERIAL AND METHODS

The experiment was carried out on 160 fattening pigs originated from Polish Large White × Polish Landrace sows mated with a Belgian Landrace × Pietrain boar. The control group was given a limited feed mixture based on soyabean meal (M1). The experimental animals were fed a limited mixture composed of organically grown cereals, legume seeds and rapeseed cake. Both mixture were isoproteinous and isoenergetic. Additionally, group II received, *ad libitum*, silage from organically cultivated whole maize plants (SM), group III the same silage and a 0.5% supplement of a herb mixture. In group IV, the maize silage was substituted by grass silage (SG), and group V received grass silage and the herb mixture. The animals were fed twice a day. After both feedings the appropriate silages were given to the experimental animals (groups II-V) and refusals was weighed before each feeding.

The animals were kept in the experiment from about 30 to 114 kg of body weight. The animals were then slaughtered and dissected. Samples of loin were taken for chemical (AOAC, 1990) and sensory analyses. The fatty acid profile of backfat was analysed using a Philips 4500 gas chromatograph. Statistical one-way analysis was carried out with the STATISTICA 5.1 software package.

## RESULTS

Table 1. Meat and fat quality

Item	Feeding groups					SEM
	I M1	II M+SM	III M+SM+H	IV M+SG	V M+SG+H	
Cold dressing yield, %	79.8 <sup>B</sup>	78.7 <sup>A</sup>	78.7 <sup>A</sup>	78.6 <sup>A</sup>	78.5 <sup>A</sup>	0.13
Meat of primal cuts, kg	25.53 <sup>aA</sup>	25.76 <sup>aA</sup>	27.38 <sup>bb</sup>	27.32 <sup>bb</sup>	26.25 <sup>aAB</sup>	0.19
Meat content in carcass, %	56.5 <sup>aA</sup>	58.0 <sup>aAB</sup>	58.6 <sup>abAB</sup>	61.13 <sup>cb</sup>	60.85 <sup>bcB</sup>	0.40
Backfat thickness, cm	2.57 <sup>cb</sup>	2.39 <sup>bbAB</sup>	2.31 <sup>abA</sup>	2.18 <sup>aA</sup>	2.13 <sup>aA</sup>	0.03
Water holding capacity, %	22.34 <sup>b</sup>	20.70 <sup>a</sup>	22.20 <sup>b</sup>	21.51 <sup>ab</sup>	22.34 <sup>b</sup>	0.21
Meat colour, Hunter						
lightness (L)	46.57 <sup>b</sup>	44.37 <sup>a</sup>	45.57 <sup>ab</sup>	44.29 <sup>a</sup>	46.57 <sup>b</sup>	0.25
redness (a)	12.90 <sup>b</sup>	12.54 <sup>ab</sup>	12.27 <sup>a</sup>	12.38 <sup>a</sup>	12.9 <sup>b</sup>	0.06
yellowness (b)	2.05 <sup>A</sup>	2.08 <sup>A</sup>	2.17 <sup>A</sup>	2.39 <sup>A</sup>	3.05 <sup>B</sup>	0.07
Chemical composition of meat, %						
dry matter	25.06 <sup>b</sup>	24.77 <sup>ab</sup>	24.69 <sup>ab</sup>	24.40 <sup>a</sup>	24.61 <sup>ab</sup>	1.12
crude protein	22.85 <sup>ab</sup>	23.14 <sup>b</sup>	22.76 <sup>ab</sup>	22.59 <sup>a</sup>	22.83 <sup>ab</sup>	0.97
crude fat	1.61	1.40	1.69	1.37	1.38	0.06
Odour	4.66	4.68	4.74	4.66	4.72	0.02
Taste	4.35 <sup>aA</sup>	4.39 <sup>aA</sup>	4.63 <sup>cb</sup>	4.41 <sup>abA</sup>	4.54 <sup>bcAB</sup>	0.02
Tenderness	4.36 <sup>aAB</sup>	4.26 <sup>aA</sup>	4.51 <sup>bb</sup>	4.30 <sup>aA</sup>	4.37 <sup>aAB</sup>	0.02
Juiciness	4.31	4.26	4.44	4.26	4.79	0.09

a,b,c -  $P \leq 0.05$ , A,B -  $P \leq 0.01$

Silage supplements lowered carcass dressing yield when compared with the control group ( $P \leq 0.01$ ). On the other hand, the experimental animals, in particular, those receiving SG, deposited more meat and had thinner backfat (Table 1). In this case the differences when compared with the controls were highly significant. Organic feeding with a herb supplement had no distinct effect on water holding capacity or colour of meat, but without these supplements the indices were better than in the control group.

Table 2. Fatty acid composition of backfat

Item	Feeding groups					SEM
	I M1	II M+SM	III M+SM+H	IV M+SG	V M+SG+H	
SFA	44.12 <sup>ab</sup>	41.74 <sup>ba</sup>	40.72 <sup>abA</sup>	39.41 <sup>aA</sup>	40.65 <sup>abA</sup>	0.34
UFA	55.87 <sup>aA</sup>	58.26 <sup>bB</sup>	59.28 <sup>bcB</sup>	60.28 <sup>cB</sup>	59.35 <sup>bcB</sup>	0.34
MUFA	43.10 <sup>abAB</sup>	42.85 <sup>aA</sup>	43.91 <sup>abcAB</sup>	44.98 <sup>cB</sup>	44.38 <sup>bcAB</sup>	0.24
PUFA	12.77 <sup>A</sup>	15.41 <sup>B</sup>	15.37 <sup>B</sup>	15.60 <sup>B</sup>	14.97 <sup>B</sup>	0.23
PUFA – n-6	10.67 <sup>A</sup>	12.77 <sup>B</sup>	12.78 <sup>B</sup>	12.96 <sup>B</sup>	12.36 <sup>B</sup>	0.21
PUFA – n-3	0.47 <sup>A</sup>	0.75 <sup>B</sup>	0.77 <sup>B</sup>	0.80 <sup>B</sup>	0.74 <sup>B</sup>	0.02
MUFA / SFA	0.98 <sup>A</sup>	1.03 <sup>AB</sup>	1.08 <sup>BC</sup>	1.14 <sup>C</sup>	1.10 <sup>BC</sup>	0.01
PUFA / SFA	0.29 <sup>A</sup>	0.37 <sup>B</sup>	0.38 <sup>B</sup>	0.40 <sup>B</sup>	0.37 <sup>B</sup>	0.01

a,b,c -  $P \leq 0.05$ , A,B -  $P \leq 0.01$

The herb mixture improved the results of sensory analysis of meat, especially its taste and tenderness. The neck backfat of the experimental animals contained significantly less saturated and more n-6 and n-3 unsaturated fatty acids (Table 2).

## DISCUSSION

The available literature data concerning the effect of organic feeding on pig carcass and meat quality are inconsistent. This is due mainly to the different composition of feed mixtures used in particular experiments. In contrast to the results obtained in the presented experiment, Soudrum et al. (2000) found a lower meat and higher fat content in the carcasses of pigs fed with a mixture containing barley, wheat, field bean and potato protein. On the other hand, Danielsen et al. (2000), when giving a mixture with red clover and red clover silage found lower weight gains and higher meat content in the carcass, which was due to the lower energy concentration in feed. It seems that in our experiment the higher meat content in the carcasses of organically fed pigs was also the result of their lower daily body weight gains. The meat of experimental pigs was more tender and tasty. After grazing, a lower carcass dressing was also found (Gustafson and Stern, 2003). Högberg et al. (2003) found a favourable effect of organic feeding on the fatty acid composition of meat fat, similarly to that found in the present experiment.

## CONCLUSIONS

Organic feeding of fattening pigs with maize or grass silages and a herb mixture increased the meat content of the carcass but lowered the carcass dressing yield. This type of feeding also improved meat quality and increased the unsaturated fatty acid profile in backfat.

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## STRESZCZENIE

**Wpływ ekologicznego żywienia tuczników na jakość tuszy i mięsa**

W doświadczeniu przeprowadzonym na 160 tucznikach badano wpływ żywienia ekologicznego na jakość tuszy i mięsa. Wszystkie zwierzęta żywiono mieszanką pełnoporcjową, w grupie kontrolnej standardową - M1, a w doświadczalnych złożoną z uprawianych ekologicznie zbóż, nasion roślin strączkowych oraz makuchu rzepakowego (M). Ponadto w grupie II i III zwierzęta otrzymywały kiszonkę z całych roślin kukurydzy uprawianej ekologicznie - SM, w IV i V kiszonkę z traw - SG, a tuczniki z grup III i V dodatek ziół - H. Ekologiczne żywienie tuczników dawkami z udziałem SM lub SG oraz z dodatkiem ziół wpłynęło korzystnie na jakość tuszy, kruchość i smak mięsa. Słonina tuczników doświadczalnych zawierała więcej kwasów PUFA.

## Relationship between mineral and protein deposition in restricted and realimented pigs

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### ABSTRACT

Mineral retention and its relation to protein deposition was measured in 54 crossbreed pigs allocated to a control and two restricted (up to 50 or 80 kg BW) groups. Control pigs were continuously fed a basal diet (B) and restricted pigs, diet B mixed with 20% grass meal. During re-alimentation, previously restricted pigs were fed diet B up to 105 kg. The animals were slaughtered at 25, 50, 80 and 105 kg BW. The previously restricted pigs showed a compensatory response with regard to protein as well as P, Ca and Mg deposition. The reaction in both protein and minerals depended on the duration of previous restriction and was full only in the pigs restricted to 50 kg BW. According to an allometric equation, the mineral content increased faster ( $b^b > 1$ ) than protein. Compensatory mineral gains in the body were closely related to protein gain and the daily mineral/protein deposition ratio was the same in all treatments.

KEY WORDS: protein, minerals, pigs

### INTRODUCTION

A positive correlation between protein and mineral deposition in the pig body has been documented (Rymarz et al., 1982; Mahan and Shields, 1998). It is also known that in temporarily restricted pigs, protein deposition is decreased, but during subsequent realimentation, it is enhanced. However, information on mineral deposition in so-treated pigs is lacking. Thus, the aim of the study was to assess mineral deposition in pigs in relation to protein accretion during restriction and realimentation.

### MATERIAL AND METHODS

Fifty-four crossbreed pigs from 25 to 105 kg body weight were kept individually and fed *ad libitum*. Two diets, basal (B) and high-fibre (F) were used.

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Diet B contained 13.1 MJ EM and 8.9 g digestible lysine, 5.29 g total phosphorus, 6.7 g calcium (with Ca:P ratio of 1.3:1), and 119 g zinc. Diet F was a mixture of diet B (80%) and grass meal (20%) and contained less energy, digestible lysine, total and digestible phosphorus, calcium and zinc by 12, 17, 8, 11, 7 and 8%, respectively. Control pigs were continuously fed the basal diet. The  $F_{50}$  and  $F_{80}$  pigs were restricted by feeding them diet F up to 50 or 80 kg, respectively. During realimentation, all previously restricted pigs were fed diet B up to 105 kg. The animals were slaughtered at 25 (n=6), 50 (n=12; 6 each from groups B and F), 80 (n=18; 6 each from groups B, F, and  $F_{50}$ ) and 105 kg (n=18; 6 each from groups B  $F_{50}$  and  $F_{80}$ ). The protein content was determined according to standard methods (AOAC, 1990), P spectrophotometrically using molybdovanadate as the colour-forming reagent, Ca and Zn from ash solutions by atomic absorption spectrophotometry. Protein and mineral retention in the body were calculated from the difference between their final and initial contents in a particular growth stage using the comparative slaughter technique.

## RESULTS AND DISCUSSION

At 25 kg the body of pigs contained 4.0 kg of protein, 580 g of ash and 115, 196, 6.58 and 0.41 g of P, Ca, Mg and Zn, respectively. The content of these components increased with growth and, in pigs fed on diet B up to 105 kg liveweight, reached levels of 15.2 kg of protein, 2.6 kg ash of ash and 485, 850, 28 and 1.9 g of P, Ca, Mg and Zn, respectively; these values are similar to those reported by Kirchgessner et al. (1994).

During restriction up to 50 kg (group  $F_{50}$ ), pigs deposited daily 24 g less protein ( $P<0.05$ ) as compared with the B pigs. This was associated with a significant decrease of ash and P deposition (16.8 vs 20.6 and 2.5 vs 3.4 g/day, respectively). Retention of the remaining minerals was also less in restricted pigs: by 0.9 g of Ca, 38 mg of Mg and 2.1 mg Zn (differences nonsignificant).

Pigs restricted from 25 to 80 kg ( $F_{80}$ ) reduced protein and phosphorus deposition by 9 and 0.4 g, and zinc retention by 2.7 mg, but the deposition of other minerals during this time was similar. However, at 80 kg they were 6 days older than their analogues from the control group. In this growth period, pigs re-alimented from 50 kg ( $F_{50}$ ) increased daily protein deposition by 19 g ( $P<0.01$ ) and P retention by 0.5 g ( $P<0.01$ ) more than restricted animals. The  $F_{80}$  and  $F_{50}$  pigs also deposited more Ca.

When the total growth period (25-105 kg) was considered, pigs from the basal group and those realimented from 50 kg deposited similar amounts of protein, ash, P, Ca and Mg. However, in pigs realimented from 80 kg, deposition of body

components was lower than in control animals, which may indicate that compensation in this group was not complete. Therefore, a certain compensatory response could be expected during subsequent growth, which was examined by an allometric model with protein content in the body as the independent variable. In calculations, 3 different paths of growth: control and two restricted/realimentd were separated. Table 2 shows that “b” (growth coefficient) is generally higher (by 3-6%) in pigs from group F<sub>50</sub> than in controls, except Ca, which indicates that compensatory retention of minerals takes place. In F<sub>80</sub> pigs, the growth coefficients were on similar levels or slightly lower as compared with control pigs, indicating that restriction prolonged up to 80 kg was too severe and the animals could not compensate deposition of body components. This suggests that the intensity of the compensatory response with regard to mineral retention depends on the duration of previous restriction.

Table 2. Relationship between protein (in kg) and mineral (Y) content in the body during growth of pigs from 25 to 105 kg as expressed by the formula:  $Y = a * \text{protein}^b$

Minerals	Group of pigs	a	b	R <sup>2</sup>
P, g	Basal (B)	19.3 ± 4.06	1.175 ± 0.08	98.1
	Compensatory F <sub>50</sub>	17.0 ± 3.55	1.213 ± 0.08	98.2
	Compensatory F <sub>80</sub>	18.9 ± 8.45	1.164 ± 0.17	98.2
Ca, g	Basal (B)	38.8 ± 6.97	1.134 ± 0.07	98.2
	Compensatory F <sub>50</sub>	40.8 ± 7.88	1.117 ± 0.08	97.9
	Compensatory F <sub>80</sub>	52.1 ± 7.60	1.013 ± 0.06	98.5
Mg, g	Basal (B)	1.7 ± 0.41	1.025 ± 0.09	95.9
	Compensatory F <sub>50</sub>	1.6 ± 0.26	1.052 ± 0.06	98.2
	Compensatory F <sub>80</sub>	2.1 ± 0.49	0.944 ± 0.09	94.8
Zn, mg	Basal (B)	76.9 ± 14.82	1.167 ± 0.07	98.7
	Compensatory F <sub>50</sub>	62.5 ± 18.80	1.237 ± 0.12	96.7
	Compensatory F <sub>80</sub>	72.9 ± 12.24	1.181 ± 0.07	98.6

The values of growth coefficients also indicate that the amount of deposited minerals increased at a faster rate (“b”>1) than the amount of protein, which is in agreement with earlier results reported by Rymarz et al. (1982). Correlation between the mineral and protein content in the body was high and ranged from 94.8 for Mg to 98.7 for Zn. This confirms earlier findings that accretion of protein and minerals is closely connected, and the ratio of daily mineral/protein deposition was the same for all treatments in the investigated growth period. This ratio was the highest for Ca/protein (average 0.060), lowest for Zn/protein (average 0.00012) and rose with increasing body weight of pigs.

## CONCLUSIONS

Pigs with temporarily restricted growth showed a compensatory response with regard to protein as well as P, Ca and Mg deposition. This response in both protein and minerals depended on the duration of previous restriction and was full only in those pigs that were restricted up to 50 kg body weight. Compensatory mineral gains in the body were closely related to protein gain. The daily mineral/protein deposition ratio was the same in all groups.

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## STRESZCZENIE

**Zależności między odkładaniem białka i składników mineralnych u świń żywionych w sposób ograniczony i podczas realimentacji**

Zależności między odkładaniem białka i składników mineralnych określono na 54 świnich podzielonych na grupę kontrolną i dwie grupy żywione restrykcyjnie (do 50 lub 80 kg m.c.). Świnie z grupy kontrolnej były żywione dietą standardową (B), z grup restrykcyjnych dietę B z 20% dodatkiem suszu z traw. Podczas realimentacji świnie z obydwóch grup restrykcyjnych otrzymywały dietę B. Zwierzęta ubito przy masie ciała 25, 50, 80 i 105 kg. U świń żywionych restrykcyjnie stwierdzono reakcję kompensacyjną w odkładaniu białka oraz P, Ca i Mg. Kompensacja zależała od długości okresu restrykcji i była pełna tylko u świń żywionych restrykcyjnie do m.c. 50 kg. Alometryczne równania wykazały, że zawartość składników mineralnych w ciele wzrasta szybciej ( $b'' > 1$ ) niż zawartość białka. Kompensacyjny przyrost składników mineralnych w ciele był ściśle zależny od przyrostu białka, a proporcja dziennego odłożenia składników mineralnych do białka była taka sama w badanych grupach.

# Alpha-ketoglutarate reduces duodenal myoelectric disturbances induced by *E. coli* enterotoxin in pigs<sup>1</sup>

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## ABSTRACT

Pigs with bipolar electrodes implanted on the antrum and duodenum were exposed to heat-labile enterotoxin from *E. coli* (LT) infused intraduodenally. The exposure was repeated after three days of supplementing feed with  $\alpha$ -ketoglutarate (AKG). 0.5  $\mu\text{g}/\text{kg}$  of LT induced prolonged changes in the migrating myoelectric complex (MMC) without inducing clinical signs of enterotoxaemia. AKG had no effect on the basic MMC pattern, but prevented LT-induced changes. The possible mechanism of AKG and LT-induced alteration may involve gamma-aminobutyric acid (GABA) synthesis and activation of GABAergic neurotransmission in the gut.

KEY WORDS: migrating myoelectric complex, enterotoxin,  $\alpha$ -ketoglutarate

## INTRODUCTION

Gastrointestinal tract motility is crucial for its functions. Heat-labile enterotoxin (LT) produced by enterotoxigenic *E. coli* is a major virulent factor responsible for infectious diarrhoea in young animals and children. Enterotoxaemia-induced ileus can lead to death due to endotoxaemia and septicaemia. It has been shown that various bacterial toxins influence the migrating myoelectric complex (MMC) in pigs, rats and calves. After administration of endotoxins, MMC cycles are shortened and more frequent. In pigs, elevated MMC migration velocity and cycling frequency are maintained one day after endotoxin administration during feeding and return to basal values 4 days later (Bruins et al., 2003).

Glutamine has a beneficial effect on gut morphology (Potsic et al., 2002). However it is debated whether glutamine has beneficial effects on the gut during disease. Since glutamine is unstable in solutions,  $\alpha$ -ketoglutarate (AKG) can be used as a glutamine

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precursor. No studies have been performed on the influence of AKG on motility or MMC during enterotoxaemia. Therefore, the present study was designed to evaluate the effect of heat-labile enterotoxin on MMC in weaned pigs, followed by investigating the effect of AKG on parameters of MMC in animals exposed to enterotoxin.

## MATERIAL AND METHODS

Castrated male pigs (10.5-13 kg BW) had surgically implanted bipolar silver electrodes on serosal membrane of the stomach antrum and duodenum, as described by Gacsalyi et al. (2000). The electrodes were connected to a telemetric transmitter permitting constant measurement of GI tract electrical activity. The animals were also fitted with a duodenal cannula.

Ten pigs were subjected to intraduodenal infusion of *E. coli* LT (Sigma, USA) in doses 0.1 and 0.5 µg/kg BW. LT was injected in a 2 ml bolus at the end of phase I of MMC between 2 and 3 p.m. Each dose was tested on the same pig with a one- or two-day interval. A further 8 pigs were used for studying the effect of AKG. The MMC in each animal was recorded under control conditions, after infusion of LT at 0.5 µg/kg BW, after 1-3 days of feeding AKT at 10 mmol/kg BW, and after LT infusion at 0.5 µg/kg BW given on the 5<sup>th</sup> day after starting AKT supplementation.

## RESULTS

The pigs were in good condition and stable during the entire experiment. LT had no effect on the duration or signal power of electromyographic events in the antrum. In the duodenum after LT infusion, the duration of the MMC cycle increased at night (postprandially). The increase was due to elongation of phase II (Figure 1). The velocity of phase III migration in the duodenum was significantly increased following LT administration (Figure 2).

Feed supplementation with AKT had no effect on basic electrical activity in the antrum. The velocity of phase III in the duodenum was unchanged after 3 days of AKG supplementation. Administration of LT on the 5<sup>th</sup> day of AKT supplementation did not affect the velocity of phase III (Figure 3)

## DISCUSSION

In the present study we have shown that low doses of LT induce disturbances in gut motility without evoking other clinical signs. Therefore, exposure to bacterial toxins even without inducing disease may cause alterations in gut function that are followed by a decrease in feed utilization. Feed supplementation with AKG has no effect on basic motility, but prevents changes

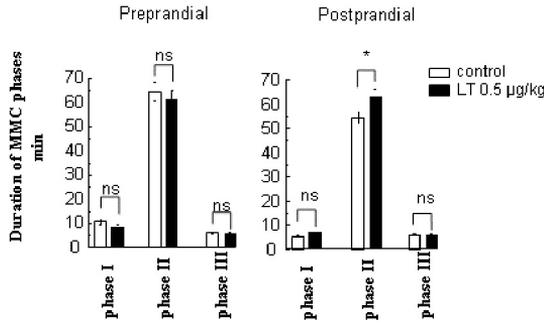


Figure 1. Duration of duodenal MMC phases in control (white columns) and after intraduodenal administration of *E. coli* heat-labile enterotoxin (LT) 0.5 µg/kg BW (black columns) recorded in 10 pigs. Data expressed as mean ± SE. \* different from respective control; P<0.05 (paired t-test)

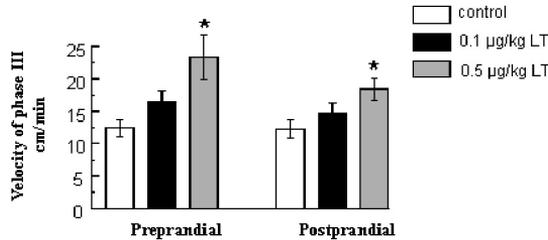


Figure 2. Velocity of phase III (cm/min) in the duodenum of weaned pigs in control (white columns), after intraduodenal administration of LT 0.1 µg/kg (black columns), and LT 0.5 µg/kg (grey columns). Data expressed as mean±SE (n=8) \* different from respective control; P<0.05 (ANOVA, Dunett post hoc test)

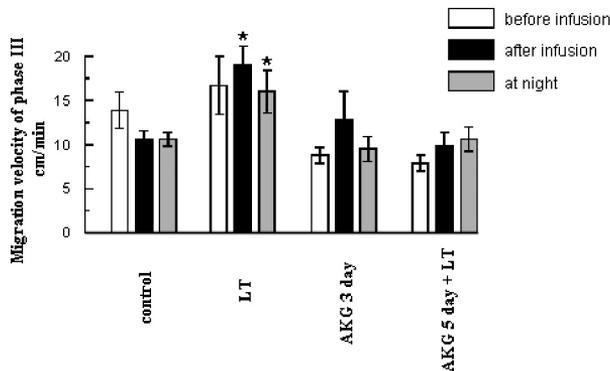


Figure 3. Migration velocity of phase III before (white columns), after infusion (black columns), and at night (grey columns) in control conditions, after administration of *E. coli* heat-labile enterotoxin (LT) 0.5 µg/kg BW infusion, after the 3rd day of  $\alpha$ -ketoglutarate (AKG) supplementation, and after LT infusion on the 5th day of AKG supplementation. Data expressed as mean±SE (n=8). \* different from respective control; P<0.05 (paired t-test)

induced by LT. It has been assumed that enhanced gamma-aminobutyric acid formation (GABA) in the intestinal mucosa by ornithine  $\alpha$ -ketoglutarate treatment might be of physiologic importance in the regulatory processes of cell growth and differentiation (Raul, 1995). The presence of GABA-positive neurons in the submucosal and myenteric plexus has been shown in pigs (Timmermans and Scheuermann, 1998). Spontaneous relaxations of the rat gastroduodenum include responses that involve a GABAergic nitric oxide-related pathway, which is targeted by VIP (Krantis et al., 1998).

## CONCLUSIONS

Feed supplementation with  $\alpha$ -ketoglutarate may have a beneficial effect on subclinical forms of enterotoxaemia, decreasing diarrhoea and improving feed utilization that leads to better health.

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## STRESZCZENIE

### **Alfa-ketoglutaran zmniejsza zaburzenia mioelektryczne wywołane przez entrotoksynę *E. coli* u świń**

Świnie z zainplantowanymi elektrodami bipolarnymi na antrum i dwunastnicy były ekspozowane na ciepłowrażliwą enterotoksynę *E. coli* (LT), która była podawana dodwunastniczo. Ekspozycję powtarzano po trzydniowym wzbogaceniu paszy w  $\alpha$ -ketoglutran (AKG). 0,5  $\mu$ g/kg LT doprowadzało do długotrwałych zmian w migrującym kompleksie mioelektrycznym (MMC) bez wywołania objawów klinicznych enterotoksemii. AKG nie miał wpływu na podstawowy zapis MMC, ale zapobiegał zmianom wywołanym przez LT. Przypuszczalny mechanizm działania AKG na zmiany w MMC wywołane LT polega na syntezie kwasu gama-amino masłowego (GABA) i aktywacji GABA-ergicznej neurotransmisji w jelicie.