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EFFECT OF FIBER CONTENT ON ABSORPTION AND DISTRIBUTION OF NITROGEN IN GROWING PIGS

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ABSTRACT

The experiment was conducted to evaluate the effect of added fiber into diets on absorption and distribution of nitrogen at different levels of dietary nitrogen. Four diets were designed with a combination of two concentrations of fiber: high (4.46 %; HF) and low (3.25 %; LF) level of fiber, and two levels of dietary nitrogen: high (18.8 %; HP) and low (14.0 %; LP) dietary nitrogen. Significant effects of fiber intake on increased dry matter intake were found only in one case. The nitrogen intake was not affected by the fiber content. Changes in the proportion of excreted nitrogen in urine and faeces were proven. Changing the nitrogen content of the feed has a more significant effect ($P < 0.03$) than changing the fiber content ($P < 0.05$). Very high coefficient of determination between nitrogen absorption and nitrogen intake calculated for the diets with low fiber content was $R^2 = 0.91$ and for diets with high fiber $R^2 = 0.97$. By comparing the differences in nitrogen digestibility expressed in g.d^{-1} nitrogen, significant deviation was found only in the HFHP group (+6.37 g.d^{-1} = +19.9 %; $P = 0.03$) among the groups with different fiber content. In retention, we found a positive change comparing the groups LFHP vs. HFHP, +6.22 g.d^{-1} = +25 % ($P = 0.05$). It is the same pair of diets in which we found also a significant difference in dry matter intake (+15 %). Absorption and retention of nitrogen, expressed as a percentage of N intakes, did not decrease in any of the experimental groups irrespective of nitrogen and fiber content in the diet. These data suggest that fiber added into a diet with higher content of CP increased nitrogen in faeces, reduced nitrogen in urine, positively affected the overall balance of N and had only weak effect on its absorption.

Key words: crude fiber; nitrogen excretion; pigs

INTRODUCTION

Fiber from various sources is a common part of monogastric animal feed. Diets with optimum amount of by-products from the food industry are an effective way to improve animal health and mitigate environmental impacts. Potential value is the opportunity to use crude fiber concentrates as 'functional' feed additives to improve young pigs' growth and welfare (Jarrett and Ashworth, 2018). Dried beet pulps, a product of processing sugar, contain crude fiber, which is useful for growing pigs. Lizardo and Aumaître (2001) introduced that the inclusion of beet pulps into a diet improved growth performance immediately after weaning and carcass composition at slaughter. Increased fiber levels promote bacteria fermentation, which is responsible for the shift of urine excretion of N in faeces (Nahm, 2003; Aarnink and Verstegen, 2007). Physical properties of fiber such as viscosity and solubility affect digestion, satiety, and transit time (Williams *et al*., 2001; Montagne *et al*., 2003). Fiber is actually a possible medium for reducing releases of nitrogen production and for improving pig intestinal health and welfare (Longland *et al*., 1994; Basset-Mens and Van der Werf, 2005). Similarly, other studies have been done to investigate the relationships between feeding and nitrogen excretion (Fernandez

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et al., 1999; Le Goff *et al*., 2003; Galassi *et al*., 2007) with particular reference to ammonia emissions.

For example, inclusion of dietary fiber can alter the gut microbiota in ways that could reduce the need for antibiotics, while controlled addition of certain fiber types may reduce nitrogen losses into the environment and so reduce the environmental cost of pig production.

The objective of this study was to verify the effect of different levels of dietary fiber on digestibility and distribution of nitrogen.

MATERIAL AND METHODS

To evaluate the effect of fiber in diets on redistribution of nitrogen eight crossbred gilts, progeny of Large White sows and Landrace boars (initial BW 29.9 ± 1.7 kg), were used. The gilts from the experimental herd of Research Institute for Animal Production Nitra were individually housed in balance cages, located in the Laboratory of digestive physiology of monogastric animals. All experimental procedures were reviewed and approved by the Animal Care Committee of the Research Institute for Animal Production Nitra. Pigs were housed individually in an environmentally controlled (21 °C) room in metabolic cages. The pigs were randomly allotted to four dietary treatments according to a 4 x 4 Latin square design.

The diet was based on wheat, corn and soybean meal. Of these, four dietary treatments were prepared and applied in the form of a Latin square with a 2 x 2 factor arrangement. The included diets were as following: Diet 1 (LFLP) with lower fiber content and low protein (CP 13.92 %) supplemented with isoleucine, lysine, methionine, threonine, tryptophan, and valine (NRC, 1998). Diet 2 (LFHP) with higher content of CP 18.38 %: the base of this diet was fortified wheat, corn, more soybean meal and crystalline amino acids (lysine and methionine); Diet 3 (HFLP, CP 14.08 %) contained similar ingredients as Diet 1 with 150 $g.kg^{-1}$ dried beet pulps. Diet 4 (HFHP, CP 19.29 %) contained similar ingredients as Diet 2 with 15 % dried beet pulps. The same energy level in the diet was reached by means of supplementation with rapeseed oil (Table 1). The pigs were fed by two equal doses at 7 a.m. and 5 p.m. for a daily rate of 90 $g.kg^{0.75}$. Water was provided *ad libitum*. Pigs were weighed at the beginning and at the end of each period.

The experimental period began after a 5-day habituation to become accustomed to the cages. The 10-day experimental period consisted of a 6-day adaptation period, within which the animals adapted to the experimental diet, and a 2 x 48 hour collection period (day 7-8 and day 9-10). During the collection period, samples of urine and faeces were collected separately. Urine was collected via catheters, without addition of sulphuric acid, and stored in ice-cooled containers. Urine pH was measured before freezing each day. 10 % aliquot was stored at -20°C. Faeces were collected, pooled, and stored at -20 °C until analysis.

Samples of diets, urine and faces were analyzed for dry matter, total N and fiber. Analyzes were performed in accordance with the standard methods of AOAC (1998). Feed and faecal samples were analyzed for dry matter (DM) after drying at 105 °C for 8 hours. Crude protein (N x 6.25) was determined by the Kjeldahl method using a Kjell-Foss 16200 auto analyzer (method 978.02). The crude fiber contents were determined using The Fibertec™ 2010 fiber analyzer Tecator, Hoganas, Sweden (method 2002.04). Chemical analyses were performed in duplicate.

The data were subjected to one-way ANOVA using Statgraphic Plus (version 3.1., Statistical Graphics Corp., Rockville, MD, USA). Differences in mean values between groups were assessed using Student's t-test of the statistical significance of the difference between the two samples. Estimation of the normal distribution of small samples and of the effect of dietary fiber and nitrogen concentration were evaluated using regression analyses.

RESULTS AND DISCUSSION

The values provided by chemical analysis of nitrogen intake and intake of pulp in the groups were affected by the methodology intent, therefore in these cases it is not possible to evaluate the differences. Inclusion of beet pulps into the diets reflected differently in all experimental groups. The dry matter intake was reduced between groups LFLP vs. HFLP - 0.09 $kg.d^{-1}$ (-7 %) and increased in LFHP vs. HFHP + 0.17 $kg.d^{-1}$ (+12 %). This significant

Table 1. Composition of basal diets and analysed content of nutrients

Ingredient *(g.kg^{-1} diet)*	Diet[a] LFLP	LFHP	HFLP	HFHP
Wheat	300.00	300.00	300.00	280.00
Maize	552.00	426.00	388.00	276.00
Soybean meal	86.90	223.00	88.80	231.00
Corn starch dextrinized	10.00	10.00	10.00	10.00
Dried beet pulp	-	-	150.00	150.00
Rapeseed oil	8.30	9.60	22.60	23.90
L-isoleucine	0.90	-	1.00	-
L-lysine.HCl	5.80	1.70	5.70	1.40
DL-methionine	0.90	-	1.20	-
L-threonine	2.20	0.30	2.40	0.40
L-tryptofan	0.40	-	0.50	-
L-valine	0.90	-	1.10	-
Monocalcium phosphate	14.10	12.70	14.10	12.70
Limestone	11.00	10.70	7.90	7.60
Salt	3.90	3.80	3.50	3.40
Vit.-min. premix[1]	3.00	3.00	3.00	3.00
Analysed nutrient contents	*(g.kg^{-1} air-dry)*			
Dry matter	883.5	886.3	887.5	890.1
Crude protein	139.2	183.8	140.8	192.9
Crude fiber	30.4	34.6	45.6	43.6

*LFLP = reduced fiber and protein diet; LFHP = reduced fiber and high protein diet; HFLP = high protein +15% dried beet pulp diet; HFHP = high protein + 15% dried beet pulp diet.

[1]Supplied per kg of diet: vit. A 9 000 IU; vit. D3 1 500 IU; α – tocopherol 35.0 mg; vit. B1 1.7 mg; vit. B2 6.0 mg; vit. B6 2.5 mg; Ca-panthothenate 15.0 mg; niacin 38.0 mg; vit. K3 2.0 mg; biotin 0.12 mg; cyanocobalamin 0.03 mg; choline 156 mg; Fe 103.0 mg; Zn 116.5 mg; Mn 49.0 mg; Cu 40.0 mg; I 1.2 mg; Co 0.4 mg; Se 0.3 mg.

difference was recorded only in the group with higher levels of nitrogen. In comparison among groups with different nitrogen content in the diet no significant differences were found. In comparison to the same groups in dry matter excretion, the difference was more pronounced, up to + 34.25 g.d^{-1}, +26.0 % (P = 0.014). We found the most excreted dry matter −165.59 g.d^{-1} in the HFHP group (received dry matter 1.29 kg.d^{-1}) In comparison with the group with low fiber content (LFHP vs. HFHP) the difference in the excreted dry matter was more pronounced up to + 34. 25 g.d^{-1}, +26.0 % (P = 0.014).

The results of the nitrogen balance influenced by the addition of pulp are shown in Table 2. Nitrogen balance was affected by the inclusion of beet pulp in all diets, but not all values were statistically significant. Nitrogen uptake was increased by including beet pulp in the HFLP diet compared to HFHP + 32% (P = 0.015), when feed LF was only +18 % (LFLP vs. HFLP). From these values, the benefit of the higher fiber content is the intake of nitrogen from the feed, as in both cases there was approximately the same increase in nitrogen in the feed between the groups.

Changes in the amount of excreted nitrogen through urine and faeces are expressed by their proportion. The largest and significant value was found in the HFLP group by + 48 %, compared to groups with different nitrogen content but the same fiber content (HFLP = 1.3 vs. HFHP = 2.5). The increased amount of total excreted nitrogen (LF + 33 % and HF + 36 %) was adequate to the increased nitrogen intake (+ 35 %, LP vs. HP). The effect of the pulp was not statistically confirmed here at any nitrogen level. However, we found significant differences between the groups in the amount of nitrogen excreted separately in urine and faeces, in the table expressed as the urine / faecal ratio (P < 0.05). Our result

Table 2. Effects of dietary protein and crude fiber level on nitrogen (N) balance

Items	LFLP* SEM	LFHP* SEM	HFLP* SEM	HFHP* SEM
DM intake, *[kg.d-1]*	1.23 ± 0.10[ab]	1.12 ± 0.08[a]	1.14 ± 0.1[ab]	1.29 ± 0.06[b]
CF intake *[g.d-1]*	42.40 ± 0.41[a]	46.75 ± 0.50[a]	58.62 ± 0.66[b]	62.19 ± 0.34[b]
N intake *[g.d-1]*	31.07 ± 0.33[a]	38.30 ± 0.45[b]	28.96 ± 0.46[a]	42.33 ± 0.29[b]
Urinary/Faecal N ratio	1.80 ± 0.11[a]	3.14 ± 0.12[c]	1.30 ± 0.11[b]	2.5 ± 0.13a
Total N excretion *[g.d-1]*	12.65 ± 0.25[a]	18.86 ± 0.28[b]	11.83 ± 0.27[a]	18.60 ± 0.23[b]
Absorbed N *[g.d-1]*	26.45 ± 0.31[a]	32.06 ± 0.47[a]	23.77 ± 0.33[a]	38.43 ± 0.37[b]
Retained N *[g.d-1]*	18.43 ± 0.29[a]	19.18 ± 0.35[a]	17.12 ± 0.29[a]	25.40 ± 0.30[b]
Absorption, % of intake	84.74 ± 0.18[a]	87.27 ± 0.12[a]	81.66 ± 0.13[a]	85.38 ± 0.13[a]
Retention, % of intake	59.01 ± 0.27[a]	52.66 ± 0.18[b]	58.72 ± 0.19[a]	56.51 ± 0.19[a]
Retention, % of absorbed	69.41 ± 0.15[a]	60.56 ± 0.29[b]	71.86 ± 0.15[b]	66.10 ± 0.14[a]

Different letters ([a, b] and [c]) on the same row indicate that treatment means are significantly different at $P < 0.05$.
*LFLP = reduced fiber and protein diet; LFHP = reduced fiber and high protein diet; HFLP = high protein +15% dried beet pulp diet; HFHP = high protein + 15% dried beet pulp diet.

with low protein diets is consistent with the findings of Galassi *et al*. (2007), who obtained similar results in a traditional Italian heavy pig on average weight (150 kg) with a diet with CP 14.0 % and CF 4.1 % compared to the control group with CP 13.5 % and CF 3.2 %.

Absorbed nitrogen calculated as a difference between nitrogen intake and nitrogen in faeces (including endogenous nitrogen) was the difference in mean values. Large differences in mean values in groups with different nitrogen content (LFLP vs. LFHP +5.61 (17 %) and HFLP versus HFHP +14.65 (38 %)) were not significant (Methodical intention). Significant evidence for better functioning of fiber on nitrogen absorption in higher nitrogen diets was found in groups with different fiber content LFHP vs. HFHP +6.37 g.d^{-1} = +19.9 % (P = 0.03). The opposite tendency was observed in the LFLP diet compared to HFLP -2.68 g.d^{-1} = -10.1 % (P = 0.13).

Similar results were observed in nitrogen retention +4 % at low fiber levels (LFLP vs. LFHP) and +33 % (+8.28 g.d^{-1}) at high fiber levels (HFLP vs. HFHP). Nitrogen retention, expressed as a percentage of N intake, was reduced in pigs fed with the high-fiber diets (-4 %) and it was even more affected by the loss of fiber on feed up to -12 %.

Nitrogen excretion was affected by the inclusion of beet pulp into the diet (Figure 1). Switch from urinary to faecal excretion was detected in all animals, while the largest and most significant it

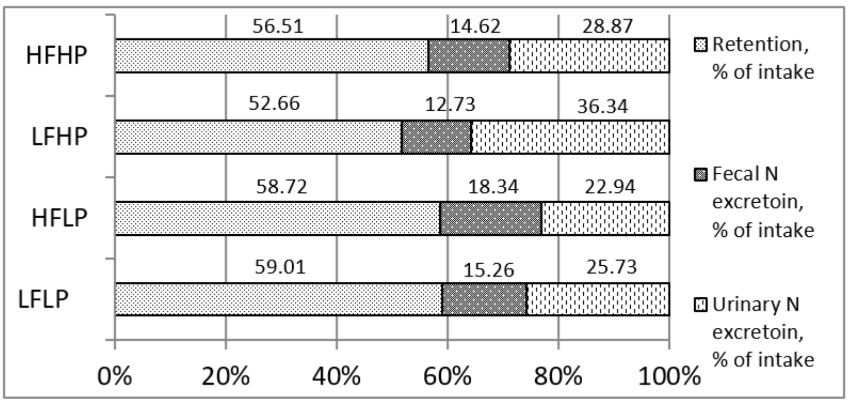

Figure 1. Distribution of nitrogen intake in to faeces and urine (expressed as % of nitrogen intake)

was in the HP diet group ($P < 0.05$). The reduction in urinary nitrogen was -7.48 % (LFHP vs. HFHP) and -2.79 % (LFLP vs. HFLP). Total N retention was detected in the higher nitrogen group of +3.85 % (LFHP vs. HFHP). In the lower nitrogen group (LFLP vs. HFLP) we found almost no change. Faecal N excretion increased by +3.1 % (LFLP vs. HFLP) and +1.89 % (LFHP vs. HFHP).

Similar results were found for Italian pigs fed with CP 14.0 % and CF 4.1 % where 12.3 %, 19.3 % faecal N was confirmed in feed with higher protein levels and increasing fiber (CF 6.7 %). Both groups were compared to CP control 13.5 and CF 3.2 % (Galassi *et al*., 2007).

A different, but not significant, effect was observed in nitrogen uptake and retention (Figure 2). A different distribution was observed in the nitrogen level in the faeces, urine, and retention in the diets with different fiber content and approximately same nitrogen content. The results of absorption and nitrogen retention after inclusion of added fiber at lower nitrogen content (LP) were worse in both cases (-1 to -2 g.d^{-1}), but in groups with higher nitrogen content (HP) it was improved at absorption by 20 % (+6. 37 g.d^{-1}) and at a retention of 32 % (+6.22 g.d^{-1}). Zervas and Zijlstra (2002) found decrease in crude protein content of 18.5 % and the addition of sugar beet pulps reduced N retention by 12.0 % compared to the control group.

In our observation at the concentration 19 % of crude protein in the diets, regression analysis showed that absorbed N (y) was a linear function of

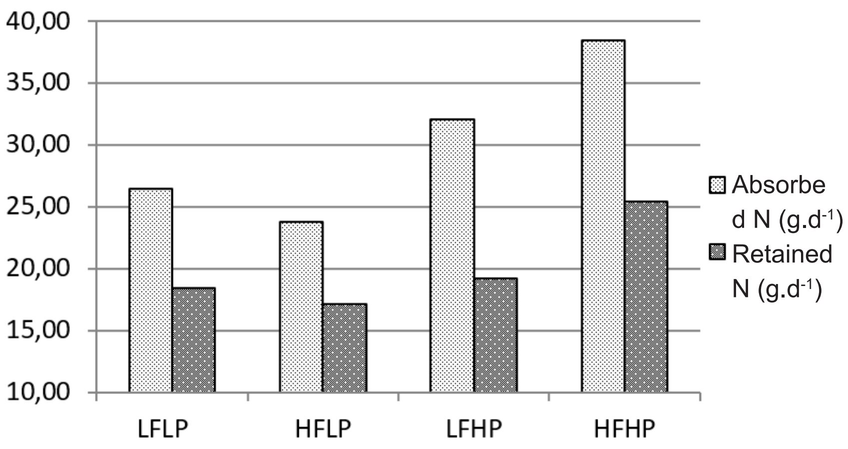

Figure 2. Amount of absorbed and retained nitrogen, according to the fiber and nitrogen content on the diet (g.d^{-1})

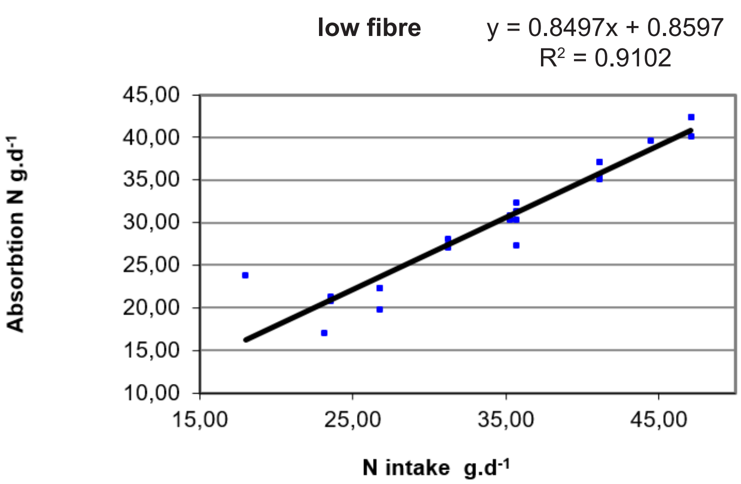

Figure 3. Relationship between nitrogen intake and nitrogen absorption in low fiber diets (LFLP and LFHP)

nitrogen intake (x), as it shown on Figures 3 and 4. The relationship depends on nitrogen content in feed. For low fiber diets it was described by an equation: $y = 0.8497 x + 0.8597$, $R^2 = 0.9102$. For the diets with high fiber content it was calculated using an equation: $y = 0.9604 x - 4.3065$, $R^2 = 0.9797$. The slope of the regression equation showed that each gram of nitrogen intake in the LF 849.7 mg and and for HF diets was absorbed up to 960.4 mg N.

The coefficient of determination (R^2) for the regression daily N absorption was predicted from N intake. The values are different according to the low fiber content in diets (LFLP + LFHP) $R^2 = 0.9102$ and (HFLP + HFHPP) $R^2 = 0.9797$. A stronger effect on nitrogen absorption was found in the group with higher fiber content.

From the calculated equations it is possible to estimate the interesting absorption at zero nitrogen intake. In case of low fiber content in feed it is about 0.8 g.d^{-1}, at higher levels of fiber it is already negative value (- 4 g.d^{-1}). A logical explanation should be the increased microbial colonization of the intestine and the transfer of nitrogen to solid excrement. This nitrogen falls into the category of endogenous production. Endogenous production and nitrogen retention values for different fiber content in feed will be the aim of future investigations. Up to date, we have found that the amount of fiber intake in experimental diets and nitrogen retention are no longer linearly dependent.

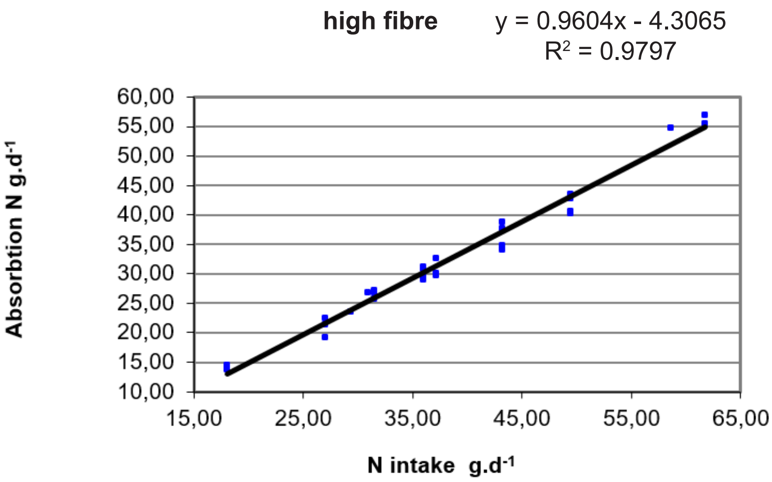

Figure 4. Relationship between nitrogen intake and nitrogen absorption in high fiber diets (HFLP and HFHP)

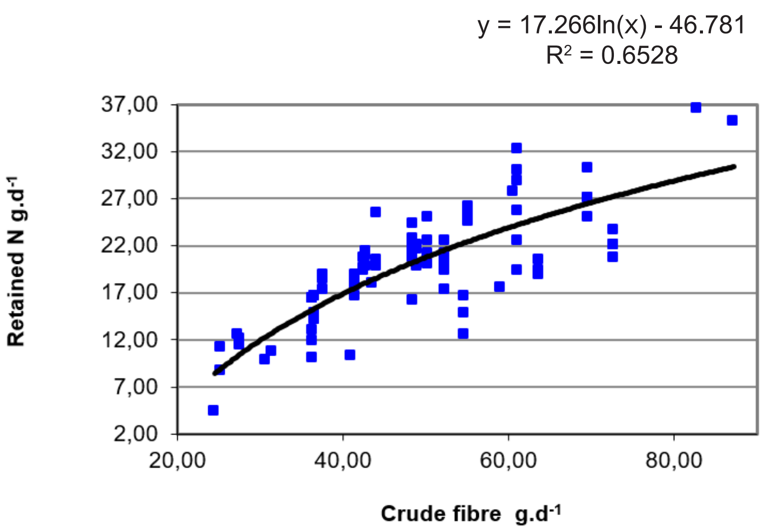

Figure 5. Relationship between crude fiber and nitrogen retention (all groups)

Dependence of nitrogen retention by the fiber content has a logarithmic character of *y = 17.266 ln (x) - 46.781*, $R^2 = 0.6528$. Figure 5 illustrates the trends in existing data and predictions of future data.

The influence of non-starch polysaccharides from beet pulps on growth performance after weaning, ileal and faecal nutrient digestibility and intestinal enzymes of piglets was investigated in several studies (Ramonet *et al*., 2000; Högberg and Lindberg, 2004). Faecal digestibility of energy and nitrogen was not affected by the presence of 6 – 12 % beet pulps in piglet diets (Lizardo and Aumaître, 2001). These, as well as our present data, suggest that fiber has a greater effect on a diet with a higher content of CP. In both cases it increased the nitrogen in faeces and reduced nitrogen in urine while had no negative effect on the absorption and total amount of nitrogen excreted. According to the logarithmic relationship for retention and fiber content, this theory is rather hypothetic. Specific equation is valid for a specific category of animals and specific quantitative values for fiber content in diets. The opportunity to use crude fiber concentrates as a functional feed additive might be of a potential value to improve young pigs' growth and welfare.

CONCLUSION

A portion of crude fiber is used as an energy source for the pig and a part of the undigested fiber serves as an energy source for microbial populations in the digestive tract. At an optimum amount it does not affect nitrogen absorption but increases microbial population. This population's use of fiber sources high in fermentable carbohydrates can shift nitrogen excretion from urea and other resources to faeces, thereby reducing chances of ammonia emission. From an environmental point of view, nitrogen excreted in the faeces in microbial proteins is more favourable, because it is not rapidly degraded compared with urinary nitrogen. Although these diets do not always maximise pig performance, they provide an effective and economical use of locally grown feedstuffs and hence contribute to sustainable production. The influence of protein and energy nutrition on health and environmental issues in various production systems still require further academic discussion.

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