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Effects of linseed and oat in antibiotic-free diets on the gut health, mucosal integrity and performance of piglets

W. Krasucki[1, 2], Z. Mroz[3] and K. Szafran[1]

[1]Agricultural University of Lublin, Institute of Animal Nutrition
Akademicka 13, 20-950 Lublin, Poland
[3]Wageningen University and Research Centre, Animal Sciences Group Lelystad
8200 AB Lelystad, The Netherlands

ABSTRACT

The infection-preventive effects of 10% linseed meal (FM) and 15% hulled oat meal (HOM) in antibiotic-free diets for weaned piglets (n=48) were studied. Compared with controls, both plants improved (P<0.01) feed intake and growth rates up to day 6 post-weaning (before infection with *E. coli*). After *E.coli* infection, all piglets showed poorer performance, irrespective of the diet. The FM and HOM exerted no clear impact on faecal consistency, *E.coli* shedding or mucosal integrity, except for reduced (P=0.003) goblet cell and increased (P=0.04) leukocyte counts.

KEY WORDS: piglets, linseed, oat, infection, gut health

INTRODUCTION

In the search for substitutes for antibiotics now banned in feeds, linseed (*Linum usitatissimum*) and oat (*Avena sativa*) have received renewed scientific interest (Morris, 2001; Oomah, 2001; Liu et al., 2002). Since prehistoric times, both plants are known for possessing, in addition to protein and energy, some specific phytochemicals and bioactive compounds. They might act to improve feed intake, protect epithelial cells against pathogenic colonization, stimulate mucosal growth and immunity in the absence of antibiotics for prophylaxis after weaning of piglets. Therefore, the aim of this study was to test the influence of linseed meal (FM) and hulled oat meal (HOM) in an antibiotic-free diet on the clinical health, performance, feacal *E. coli* shedding, haematological indices and gut morphology of weaned piglets before and after exposure to a controlled infection with enterotoxigenic *E. coli* K88 (ETEC).

[2] Corresponding author: e-mail: wiliam@ursus.ar.lublin.pl

MATERIAL AND METHODS

Forty-eight crossbred piglets of 8 kg initial BW were studied during 19 days post-weaning (5 d pre-infection and 14 d post-infection with ETEC with a single oral dose of 5×10^9 CFU *E. coli* O149K91F4 K88ac). They were kept individually at an ambient temperature of 25°C and allotted to 4 isonitrogenous and isoenergetic treatments: 1. basal, no ETEC infection (B); 2. B + ETEC infection (B+); 3. B+10% FM +ETEC infection (BF+); 4. B+15% HOM + ETEC infection (BO+). The antibiotic-free, basal diet contained wheat (32.6%), barley (30.0%) and soyabean meal (17.8%) as the primary ingredients. The contents of CP, ileal digestible lysine and NE_f were 20.3%, 0.98%, and 10.0 MJ/kg, respectively. The FM and HOM were incorporated at the expense of wheat and soyabean meal. The diets and faeces were analysed for the contents of DM, ash, and N (AOAC, 1984). Also, dietary minerals were determined by using inductively coupled plasma atomic emission spectrometry (ICP-AES). Samples of faeces were taken daily directly from the rectum for: 1. enumeration of haemolytic *E. coli* on sheep blood agar plates (after incubation for 24 h at 37˚C); 2. faecal score index (FSI, scale 0 to 3). The body weight of piglets and feed intake were recorded on d 0 (weaning), 6 (before ETEC infection) and 19 (slaughter). On d 11 and 19 after weaning, blood samples from *v. jugularis* were taken to assay the haemogram and IgG titers using a CellDyn 4000 automated haematology analyser. Post-mortem examination encompassed intestinal size, weight and histology. Jejunal sections were stained with haematoxylin and eosin to measure villous/crypt shapes using light and scanning electron microscopy. Also, staining with high-iron diamin (HID) blue was performed to distinguish sialomucins and sulphomucins. pH and VFA contents were measured in the digest by HPLC. The data were subjected to ANOVA analysis of variance for a randomized block design, with treatments and *E.coli* infection as experimental factors. The significance of the differences between treatments was tested with Student's t-test at $P<0.05$ or 0.01.

RESULTS AND DISCUSSION

There were no clinical health problems or piglet mortality before the *E. coli* challenge (up to d 5 post-weaning), whereas 3 d after the challenge, two piglets died (one in BF+, and another in BO+) due to severe diarrhoea. Compared with controls (B), the piglets fed 10% FM and 15% HOM consumed more ($P<0.01$) feed and grew faster ($P<0.01$) before the *E. coli* challenge, but not afterwards (Table 1). The piglets challenged with ETEC K88 grew significantly slower than the control (B), non-challenged piglets. However, over the whole post-weaning

period of 19 d, the ADG of piglets were similar (P=0.31), i.e. 392, 311, 363, and 340 g/d for B, B+, BF+, and BO+, respectively. Faecal shedding of haemolytic *E. coli* highly corresponded to the ETEC *K88* infection, whereas the differences among the dietary treatments were less evident (Table 1).

Table 1. Feed intake (FI), growth rates and post-infective shedding of haemolytic *E. coli* (lg CFU/g) in faeces of piglets as affected by the treatments

Item	B	B+	BF+	BO+	P-value
Pre-infective FI (days 0-5), g/d	141^{ab}	123^{a}	204^{b}	201^{b}	<0.01
Post-infective FI (days 6-11), g/d	501	395	443	471	0.30
Pre-infective daily gain, g	24^{ab}	-20^{a}	92^{b}	58^{ab}	<0.01
Post-infective daily gain, g	533	439	431	449	0.12
Post-infective shedding of haemolytic E.coli (lg CFU/g) in faeces					
day 1 post-infection with ETEC	0.0^{a}	6.2^{b}	7.6^{c}	7.1^{bc}	<0.001
day 2 post-infection with ETEC	0.0^{a}	5.7^{b}	5.7^{b}	5.7^{b}	<0.001
day 3 post-infection with ETEC	0.0^{a}	5.6^{b}	6.0^{b}	5.0^{b}	<0.001
day 4 post-infection with ETEC	0.2^{a}	5.1^{b}	5.0^{b}	5.6^{b}	<0.001

B (basal); B+ (B+infection); BF+ (B+10%FM+infection); BO+ (B+15% HOM+infection)
^{a,b} means in rows with different superscripts differ at P<0.05

The FSI and DM contents in faeces were affected (P<0.05) by the *E. coli* challenge, but not by the diets. Neither the length nor weight of the duodenum differed among the treatments. However, the jejunum of piglets fed both plants was longer (P=0.04) and tended (P=0.08) to contain less NH_3 in digesta than in B+. Besides, their ileum tended (P=0.06) to be lighter, whereas the contents of VFA were similar among the treatments. Jejunal histology and HID-values were similar, except for the goblet cells and mucosal thickness (Table 2).

Table 2. Jejunal histology and digesta composition on day 13 post-infection in piglets

Item	B	B+	BF+	BO+	P-value
Villi/crypt ratio	1.60	1.44	1.40	1.31	0.12
No. of goblet cells, per 100 µm crypts	8.91^{b}	8.03^{ab}	7.21^{a}	7.52^{a}	0.003
Mucosal thickness, µm	834^{ab}	879^{b}	775^{a}	899^{b}	0.01
HID negative, %	26.9	22.7	17.9	22.4	0.39
Digesta DM, g/kg	115^{b}	89^{a}	88^{a}	92^{a}	0.04
Digesta NH_3, mg/kg	263	230	211	235	0.08

^{a,b} means in rows with different superscripts differ at P<0.05

Of the total haemogram, only leukocytes, basophiles (as % of leukocytes) and IgG-titers were significantly affected by the dietary treatments in the post-infection period (Table 3).

Table 3. Blood indices in weaned piglets as affected by the treatments and *E.coli* infection

Blood indices	B	B+	BF+	BO+	P-value
Leukocytes, $10^9/L^1$	14.4^a	14.6^a	16.4^{ab}	18.9^b	0.04
Basophiles, %[1]	0.84^a	1.47^b	0.68^a	1.12^{ab}	0.04
Monocytes, %[1]	5.4	7.5	4.6	5.5	0.06
IgG, ^{10}log titer[1]	0.23^a	0.77^b	0.41^a	0.36^a	0.02
IgG, ^{10}log titer[2]	0.29^a	1.12^b	1.18^b	1.07^b	<0.01

[a,b] means in rows with different superscripts differ at P<0.05
[1]day 5 post-infection, [2]day 13 post-infection

CONCLUSIONS

Adding 10% of linseed meal and 15% of hulled oat meal significantly improved feed intake and growth of weaned piglets before *E. coli* infection (up to 5 d post-weaning), but not afterwards. Based on all the response criteria, the anti-pathogenic potency of both plants in a weaner diet was less evident.

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STRESZCZENIE

Wpływ śruty lnianej i owsianej w dietach bez antybiotyków na zdrowotność jelita, integralność błony śluzowej oraz rozwój prosiąt

Przeciw infekcyjne właściwości dodatku mączki z siemienia lnianego (15%) lub z obłuszczonego owsa (10%) do dawek pokarmowych testowano na 48 prosiętach. W 6-tym dniu po odsadzeniu prosięta zostały doustnie zainfekowane *E. coli*. Oznaczano konsystencję kału, wydalanie *E. coli* w kale, cechy funkcyjne mukozy jelitowej oraz wskaźniki krwi. Dodatek mączek wpłynął istotnie (P<0,01) na poprawę pobrania paszy i przyrosty prosiąt przed infekcją. Po infekcji zmniejszyło się spożycie paszy i pogorszyły się przyrosty, niezależnie od dawki. Konsystencja kału, wydalanie *E. coli* i histologia jelita biodrowego były mało zróżnicowane miedzy grupami, ale liczba komórek gobletowych była mniejsza (P=0,003) i leukocytów większa (P=0,04) u prosiąt otrzymujących mączki w porównaniu z kontrolnymi.