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Reducing the dietary omega-6 to omega-3 polyunsaturated fatty acid ratio attenuated inflammatory indices and sustained epithelial tight junction integrity in weaner pigs housed in a poor sanitation condition

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ABSTRACT

The present study was conducted to determine the effect of reducing dietary n-6:n-3 PUFA ratio on the performance, inflammatory response and gut morphology of PWD challenged with sanitary and poor sanitary conditions in weaned pigs, and to test the hypotheses that (1) exposure to an poor sanitary environment will increase indices for inflammatory response; and (2) reducing n-6:n-3 PUFA ratio in diets for weaned pigs will attenuate the inflammatory response induced by the environmental challenge. A total of 108 male pigs [Duroc × (Yorkshire × Landrace); initial BW 7.1 ± 0.5 kg] weaned at 21 days of age were randomly allocated to one of 3 dietary treatments and 2 environmental conditions (sanitary vs. poor sanitary) to give 6 replicate pens per treatment with 3 pigs per pen. The dietary treatments were 3 graded levels of n-6:n-3 PUFA ratio (i.e.,20:1, 10:1 and 4:1) formulated using tallow, safflower oil, and a vegetable and fish oil blended product. One pig per pen (n = 6) was euthanized on d 0, d 7 and d 14, to collect blood and small intestinal tissue samples. Pigs exposed to a poor sanitary environment tended ($P < 0.10$) to grow more slowly and utilized feed less efficiently ($P < 0.05$) compared with the pigs housed in sanitary conditions. Housing weaned pigs in a poor sanitary environment increased ($P < 0.05$) the incidence of diarrhoea. Furthermore, a poor sanitary environment increased ($P < 0.001$) the occludin diffusion in the ileal epithelium of weaned pigs and increased plasma concentrations of TNF-α ($P < 0.05$), COX-2 ($P < 0.05$), PGE2 ($P < 0.01$) and LTB4 ($P < 0.05$) on d 14. Reducing the n-6:n-3 PUFA ratio improved ($P < 0.05$) both ADG and FCR but reduced ($P < 0.01$) the incidence of diarrhoea over 14 days after weaning, and they tended to attenuate ($P < 0.10$) the diffusion of the transmembrane tight junction protein occludin at the apical intercellular region of the ileal epithelium. Moreover, reducing the n-6:n-3 ratio in the diet attenuated the increased inflamma-

Abbreviations: ADFI, average daily feed intake; ADG, average daily gain; ANOVA, analysis of variance; BW, body weight; COX-2, cyclooxygenase-2; CP, crude protein; D, dietary treatments; DE, digestible energy; DHA, docosahexaenoic acid; DM, dry matter; E, environmental condition (sanitary vs poor sanitary); ELISA, enzyme linked immunosorbent assay; EPA, eicosapentaenoic acid; FCR, feed conversion ratio; GE, gross energy; IL-1β, interleukin 1β; LTB_4, leukotriene B_4; N, nitrogen; n-6:n-3, omega-6 to omega-3; PGE_2, prostaglandin E_2; PPARγ, peroxisome proliferator-activated receptor-γ; PUFA, polyunsaturated fatty acid; PWD, post-weaning diarrhoea; SEM, standard error of mean; SID, standardised ileal digestible; TNF-α, tumor necrosis factor α; V:C, villous height crypt depth

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tory indices induced by the environmental challenge. Correlation analysis indicated that n-6 PUFA intake of individual pigs positively correlated with plasma concentrations of IL-1β ($P < 0.01$), TNF-α ($P < 0.05$), PGE_2 ($P < 0.01$) and COX-2 ($P < 0.05$). Our results indicated that housing pigs in a poor sanitary environment after weaning increased inflammatory responses and reduced growth performance. Reducing the n-6:n-3 PUFA ratio to 4:1 attenuated the inflammatory responses observed after weaning in both environment on d 7 and in the poor sanitary environment on d 14.

1. Introduction

Weaner pigs are exposed to a number of stressors including microbial and (or) viral challenges. The association between stress and continuous exposure to pathogens can damage the intestinal epithelium, and lead to secondary inflammation and activation of the immune system (Lallès et al., 2007; Kim et al., 2012). Inflammation and subsequent immune system activation stimulates release of pro-inflammatory cytokines (Kim et al., 2013) that in turn stimulate the neurological lipid (eicosanoid) signaling system, which produces prostaglandin E_2 (PGE_2) and leukotriene B_4 (LTB_4) through actions of cyclooxygenase and lipoxygenase. Production of PGE_2 and LTB_4 is known to increase metabolic rate and heat production and trigger anorexia (Rakhshandeh and de Lange, 2011).

Caughey et al. (1996) demonstrated an inverse exponential relationship between eicosapentaenoic acid (EPA; 20:2n-3) content in

Table 1
Composition of the experimental diets (g/kg, as-fed basis).

Item	Dietary treatment (n-6:n-3 ratios)		
Ingredients, g/kg	20:1	10:1	4:1
Barley	50.0	50.0	50.0
Maize	308.6	306.2	305.2
Wheat	100.0	100.0	100.0
Rolled oats	100.0	100.0	100.0
Soybean meal	150.0	150.0	150.0
Full fat soya	50.0	50.0	50.0
Blood meal	20.0	20.0	20.0
Fishmeal	67.1	67.3	67.4
Whey powder	100.0	100.0	100.0
Tallow	14.8	17.0	17.9
Safflower oil	17.0	4.8	0.0
Fish oil[a]	0.0	12.2	17.0
Lysine-HCl	3.1	3.1	3.1
DL-Methionine	2.3	2.3	2.3
L-Threonine	1.3	1.3	1.3
L-Tryptophan	0.7	0.7	0.7
Vitamin-mineral premix[b]	1.0	1.0	1.0
Limestone	4.1	4.1	4.1
Dicalcium phosphors	7.7	7.6	7.6
Salt	2.0	2.0	2.0
Choline Chloride	0.4	0.4	0.4
Calculated analysis			
CP, g/kg	222.0	222.0	221.0
DE, MJ/kg	15.5	15.5	15.5
SID Lysine, g/MJ DE	0.9	0.9	0.9
Crude fat, g/kg	76.2	76.2	76.2
Analysed chemical composition			
CP, g/kg	218.8	229.2	215.1
DM, g/kg	887	897	901
GE, MJ/kg	16.9	17.1	17.2
Total n−6[c], g/kg	54.8	53.8	44.5
Total n−3[d], g/kg	2.8	5.1	11.3
n-6:n-3 ratio	19.4:1	10.6:1	3.9:1

[a] REI 3• (Morning Bio Ltd., Cheonan, South Korea) supplied per kilogram of total diets: omega 3 fatty acid 160 g (α-linolenic acid 147 g, docosahexaenoic acid 9 g and eicosapentaenoic acid 4 g).

[b] Provided the following nutrients (per kg of air-dry diet): Vitamins: A 7000 IU, D_3 1400 IU, E 20 mg, K 1 mg, B_1 1 mg, B_2 3 mg, B_6 1.5 mg, B_{12} 15 μg, calcium pantothenate 10.7 mg, folic acid 0.2 mg, niacin 12 mg, biotin 30 μg. Minerals: Co 0.2 mg (as cobalt sulphate), Cu 10 mg (as copper sulphate), iodine 0.5 mg (as potassium iodine), iron 60 mg (as ferrous sulphate), Mn 40 mg (as manganous oxide), Se 0.3 mg (as sodium selenite), Zn 100 mg (as zinc oxide). (BJ Grower 1, BioJohn Pty Ltd., WA, Australia).

[c] Total n-6; C18:2 + C18:3 + C20:4.

[d] Total n-3; C18:3n3 + C18:4 + C20:5 + C22:5 + C22:6.