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Feeding a diet with a decreased protein content reduces both nitrogen content in the gastrointestinal tract and post-weaning diarrhoea, but does not affect apparent nitrogen digestibility in weaner pigs challenged with an enterotoxigenic strain of *Escherichia coli*

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ABSTRACT

This study evaluated possible nutritional and physiological mechanisms to explain why feeding a diet of decreased protein content reduces PWD. A total of 48 male pigs weaned at 21 d (initial BW 6.9 ± 0.11 kg; mean ± SEM) was used in a 2 × 2 × 2 factorial arrangement of treatments with the respective factors being: (1) PL (HP 239 g/kg *vs.* LP 190 g/kg CP); (2) presence or absence of an ETEC challenge; and (3) duration of feeding after weaning until euthanasia (D; 7 d *vs.* 14 d). No dietary antimicrobial compounds were used, and diet LP contained crystalline AA including isoleucine and valine to achieve an ideal AA pattern. Pigs were offered the experimental diets on an *ad libitum* basis. Feeding a LP diet decreased total N intake, ileal N flow, PUN and NH_3-N contents at the ileum and all sites in the large intestine ($P<0.05$–0.001), but did not alter ($P>0.05$) the AID of N and AA at either d 7 or d 14, except for serine which was lower in pigs fed the LP diet ($P<0.001$). Feeding diet HP increased the incidence of PWD, and ETEC infection increased PWD only in pigs fed the HP diet (PL × ETEC interaction, $P<0.05$). Pigs fed diet HP had more PWD at d 7 but not at d 14 after weaning (PL × D interaction, $P<0.05$). Experimental ETEC infection increased ($P<0.001$) faecal *Escherichia coli* score compared to non-infected pigs, and decreased AID of some AA at d 7 (ETEC × D interaction, $P<0.05$–0.001). Feeding diet LP reduced the molar proportion of BCFA in the caecum and proximal colon ($P<0.001$ and $P<0.05$, respectively), but total VFA concentrations in this organ were unaffected by PL ($P>0.05$). Pigs fed diet LP had decreased pH in the jejunum and ileum ($P<0.05$ and $P<0.01$, respectively), while ETEC infection increased pH in the caecum and proximal colon at d 7 (ETEC × D interaction, $P<0.05$). Feeding diet LP did not

Abbreviations: AA, amino acid(s); ADF, acid detergent fibre; ADG, average daily gain; ADFI, average daily feed intake; AID, apparent ileal digestibility; ANOVA, analysis of variance; BCFA, branched-chain fatty acid(s); BW, body weight; cfu, colony-forming units; CP, crude protein; D, duration of feeding after weaning; d, day(s); DE, digestible energy; DM, dry matter; EBW, empty body weight; ETEC, enterotoxigenic *Escherichia coli*; FC, faecal consistency; GE, gross energy; GIT, gastrointestinal tract; h, hour(s); GLM, general liner model; HP, high protein; LP, low protein; LSD, least significant difference; ME, metabolisable energy; N, nitrogen; NDF, neutral detergent fibre; NE, net energy; NH_3-N, ammonia nitrogen; PL, protein level; PUN, plasma urea nitrogen; PWD, post-weaning diarrhoea; SEM, standard error of mean; SID, standardized ileal digestible; VFA, volatile fatty acid(s).

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alter GIT weight, but ETEC infection adversely affected the proportional weight of the GIT at d 7 (ETEC × D interaction, P<0.01). The PL did not alter small intestinal morphology and growth. These results suggest that feeding a LP diet immediately after weaning reduces the flow of N into the large intestine, thereby decreasing protein fermentation without altering apparent AA digestibility at the ileum.

1. Introduction

Post-weaning diarrhoea (PWD) is an enteric disorder that can negatively influence production, and is usually characterized by frequent discharge of watery faeces associated with dehydration, lethargy, and sometimes mortality in the first 2 weeks of weaning (Hampson, 1994; Pluske et al., 2002). PWD is usually associated with colonisation of the small intestine with toxin-producing β-haemolytic ETEC (Nagy and Fekete, 1999; Fairbrother et al., 2005), however some recent studies have shown that feeding a low protein diet for either 7 d or 14 d after weaning decreased the incidence of PWD commensurate with decreased indices of protein fermentation in the large intestine, although it was evident that feeding a low protein diet for as little as 7 d was sufficient to reduce PWD (Heo et al., 2008, 2009). Numerous authors also have demonstrated that feeding a low protein diet reduces PWD associated with reductions in indices of protein fermentation (*i.e.*, NH_3-N and PUN) in the GIT (Nyachoti et al., 2006; Wellock et al., 2006; Htoo et al., 2007; Yue and Qiao, 2008; Opapeju et al., 2008, 2009).

At present, there is a lack of evidence underpinning how feeding a diet of decreased protein content might reduce PWD. Pigs eating less protein will have a smaller quantity of protein entering the large intestine, which in turn most likely explains the decreased indices of protein fermentation that, in turn, influences the expression of PWD. With this in mind, the current experiment tested the hypothesis that feeding a diet of decreased protein content would cause a comparable N digestibility at the terminal ileum and hence decrease the intestinal flow of N. Such a mechanism would potentially decrease protein fermentation indices in the large intestine thereby reducing the risk of PWD. This experiment also offered an opportunity to examine the effects of infection with ETEC on apparent digestibility of AA measured at the terminal ileum.

2. Materials and methods

This study was reviewed and approved by the Murdoch University Animal Ethics Committee (R2094/07). Animals were cared for according to the Australian Code of Practice for the Care and Use of Animals for Scientific Purposes (CSIRO, 2008).

2.1. Experimental design, animals, housing and diets

The experiment was designed as a 2 × 2 × 2 factorial arrangement of treatments (*n* = 6), with the respective factors being: (1) 2 dietary PL (HP; 239 g/kg *vs.* LP; 190 g/kg), (2) presence or absence of an ETEC challenge (serotype O149; K91; K88), and (3) duration of feeding after weaning until euthanasia (D; 7 d *vs.* 14 d). This factor examined first, whether a LP diet could be fed for a shorter period after weaning to achieve the same effects, and second, the effects of time of feeding after weaning on protein digestion in the GIT.

Forty-eight male pigs (Large White × Landrace) aged 21 d and weighing 6.9 ± 0.11 kg (mean ± SEM) were used. Pigs were obtained from a commercial supplier (Wandalup Farms Ltd., Mandurah, WA, Australia) on the day of weaning and transported to the experimental facility at Murdoch University, where each pen was equipped with a nipple bowl drinker and a metal trough. Pigs were allocated to their experimental feeding regimen based on initial BW and block within the room in the animal facility. Infected and non-infected pigs were separately housed in rooms (after Ding et al., 2006) with eight pens of three pigs each (space allowance of 0.88 m^2 per pig, and a feeder space allowance of 7.8 cm per pig). The pigs were offered their respective experimental diets *ad libitum* for 2 weeks. Cleaning and feeding schedules were implemented to ensure that movement between rooms was conducted in the order from non-infected to infected groups.

Diets were formulated to contain similar DE content and NDF contents but a different PL (Tables 1 and 2), and were formulated using standardized ileal digestible AA contents (Sauvant et al., 2004). Diets were fed in mash form and were formulated to contain at least an ideal pattern of ileal digestible AA (Chung and Baker, 1992). Crystalline AA (lysine, methionine, tryptophan and threonine) were added to the LP diets, with crystalline isoleucine and valine also added to achieve the ideal pattern of essential AA (Chung and Baker, 1992). The ambient temperature was maintained at 29 ± 1 °C for the initial week, and then decreased by 2 °C in the second week.

2.2. Infection procedure

Experimental infection with ETEC was conducted at 76, 96 and 120 h after weaning, using the procedures described previously (Heo et al., 2009). Each pig in the infection group received 8 mL of freshly prepared broth for the respective time-set, providing approximately 1.84×10^8 cfu/mL of ETEC per pig per day.