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Effects of chitooligosaccharide supplementation on growth performance, nutrient digestibility, blood characteristics and immune responses after lipopolysaccharide challenge in weanling pigs

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ABSTRACT

The objective of this study was to determine the effects of dietary supplementation with chitooligosaccharide (COS) on growth performance, nutrient digestibility, blood characteristics and immune response in lipopolysaccharide-challenged weanling pigs. A total of 90 crossbred weanling pigs (5.44 ± 0.50 kg BW) were employed in Exp. 1. The three dietary treatments were basal diets supplemented with 0, 2.5, and 5 g COS/kg, and fed for 28 d. Each treatment had 6 replications with 5 pigs per pen. Increasing the level of supplemental COS tended to linearly ($P<0.10$) improve ADG and ADFI during phase 2 and overall period, while there were no differences in G:F. The linear improvement in the apparent DM ($P<0.05$) and N ($P<0.10$) digestibility in pigs fed COS supplemented diets was noticed. The tested blood characteristics were not influenced under non-challenge conditions. In Exp. 2, a total of 20 pigs (5.22 ± 0.31 kg BW) were initially assigned to two dietary treatments and fed basal diets supplemented with 0 or 0.5 g COS/kg for 28 d. At the end of d 28, half of the pigs in each treatment ($n=5$) were injected i.p. with *Escherichia coli* lipopolysaccharide at a concentration of 100 μg/kg of BW. The other half of the pigs in each treatment were injected with sterile saline solution at a concentration of 100 μg/kg of BW. This arrangement resulted in a 2 × 2 factorial design with diet and LPS challenge as the main effects. Blood sample and rectal temperature data were collected at 0, 2, 4 and 12 h post-challenge. Rectal temperatures increased as the result of LPS injection at 4 and 12 h post-challenge ($P<0.05$). Serum cortisol, IGF-1, and TNF-α concentration were also increased as the result of LPS challenge ($P<0.05$). The COS treatments resulted in lower cortisol concentrations at 2 h and higher IGF-1 concentrations at 4 h post-challenge ($P<0.05$). COS and LPS interactions were also observed on cortisol and IGF-1 when the COS effects were presented ($P<0.05$). Haptoglobin concentrations remained unaffected throughout the challenge period. White blood cell counts were increased in the LPS-treated pigs at 2 and 4 h post-challenge ($P<0.01$). Lymphocyte count was elevated at 2 h and reduced at 12 h post-challenge as the result of LPS challenge ($P<0.05$). However, there were no COS main effects observed on lymphocyte count throughout the challenge period. The comparison between two LPS challenged treatments also indicated that COS treatment has beneficial effects on rectal temperature, cortisol and IGF-1 concentrations. In conclusion, dietary supplementation with COS had little effect on nutrient digestibility and inflammatory stress markers in weanling pigs.

1. Introduction

In recent years, the development of functional oligosaccharide products has burgeoned into an important industry, which encompasses medicine, chemistry, agriculture, etc. In the field of animal nutrition, several studies have already been published

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Table 1
Compositions of experimental diets (as-fed basis).

Ingredients (g/kg)	Phase 1 (day 0 to 14)	Phase 2 (day 15 to 28)
Expanded corn	66.5	357.2
Expanded oat	100.0	–
Biscuit meal [a]	–	50.0
Soybean meal	80.0	200.0
Fermented soybean meal	78.0	82.0
Fish meal	50.0	40.0
Soy oil	41.5	48.0
Lactose	100.0	60.0
Whey	165.0	100.0
Milk product [a]	130.0	20.0
Lecithin	5.0	–
Monocalcium phosphate	12.5	10.0
Glucose	50.0	–
Sugar	40.0	20.0
Plasma powder	65.0	–
L-lysine-HCl	01.2	2.5
DL-methionine	02.6	1.5
L-Threonine	7.7	0.8
Choline chloride	2.0	1.0
Vitamin premix [b]	1.0	1.0
Mineral premix [c]	2.0	2.0
Limestone	–	2.0
Salt	–	2.0
Chemical composition [d]		
ME (MJ/kg)	14.8	14.8
Crude protein (g/kg)	220.0	210.0
Lysine (g/kg)	15.7	14.1
Methionine (g/kg)	6.0	4.9
Calcium (g/kg)	8.0	7.8
Phosphorus (g/kg)	7.6	7.6

[a] Mainly contains 210 g/kg fat and 220 g/kg protein.
[b] Provided per kg of complete diet: vitamin A, 11,025 IU; vitamin D_3, 1103 IU; vitamin E, 44 IU; vitamin K, 4.4 mg; riboflavin, 8.3 mg; niacin, 50 mg; thiamine, D-pantothenic acid, 29 mg; choline, 166 mg; and vitamin B_{12}, 33 μg.
[c] Provided per kg of complete diet: Fe, 200 mg; Cu, 12 mg; Zn, 200 mg; Mn, 8 mg; I, 0.28 mg; Se, 0.15 mg.
[d] Calculated values.

regarding the application of oligosaccharide products, including fructo-oligosaccharides, galacto-oligosaccharides, and mannanoligosaccharides and so on (Houdijk et al., 1998; Rozeboom et al., 2005). Previous studies have shown that the primary functions of this type of physiologically active material include the reduction of pathogen levels in the intestine, as well as improvements in gut health, increases in nutrient digestibility, and enhancements in immune function, etc. (LeMieux et al., 2003; Rozeboom et al., 2005). In addition, oligosaccharide products influence lipid and growth hormone metabolism (Sugano et al., 1988; Tang et al., 2005).

Chitooligosaccharide (COS) is one type of the above mentioned oligosaccharides. Unlike other oligosaccharide products, such as mannanoligosaccharides obtained from the yeast cell walls of *Saccharomyces cerevisiae*, COS is produced from chitin or chitosan by chemical or enzymatic decomposition methods (Li et al., 2007). COS, which has a far lower molecular weight than chitosan, has been suggested to present higher water solubility and activity. Due to the fact that the chitosan is found abundantly in natural sources, it is produced commercially on a large scale in different countries (Singla and Chawla, 2001). Therefore, the development potential of COS is a matter of great interest to researchers. In fact, some studies regarding COS have already been conducted in either pigs or broilers, and beneficial effects on growth performance, immunity, or blood profiles have been reported (Han et al., 2007; Li et al., 2007). However, such effects were not always consistently reported. One of the reasons was considered to be that result to this discrepancy was considered to be the hygienic condition of most experiments were much better than the situation in real practise farms. Therefore, we hypothesized that the beneficial effects of COS may present more obviously under a bacteria challenge condition. Study conducted by Wright et al. (2000) suggested that the lipopolysaccharide (LPS) challenge decreased feed intake, stimulated a febrile response and activated the hypothalamic–pituitary–adrenal axis as demonstrated by increased cortisol levels. This provided an ideal experimental model for the immunity related study.

The objective of the current study was to further evaluate the efficacy of COS on growth performance, nutrient digestibility, blood characteristics, and immune response under inflammatory challenge conditions.

2. Materials and methods

The Animal Care and Use Committee of Dankook University approved all of the experimental protocols used in the current study. The primary composition of COS used in the current study includes 186 g/kg crude protein, 145 g/kg crude fat, 107 g/kg crude fiber, 90 g/kg moisture, >40 g/kg chitin·chitosan, >30 g/kg chitooligosaccharide, >90 mg/kg carotenoid and others (company specification). The average molecular weight of COS was about 2000 Da. This product was manufactured by EASY BIO System Inc. (Korea).

2.1. Experiment 1

2.1.1. Animals, housing, and treatments

A total of 90 gilts ([Landrace × Yorkshire] × Duroc) with an average initial BW of 5.44 ± 0.50 kg were assigned to one of three dietary treatments. Six replicate pens consisting of five pigs per pen were used in a randomized complete block design. The dietary treatments included a corn–soybean meal-based control diet or control diet supplemented with 2.5 and 5 g/kg COS. The COS was added at the expense of corn. A two-phase nursery feeding program was employed in the experiment, with diets formulated to provide all of the nutrients to meet or exceed NRC (1998) requirements (Table 1). The experiment lasted for 28 d and all of the pigs received the same phase 1 control diet for 2 d of acclimation prior to the experiment. The pigs were switched to phase 2 experimental diets during d 15–28.

All of the pigs were housed in an environmentally-controlled nursery facility with slatted plastic flooring and a mechanical ventilation system. The environmental temperature was maintained at 30 °C for the first week of the experiment, and was then reduced by 1 °C per week over the next three weeks. Each pen (1.8 × 1 m) was provided with a stainless steel feeder and one nipple waterer, which allowed *ad libitum* access to feed and water throughout the experiment.

2.1.2. Experimental procedures, sampling, and analysis

Individual pig BW was measured initially and on d 14 and 28 of the experiment. Feed disappearance per pen was also assessed on d 14 and 28 of the experiment. The ADG, ADFI,