

The effect of diets containing soybean meal, soybean protein concentrate, and soybean protein isolate of different oligosaccharide content on growth performance and gut function of young turkeys

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ABSTRACT An experiment was conducted to investigate the effects of diets containing soybean meal (SBM), soybean protein concentrate (SPC), and soybean protein isolate (SPI) on growth performance and gut function of the young turkey. A total of 812 one-day-old male turkey poults were randomly assigned to 4 dietary treatments, with 7 pens per treatment and 29 birds per pen. The 4 experimental diets contained SBM, SBM-SPC, SPC, and SPI and were fed throughout the two 4-wk experimental periods. In each period, the diets were isonitrogenous and isocaloric and contained similar amounts of total and water-soluble nonstarch polysaccharides. The content of oligosaccharides differed among the diets and averaged 2.4, 1.9, 0.9, and 0.1% for SBM, SBM-SPC, SPC, and SPI, respectively. When compared with SBM, birds consuming the SBM-SPC and SPC diets had higher ($P < 0.05$) final BW (4.32 vs. 4.45 and 4.46 kg, respectively). Incorporation of SPI as a substitute for SBM resulted in improved ($P < 0.05$) feed utilization (from 1.76 to 1.67) but did not affect

the final BW. Significant changes in cecal concentrations of short-chain fatty acids were observed and averaged 130, 103, and 89 $\mu\text{mol/g}$ of digesta for the SBM, SBM-SPC, and SPC diets, respectively. This coincided with the proportional decrease in dietary oligosaccharide content (from 2.4 to 0.9%) and was further substantiated by a significant decrease in ileum weights. Feeding the SPI diet resulted in the lowest ileal and cecal tissue weights as well as the lowest cecal short-chain fatty acids concentration. There was no effect of diet on digesta pH, viscosity, and mucosal sucrase and maltase activities. Bacterial β -glucuronidase activity was decreased ($P = 0.08$) in the cecum (from 0.98 to 0.60 U/g) with decreased dietary oligosaccharide content. In conclusion, partial or almost complete substitution of SBM with SPC suppressed the fermentation processes in the ceca but enhanced the growth rate. Substitution of SBM with SPI significantly improved feed utilization but decreased BW of 4-wk-old turkeys with no effect on growth rate of older 8-wk-old birds.

Key words: soybean protein concentrate, soybean protein isolate, oligosaccharide, gastrointestinal tract, turkey

2009 Poultry Science 88:2132–2140

doi:10.3382/ps.2009-00066

INTRODUCTION

Soybean meal (SBM) is a common protein source in animal feeds. Processed soybean products [e.g., soy protein concentrate (SPC) or protein isolate (SPI)] are of lesser significance in poultry feeding but are recommended as dietary components with functional properties for human nutrition and have been implicated in cancer prevention (Bennink, 2001; Linz et al., 2004) and prevention of many other diseases, including degenerative changes in the liver (Gudbrandsen et al., 2006). For

this reason, SPC and SPI may be used as value-added feed components for young animals with underdeveloped gastrointestinal tract. Sohn et al. (1994) reported that in the 21- to 35-d-old pigs, SPI as well as SPC, but not SBM, were of equal quality as substitutes for milk protein. In another study, partial replacement of SBM with extruded SPC in the high-SBM diet (40%) significantly improved pig performance; however, growth performance was not affected when nonextruded SPC was fed (Lenehan et al., 2007). In similar experiments with pigs, the digestibility of CP and amino acids of SPC and SPI were demonstrated to be higher than those of SBM (Sohn et al., 1994; Grala et al., 1998). In a similar study, BW gain of broiler chickens fed diets containing SPC and SPI were equal to those fed a casein-based diet but significantly lower than those fed

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Received February 5, 2009.

Accepted June 5, 2009.

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SBM (Batal and Parsons, 2003). Therefore, the processes involved in SPC and SPI preparation may result in improved energy and amino acid availability, which, in turn, may have some positive effects on chick growth, especially during the first 3 wk posthatch, because the energy and amino acid digestibility would increase with age (Batal and Parsons, 2002, 2003). Growth rates of chicks fed the SPC- and SPI-containing diets may in some cases be decreased due to amino acid imbalance caused by insufficient concentrations of sulfur amino acids and threonine (Emmert and Baker, 1995). This problem can be overcome by dietary supplementation with synthetic amino acids. On the other hand, during the production process, some bioactive components may be removed, which, in turn, might influence the physiological properties of the final products with some repercussions associated with the gastrointestinal tract (GIT) function (Li et al., 1991; Liener, 1994; Grala et al., 1998).

In addition to the high protein content, SBM is a rich source of carbohydrates (32 to 34%), with about half of these being nonstructural in nature and representing simple sugars, sucrose, oligosaccharides, and starch (Bach Knudsen, 1997). The other portion represents the structural polysaccharides with pectic polysaccharides predominating (Bach Knudsen, 1997; Karr-Lilienthal et al., 2005; Meng et al., 2005). Technological processes involved in SPC and SPI production may modify the content and the properties of the carbohydrate fractions. Oligosaccharides (raffinose family oligosaccharides, galactooligosaccharides, α -D-galactosides, galactosyl-sucrose oligosaccharides), which comprise approximately 5 to 6% of the soybean meal DM, are extensively fermented by bacterial population in the lower gut, and to some extent in the distal ileum of nonruminants (Liyang et al., 2003). This fermentation pattern may result in both positive and negative effects (Karr-Lilienthal et al., 2005). At this point, some authors have recognized α -galactosides raffinose, stachyose, and verbascose as oligosaccharides favorably stimulating the GIT ecosystem, especially in its lower segments (Chow, 2002). However, other investigations demonstrated the opposite effects of soybean oligosaccharides (Liyang et al., 2003). Fernandez-Quintela et al. (1998) suggested that protein isolates obtained from legume seeds may have a positive effect on glucose and triglyceride plasma levels and reduce, to some extent, some undesirable effects associated with legume consumption.

The ban on the use of meat and meat-bone meal (except fishmeal) in animal feeds in the European Union has made meeting the nutritional requirements of young and fast-growing birds increasingly difficult, especially of young turkeys of heavy lines that require 28% of CP in their diet in the first period of nutrition (NRC, 1994). In this case, the content of protein supplements, including SBM, may exceed 50%. As a consequence, such diets may contain high quantities of oligosaccharides (Baker, 2000). On the other hand, extraction of SBM with various solvents may result in the production

of SPC containing only 3% of oligosaccharides or SPI with no oligosaccharides present (Refstie et al., 1999; Peisker, 2001). Although these types of products have already been studied with broiler chickens (Coon et al., 1990; Refstie et al., 1999), very little information is available on their usefulness in young turkey nutrition.

Therefore, the main goal of the present study was to better understand the physiological response of young turkeys to diets containing SPC and SPI of different carbohydrate content and composition. Moreover, an attempt has been made to assess the effectiveness of substituting SBM with SPC and SPI.

MATERIALS AND METHODS

Soybean Products

Soy protein concentrate Supro 500E IP and SPI Procon 2100 IP were purchased from Solae Company (St. Louis, MO). Soy hulls were obtained from a feed mill in Coefeld-Lette (Germany). Soybean meal was obtained from a local feed company. Chemical composition of soybean products used in the study is presented in Table 1.

Diets

The 4 experimental wheat-based diets contained SBM, SBM-SPC, SPC, and SPI (Table 2). All diets were isonitrogenous (27 and 25% CP in 1 to 4- and 5 to 8-wk experimental periods, respectively), isocaloric (2,900 kcal/kg of ME), contained similar amounts of crude fiber (CF) and total and water-soluble NSP, and differed in the content of oligosaccharides. Dietary CF content was adjusted using soybean hulls. The contents of amino acids, minerals, and vitamins were similar in all diets and met or exceeded NRC specifications. Antibiotic was excluded from all diets.

Birds and Housing

A total of 812 one-day-old BIG-6 (Hatchery Grelavi Co., Ketrzyn, Poland) male turkey poults were randomly assigned to 4 dietary treatments, each consisting of 7 pens of 29 birds per pen. The birds were vaccinated at 1 d of age using Aviffa-RTI (Rhone Merieux, Lyon, France), a vaccine against turkey rhinotracheitis (an aerosol-spraying method). The poults were raised in the floor pens and were provided with 16L:8D per day. Room temperature was maintained at 32°C for the first 5 d and was gradually decreased according to normal management practices until a temperature of 22°C was achieved. The birds were given free access to mash diets. All diets were fed throughout the 2 experimental periods of 4 wk each (i.e., 1 to 4 and 5 to 8 wk). At the end of each 4-wk period, the birds were weighed and feed intake was recorded. Weight gain and feed efficiency for each period were determined and each pen was considered an experimental unit. The animal

Table 1. Chemical composition of soybean products used in the study (mg/g, as-fed basis)

| Component | Soybean meal | Protein concentrate | Protein isolate | Hulls |
|---------------------------------|--------------------|---------------------|-----------------|-------|
| DM | 896.2 ¹ | 955.9 | 952.4 | 898.4 |
| Ash | 64.2 | 70.9 | 36.8 | 51.0 |
| CP | 470.1 | 649.2 | 858.8 | 126.8 |
| Ether extract | 19.8 | 13.2 | 4.8 | 14.7 |
| Carbohydrates | | | | |
| Monosaccharides ² | 9.5 | 5.2 | 6.8 | 13.4 |
| Sucrose | 56.9 | 10.1 | 1.3 | 14.6 |
| Oligosaccharides | 53.0 | 24.6 | 3.7 | 8.0 |
| Raffinose | 10.4 | 4.0 | 0.1 | 3.0 |
| Stachyose | 40.1 | 19.8 | ND ³ | 4.6 |
| Verbascose | 2.5 | 0.8 | 3.6 | 0.4 |
| Dietary fiber fractions | | | | |
| Crude fiber | 34.8 | 28.1 | 1.6 | 364.0 |
| Acid detergent fiber | 55.2 | 44.2 | ND | 423.3 |
| Neutral detergent fiber | 76.5 | 70.7 | 1.9 | 550.2 |
| Nonstarch polysaccharides (NSP) | 125.9 | 129.2 | 12.9 | 461.0 |
| NSP component sugars | | | | |
| Rhamnose | ND | ND | ND | 3.9 |
| Arabinose | 20.2 | 23.7 | ND | 33.9 |
| Xylose | 9.2 | 8.3 | ND | 67.4 |
| Mannose | 5.0 | 5.1 | 5.6 | 22.7 |
| Galactose | 38.9 | 48.2 | 3.3 | 16.3 |
| Glucose | 31.2 | 25.3 | 1.6 | 263.1 |
| Uronic acids | 21.4 | 18.7 | 2.4 | 53.7 |
| Water-insoluble NSP | 105.4 | 109.7 | 10.0 | 395.3 |
| Water-soluble NSP | 20.5 | 19.5 | 2.9 | 65.7 |

¹Values are means of duplicate determinations.

²Includes glucose and fructose.

³Not detected.

protocol for this study was approved by the Animal Care and Use Committee of the University of Warmia and Mazury.

Sample Collection

After 4 and 8 wk of feeding, 10 birds representing an average BW of each group were killed by cervical dislocation according to the recommendations for euthanasia of experimental animals (Close et al., 1997). After laparotomy, segments of the digestive tract (small intestine and ceca) were removed, emptied, and weighed. As soon as possible after euthanasia (ca. 20 min), ileal and cecal pH was measured using a microelectrode and pH-ion meter (model 301, Hanna Instruments, Vila do Conde, Portugal). Samples of ileal (middle, 1/3 section of ileum) and cecal contents were used for immediate analysis of DM, viscosity, and short-chain fatty acids (SCFA), whereas the rest of the cecal digesta was transferred to the test tubes and stored at -70°C until needed. The ceca were flushed with water, blotted on filter paper, and weighed. The small intestine was divided into 4 equal parts, and then the second part (jejunum) from the duodenum end was rinsed with ice-cold physiological saline and cut open. The mucosal samples were collected by scraping with glass slides on an iced glass plate, weighed, and subsequently stored at -70°C .

Chemical Analyses

Feed ingredient, diet, or digesta samples were analyzed in duplicate for DM, CP, fat, CF, neutral deter-

gent fiber, acid detergent fiber, and ash using AOAC (2005) methods 934.01, 976.05, 920.39, 978.10, 2002.04, 989.03, and 942.05, respectively. For chemical analysis, the samples were ground to pass through a 0.5-mm sieve.

Carbohydrates (glucose, fructose, sucrose, raffinose, stachyose, verbascose) were analyzed using the procedure described by Muzquiz et al. (1992) with some modifications. Briefly, 0.5 g of sample was homogenized with 5 mL of 50% ethanol for 1 min at 21°C using an Ultra-Turrax homogenizer (M. Zipperer GmbH, Staufen, Germany). The mixture was centrifuged for 5 min at $669 \times g$, the supernatant was decanted, and the residue was extracted with a fresh portion of 50% ethanol. This procedure was repeated twice. The combined extracts were purified using C_{18} cartridges (500 mg/6 mL) connected to a vacuum system. The effluent was evaporated to dryness, redissolved in deionized water (1 mL), and centrifuged at $2,676 \times g$ for 8 min at 21°C . The analysis of sugars was carried out by HPLC using a Merck Hitachi La Chrom 7000 HPLC and a L-7490 refractive index detector (Merck Hitachi, Darmstadt, Germany). For sugar separation, a Bondapak NH_2 125Å 10- μm column (3.9×300 mm; Waters, Milford, MA) and acetonitrile:water 79:21 (vol/vol) as mobile phase were used. Injection volume was 20 μL . Quantification of each sugar was performed by comparing the peak areas of the samples with those of the known amounts of sugar standards over the range of 0 to 4 mg/mL.

Nonstarch polysaccharides (NSP) were determined by gas-liquid chromatography (component neutral sugars) and by colorimetry (uronic acids) using the proce-

diure described by Englyst and Cummings (1984, 1988) with some modifications (Slominski and Campbell, 1990). Briefly, 100-mg samples were boiled with 2 mL of dimethylsulfoxide for 1 h and then incubated at 45°C overnight with a sodium acetate buffer solution (pH 5.2) of starch-degrading enzymes amylase, pullulanase, and amyloglucosidase (Sigma Chemical Co., St Louis, MO). Ethanol was then added and the mixture was left for 1 h at room temperature before being centrifuged at $1,990 \times g$ for 10 min at 21°C. The supernatant was discarded and the dried residue was dissolved in 1 mL of 12 *M* sulfuric acid and incubated for 1 h at 35°C. Six milliliters of water and 5 mL of myo-inositol (internal standard) solution were then added and the mixture was boiled for 2 h. One milliliter of the hydrolysate was taken and neutralized with 12 *M* ammonium hydroxide, reduced with sodium borohydride, and acetylated with acetate anhydride in the presence of 1-methylimidazole. Component sugars were separated using a SP-2340 column and a Varian CP 3380 Gas Chromatograph (Varian Canada Inc., Mississauga, Ontario, Canada). Uronic acids were determined using the procedure described by Scott (1979). Water-soluble NSP content was deter-

mined according to the method described by Slominski et al. (1993).

For digesta viscosity measurements, the contents of the small intestine were collected, mixed on a vortex mixer, and centrifuged at $7,211 \times g$ for 10 min at 21°C. The supernatant (0.5 mL) was placed in a Brookfield LVDV-II+ cone-plate rotational viscometer (CP40, Brookfield Engineering Laboratories Inc., Stoughton, MA) and the viscosity was measured at a fixed temperature of 39°C and a shear rate of 60 per minute.

The mucosal sucrase and maltase activities were assayed by the method of Dahlqvist (1964). The amount of liberated glucose was measured spectrophotometrically and the enzyme activity was expressed as micromoles of disaccharide hydrolyzed per minute and gram of protein. Protein content in the ileal mucosa was determined by the Lowry method (Lowry et al., 1951) using BSA as a standard.

The activity of bacterial β -glucosidase and β -glucuronidase in the cecal digesta was measured by the rate of *p*-nitrophenol release from the nitrophenylglucosides according to the modified method of Djouzi and Andrieux (1997) as described by Juskiewicz and

Table 2. Composition of experimental diets (as-fed basis)

| Item | Starter (1 to 4 wk) | | | | Grower (5 to 8 wk) | | | |
|---|---------------------|---------|-------|-------|--------------------|---------|-------|-------|
| | SBM | SBM-SPC | SPC | SPI | SBM | SBM-SPC | SPC | SPI |
| Ingredient, g/kg | | | | | | | | |
| Wheat | 423.0 | 499.4 | 614.9 | 699.4 | 488.1 | 529.7 | 643.8 | 729.0 |
| Soybean meal (SBM) | 443.0 | 283.0 | 39.0 | — | 393.9 | 304.8 | 62.1 | — |
| Soybean protein concentrate (SPC) | — | 100.0 | 253.0 | — | — | 55.8 | 208.0 | — |
| Soybean protein isolate (SPI) | — | — | — | 195.0 | — | — | — | 174.3 |
| Soybean hulls | 25.0 | 28.0 | 32.0 | 48.0 | 25.5 | 27.1 | 31.3 | 46.4 |
| Soybean oil | 55.0 | 36.0 | 7.0 | — | 46.4 | 36.0 | 7.7 | — |
| Sodium bicarbonate | 1.0 | 1.0 | 1.0 | 1.0 | 1.0 | 1.0 | 1.0 | 1.0 |
| Sodium chloride | 3.0 | 3.0 | 3.0 | 3.0 | 2.6 | 2.6 | 2.6 | 2.7 |
| Limestone | 16.0 | 15.5 | 15.8 | 16.3 | 13.2 | 13.4 | 13.7 | 14.1 |
| Monocalcium phosphate | 18.0 | 18.2 | 18.6 | 18.8 | 14.3 | 14.5 | 14.9 | 15.1 |
| DL-Methionine | 2.6 | 2.5 | 2.2 | 3.0 | 1.9 | 1.9 | 1.7 | 2.3 |
| L-Lysine-HCl | 2.4 | 2.5 | 2.7 | 3.8 | 2.6 | 2.7 | 2.8 | 3.8 |
| L-Threonine | 1.0 | 0.9 | 0.9 | 1.7 | 0.5 | 0.5 | 0.4 | 1.2 |
| Mineral and vitamin premix ¹ | 10.0 | 10.0 | 10.0 | 10.0 | 10.0 | 10.0 | 10.0 | 10.0 |
| Total | 1,000 | 1,000 | 1,000 | 1,000 | 1,000 | 1,000 | 1,000 | 1,000 |
| Calculated composition | | | | | | | | |
| CP, % | 27.0 | 27.0 | 27.0 | 27.0 | 25.5 | 25.5 | 25.5 | 25.5 |
| ME, kcal/kg | 2,878 | 2,898 | 2,897 | 2,900 | 2,900 | 2,899 | 2,900 | 2,904 |
| Calcium, % | 1.23 | 1.21 | 1.21 | 1.21 | 1.05 | 1.05 | 1.05 | 1.05 |
| Available phosphorus, % | 0.60 | 0.60 | 0.60 | 0.60 | 0.52 | 0.52 | 0.52 | 0.52 |
| Methionine, % | 0.61 | 0.61 | 0.60 | 0.64 | 0.53 | 0.53 | 0.53 | 0.55 |
| Methionine + cystine, % | 1.05 | 1.05 | 1.05 | 1.05 | 0.95 | 0.95 | 0.95 | 0.95 |
| Lysine, % | 1.60 | 1.60 | 1.60 | 1.60 | 1.50 | 1.50 | 1.50 | 1.50 |
| Threonine, % | 1.06 | 1.06 | 1.06 | 1.06 | 0.95 | 0.95 | 0.95 | 0.95 |
| Sodium, % | 0.16 | 0.16 | 0.16 | 0.16 | 0.15 | 0.15 | 0.15 | 0.15 |
| Analyzed composition | | | | | | | | |
| CP, % | 26.9 | 27.0 | 27.0 | 27.1 | 25.4 | 25.5 | 26.0 | 25.6 |
| Crude fiber, % | 3.4 | 3.5 | 3.4 | 3.2 | 3.7 | 3.6 | 3.7 | 3.7 |
| NSP, ² % | 10.2 | 10.2 | 10.1 | 8.0 | 10.1 | 10.1 | 10.0 | 8.2 |
| Water-soluble NSP, % | 2.20 | 2.25 | 2.32 | 2.31 | 2.26 | 2.28 | 2.25 | 2.38 |
| Oligosaccharides, ³ % | 2.61 | 2.03 | 0.94 | 0.24 | 2.27 | 1.70 | 0.93 | 0.05 |

¹Provided the following per kilogram of diet in the starter (1 to 4 wk) and grower (5 to 8 wk) diets: vitamin A, 15,000 and 13,000 IU; vitamin E, 40 and 35 mg; respectively. For 1 to 8 wk of feeding, the mineral and vitamin premix supplied the following per kilogram of diet: Se, 0.3 mg; Mn, 150 mg; Zn, 90 mg; Fe, 60 mg; Cu, 15 mg; I, 1 mg; diclazuril (coccidiostat), 1 mg; vitamin D₃, 4,500 IU; vitamin K₃, 2.5 mg; thiamin, 3.5 mg; riboflavin, 10 mg; vitamin B₆, 6 mg; vitamin B₁₂, 0.03 mg; folic acid, 2 mg; biotin, 0.36 mg.

²Nonstarch polysaccharides.

³Includes raffinose, stachyose, and verbascose.

Zdunczyk (2002). The following substrates were used: *p*-nitrophenyl- β -D-glucopyranoside for β -glucosidase and *p*-nitrophenyl- β -D-glucuronide for β -glucuronidase. The reaction mixture contained 0.3 mL of substrate solution (5 mM) and 0.2 mL of a 1:10 (vol/vol) dilution of the cecal sample in 100 mM phosphate buffer (pH 7.0). Incubation was carried out at 39°C, and following the addition of 2.5 mL of 0.25 M cold sodium carbonate and centrifugation at $7,211 \times g$ for 15 min at 21°C, the absorbance was measured at 400 nm. Enzyme activity (IU) was expressed as micromoles of *p*-nitrophenol formed per minute and per gram of digesta.

Cecal digesta samples were subjected to SCFA analysis using gas-liquid chromatography (Shimadzu GC-14A, Shimadzu Co., Kyoto, Japan). The samples (0.2 g) were mixed with 0.2 mL of formic acid, diluted with deionized water, and centrifuged at $7,211 \times g$ for 5 min at 20°C. Supernatant was loaded onto the chromatography glass column (2.5 m \times 2.6 mm) packed with 10% SP-1200-1% H₃PO₄ on 80/100 Chromosorb W AW (Supelco Co., Bellefonte, PA). The chromatograph was coupled to a flame ionization detector. Column, injector, and detector temperatures were 110, 195, and 180°C, respectively.

Statistical Analysis

The results of the experiment were analyzed using a 1-way ANOVA test, and significant differences between groups were determined by Duncan's multiple range test. Differences were considered significant at $P < 0.05$ (Snedecor and Cochran, 1989). The Statistica software package version 6.0 (StatSoft Corp., Krakow, Poland) was used for statistical calculations.

RESULTS AND DISCUSSION

The contents of CP in SBM, SPC, and SPI were 470.1, 649.2, and 858.8 g/kg, respectively (Table 1). In comparison with SBM, which contained 19.8 g/kg of crude fat and 34.8 g/kg of CF, the content of these components in SPC was lower by about 30 and 20%, respectively. Overall, the contents of CP, crude fat, and CF were in agreement with some earlier reports of these product evaluations (Grieshop and Fahey, 2000).

The content of total and water-soluble NSP in both SBM and SPC was similar and about 10 times higher than that of SPI. The content of NSP in SBM and SPC corresponded well with the data of Huisman et al. (1998) and Meng et al. (2005). Nonstarch polysaccharide component sugar composition revealed the presence of large amounts of galactose, glucose, uronic acids, and arabinose, indicating that the main portion of soybean dietary fiber is composed of galactan, arabinogalactan, pectic, and cellulose polysaccharides as reported earlier (Huisman et al., 1998; Karr-Lilienthal et al., 2005). The SPI preparation was low in NSP and contained 10 times less NSP than SBM and SPC. When compared with the total NSP content, the amount of

water-soluble NSP was lower by 17, 15, and 23% in SBM, SPC, and SPI, respectively. Previous research has indicated that the content of water-soluble NSP in soybeans may vary considerably and range between 10 and 30% of the total NSP content (Huisman et al., 1998; Karr-Lilienthal et al., 2005).

The soybean products varied considerably in the amount of soluble sugars, including oligosaccharides. The sum of oligosaccharides averaged 53.0 g/kg for SBM and was much higher than the values of 24.6 and 3.7 g/kg, respectively, for SPC and SPI. The contents of oligosaccharides determined in the current study are in agreement with the data reported for the conventional SBM (Grieshop et al., 2003; van Kempen et al., 2006) and the SPC samples from different sources (Refstie et al., 1999; Peisker, 2001). Soy protein isolate contained small amounts of nonprotein ingredients, including soluble and structural carbohydrates, most of which were removed during the protein isolation process (Grieshop and Fahey, 2000). The differences in the carbohydrate fraction of soybean products determined the composition of dietary fiber and oligosaccharides in the experimental diets (Table 2). Diets containing SBM, SBM-SPC, and SPC were similar in total NSP and contained about 10% of this fiber component. The SPI diet contained less NSP (8.0 and 8.2% in the first and second 4-wk periods, respectively). It is of importance to note that the content of water-soluble NSP was similar for all diets and ranged between 2.2 and 2.4%. The content of oligosaccharides differed among the diets and averaged 2.4, 1.9, 0.9, and 0.1% for SBM, SBM-SPC, SPC, and SPI, respectively. In the present study, isonitrogenous diets balanced for methionine, lysine, and threonine, and with similar total and water-soluble NSP contents, were used. Therefore, the main dietary factor affecting the GIT function was most likely the dietary concentration of oligosaccharides. Varied application of SPC in experimental diets as a partial or almost total replacement of SBM enabled us to decrease the content of dietary oligosaccharides from 2.5% in the SBM diet to about 2 and 1% in the SBM-SPC and SPC diets, respectively (Table 2). Furthermore, dietary incorporation of SPI, which was associated with a complete withdrawal of SBM, resulted in decreased dietary oligosaccharide concentration to about 0.1% in the SPI diet.

In comparison to other treatments, feeding the SPI diets resulted in a significant decrease in BW after 4 wk of experiment (Table 3). During the next period (5 to 8 wk), the birds from the SBM-SPC and SPC diets gained significantly more than those fed diets containing SBM and SPI. In both experimental periods (1 to 4 and 5 to 8 wk), the lowest ($P < 0.05$) feed conversion ratio was observed for birds fed the SPI-containing diet. A partial or almost complete substitution of SBM with the SPC product resulted in an increased BW of turkeys at 8 wk of age. When compared with the control SBM birds, the SPI diet with very little oligosaccharides had no positive effect on BW (Table 3). However, feed con-

version ratio was significantly lower in the birds from the SPI group than in those of the other groups. This is in agreement with previous research indicating that soybean oligosaccharides may decrease energy utilization (Coon et al., 1990; van Kempen et al., 2006) and may have some negative effect on ileal DM digestibility in piglets (Wiggins, 1984).

After 4 wk of experiment, the highest mass of ileum was observed in turkeys fed the SBM diet (Table 4). For other groups, the ileal tissue mass was significantly decreased and proportional to the decreased level of dietary oligosaccharides. A similar decrease was observed for the 8-wk-old birds. In both experimental periods (i.e., after 4 and 8 wk), similar pH and viscosity values for ileal digesta were observed for all dietary treatments. The activities of mucosal sucrase and maltase were similar for all treatments during the entire study. Earlier research has indicated that the hydroscopic properties of SBM oligosaccharides may affect transit of digesta in the GIT (Bedford, 1995) by elevating intestinal osmolarity (Wiggins, 1984) as well as by increasing digesta viscosity (Smits and Annison, 1996). Our previous research has shown that oligosaccharides extracted from lupine seeds can reduce water absorption in the perfused small intestine of rats (Zdunczyk et al., 1998). In the present study, application of a diet containing very little oligosaccharides decreased the mass of ileum tissue but did not significantly influence the activity of mucosal glycolytic enzymes; however, a decrease (albeit $P > 0.05$) in sucrase and maltase activities was observed (Table 4). These results are in line with the fact that the mucosal disaccharidase activity when expressed per mass of mucosa (or intestine) is closely correlated with the number of enterocytes per villus in all regions of the poultry small intestine, and the mucosal enzyme activity would be highly correlated with the BW of the host (Sklan, 2001). Indeed, the lowest BW, both in 4- and 8-wk-old turkeys, was observed for the SPI diet, which contained only trace amounts of oligosaccharides. One could assume that a total withdrawal of oligosaccharides from a diet may result in undesirable hypotrophy of small intestinal tissue and associated decrease in mucosal disaccharidase activity, which, in turn, may decrease the growth rate. Contrary to previous research, the dietary treatments associated

with decreased oligosaccharide content showed a decreased concentration of DM in ileal digesta after 4 wk ($P = 0.067$) and 8 wk ($P = 0.064$) and increased ($P = 0.060$) ileal digesta viscosity at 4 wk. This result suggests that water-soluble NSP rather than soybean oligosaccharides are responsible for this slight increase in intestinal viscosity. It must be emphasized, however, that the viscosity values observed in the current study were relatively low and lower than the values of 3.0 to 5.0 mPa·s observed in broiler chickens fed wheat-based diets (Boros et al., 2004; Meng et al., 2004, 2005). Furthermore, such low intestinal viscosity values have been reported to have a minimal, if any, effect on growth performance of broilers (Liang and Liu, 1988).

The cecal environment was significantly affected by dietary treatments (Table 5). After 4 wk, a significantly decreased ($P < 0.05$) cecal tissue mass and increased amounts of cecal digesta and digesta DM contents were observed for birds in the SPC and SPI groups. In the next period, differences in the aforementioned indices of cecal metabolism, except for digesta DM content, were not statistically significant. At 4 wk of age, the highest activity of cecal β -glucuronidase was observed in birds of the SPC group and it was significantly higher than the activity observed in the birds in the SBM and SBM-SPC groups. The lowest activity of this bacterial enzyme was observed for the birds from the SBM-SPC treatment.

Carbohydrates undigested in the upper part of the GIT are known to be fermented in the large intestine of poultry by the cecal bacterial population (Patterson and Burkholder, 2003; Juskiewicz et al., 2006). When large amounts of fermentable oligosaccharides were fed, the increased ceca weights (especially tissue) were considered as a positive physiological response associated with some beneficial changes in the large intestine (e.g., acidification of digesta, decreased ammonia concentration, and increased SCFA contents; WHO, 1987).

Dietary oligosaccharides may stimulate the enzyme production by cecal bacterial flora, particularly the glycolytic ones, thus facilitating the hydrolysis of carbohydrate polymers (Monsan and Paul, 1995). The main end products of bacterial fermentation are SCFA, mainly acetate, propionate, and butyrate (Kleessen et al., 2001; Jamroz et al., 2002). The presence of readily

Table 3. The effect of soybean meal (SBM)-, soybean protein concentrate (SPC)-, and soybean protein isolate (SPI)-based diets of different oligosaccharide contents on growth performance of turkeys at 4 and 8 wk of age

| Item | Diet | | | | SEM | P-value |
|--|----------------------|--------------------|--------------------|--------------------|-------|---------|
| | SBM | SBM-SPC | SPC | SPI | | |
| BW, kg/bird | | | | | | |
| 4 wk of age | 1.074 ^{a,1} | 1.078 ^a | 1.075 ^a | 1.038 ^b | 0.022 | 0.001 |
| 8 wk of age | 4.324 ^b | 4.452 ^a | 4.459 ^a | 4.282 ^b | 0.068 | 0.001 |
| Feed conversion ratio, g of feed:g of gain | | | | | | |
| 0 to 4 wk | 1.416 ^b | 1.469 ^b | 1.416 ^b | 1.368 ^a | 0.009 | 0.006 |
| 0 to 8 wk | 1.760 ^b | 1.787 ^b | 1.742 ^b | 1.667 ^a | 0.010 | 0.004 |

^{a,b}Means within a row with no common superscript differ significantly ($P < 0.05$).

¹Values are means of 7 observations per treatment.

Table 4. The effect of soybean meal (SBM)-, soybean protein concentrate (SPC)-, and soybean protein isolate (SPI)-based diets of different oligosaccharide contents on ileum weight, digesta viscosity, pH values, and mucosal enzyme activities in turkeys at 4 and 8 wk of age

| Item | Diet | | | | SEM | P-value |
|---------------------------------|---------------------|-------------------|-------------------|-------------------|------|---------|
| | SBM | SBM-SPC | SPC | SPI | | |
| 4 wk of age | | | | | | |
| Ileum tissue weight, g/kg of BW | 72.5 ^{a,1} | 66.2 ^b | 52.2 ^c | 50.2 ^d | 1.85 | 0.001 |
| DM of ileal digesta, % | 18.6 | 16.5 | 14.4 | 15.7 | 0.40 | 0.067 |
| pH of ileal digesta | 6.9 | 7.1 | 7.2 | 7.3 | 0.08 | 0.418 |
| Viscosity, mPa·s | 1.8 | 2.0 | 2.1 | 2.2 | 0.07 | 0.181 |
| Sucrase activity, U/g | 14.1 | 13.8 | 12.3 | 12.2 | 0.70 | 0.697 |
| Maltase activity, U/g | 45.0 | 40.8 | 40.3 | 37.6 | 1.08 | 0.112 |
| 8 wk of age | | | | | | |
| Ileum tissue weight, g/kg of BW | 40.5 ^a | 40.2 ^a | 37.2 ^b | 32.2 ^c | 0.67 | 0.026 |
| DM of ileal digesta, % | 17.8 | 16.3 | 15.1 | 15.9 | 0.40 | 0.064 |
| pH of ileal digesta | 7.1 | 6.4 | 7.1 | 6.8 | 0.10 | 0.085 |
| Viscosity, mPa·s | 2.0 | 2.2 | 2.5 | 2.5 | 0.13 | 0.060 |
| Sucrase activity, U/g | 13.9 | 12.3 | 11.7 | 11.6 | 0.57 | 0.471 |
| Maltase activity, U/g | 39.4 | 38.7 | 36.6 | 37.4 | 0.95 | 0.754 |

^{a-d}Means within a row with no common superscript differ significantly ($P < 0.05$).

¹Values are means of 10 observations per treatment.

fermentable oligosaccharides in a diet, including galactooligosaccharides, may increase accumulation of cecal digesta composed of undigested constituents and increase bacterial mass (Józefiak et al., 2004). Our results indicate that the experimental diets with different oligosaccharide concentrations significantly affected ceca weights in the 4-wk-old but not in the 8-wk-old birds (Table 5). The SBM and SBM-SPC diets containing, respectively, 2.5 and 2% of oligosaccharides increased cecal mass and decreased digesta mass in the 4-wk-old birds.

Irrespectively of the experimental period, the highest concentrations of total SCFA, in particular acetic and butyric, were observed in the SBM group fed 2.5% of dietary oligosaccharides (Table 6). The lowest concentrations of cecal SCFA, especially acetic and butyric acids, were observed for the diets containing SPC and SPI. The total production of SCFA in the ceca of

8-wk-old turkeys fed the SBM diet was significantly higher ($P < 0.05$) than that of the other groups. There were no differences in the profile of SCFA among the treatments. These results may explain differences in cecal tissue and digesta weights among the treatments. Butyric acid appears to be essential in maintaining a healthy ceca and colon; it is the preferred energy source for the mucosa cells and has been suggested to be beneficial for gut cell proliferation (Józefiak et al., 2004). On the other hand, increased bacterial fermentation of oligosaccharides may enhance digesta passage rate, thereby leading to a faster emptying of the ceca (Coon et al., 1990).

In the presence of different fermentable carbohydrates, the SCFA pool size rather than the individual fatty acid concentration has been postulated to be the best indicator of the intensity of the large intestine fermentation (Campbell et al., 1997). As far as the cecal

Table 5. The effect of soybean meal (SBM)-, soybean protein concentrate (SPC)-, and soybean protein isolate (SPI)-based diets of different oligosaccharide contents on ceca weight, digesta viscosity, pH values, and bacterial enzyme activities in turkeys at 4 and 8 wk of age

| Item | Diet | | | | SEM | P-value |
|--------------------------------------|----------------------|--------------------|--------------------|--------------------|-------|---------|
| | SBM | SBM-SPC | SPC | SPI | | |
| 4 wk of age | | | | | | |
| Ceca tissue weight, g/kg of BW | 7.88 ^{ab,1} | 8.20 ^a | 6.81 ^{bc} | 6.73 ^c | 0.217 | 0.031 |
| Digesta weight, g/kg of BW | 1.91 ^c | 2.19 ^{bc} | 2.94 ^a | 2.83 ^{ab} | 0.138 | 0.015 |
| pH of cecal digesta | 6.0 | 6.0 | 6.2 | 5.7 | 0.09 | 0.354 |
| DM of cecal digesta, % | 14.7 ^c | 14.6 ^c | 19.4 ^a | 18.8 ^b | 0.48 | 0.044 |
| β -Glucosidase activity, U/g | 0.20 | 0.14 | 0.27 | 0.28 | 0.022 | 0.079 |
| β -Glucuronidase activity, U/g | 0.34 ^{bc} | 0.29 ^c | 0.50 ^a | 0.45 ^{ab} | 0.028 | 0.024 |
| 8 wk of age | | | | | | |
| Ceca tissue weight, g/kg of BW | 6.16 | 6.17 | 5.95 | 5.21 | 0.116 | 0.416 |
| Digesta weight, g/kg of BW | 2.98 | 2.71 | 2.48 | 2.52 | 0.120 | 0.462 |
| pH of cecal digesta | 5.9 | 6.1 | 6.0 | 6.0 | 0.11 | 0.934 |
| DM of cecal digesta, % | 15.8 ^b | 17.9 ^b | 17.3 ^b | 21.6 ^a | 0.58 | 0.036 |
| β -Glucosidase activity, U/g | 0.47 | 0.43 | 0.43 | 0.42 | 0.053 | 0.987 |
| β -Glucuronidase activity, U/g | 0.98 | 0.69 | 0.65 | 0.60 | 0.080 | 0.355 |

^{a-c}Means within a row with no common superscript differ significantly ($P < 0.05$).

¹Values are means of 10 observations per treatment.

Table 6. The effect of soybean meal (SBM)-, soybean protein concentrate (SPC)-, and soybean protein isolate (SPI)-based diets of different oligosaccharide contents on short-chain fatty acids (SCFA) content in the ceca of turkeys at 4 and 8 wk of age

| Item | Diet | | | | SEM | P-value |
|--|----------------------|--------------------|--------------------|--------------------|-------|---------|
| | SBM | SBM-SPC | SPC | SPI | | |
| 4 wk of age | | | | | | |
| SCFA, $\mu\text{mol/g}$ of fresh digesta | | | | | | |
| Total SCFA | 113.2 ^{a,1} | 97.3 ^{ab} | 78.3 ^c | 75.2 ^c | 3.96 | 0.001 |
| Acetate | 86.5 ^a | 72.0 ^b | 58.0 ^c | 59.6 ^c | 3.03 | 0.001 |
| Propionate | 4.5 | 4.7 | 4.4 | 3.6 | 0.28 | 0.616 |
| Isobutyrate | 0.4 | 0.3 | 0.4 | 0.2 | 0.051 | 0.689 |
| Butyrate | 20.8 ^a | 19.9 ^a | 14.8 ^b | 11.2 ^b | 1.18 | 0.007 |
| Isovalerate | 0.5 | 0.5 | 0.4 | 0.2 | 0.09 | 0.448 |
| Valerate | 0.4 | 0.2 | 0.4 | 0.4 | 0.05 | 0.234 |
| SCFA pool, $\mu\text{mol/kg}$ of BW | 208.6 | 208.2 | 232.1 | 204.5 | 8.42 | 0.654 |
| 8 wk of age | | | | | | |
| SCFA, $\mu\text{mol/g}$ of fresh digesta | | | | | | |
| Total SCFA | 130.0 ^a | 103.3 ^b | 89.3 ^b | 86.4 ^b | 4.60 | 0.001 |
| Acetate | 85.7 ^a | 68.5 ^b | 63.8 ^b | 58.7 ^b | 2.89 | 0.001 |
| Propionate | 22.9 | 18.6 | 12.4 | 13.7 | 1.69 | 0.099 |
| Isobutyrate | 0.5 | 0.4 | 0.3 | 0.6 | 0.09 | 0.714 |
| Butyrate | 17.2 | 13.7 | 11.2 | 10.2 | 1.05 | 0.080 |
| Isovalerate | 0.9 | 0.5 | 0.3 | 0.6 | 0.17 | 0.677 |
| Valerate | 2.7 | 1.6 | 1.4 | 2.7 | 0.30 | 0.244 |
| SCFA pool, $\mu\text{mol/kg}$ of BW | 380.6 ^a | 255.5 ^b | 218.0 ^b | 214.7 ^b | 15.02 | 0.001 |

^{a-c}Means within a row with no common superscript differ significantly ($P < 0.05$).

¹Values are means of 10 observations per treatment.

SCFA pool size is concerned, the results of the present study support this hypothesis. In the 8-wk-old turkeys, the SCFA pool in the birds fed the SBM diet was almost 2-fold higher than that of the birds fed the SPC and SPI diets. Despite different cecal concentrations of SCFA, there were no significant differences in pH of cecal digesta among the treatments. A probable explanation for this would be a decreased concentration of DM in groups with an increased content of galactooligosaccharides and enhanced buffering capacity of watery digesta.

In conclusion, partial or almost complete substitution of SBM with SPC in turkey poult diets, which was associated with the decreased dietary oligosaccharide content, suppressed the fermentation processes in the ceca but enhanced BW of 8-wk-old turkeys. The data showed that the concentration and the pool of SCFA in the ceca were positively correlated with the content of dietary oligosaccharides. Almost complete removal of oligosaccharides from the diet, which was accomplished by substitution of SBM with SPI, was associated with improved mean feed utilization but decreased mean BW in the 4-wk-old turkeys and had no effect on the overall growth rate of turkeys up to 8 wk of age.

ACKNOWLEDGMENTS

This study was supported by the Polish Committee for Scientific Research (grant no. 311 058 31).

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