The effect of butyric acid salts on feed palatability, growth performance and gut health in weanling piglets

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The aim of this study was to investigate the effect of butyric acid salts on growth performance, blood hematological and biochemical parameters, *Escherichia coli* counts in the duodenum, intestinal morphometry, histology of selected organs and feed palatability in weaned piglets.

Two experiments involving a total of 72 piglets (Polish Large White x Duroc) weaned at 28 days of age were conducted during the study. In both experiments, piglets received a basal diet (control group - C) or diets supplemented with 4000 mg/kg of sodium butyrate (C+SB), 2000 mg/kg of coated sodium butyrate (C+SBE), and 2000 mg/kg of a blend of coated calcium butyrate and sodium butyrate (C+CBSBC) (*ad libitum* access to feed and water).

In the first 8-d experiment, 32 piglets (homogenous in terms of weight and sex) were divided into 4 treatment groups (n=8), with 4 replicates per group and 2 piglets per replicate, to determine the effect of sodium butyrate on feed palatability. The animals received one of the four experimental diets according to the 4x4 Latin Square design. The diets were fed to piglets 4 times a day, at 8.00 a.m., 11.00 a.m., 2.00 p.m. and 5.00 p.m., by the single stimulus method, with free access to one of the diets for 20 minutes during each feeding session (*ad libitum* access to water). Feed intake was monitored daily. Piglets were weighed on days 1 and 8.

The four diets used in the first experiment were also administered to piglets in the second 28-d experiment (*ad libitum* access to feed and water); 40 piglets (homogenous in terms of weight and sex) were weighed and assigned to 4 treatment groups (n=10), with 5 replicates per group and 2 piglets per replicate. The piglets were weighed individually at weekly intervals. The amounts of feed offered and consumed per pen were recorded daily. Blood samples were collected for hematological (WBC, RBC, HGB, HCT, MCV, MCH, MCHC, PLT, NEUT, LYM, MONO, EOS, BASO, IG) and biochemical (TC, HDL, LDL, TG, GLU) analyses on the last day of the experiment (from 6 animals in each group). Serum zonulin concentration (a marker of intestinal permeability) was analyzed using the enzyme-linked immunosorbent assay (ELISA) kit. At 57 days of age, the piglets (6 animals from each

group) were slaughtered. Duodenal digesta samples were collected for analysis, and the most-probable-number of *Escherichia coli* was determined. Tissue samples of the intestines (duodenum, jejunum, ileum, colon), stomachs, livers, pancreases and kidneys were collected for analysis. Intestinal morphometry and the histology of selected organs were evaluated. The results were processed statistically by one-way analysis of variance (ANOVA) with the use of Statistica StatSoft 13.3. software and Tukey's post hoc test. All data are presented as means ± SE. Differences were considered to be significant at $P \leq 0.05$ and $P \leq 0.01$.

The analyzed additives had no effect on feed palatability. Sodium butyrate in the amount of 4000 mg/kg of feed significantly influenced the final body weight of piglets ($P \leq 0.05$). The highest total body weight gains were observed in all piglets receiving diets supplemented with sodium butyrate (C+SB) and a blend of coated calcium butyrate and sodium butyrate (C+CBSBC), with no effect on average daily feed intake. The addition of 4000 mg/kg of sodium butyrate had a more beneficial influence ($P \leq 0.05$) on total body weight gains than the addition of 2000 mg/kg of coated sodium butyrate. Differences ($P \leq 0.05$) in the serum levels of total cholesterol, HDL cholesterol and triglycerides were noted in two groups. The addition of 2000 mg/kg of coated sodium butyrate decreased total cholesterol concentration. Moreover, a blend of coated calcium butyrate and sodium butyrate as well as coated sodium butyrate, both at 2000 mg/kg, decreased the serum levels of HDL cholesterol and triglycerides. The applied additives led to an increase in glucose levels above the normal reference range. The addition of butyric acid salts had no significant effect on the blood hematological parameters of piglets, but some of their average values were beyond the normal physiological ranges for the species. Sodium butyrate increased the white blood cell count (WBC) and the percentage of monocytes (MONO). The inclusion of sodium butyrate and a blend of coated calcium butyrate and sodium butyrate in piglet diets decreased mean corpuscular hemoglobin (MCH) levels. In addition, coated sodium butyrate and a blend of coated calcium butyrate and sodium butyrate caused thrombocythemia. Coated sodium butyrate contributed to increasing the thickness of the smooth muscle layer in the stomach ($P \leq 0.01$). Dietary supplementation with sodium butyrate and coated sodium butyrate led to an increase ($P \leq 0.05$) in the thickness of the muscle layer in the ileum. Moreover, the applied feed additives significantly ($P \leq 0.01$ and $P \leq 0.05$) increased the length of duodenal villi, with no effect on crypt depth. A significant ($P \leq 0.05$) increase in the length of intestinal villi was also observed in the jejunum in response to sodium butyrate. Butyric acid derivatives added to piglet diets alleviated morphological changes in the stomach and duodenum. All additives caused an increase in the number of intraepithelial lymphocytes (IEL) in the duodenum and

jejunum, which indicates that they stimulated the immune response. Normal histology of the liver and kidneys was observed after the administration of sodium butyrate and coated sodium butyrate. The tested additives did not affect *Escherichia coli* counts in the duodenum of experimental animals. Blood zonulin concentration decreased in experimental groups, but the decrease was not statistically significant.

Key words: sodium butyrate, calcium butyrate, weanling piglets, feed palatability, blood parameters, intestinal morphometry, organ histology, *Escherichia coli*, serum zonulin