Poultry Science Journal
ISSN: 2345-6604 (Print), 2345-6566 (Online) http://psj.gau.ac.ir
DOI: 10.22069/PSJ.2016.11164.1191

Effects of Lactobacillus-Based Probiotic on Performance, Gut Microflora, Hematology and Intestinal Morphology in Young Broiler Chickens Challenged with *Salmonella* Typhimurium

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Poultry Science Journal 2016, 4(2): 157-165

Keywords
Broiler
Heterophil
Lactobacillus
Villus surface
Salmonella Typhimurium

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Article history
Received: May 16, 2016
Revised: June 24, 2016
Accepted: October 3, 2016

Abstract

This experiment was conducted to compare the effects of multi-strain probiotic at various inclusion levels on performance, intestinal morphology, gut microflora and hematology in broiler chickens infected with *Salmonella* Typhimurium (ST). A total of 120 1-day old Ross 308 broiler chickens were distributed into 20 floor pens and reared for 10 days under 5 experimental treatments including a corn-soy basal diet with no probiotic (Control), or 0.5 g (0.05%), 1 g (0.1%), 1.5 g (0.15%) and 2 g (0.2%) probiotic/kg diet. Chickens were infected orally with ST at the second day of the experiment. Broilers under 0.15% of probiotic had higher body weight gain compared to other treatments. Probiotic supplementation, except at 0.05%, significantly improved feed conversion ratio. The use of 0.1 and 0.15% probiotic reduced the population of Salmonella in the ileum. The lowest heterophil: lymphocyte ratio was observed in 0.15% of probiotic, although 0.1 and 0.2% of probiotic significantly reduced this ratio compared to control group as well. Adding 0.15% of probiotic to the basal diet increased ($P < 0.05$) the ileal villus height to crypt depth ratio as well as villus height in the ileum, jejunum and duodenum. According to the present results, especially for body weight gain and salmonella counts in the ileum, adding 0.15% of probiotic in broiler chickens diet can be used for effective control of ST infection.

Introduction

Probiotics reduce the population of pathogenic bacteria and consequently beneficially change the microflora of the gastrointestinal tract (Cao *et al*., 2013). At the time of introduction of a new probiotic, probiotic bacteria may be evaluated *in vitro* and/or *in vivo*, individually and/or in groups against pathogenic bacteria such as Salmonella, *E. coli* and *Clostridium perfringens*

Please cite this article as: Ashayerizadeh O, Dastar B, Samadi F, Khomeiri M, Yamchi A4 & Zerehdaran S. 2016. Effects of Lactobacillus-Based Probiotic on Performance, Gut Microflora, Hematology and Intestinal Morphology in Young Broiler Chickens Challenged with *Salmonella* Typhimurium. Poult. Sci. J. 4(2): 157-165

(Loh *et al*., 2010).

However, as reported by Gonzalez-Gil *et al*. (2014), results of *in vitro* or individual experiments could vary due to the absence of effects of diet, host, and interactions between probiotic bacteria and between probiotics, pathogens, diet, and hosts (Torok *et al*., 2008). Some studies were performed to evaluate the competitive exclusion potential of probiotic strains in *in vivo* experiments against pathogenic bacteria. Intestinal mucosa is not only the place of nutrient digestion and absorption but also acts as a natural defense against pathogens in the gut. *Salmonella enterica* serotype Typhimurium could invade and deploy in the digestive tract, cause inflammation, and also cause the presence of bacteria in blood by destructing immune system activities. In addition, Salmonella can reduce efficiency and increase mortality in the poultry industry and contaminate poultry products for human consumption (Shao *et al*., 2013). *Salmonella* Typhimurium (ST) is resistant to antimicrobial proteins that are secreted by the host as part of the nutritional immune response (Deriu *et al*., 2013).

Lymphoid tissue of broiler chicken's gastrointestinal tract is relatively disorganized and cecum lymph tissue and peyer's patches in the small intestine do not develop until two weeks of age. So, Salmonella would be able to penetrate and enter intestinal epithelium cells and macrophages and then use them as a means to transfer and/or proliferate in follicles of organs such as bursa of fabricius, liver and spleen (Henderson *et al*., 1999). On the other hand, 24-72-hour food deprivation – which will occur due the delay between hatch and access to feed and water in farms – negatively affects yolk usage, gut development, growth performance, and slaughter weight. Ultimately, this would increase the bird's susceptibility to pathogenic bacteria infection (e.g. salmonella) after depressing the development of the immune system (Biloni *et al*., 2013). Lactobacillus-based probiotic could significantly reduce the population of Salmonella in the intestine of broiler chickens (Higgins *et al*., 2007; Menconi *et al*., 2011). Our study was carried out *in vivo* to determine the inhibitory effects of different dosage of Lactobacillus-based probiotic on intestinal ST colonization and also on growth, intestinal morphology, white blood cells count, and digestive enzyme activity of young broiler chickens after 24 hrs feed restrictions at the beginning of the experiment.

Materials and Methods

All of procedures, animal ethics and welfare were performed according to ethic committee of Gorgan University of Agricultural Sciences and Natural Resources, Gorgan, Iran

Salmonella Typhimurium

A primary poultry isolates of ST (ATCC 14028) was obtained from the Iranian Research Organization for Science and Technology (IROST). To prepare the inocula, bacteria were grown in a nutrient broth (Quelab, Montreal, Canada) at 37°C for 24 hrs. The numbers of colony-forming units (CFU) were determined by counting serial 10-fold dilutions of the bacterial suspension, on brilliant-green phenol-red lactose sucrose (BPLS; Merck, Germany) agar plates (Ribeiro *et al*., 2007). Subsequent, bacterial suspensions were diluted to the necessary concentrations for oral inoculation in broiler chickens.

Chickens and dietary treatments

120 one-day-old male Ross 308 broiler chickens were distributed equally among five treatments with four replicates in each treatment and then reared for ten days. A completely randomized design was used. A coccidiostat-free basal diet was formulated according to the recommendation from Ross Broiler Nutrition Specification (2009) for days one to ten of the experiment. The compositions of the diets are shown in Table 1. Five tested diets were formulated by supplementing the basal diet (per kg) without probiotic (control), or with 0.5 g (0.05%), 1 g (0.1%), 1.5 g (0.15%) and 2 g (0.2%) probiotic containing (about 65×10^8 CFU/g) *Lactobacillus plantarum*, *Lactobacillus fermentum* and *Lactobacillus rhamnosus*. All birds received feed and water *ad libitum*. Before ST inoculation, birds from each experimental group were tested to be negative for Salmonella contamination. On day two and eight post-hatch, all the birds were orally administered with 0.25 mL (5×10^5 CFU/mL) of ST (ATCC 14028).

Growth performance evaluation

Birds were weighed individually after their arrival at the experimental farm from the hatchery (initial weight) and also on day 10. Feed intake was recorded during the

experimental period for each treatment and the feed conversion ratio was calculated.

Microbiological analysis

On day 10, one bird from every cage was euthanized by cervical dislocation. About 5 cm of the ileum (from the Meckel's diverticulum to the cecal junction) and the crop contents and mucosa were sampled. To determine the microbial population, one gram of crop and ileum content was used to make serial 10-fold dilutions using buffered peptone water. In crop, lactic acid bacteria and Lactobacillus were quantified on de Man Rogosa and Sharpe (MRS; Merck, Germany) agar and Rogosa agar (Quelab, Montreal, Canada), respectively (Engberg *et al*., 2000). In ileum, lactic acid bacteria, Salmonella, and coliforms (Ribeiro *et al*., 2007; Morey *et al*., 2012) were quantified on MRS agar (Merck, Germany), BPLS agar, and violet red bile (VRB; Merck, Germany) agar, respectively. All plates were incubated in an anaerobic cabinet at 37° C for 24 hrs.

pH measurement

To measure pH, 1 g of crop and ileum content from each chicken was collected and transferred into 2 mL distilled water. pH was measured using a pH meter (Izat *et al*., 1990).

Table 1. Ingredients and chemical composition of basal diet (%)

Corn	55.89
Soybean meal	37.17
Vegetable oil	2.21
Dicalcium phosphate	1.79
Oyster shell	1.3
Vitamin mixture [1]	0.25
Mineral mixture [2]	0.25
Salt	0.5
L-Lysine	0.29
DL-Methionine	0.35
Calculated composition	
ME (Kcal/Kg)	2900
Crude protein	21.1
Calcium	1.0
Available phosphorus	0.48
Lysine	1.37
Methionine	0.68
Methionine+Cystine	1.02

[1] Contained per kilogram of diet: vitamin A (trans - retinyl acetate), 10,000 IU; vitamin D_3 (cholecalciferol), 2,000 IU; vitamin E (DL-α- tocopherol acetate), 10 mg; vitamin K (bisulfate menadione complex), 1 mg; vitamin B_1 (thiamin mononitrate), 1 mg; vitamin B_2 (riboflavin), 5 mg; vitamine B_3 (Niacin), 30 mg; vitamine B_6 (pyridoxine-hydrochloride), 1.5 mg; vitamine B_8 (biotin), 0.05 mg; vitamin B_5 (D - calcium pantothenate), 10 mg; vitamine B_9 (folic acid), 1 mg; and antioxidant (butylated hydroxytoluene), 10 mg.

[2] Contained per kilogram of diet: Mn (manganese sulfate), 60 mg; Zn (zinc sulfate), 50 mg; Fe (ferrous sulfate), 30 mg; Cu (copper sulfate), 4 mg; I (potassium iodide), 3 mg; Se (sodium selenite), 0.1 mg; and Co (cobalt carbonate), 0.1 mg.

Hematological measurements

To obtain leukocyte count, eight birds from each treatment were carried to a separate room, and their blood (0.3 mL) was collected immediately (*via* wing vein) in tubes containing EDTA as an anticoagulant. After collection, one blood drop was smeared on a glass slide and stained using May-Grunwald and Giemsa stains (Lucas and Jamroz, 1961) approximately 2 to 4 hrs after fixation with methanol. Then, 100 leukocytes (i.e. heterophils, eosinophils, basophils, lymphocytes and monocytes) were counted on one slide from each broiler chicken to calculate the heterophil to lymphocyte (H: L) ratio (Campo and Davila, 2002).

Tissue sampling

At the end of the feeding period, another eight birds from each treatment were randomly selected, and euthanized by cervical dislocation. The whole intestinal tract was removed, and segments of approximately 3 cm were taken

from the midpoint of the duodenum, jejunum (between the bile duct entry and Meckel's diverticulum) and ileum (between the Meckel's diverticulum and cecum). Segments were fixed in 10% neutral buffered formalin solution and embedded in paraffin wax. All histological morphometric studies were performed on 5 µm sections, stained with haematoxylin and eosin, and examined by a light microscope (Zentek *et al*., 2002). The slides were examined with an Olympus AX70 microscope (Olympus Corporation, Tokyo, Japan) fitted with a digital video camera (Nikon Eclipse TS100, Japan). The images were analyzed using Image J analysis software V 1.32j (ImageJ, National Institute of Mental Health, Bethesda, MD, USA) according to Abràmoff *et al*. (2004).

Histomorphological measurements

The unscathed crypt-villus units were selected in triplicate for each intestinal sample, according to the presence of intact lamina propria. Villus height was measured from the tip of the villus to the villus-crypt junction; crypt depth was defined as the depth of the fold between two interval villus-crypt junctions, and villus height to crypt depth ratio was calculated. Villus surface area was calculated as: (2π)(Villus width/2)(Villus length) (Solis de los Santos *et al*., 2005).

Statistical analysis

Data collected were subjected to one-way ANOVA using General Linear Model (GLM) procedure of SAS (SAS Institute, 2003). Significant differences among treatments were identified at 5% level by Duncan's multiple range tests.

Results

Growth performance

The effects of probiotic treatments on the performance of broiler chickens are given in Table 2. Chickens fed a diet containing 0.15% probiotic exhibited significantly higher body weight gain than those fed a non-supplemented control diet. All levels of probiotic supplementation, except 0.05%, improved ($P < 0.05$) feed conversion ratio when compared to the control group. Feed intake was not affected by treatments.

Table 2. Effects of dietary treatments on growth performance of broiler chicks infected with ST[1] from day 0-10.

Treatment	Body weight gain (g)	Feed intake (g)	Feed conversion ratio (g/g)
Control	152.91^{b}	266.67	1.75^{b}
0.05% probiotic	155.37^{ab}	257.71	1.66^{ab}
0.1% probiotic	162.58^{ab}	257.09	1.58^{a}
0.15% probiotic	170.62^{a}	269.58	1.58^{a}
0.2 %probiotic	164.37^{ab}	268.54	1.63^{a}
SEM	5.02	7.96	0.03
P-value	0.048	0.681	0.024

^{a,b} means in each column with different superscripts are significantly different ($P < 0.05$).

[1] ST = *Salmonella* Typhimurium

Microbiological findings

The composition of earlier GI microflora of broilers is shown in Table 3. Lactic acid bacteria and lactobacillus populations in the crop of broilers fed 0.15% probiotic-incorporated feed were significantly higher ($P < 0.05$) than the control. Also, lactobacillus count in the crop of birds fed 0.2% probiotic was greater ($P < 0.05$) than in control. The ratio of lactic acid bacteria in ileum was positively influenced ($P < 0.05$) by the administration of 0.05, 0.1 and 0.15% probiotic in chicken diets compared to the control and 0.2% probiotic treatments. Supplementation of 0.1 and 0.15% probiotic to the basal diet decreased the Salmonella present in the ileum when compared to control birds. Coliform bacteria in the ileum of birds fed 0.15% probiotic treatment had a significant reduction ($P < 0.05$) than birds in the control group.

pH measurements

The effects of experimental treatments on the pH of the crop and ileum of broiler chickens are given in Table 3. The pH of ileum was not affected ($P > 0.05$) by experimental treatments. The crop pH was significantly reduced ($P < 0.05$) in birds fed 0.2% probiotic compared to the control.

Table 3. Effects of dietary treatments on gut microflora (log_{10} CFU/g) and pH of broiler chicks infected with ST[1]

	Treatments					*SEM*	*P*-value
	Control	0.05% probiotic	0.1% probiotic	0.15% probiotic	0.2% probiotic		
Crop							
Lactic acid bacteria	6.92^{b}	7.39^{ab}	7.38^{ab}	7.62^{a}	7.57^{ab}	0.20	0.044
lactobacillus	6.84^{b}	7.33^{ab}	7.32^{ab}	7.57^{a}	7.52^{a}	0.19	0.036
pH	5.06^{a}	4.95^{ab}	4.69^{ab}	4.68^{ab}	4.59^{b}	0.10	0.035
Ileum							
Lactic acid bacteria	6.48^{c}	7.26^{b}	7.52^{ab}	7.92^{a}	6.55^{c}	0.19	0.004
Salmonella	3.92^{a}	3.87^{ab}	3.18^{b}	3.16^{b}	3.58^{ab}	0.21	0.010
Coliforms	5.72^{a}	5.68^{a}	4.69^{ab}	4.47^{b}	4.64^{ab}	0.34	0.027
pH	6.44	6.43	6.35	5.93	6.34	0.14	0.274

^{a,b,c} means in each row with different superscripts are significantly different ($P < 0.05$).
[1] ST = *Salmonella* Typhimurium

Hematological results

The heterophil, lymphocyte and H: L ratio in 0.1, 0.15 and 0.2% probiotic were significantly lower ($P < 0.05$) than the control group (Table 4). Furthermore, birds in 0.15% probiotic had a significantly lower percent of heterophil and H: L ratio in blood than 0.05% probiotic group. The use of probiotic led to the reduction of monocyte compared to control ($P < 0.05$). The percent of eosinophil and basophil were not affected by treatments ($P > 0.05$).

Table 4. Effects of dietary treatments on hematology (%) of 10 day old broiler chicks infected with ST[1]

	Treatments					*SEM*	*P*-value
	Control	0.05% probiotic	0.1% probiotic	0.15% probiotic	0.2% probiotic		
Heterophil	34.50^{a}	32.37^{ab}	29.25^{c}	28.62^{c}	31^{bc}	1.46	0.001
Lymphocyte	52.50^{c}	54.75^{bc}	58.37^{ab}	60.12^{a}	57.87^{ab}	1.85	0.010
Monocyte	8.25^{a}	6.87^{b}	7.00^{b}	6.62^{b}	6.75^{b}	0.57	0.049
Eosinophil	2.25	3.75	3.37	3.75	2.62	0.85	0.300
Basophil	2.50	2.25	2.00	0.87	1.75	0.80	0.321
H: L ratio	0.66^{a}	0.59^{ab}	0.50^{bc}	0.47^{c}	0.53^{bc}	0.04	0.001

^{a,b,c} means in each row with different superscripts are significantly different ($P < 0.05$).
[1] ST = *Salmonella* Typhimurium.

Intestinal morphology

The results of histomorphological measurements of the small intestine are presented in Table 5. The use of 0.15 and 0.2% probiotic had beneficial effects on duodenal and ileal villus height ($P < 0.05$). In addition, birds fed 0.15% probiotic had longer jejunal villus height than those control treatments ($P < 0.05$). The levels of 0.1, 0.15 and 0.2% probiotic in the duodenum, 0.1 and 0.15% in jejunum significantly reduced ($P < 0.05$) Villus width compared to control group. The jejunal crypt depth was significantly decreased for 0.2% probiotic supplementation compared to control ($P < 0.05$). The ratio of villus height: crypt depth in ileum of birds receiving 0.15% probiotic in the diet was improved ($P < 0.05$) when compared to the control treatment. Also, this ratio in duodenum of birds under 0.2% probiotic was higher ($P < 0.05$) than control group.

Discussion

In broiler chickens, the first 12 days of life are important in digestive tract development and changes during this time could affect the early and final body weight. Though it has been reported that ST in the intestines of 3-d old or older broiler chickens were not associated with the disease and had no effect on growth performance (Knap *et al*., 2011; Withanage *et al*., 2004), adding probiotics to diets, especially 15% probiotic, could nonetheless be effective in improving growth. In contrast, Ribeiro *et al.* (2007), reported that the age of birds, strain and dosage of inoculated Salmonella are the major factors that affect the severity of Salmonella infection and mortality of broilers. *Salmonella* Typhimurium act through the destruction and inflammation of the intestinal villi, which will lead to heterophil accumulation in the affected

areas (Withanage *et al*., 2004; Henderson *et al*., 1999). In the present study, feed conversion ratio of control birds seems to have been affected by Salmonella invasion of intestinal tract cells. In the study of Biloni *et al*. (2013), which was similar to our experimental procedure, the use of probiotics in ST-infected broiler chickens insignificantly increased body weight gain on 7 and 14 days of age. On the other hand, increased levels of prebiotics in the diet are not necessarily associated with improved bird performance (Wang and Gu, 2010; Apata, 2008; Bai *et al*., 2013). Huang *et al*. (2004) showed that there is an optimal concentration of Lactobacillus-based probiotics associated with the bacteria strain and higher dosages do not always lead to better performance.

Table 5. Effects of dietary treatments on intestinal histology of 10 day old broiler chicks infected with ST[1]

	Treatments					*SEM*	*P*-value
	Control	0.05% probiotic	0.1% probiotic	0.15% probiotic	0.2 % probiotic		
Villus height (μm)							
Duodenum	764.6^{b}	839.8^{b}	957.2^{ab}	1168.7^{a}	1142.5^{a}	42.78	0.003
Jejunum	584.8^{b}	634.3^{b}	877.0^{ab}	690.6^{a}	793.9^{ab}	47.75	0.050
Ileum	548.0^{b}	586.6^{ab}	666.0^{ab}	770.8^{a}	779.8^{a}	30.29	0.039
Villus width (μm)							
Duodenum	179.87^{a}	156.25^{ab}	136.47^{b}	140.19^{b}	136.23^{b}	5.53	0.038
Jejunum	168.20^{a}	163.86^{a}	125.86^{b}	124.08^{b}	139.38^{ab}	5.06	0.004
Ileum	170.36	148.77	140.49	137.64	139.19	6.48	0.491
Crypt depth (μm)							
Duodenum	135.71	131.76	122.81	119.94	122.44	4.38	0. 772
Jejunum	160.48^{a}	150.25^{ab}	141.30^{ab}	136.80^{ab}	120.20^{b}	5.16	0.014
Ileum	156.94	151.08	152.22	152.97	150.63	5.52	0.997
Villus surface area (mm^{2})							
Duodenum	0.425	0.411	0.420	0.478	0.474	0.01	0.773
Jejunum	0.310	0.306	0.350	0.383	0.344	0.02	0.764
Ileum	0.294	0.270	0.292	0.333	0.329	0.01	0.724
Villus height:crypt depth							
Duodenum	6.05^{b}	6.95^{ab}	7.91^{ab}	9.08^{ab}	10.35^{a}	0.52	0.045
Jejunum	4.12	4.71	6.79	6.81	6.88	0.46	0.150
Ileum	3.65^{b}	4.07^{ab}	4.39^{ab}	5.66^{a}	5.15^{ab}	0.25	0.046

^{a,b,c} means in each row with different superscripts are significantly different ($P < 0.05$).

[1] ST = *Salmonella* Typhimurium.

In the study of Menconi *et al*. (2011) which followed a similar purpose to this experiment, 1-d old broiler chickens, 1 hr after inoculation with Salmonella Heidelberg, orally received 10^{6} CFU of multistrain-probiotics that significantly reduced the Salmonella numbers in the intestinal contents after 24 and 72 hrs. Also Revolledo *et al*. (2009) found that after seven days after inoculating day-old chickens with ST, an 11-strain probiotic (1.6×10^{11} CFU/g) in the diet significantly reduced cecal Salmonella populations. These results were also observed in the study of Knap *et al*. (2011) through the addition of *Bacillus subtilis* (8×10^{5} CFU / g) in the diet of broiler chickens contaminated with Salmonella. Despite a lot of information about the effects of probiotics in reducing the colonization of pathogenic bacteria in the gut, it is still not fully understood how probiotics work (Menconi *et al*., 2011). However, the production of lactic acid and volatile fatty acids and therefore, lowering the gut pH play an important role in the control of the gastrointestinal tract microflora and provide the context for increases in lactic acid bacteria population. It is reported that relatively gradual increases of volatile fatty acids compared to their sudden increase in the digestive tract is the main reason for the removal of Salmonella in ceca of broiler chickens (Van der Wielen *et al*., 2001). Although bacterial interactions (competitive exclusion) are the most well-accepted mechanism for the reduction of Salmonella, stimulation of innate immune response is also possible. According to our results on ileum and cecum pH in the present study, the declines of

coliform and Salmonella populations in our study demonstrates the potential of lactic acid bacteria in reducing pH in these segments.

The interaction of probiotics and host immune system could lead to probiotic immunomodulatory activities (Ashraf *et al*., 2013; Menconi et al., 2011). This interaction could also lead to an increase in antibodies, T cell activation or suppression, and changes in the rate of gene expression of cytokines (Menconi *et al*., 2011). The presence of lactic acid bacteria in the diet stimulates the production of lymphocytes, especially type B that produce antibodies to create humoral immunity (Apata, 2008). In addition, the heterophil: lymphocyte ratio is an appropriate indicator to show stress in poultry (Ghareeb *et al*., 2008). Henderson *et al*. (1999) reported that in the presence of ST, heterophils migrate from mucosal epithelial cells into the intestinal lumen; so three days after inoculation with Salmonella, a large number of heterophil will be found in the intestinal contents and only a few heterophil remain in the epithelium. Though some studies show a significant antibody response in Salmonella-infected chickens, Beal *et al*., (2006) found that the B cell and antibody responses were not active to eliminate Salmonella cells, and could not explain which mechanism (apart from B cell activity) is responsible for this cleanup. According to these reports, the use of probiotics could decrease the energy demand for stimulation of the immune system (increase heterophil and monocyte production and secretion into the gastrointestinal tract) by lowering pathogens (i.e. Salmonella and coliforms).

Improvement of feed efficiency and body weight gain could be related to the increase of villus height and surface area of the small intestine (Awad *et al*., 2009). It is reported that salmonella infection had an adverse effect on villus height and crypt depth in the duodenum (Ribeiro *et al*., 2007). However, Biloni *et al*. (2013) found that probiotic supplementation significantly improved most of the villus and crypt characteristics in the duodenum and ileum of ST-infected broiler chickens on days 7 and 14 of age. However, our findings on morphology in the duodenum were in contrast with this report. As we expected, comparison of the results from Tables 2 and 5 indicated a direct relationship between body weight gain and intestinal morphology improvement. Increasing crypt depth may be indicative of a faster turnover of the intestinal mucosa layer for villus renewal, while the intestinal response mechanism tries to mend the atrophy and/or natural sloughing caused by pathogenic bacteria and their toxins. This faster turnover needs more energy and therefore a significant amount of dietary energy is invested into the process (Haldar *et al*., 2011). The increase in villus height will increase the intestinal absorptive surface area for available nutrients. Therefore, the greater villus height to crypt depth ratio will lead to better growth performance in broiler chickens (Awad *et al*., 2009; Awad *et al*., 2010). In the present study, the decrease of villus height and increase of crypt depth and villus width are associated with the increase of feed intake in control treatment, showing that this group was more affected by the Salmonella-induced stress. The use of probiotic lowered the intensity of this stress in different parts of the small intestine in varying degrees. Also, the lack of significant differences in villus surface area among treatments could be due to villus inflammation.

Conclusion

It was concluded that the use of 0.15% probiotic containing *Lactobacillus plantarum*, *Lactobacillus fermentum*, and *Lactobacillus rhamnosus* may substantially improve growth performance in ST-infected broiler chickens. The responses were mediated mostly through beneficial effects on intestinal villi structure, hematological responses, and by altering gut microflora. Therefore, this probiotic composition and dosage may be explored as a dietary tool in the improvement of broiler production and flock immunity to ST.

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Poultry Science Journal
ISSN: 2345-6604 (Print), 2345-6566 (Online)
http://psj.gau.ac.ir
GUASNR

تاثیر سطوح مختلف پروبیوتیک بر پایه لاکتوباسیلوس‌ها بر عملکرد، جمعیت میکروبی دستگاه گوارش، خون‌شناسی و ریخت‌شناسی روده جوجه‌های گوشتی جوان آلوده به *سالمونلا تیفیموریوم*

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Poultry Science Journal 2015, 4(2): 157-165
DOI: 10.22069/PSJ.2016.11164.1191

چکیده

این آزمایش به منظور مقایسه اثر سطوح مختلف پروبیوتیک چند سویه‌ای بر عملکرد، خصوصیات پرزها و فعالیت آنزیم‌های گوارشی در روده و جمعیت میکروبی دستگاه گوارش جوجه‌های گوشتی آلوده به *سالمونلا تیفیموریوم* انجام شد. ۱۲۰ قطعه جوجه یکروزه گوشتی سویه تجاری راس ۳۰۸ در ۲۰ واحد آزمایشی توزیع شدند و تا سن ۱۰ روزگی در ۵ تیمار آزمایشی شامل یک جیره پایه ذرت- سویا فاقد پروبیوتیک (شاهد)، یا دارای ۰/۰۵، ۰/۱، ۰/۱۵ و ۰/۲ درصد پروبیوتیک در هر کیلوگرم جیره پرورش یافتند. ۵ تیمار و ۴ تکرار توزیع شدند و تا سن ۱۰ روزگی بر روی بستر پرورش یافتند. تمام پرندگان در روز دوم آزمایش از طریق دهان با *سالمونلا تیفیموریوم* تلقیح شدند. جوجه‌های گوشتی در تیمار ۰/۱۵ درصد پروبیوتیک در مقایسه با سایر تیمارها افزایش وزن بالاتری داشتند ($P < ۰/۰۵$). مکمل پروبیوتیک، به جز در سطح ۰/۰۵ درصد، ضریب تبدیل خوراک را به‌صورت معنی‌دار بهبود داد. استفاده از ۰/۱ و ۰/۱۵ درصد پروبیوتیک جمعیت سالمونلاها را در ایلئوم کاهش داد. کمترین نسبت هتروفیل به لمفوسیت در تیمار ۰/۱۵ درصد پروبیوتیک مشاهده شد؛ با این حال، تیمارهای حاوی ۰/۱ و ۰/۲ درصد پروبیوتیک نیز این نسبت را در مقایسه با تیمار شاهد به‌صورت معنی‌دار کاهش دادند. افزودن ۰/۱۵ درصد پروبیوتیک به جیره پایه نسبت طول پرز به عمق کریپت را در ایلئوم و همچنین طول پرز را در ایلئوم، ژژنوم و دودنوم افزایش داد ($P < ۰/۰۵$). با توجه به نتایج این آزمایش، به‌ویژه افزایش وزن بدن و جمعیت سالمونلاها در ایلئوم، افزودن ۰/۱۵ درصد پروبیوتیک به جیره جوجه‌های گوشتی می‌تواند به‌منظور کنترل موثر عفونت *سالمونلا تیفیموریوم* استفاده شود.

کلمات کلیدی
جوجه گوشتی
هتروفیل
باکتری لاکتوباسیل
پرز روده
سالمونلا تیفیموریوم

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تاریخچه مقاله
دریافت: ۱۶ می ۲۰۱۶
ویرایش: ۲۴ ژوئن ۲۰۱۶
پذیرش: ۳ اکتبر ۲۰۱۶

Please cite this article as: Ashayerizadeh O, Dastar B, Samadi F, Khomeiri M, Yamchi A4 & Zerehdaran S. 2016. Effects of Lactobacillus-Based Probiotic on Performance, Gut Microflora, Hematology and Intestinal Morphology in Young Broiler Chickens Challenged with *Salmonella* Typhimurium. Poult. Sci. J. 4(2): 157-165.