





Effect of Dietary Supplementation with Rosemary Complex Powder on the Growth Performance of Native Chickens

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ABSTRACT

This study investigated the effects of dietary supplementation with rosemary complex powder on the growth performance of native chickens. In total, 180 one day-old native chicks were assigned to one of three dietary groups (60 birds each). The control group (Group A) received the basal diet. In addition to the basal diet, the two experimental diets (Groups B and C) were supplemented with 0.2% and 0.4% rosemary complex powder, which contained rosemary leaves, sweet basil, pineapple sage and sweet lavender. Over 19 weeks, feed intake was recorded to determine the average daily gain and feed conversion ratio. In weeks 10 and 15, blood samples were taken for serum antibody titer analysis. At the end of the experiment, serum immunoglobulin A (IgA) and serum immunoglobulin G (IgG) concentrations were examined. No differences were observed among the groups in terms of the starting weight, weight in week 19, average daily feed intake, average daily gain, or average feed conversion ratio. The addition of rosemary complex powder improved total weight gain by 1.52%. Serum IgA and serum IgG concentrations were significantly lower in the experimental groups in comparison to the control group ($p < 0.05$). Villus height, villus width, crypt depth, and villus height/crypt depth ratio were significantly higher in group B than in group A ($p < 0.05$). In summary, rosemary complex powder improved the intestinal absorption capacity of chickens and significantly reduced their immunoglobulin concentrations.

INTRODUCTION

In December 2015, the World Organization for Animal Health (OIE) held the Second OIE International Symposium—Substituting Animal Health Products for Antibiotics in Paris. The issue of antibiotic abuse on animal husbandry products was discussed. Tilburt & Kaptchuk (2008) demonstrated that herbs can invigorate the stomach, aid digestion, calm the nerves, and also be used for sterilization in both traditional Chinese and Western medicine.

Christaki *et al.* (2004) reported that Chinese herbal crude extracts from *Lentinus edodes*, *Tremella fuciformis*, or *Astragalus membranaceus*, significantly increased the levels of cecal-specific IgA and IgG, and antigen-specific proliferation of splenocytes in young chickens. Chen *et al.* (2003) examined the feasibility of using Chinese medicinal herbs as feed additives. It was found that the large-molecular-weight Chinese herbal polysaccharide (Achyranthan) was not as effective as the small molecular weight polysaccharide (Astragalan) in eliciting the immunity of broilers. Changes in the number and concentration of immunoglobulin in chicken serum can reveal whether disease interference is a concern. IgA is secreted from chickens' gallbladder and small intestine and



spreads in the mucosal area. It can protect the mucosa in the oral cavity, respiratory tract, and digestive tract against pathogen colonization. Trochimiak & Hübner-Woźniak (2012) indicated that IgA in human mucosa is the first line of defense against external harmful substances. Higher serum IgA levels are associated with higher risk of autoimmune diseases such as rheumatoid arthritis and systemic lupus erythematosus, as well as liver cirrhosis and chronic hepatitis. The risk of other diseases is also higher. Breedveld & Egmond (2019) revealed that individuals with both alcoholism and metabolic syndrome exhibited particularly high serum IgA concentrations, suggesting that increases in the secretion of individual immunoglobulins may be the result of resistance to autoantigen interference or the body's efforts to protect itself from pathogen colonization. Alagawany & El-Hack (2015) added 0, 3, 6, and 9 g per kilogram of rosemary powder to the diets of laying hens. Both the number of eggs laid and the weight of the eggs increased with increases in the dose of rosemary powder. Moreover, the weight of the egg yolk and its ratio to protein were augmented. In addition, the concentrations of triglyceride and cholesterol in the blood decreased with increases in the dose of rosemary powder. The concentration of IgM increased, but the concentration of IgA decreased. Overall, the results indicate that rosemary can be used as a feed additive for enhancing the laying traits and immunity of laying hens.

Hammershøj & Johansen (2016) compiled a report on the effects of forage and herbage on the egg quality of laying hens. The type and content of forage affected the color and fatty acid composition of the egg yolk, including the high proportion of unsaturated fatty acids. Kiczorowski *et al.* (2016) supplemented the diets of broiler chickens with 3-4% resin extracted from the fruit of *Boswellia serrata*. No significant effect on growth performance was observed, but the inclusion significantly improved dry matter and organic matter digestibility, reduced the proliferation of *Escherichia coli* and *Clostridium* colonies, as well as increased the population of lactic acid bacteria in the intestinal tract.

Experimental evidence on the use of herb- or plant-based feed additives in chicken diets in Taiwan is scant. Dietary supplementation with herbs can be used to improve chicken immunity, thereby reducing chicken mortality and increasing the income of chicken producers. Rosemary complex powder was herein employed as a feed additive for improving the growth performance, immunity, and intestinal traits of native chickens.

MATERIALS AND METHODS

Experimental Animals and Experimental Design

The protocol for the use, breeding, and management of the animals in the experiment was approved by the Laboratory Animal Care and Use Team of the Hualien Breeding Farm of the Animal Production Laboratory of the Executive Yuan Agriculture Committee (approval No. HUA IACUC10706). The experimental hybrid native chicken was bred crossing the Taiwan native chicken hybrid with fighting cock at the Hualien Animal Propagation Station. In total, 180 one-day-old chicks bred at the Hualien Propagation Station of the Taiwan Livestock Research Institute were used. The chicks were divided randomly into 3 groups, with 4 duplications per group and 15 birds per duplication. The chicks were raised in a flat feeding pen that was 300 x 250 cm wide. Rosemary leaves (*Rosmarinus officinalis*), Sweet basil (*Ocimum basilicum*), Pineapple sage (*Salvia elegans*), and sweet lavender (*Lavandula x heterophylla*) were purchased from Yinsherb Corp. Ltd. (Taipei City, Taiwan (R.O.C.)) as dried herbs and were mixed into rosemary complex powder at the ratio of 40, 20, 20 and 20%, respectively. The conditions of the three groups are listed as follows. Group A (the control group) was fed a basal diet. Group B was fed a basal diet supplemented with 0.2% rosemary complex powder, and Group C was fed a basal diet supplemented with 0.4% rosemary complex powder.

The base of each rosemary plant (approximately 15 cm in height) was retained, and the entire upper plant was harvested before blooming occurred. The leaves and stems were dried at 55 °C for 48 h, and then ground for later use. The effective essential oil content of each batch of rosemary plants collected by the company was quantitatively analyzed on a regular basis. During the 19-week experiment, the chickens were given access to feed and water *ad libitum*. The basal diet was formulated considering the climate of the breeding area and with reference to manuals published by the National Research Council (1994). The nutritional composition of the basal diet is presented in Table 1. The feed administered when the chickens were aged 0 - 4, 5 - 8, and 9 - 19 weeks old contained 23.0% crude protein (CP), 3,177 kcal/kg metabolizable energy (ME), 19.5% ME and 3,114 kcal/kg, and 15.0% CP and 3,071 kcal/kg ME, respectively. The 1-day-old chicks were administered the Marek vaccine in sequence under the native chicken vaccination schedule formulated by the on-site veterinarian (Table 2).


Table 1 – Nutritional composition of the basal diet (weeks 0 - 19).

Ingredients	0 - 4 wk	5 - 8 wk	9 - 19 wk
Corn	56.0	66.0	78.8
Soybean meal	30.0	25.0	16.0
Fish meal	7.0	4.0	2.0
Dicalcium phosphate	1.0	1.0	1.2
Oyster Shell	1.2	1.2	1.3
Oil	4.0	2.0	0.0
Salt	0.5	0.5	0.5
Vitamin premixa	0.1	0.1	0.1
Mineral premixb	0.1	0.1	0.1
Methionine	0.1	0.1	0.0
Total	100.0	100.0	100.0
Analyzed			
Dry matter, %	89.9	89.6	89.4
Crude protein, %	23.0	19.5	15.0
ME, kcal/kg	3177.0	3114.0	3071.0
Calcium %	1.1	0.97	0.94

^aOne kilogram of feed provided vitamin A, 10000 IU; vitamin D3, 1000 IU; vitamin E, 25 IU; vitamin K, 3 mg; thiamin, 3 mg; riboflavin, 5 mg; pyridoxine, 3 mg; vitamin B12, 0.03 mg; Ca-pantothenate, 10 mg; niacin, 50 mg; biotin (1.0%), 0.1 mg; folic acid, 3 mg.

^bOne kilogram of feed provided Mn (MnSO₄ · H₂O), 60 mg; Zn (ZnO), 60 mg; Cu (CuSO₄ · 5H₂O), 5 mg; Fe (FeSO₄ · 7H₂O), 70 mg; Se (Na₂SeO₃), 0.1 mg.

Table 2 – Vaccination program of native chickens in Taiwan.

Age	The vaccination program of Taiwan native chickens
1 day	MD Live Vaccine + Chick-N-Pox Liv Vaccine (Subcutaneous injection)
1 days	ND + IB Two in one- Live Vaccine (Eye drops)
3 days	IBD Live Vaccine (Drinking water)
10 days	IBD Live Vaccine (Drinking water)
2 wks	Mycoplasma vaccine (Eye drops)
2 wks	Chick-N-Pox Live Vaccine (Subcutaneous injection)
3 wks	ND + IB Two in one- Live Vaccine (Drinking water)
3 wks	IBD Live Vaccine (Drinking water)
4 wks	ND Live Vaccine (Drinking water)
4 wks	Rio virus (Drinking water)
7 wks	Chicken infectious laryngotracheitis (Eye drops)
12 wks	ND × IC Two in one- Inactivated Vaccine (Intramuscular injection)
16 wks	ND + IB Two in one- Live Vaccine (Drinking water)

MD, Marek's disease; ND, Newcastle disease; IBD, infectious bursal disease; IB, infectious bronchitis; CRD, chronic respiratory disease; IC, infectious coryza.

Testing Items and Methods

The general composition of the test diet was analyzed with reference to the Official Methods of Analysis of AOAC International (1995).

Growth Traits

The 90 male and 90 female chickens were raised separately. Their feed intake was recorded, and their average daily gain, average feed intake, and feed conversion ratio (feed amount/weight gain) were calculated. In weeks 0, 4, 8, 12, 16, and 19, the

chickens were weighed. In weeks 10 and 15, blood samples were taken for serum antibody analysis. Repeated sampling was conducted. Specifically, the testing units comprised one male and one female chicken in each group, eight chickens in each test group, a total of 48 chickens, and individual chickens. Using a syringe, 5 mL of blood was collected from the underwing vein. The blood was centrifuged in a telecentric separator (Kubota 5800, Taiwan) at 3,000 rpm for 15 min at 4 °C, and the plasma obtained was stored in a –20 °C freezer. The serum was analyzed for IBD, IB, and ND antibody titers at the Southern District Laboratory of the Poultry Health Center of the Central Animal Products Association. The detection of ND antibody titers involved the hemagglutination inhibition test, whereas the detection of IBD and IB antibody titers involved enzyme-linked immunosorbent assay (ELISA) performed using a kit produced by Kirkegaard & Perry Laboratories, Inc.

Biochemical Analysis of Blood Samples

In week 19, blood samples from one male and one female chicken from each repeated sampling (8 chickens from each group, for a total of 24 birds) were subjected to biochemical analysis, and individual chickens were used as the test unit. Heparin-treated injection syringes were used to collect 2 mL of blood from the underwing vein. The collected blood was centrifuged in a telecentric separator at 3,000 rpm for 15 min at 4 °C, and the serum obtained was stored in a –20 °C freezer. A biochemistry analyzer (Spotchem SP-4410; Arkray, Japan) and its various kits were employed to examine the total bilirubin concentrations in the liver and gallbladder and alanine aminotransferase (ALT) activity, as well as creatinine and amylase concentrations (indicators of renal and pancreatic function, respectively).

Serum IgG and Serum IgA Concentrations

Serum immunoglobulin tests were performed in weeks 12 and 19. Repeated sampling was conducted. Specifically, one male and one female chicken were sampled from each group (8 from each group, totaling 48 chickens), and individual chickens were used as the test unit. The serum was diluted 80,000 and 4,000 times with a buffer solution and analyzed using the Chicken ELISA IgG and IgA Kit (Bethyl), followed by analysis on an ELISA reader (Bio-Rad Laboratories Inc., Irvine, CA, USA) at a wavelength of 450 nm.

Observation of Jejunum Villi Tissue Sections

In week 19, 24 chickens (one male and one female chicken from each repeated sampling, eight from each



group) were killed. Their intestinal tissue was sectioned and stained for the observation of intestinal villi patterns (Uni *et al.*, 1995). Specifically, approximately 2 cm of the middle part of the jejunum was removed. After the contents were washed, the jejunum was placed in neutral formalin solution for over 24 h. Subsequently, it was embedded in paraffin and sectioned. Hematoxylin and eosin staining was performed, and the sections were observed under a microscope (Labophot-2; Nikon) in accordance with the measurement standards established by Uni *et al.* (1995). For each section, the height and width of the jejunum villi and the depth of the crypts were examined and scored out of 10 location points.

Statistical Methods

A completely randomized study design was used to evaluate the effects of vanilla powder–rosemary compound supplementation on the physicochemical

properties of hybrid chicken meat. Data were analyzed using analysis of variance, and significant differences were determined using Duncan's multiple range test. Statistical significance was set at $p < 0.05$. The analyses were performed using SAS program (ver. 8.0, SAS Institute, 2002).

RESULTS AND DISCUSSION

The effects of dietary supplementation with 0.2% and 0.4% rosemary complex powder on the chickens' growth traits are presented in Table 3. The average daily feed intakes of groups A, B, and C were 84.7, 85.8, and 88.1 g, respectively, and significant among-group differences were observed ($p < 0.05$). A three-stage feed intake survey was performed. In weeks 0 - 8, the average daily feed intake of the groups did not differ significantly. However, a significant difference was noted between the feed intake of the group C

Table 3 – Effect of rosemary complex powder addition on growth performance.

Items	0%	0.2%	0.4%	SEM1	<i>p</i> -value
Number	60	60	60		
days	133	133	133		
0 - day BW, g	25.4	25.4	25.1	2.1	0.7
133 - day BW, g	2657.3	2694.2	2627.5	534.8	0.8
Average daily gain, g	19.8	20.1	19.6	4.0	0.8
Daily feed intake, g/bird/day	84.7c	85.8b	88.1a	2.8	<.0001
Feed conversion ration	4.3	4.3	4.5	1.0	0.4

^{abc}Means with different superscripts indicate a significant difference ($p < 0.05$).

¹SEM: standard error mean.

chickens aged 9 to 19 weeks and group A chickens aged 0 to 19 weeks ($p < 0.05$; Table 4). A possible reason is that the addition of plant debris slightly increased the fiber content of the feed, stimulating appetite and enhancing both the digestibility of the feed and the emptying of the digestive tract, which makes feed intake seem to be higher than the control. The bitter astringent taste of the saponin contained in alfalfa may affect the feed intake of laying hens (Deng *et al.*, 2011). The rosemary complex powder used herein does not contain such components affecting the feeding of laying pens.

The average daily gains of groups A, B, and C were 19.8, 20.1, and 19.6 g, respectively, and the average feed conversion ratios were 4.3, 4.3, and 4.5, respectively. No significant differences were observed between the two experimental groups and the control group (Table 3). However, group B exhibited slight improvements in the total weight gain (1.41%) and feed conversion ratio (0.23%). The body weights of

the male and female chickens aged 19 weeks are as follows: 3085.0 and 2168.5 g in group A, 3128.8 and 2259.7 g in group B, and 2992.0 and 2202.3 g in group C, respectively. As shown in Figure 1, no significant among-group differences were noted, but group B tended to have a higher body weight. Similar findings have been reported by Sierżant *et al.* (2021), who confirmed no significant effect on the average body weight of broilers after supplementation of diets with two concentrations (0.25 and 0.5 %) of rosemary and blackcurrant extracts. Petricevic *et al.* (2018) reported that the use of 0.2% rosemary in diets caused no significant differences in average daily gain, but 0.4 and 0.6% supplemented rosemary powder in broiler feed could improve average daily gain and feed conversion ratio. In summary, dietary supplementation with rosemary complex powder did not increase the market weight of the native chickens, but it also did not have adverse effects on their growth performance (Table 3).



Table 4 – Effect of rosemary complex powder addition on growth performance by week.

Items	0 %	0.2 %	0.4 %	SEM1	p-value
Weight gain, g/bird					
0 - 8 wk	1041.2	1005.5	1014.9	149.0	0.4
8 - 19 wk	1591.5	1663.4	1587.5	427.8	0.6
0 - 19 wk	2631.9	2668.8	2602.4	534.5	0.8
Daily feed intake, g/bird/day					
0 - 8 wk	46.4ab	47.4a	45.3b	1.7	<.0001
8 - 19 wk	112.5b	113.8b	119.2a	4.2	<.0001
0 - 19 wk	84.7c	85.8b	88.1a	2.7	<.0001
Feed conversion ratio, feed/gain					
0 - 8 wk	2.5	2.6	2.5	0.4	0.5
8 - 19 wk	5.4	5.3	5.8	1.7	0.3
0 - 19 wk	4.3	4.3	4.5	1.0	0.4

^{abc}Means with different superscripts indicate a significant difference ($p < 0.05$).
¹SEM: standard error mean.

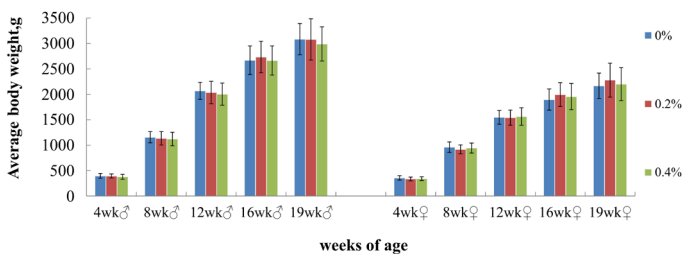


Figure 1 – Effect of rosemary complex powder addition to weight gain in native chickens in various growth stages.

Regarding the blood biochemistry of the 19-week-old chickens, no significant differences were found in the concentrations of total bilirubin and creatinine in the serum or in ALT and pancreatic amylase activities (Table 5). Bilirubin and ALT are often selected as parameters of liver and gallbladder function. Creatinine and pancreatic amylase are the parameters of kidney and pancreas function. When kidney function is poor, the elimination rate of creatinine decreases and its content in the blood increases. Mahgoub *et al.* (2019) investigated the impacts of rosemary cold-pressed oil (0, 1 and 2 mL/kg diet) on total bilirubin in Japanese quails. The results showed total bilirubin concentrations have no significant differences and coincided with our current consequence. These results came in accordance with those obtained in the study of Ghozlan *et al.* (2017). The non-significant alterations in serum ALT activities and creatinine levels indicated

Table 5 – Effect of rosemary complex powder addition on blood biochemistry parameters.

Items	0 %	0.2 %	0.4 %	SEM1	p-value
Number	8	8	8		
T-Bilirubin, mg/dL	0.49	0.50	0.44	0.17	0.8
ALT, U/L	1.00	1.38	1.13	0.47	0.5
Creatinine, mg/dL	0.29	0.28	0.29	0.05	0.8
Amylase, U/L	451.38	446.63	440.13	143.42	0.9

¹SEM: standard error mean.

the safe use of rosemary as a feed additive in broiler chickens diet in regards to liver and kidney functions. Amylase is the most important enzyme in the body for the digestion of carbohydrates. Pancreatic enzyme secretion is stimulated by plant extracts. It is assumed that increased pancreatic enzyme secretion would improve chicken performance (Puvača *et al.*, 2022).

Serum IgA concentrations and serum IgG concentrations can be employed as indicators of immune function (Hung *et al.*, 2011). As presented in Figure 2, the average concentrations of serum IgA and serum IgG in groups A, B, and C in week 12 were 56.2 and 752.6 mg/dL, 43.6 and 553.0 mg/dL, and 34.7 and 509.2 mg/dL, respectively. In week 19, the corresponding concentrations were 63.6 and 798.0 mg/dL, 43.1 and 518.0 mg/dL, and 34.0 and 516.4 mg/dL, respectively.

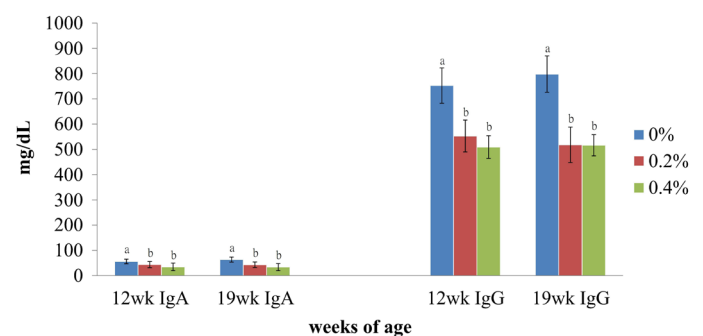


Figure 2 – Effect of rosemary complex powder addition on IgA and IgG concentrations in the blood of native chickens.



The results demonstrated that serum IgA and serum IgG concentrations significantly differed between the experimental and control groups ($p < 0.05$). Specifically, serum IgA and serum IgG concentrations of the experimental groups were significantly lower than those of the control group. Most pathogenic bacteria invade the host through mucosal surfaces, especially those in the gastrointestinal, respiratory, and genitourinary tracts. Mammals and birds produce IgA to generate a primary immune response (Mostov & Kaetzel, 1999). IgA plays an important defensive function in preventing pathogens attaching to the mucosal cells and inhibiting bacterial colonization and viral infection (Pabst *et al.*, 2016). But excessive concentrations of IgA and IgG induce inflammatory responses. Chronically high IgA concentrations are associated with various diseases, including nephropathy, dermatitis herpetiformis, vasculitis, and rheumatoid arthritis (Gonzalez-Quintela *et al.*, 2008; Breedveld & Egmond, 2019). The low serum IgA and serum IgG concentrations observed in the present

experimental groups indicated that the addition of rosemary complex powder enhanced the chickens' initial immune defense mechanism and reduced the probability of their body mucosa being colonized by viruses. By proving the presence of this IgG antibody, it can be shown that there was a previous disease or that immunization was performed (Puvača *et al.*, 2022). Overall, the experimental groups secreted significantly less serum IgA and serum IgG than the control group.

Table 6 shows the effect of adding rosemary complex powder on chickens vaccinated with Newcastle disease (ND), infectious bronchitis (IB) and infectious bursal disease (IBD). Serum from vaccinated chicks did not show significant differences in humoral immune responses and antibody concentrations. Contrary to this, Abo Ghanima *et al.* (2020) found that all immunity indices (Newcastle disease, Avian influenza H5, and Avian influenza H9) between the control and 0.03 % diet of rosemary and cinnamon essential oils groups had significant differences.

Table 6 – Effect of rosemary complex powder addition on ND, IBD, and IB antibody titer concentrations in the blood.

Items	0 %	0.2 %	0.4 %	SEM ⁴	p-value
10thWeek					
Number	8	8	8		
ND ¹ (log ₂)	3.1	3.3	3.2	0.2	0.3
IBD ² (×10 ³)	3.1	3.3	3.6	1.2	0.7
IB ³ (×10 ³)	2.2	2.2	2.3	0.6	0.9
15thWeek					
Number	8	8	8		
ND ¹ (log ₂)	2.0	2.0	2.0	0.2	1.0
IBD ² (×10 ³)	2.6	2.6	2.7	0.5	0.9
IB ³ (×10 ³)	2.2	2.3	2.2	0.7	0.9

¹ND, Newcastle disease; ²IBD, infectious bursal disease; ³IB, infectious bronchitis. ⁴SEM: standard error mean.

In general, physiological changes in chickens fed components conferring health benefits are often caused by changes in intestinal factors. The development of intestinal villi can induce changes in the intestinal microbiome and constitutes a key limiting factor in growth and nutrient utilization (Noy & Sklan, 1995). The characteristics of the jejunum villi of the chickens in the three groups are displayed in Table 7. In this trial, the length and width of the villi in groups A, B, and C were 868.4 and 203.3 μm , 1064.1 and 348.3 μm , and 1026.4 and 271.2 μm , respectively. Groups A and B differed significantly ($p < 0.05$), demonstrating that a basal diet supplemented with 0.2% rosemary complex powder yielded the greatest health benefits. A gastrointestinal profile characterized by deeper crypts and shorter villi is associated with the presence of toxins (Yason *et al.*, 1986). Herein, villus width, crypt depth,

and villus height/crypt depth ratio were significantly higher in group B than in group A ($p < 0.05$). Our results agree with Mousapour *et al.* (2022), who found villus height was increased ($p < 0.01$) in birds supplemented with 0.015-0.03% rosemary essential oils, as compared to control and antibiotic diets. Development in villus height and width is directly correlated with an increased epithelial turnover that leads to an increase in the absorption of available nutrients. Amiri *et al.* (2022) showed that using 0.01-0.02% garlic in broiler diet significantly enhanced villi length and villi width. Villi development provided a greater absorption surface for available nutrients (Karangiya *et al.*, 2016). Our result was consistent with the observation of higher body weight gain and feed conversion ratio in group B (Table 3). However, whether this result is attributable to the addition of rosemary complex powder warrants


Table 7 – Effect of rosemary complex powder addition on the characteristics of jejunum villi.

Items	0 %	0.2 %	0.4 %	SEM ¹	p-value
Number	8	8	8		
Villus height, μm	830.3 ^b	1108.6 ^a	1026.4 ^a	173.1	0.02
Villus width, μm	179.6 ^b	373.9 ^a	271.2 ^{ab}	125.0	0.03
Crypt depth, μm	236.5 ^a	136.4 ^b	273.9 ^a	64.7	0.001
Villus height / Crypt depth, μm	3.9 ^b	9.2 ^a	3.9 ^b	2.4	<.0001

^{abc}Means with different superscripts indicate a significant difference ($p < 0.05$). ¹SEM: standard error mean.

confirmation through further experiments, as does whether such addition strengthens chickens' immunity and mitigates reductions in villus length ascribable to toxin interference.

CONCLUSION AND RECOMMENDATIONS

The problem of pathogenic microorganisms developing resistance against antibiotics used in animal husbandry is serious and merits urgent attention. Herbs exert various pharmacological effects, including antibacterial and immunity-enhancing effects. Herein, dietary supplementation with rosemary complex powder improved the weight gain of native chickens by 1.52%, and significantly increased the beneficial effects on in vivo conditions (immunity, performance and gut health) in chickens. More studies are needed to explain the mode of action of the active components of such rosemary complex powder.

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