Available online at www.scholarsresearchlibrary.com

Scholars Research Library

Annals of Biological Research, 2011, 2 (5) :291-298
(http://scholarsresearchlibrary.com/archive.html)

Annals of Biological Research
Scholars Research Library
ISSN 0976-1233
CODEN (USA): ABRNBW

The Effect of Different Levels of Virginiamycin on Performance, Immune organs and Blood Metabolite of Broiler Chickens

Farhad Ahmadi

Departments of Animal Science, Shabestar branch, Islamic Azad University, Shabestar, Iran

---

ABSTRACT

The objective of this study was to investigate the effect of different level of virginiamycin (VIR) on Growth performance, biochemistry, and hematology blood parameters broiler chickens. A total of 240 day-old broilers (Ross 308) were randomly allocated to four groups of 15 chicks each. Each four groups assigned in completely randomized design with four replicate. Experimental groups consisted of: T1) Basal diet (control), and others groups T2, T3, and T4 with 10, 20, and 30 ppm VIR kg^{-1} diet respectively added to the basal diet. Body weights and, feed intake recorded as weekly and used for calculated FCR at 42 days. At the end of research four birds (one bird per replicate) selected and after sampling blood, slaughtered for determination organ weights. About 4 ml drawing blood sample from wing vein. Serum removed by centrifuge and stored in -20°C until analysis. The results indicated that supplemented diet with 20 ppm VIR kg^{-1} diet had significantly increased body weight ($P<0.05$), also had significantly decreased FCR compared with control and other groups ($P<0.05$). Relative weight of small intestine and abdominal fat had significantly decreased in T2 and T3 compared with control ($P<0.05$). Value of Ca, P, and Mg in serum had a trend increasing parallel with increased level of virginiamycin in diet ($P>0.05$). Concentrations of Triglyceride, and cholesterol showed decreasing trend among treatment compared with control group, but had not significant effect ($P>0.05$). In addition, the results current research showed that: 1) to add virginiamycin in diet was improved performance traits and important blood parameters, and 2) the best level of VIR to applied nutrition was 20 ppm kg^{-1} diets.

Key words: Broiler, Blood metabolites, Performance, Virginiamycin.

INTRODUCTION

In poultry nutrition, the growth-promoting effect of antibiotics was noticed in the early 1950s [15]. The positive effect on growth was mainly related to the ‘‘microflora-management’’ theory

based on three mechanisms: *The first* mechanism is the control of gut micro biota. It results in decreased competition for nutrients and a reduction in microbial metabolites depressing growth. *The second* mechanism is the reduction of gut size [9, 18]. This condition caused a lower production of luminal short-chain fatty acids (SCFA) derived from microbial fermentation reduces mucosa cell proliferation and induces thinner villi lamina and gut wall, providing enhanced nutrient digestibility, and *The third* mechanism is the reduction in opportunistic pathogens and subclinical infection. Although, usage of AGPs have been banned by European Union since 2006 Jan 01, as a preventive measure to avoid antimicrobial resistance, But, in Iran and some of other country still widely used as feed additives in applied poultry nutrition and, these substances are considered to have growth-promoting properties [10], and they are used to selectively target, kill, or inhibit the growth of microorganisms [5].

Virginiamycin (VIR) is an antibiotic that was produced by a variant of the *Streptomyces virginiae* species. It was first isolated in 1955 by De Somer and Van Dijck [17]. Virginiamycin marketed for use in domestic animals is actually a combination of two antibiotics, $VIR_{m1,}$ and $VIR_{s1.}$ Virginiamycin m_1 binds to ribosome's and inhibits translation by itself, but it is more effective in combination with VIR_{s1} because cooperative binding of these two antibiotics acts synergistically to prevent protein synthesis within bacteria [16, 12]. This antibiotic is commonly used to treat Gram-positive organism infections. Also, has reported to have growth-promoting effects at sub therapeutic levels in diets for cattle, swine, and poultry [14, 19, 21]. Parks *et al*. [20] suggested that VIR controls microbial growth by acting on the mircoflora's biochemical processes in the cell, such as protein synthesis, by inhibiting the elongation of *Methonobacterium* and *Escherichia coli*, or by reducing lactic acid producing bacteria by 10 to 20 fold in the stomach.

Cummings [8] reported that antibiotics, such as VIR, reduce lactic-acid-producing bacteria, which predominate in the upper gastrointestinal tract of the broiler. While lactic acid producing bacteria (*Lactoballus*, *Stretocci*, and *Staphylococci*) help prevent *Salmonella*, they also are largely responsible for retarded growth seen in pigs and chickens. The reduction in bacterial count may increase nutrient availability of the feed because there is less competition for the nutrient between the animal and the micro flora. Antibacterial agents may improve growth performance and nutrient utilization by thinning the small intestinal epithelium or by decreasing the production of growth depressing toxins or metabolites by intestinal microflora [11]. Several researchers have reported the nutrient sparing effect of VIR on crude protein, Ca, and P in pigs and poultry [1, 4, 23]. The objective of the present research was: 1) to evaluate the use of different level VIR on performance, relative weight of visceral tissue and immune organs, and some of parameters in serum of blood broilers, 2) Introduced an optimum level to procedures that using as applied dosage because of decreasing diet of cost.

MATERIALS AND METHODS

Birds, diets and housing

A total of 240 one-day-old broiler chicks (Ross 308) were obtained from a local of commercial breeder farm hatchery. The birds were weighed at the beginning of the experiment, randomly divided four groups (60 birds per each group), and housed in pens of identical size (1.75 × 1×1 m) in a deep litter system that covered with wood shavings as the litter material. Each group had

four replicates (15 birds per experimental pen). The conditions of production were the same for total of population of statistical. Environmental temperature in the first week of life was 33°C, and decreased to 24 °C until the end of experiment. The control group was fed starter (1-21) and grower (22-42) diets basis of corn- soybean meal. Basal diet current study formulated to meet or excess according to national research council [18]. Ingredients and composition of diet presented in Table1. The birds had accessed *as ad- libitum* to water and feed during research. Basal diets was the following: T1: control (without VIR); T2: basal diet +10 ppm VIR kg^{-1}; T3: basal diet +20 ppm VIR kg^{-1}; T_4: basal diet+30 ppm VIR kg^{-1}.

Data Collection

Growth performance

After provided broiler chickens and transfer to farm, initial body weight recorded individually at the beginning of the experiment as well as at the final of studying period (42 days). The feed consumption was measured as weekly throughout experiment. Cumulative weight gain; feed consumption and food conversion ratio (food intake ÷ weight gain, FCR) were calculated. The mortality and live ability percentage at the end of the feeding period were determined.

Visceral organs

The birds, immediately, after sampling blood slaughtered and visceral organs such as liver, gizzard, pancreases, small intestine, abdominal fat and immune organs as bursa Fabricious, spleen, and thymus removed and relative weight of those calculated based on live body weight (LBW) by the following formula: [(Weight of organs ÷ LBW) ×100].

Table1: Ingredients and composition of broiler basal diets at starter and grower period[1, 2]

Ingredients (%)	Starter (1-21d)	Grower (221-42d)
Corn grain	60.73	59.30
Soybean meal	32.42	28.19
Soybean oil	4.66	4.62
DL-Methionine	0.22	0.192
Lysine HCl	0.22	0.218
Calcium carbonate	1.36	1.20
Dicalcium phosphate (DCP)	1.63	1.53
Sodium chloride	0.25	0.2
Vitamin and mineral premix[1]	0.6	0.6
Salinomycine	0.05	0.05
Calculated compositions		
ME (kcal kg^{-1})	3000	3050
CP (N×6.25)	22.0	21.0
Lysine	1.25	1.15
Met+cys	0.95	0.90
Ca	1.1	1.0
Available P	0.42	0.4

[1]supplement supplied the following per 3 kilogram of diet: Provided per kilogram of diet: vitamin A, 8,000 IU; vitamin D3 (cholecalciferol), 3,200 IU; vitamin E, 25 IU; vitamin K3, 1.5 mg; vitamin B12 (cyanocobalamin), 0.02 mg; biotin, 0.1 mg; folacin (folic acid), 1 mg; niacin (nicotinic acid), 50 mg; pantothenic acid, 15 mg; pyridoxine, 4 mg; riboflavin, 10 mg; and thiamin (thiamin mononitrate), 3 mg, and mineral included: copper, 7.0 mg; iodine (potassium iodate), 1.0 mg; iron (ferrous sulfate+7 H2O), 50 mg; manganese (manganese sulfate), 100 mg; selenium (sodium selenite), 0.15 mg; and zinc (zinc sulfate), 75 mg.

Immune Organs

Immediately after blood sampling, birds slaughtered, and then immune organs such as spleen, thymus and bursa of Fabricius were removed, cleansed of adhering material, and those related weight was calculated by the following formula:

Related organ weight = organ weight (g) ×100 / Live body weight (g)

Relative organs weights were expressed as percentage of live body weight (%LBW).

Blood Collection and Analysis

At the end of experiment, four birds were randomly selected from each replicate and 4mL of blood withdrawn from the wing vein of each bird. Freshly collected blood was left at room temperature for 45 min prior to placing on ice for 1 h to shrink the clot. Then the sample was centrifuged at 2000×g for 15 minutes and sera was separated and stored at -20°C until subsequent analysis. Cholesterol, triglyceride, Ca, P, and Mg in the serum were determined enzymatic ally using an automatic biochemistry standard kit (Model CL-8000, Shimadzu Co. Japan).

Statistical Analysis

Data were subjected to ANOVA using the general linear model procedure of SAS Institute [22]. Means were compared using Duncan's test and were considered statistical significance based on $P<0.05$. The data were analyzed according to the following model:

$$Yij = \mu + \alpha_i + e_{ij}$$

Where:
Y_{ij}= All dependent variable
μ= overall mean, Ti = the fixed effect of VIR levels
(i=0, 10, 20, and 30 ppm VIR kg^{-1} diet), and e_{ij} = the random error term.

RESULTS AND DISCUSSION

Growth performance

The influences of treatments contain different levels of VIR on the performance traits of broilers were summarized in Table 2. Such as data was indicated in the final of trial period (1-42d), there were significant differences among treatments in the LBW, and feed conversion ratio of broilers fed on diets with different levels of VIR ($P<0.05$). The best result related to LBW and FCR observed in the fed on basal diet that supplemented with 20 ppm VIR (T3) as compared with control (T1), T2, and T4. The results current research are in agree with the findings of Buresh *et al*. [4] who reported that the addition of VIR to the diet improved body weight, feed efficiency, and dietary energy utilization in turkey poults with restricted access to feed. Feighner and Dashkevicz. [11] Reported that antibacterial agents may improve growth performance and nutrient utilization by thinning the small-intestinal epithelium or by decreasing the production of growth depressing toxins or metabolites by intestinal micro flora. Cervantes *et al*. [7] reported that the addition of VIR to marginally deficient P diets for broilers increased body weight,

improved feed conversion, and decreased mortality. Belay and Teeter. [3] Reported that VIR increased dressing percentage, which was attributed to the decrease in intestinal weight.

Relative weight of visceral tissues

The effect of different levels of VIR on the relative weight of visceral organs at 42 days of age of broilers showed in Tables3. Also, they indicated that a decrease in intestinal mass was important because the small-intestinal mucosa is the most rapidly regenerating tissue in the body, and maintenance of a greater intestinal mass would result in a greater utilization of nutrients by the intestinal mucosa.

Table 2: Effect of different levels of VIR on growth performance at 42 days of age

Treatment*	N[1]	LBW (g)	Carcasses yield (%)	FI (g)	FCR	Mortality (%)
T1	4	2232.58[b]	70.23	4453.05	1.99[b]	20.00
T2	4	2297.08[b]	70.58	4400.15	1.93[b]	13.33
T3	4	2442.98[a]	75.84	4381.25	1.79[a]	6.66
T4	4	2399.98[a]	74.94	4353.12	1.81[a]	6.66
Mean ± SE		2323.17±116.31	72.89±0.4	4320.21±138.27	1.86±0.19	11.62±0.18

Means with in columns with different superscript differ significantly (P<0.05); N= observation
[1]Data are means of four replicates of four broilers per replicate.
**T1=Control; T2= Control + 10 ppm VIR; T3= Control + 20 ppm VIR; T4= Control + 30 ppm VIR*

VIR can decrease thickness of the mucosa and thinning of the intestinal wall due to reducing intestinal micro flora [11]. Henry *et al*. [13] reported that the addition of VIR to a diet reduced intestinal weight in broilers, which was attributed to a thinning of the intestinal wall. Therefore, this condition may be reason for decreasing weight of SI.

Table 3: Effect of VIR on relative weight of broiler visceral organs at 42 days of age (%LBW) [1]

Treatments[2]	N[3]	Gizzard	pancreas	Liver	Heart	Abdominal fat	Small intestine
T1	4	1.19	0.16	2.84	0.60	2.92a	2.56a
T2	4	1.21	0.16	2.78	0.60	2.12b	2.10b
T3	4	1.21	0.19	2.82	0.57	2.05b	2.19b
T4	4	1.20	0.20	2.74	0.59	2.08b	2.21b
Mean ± SE		1.19±0.08	0.18±0.02	2.12±0.16	0.59±0.06	2.28±0.13	2.26±0.14

[1]Means with different superscripts within the same column differ significantly (P<0.05).
[2]T1=Control; T2= Control + 10 ppm VIR; T3= Control + 20 ppm VIR; T4= Control + 30 ppm VIR
[3]Data are means of four broilers per replicate.

Relative weight of immune organs

The result related to effect of VIR on relative weight of immune organs presented in table 4 and figure1.The relative weights of immune organs weren't significantly (P>0.05) effected in the birds that fed supplemented diet with different levels of VIR as compared with control treatment, but observed a trend of increasing relative weight of bursa Fabricious. The highest weight belong to treatment T3 contain 20 ppm VIR (0.20%) from 1 to 42 days of age. The reason of this situation may be due to change of profile intestinal microflora. Zhou *et al*. [24] reported that VIR had altered composition of the intestinal micro biota of broiler chickens. Therefore, change of gut microflora can affect on immune system and bursa Fabricious development. Also, many other studies using germ-free animal models have shown that the gut micro biota is essential for

the development of the mucosal immune system [2, 6]. Especially during the development of the chicken mucosal immune system, it can be hypothesized that in-feed antibiotics could affect the quality and quantity of immune response, both locally and systemically.

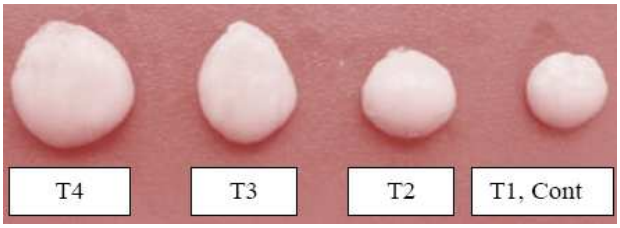

Fig1. Effect of different VIR on the relative weight of bursa Fabricious of broiler at 42 days of ages

Table4: Effect of different levels of VIR on relative weight of immune organs

Treatments	N	Bursa	spleen	Thymus
T1	4	1.17	0.41	0.68
T2	4	1.18	0.41	0.67
T3	4	1.20	0.40	0.68
T4	4	1.19	0.41	0.66
Mean ± SE		0.21±0.008	0.27±0.007	0.68±0.004

Serum metabolites

The results related to affect of different levels of virginiamycin on the sera parameters of broiler presented in table5. In current research, had not observation any statistical significantly difference in comparison with control treatment, also among other treatments (P>0.05), But detected increasing trend of concentration serum parameters such as Ca, P, and Mg. This finding is agree with results of other researcher who reported that that the addition of VIR improved P, Ca, and Zn digestibility by 28, 11, and 19%, respectively, and absolute retention by 33, 19, and 21%, respectively Agudelo et al. [1]. Several researchers have reported the nutrient sparing effect of VIR on Ca and P in pigs and poultry [1, 4, 23]. Antibacterial agents may improve growth performance and nutrient utilization by thinning the small-intestinal epithelium or by decreasing the production of growth depressing toxins or metabolites by intestinal microflora, also the nutrient sparing effects of VIR on crude protein, energy, Ca, Mn, and P in pigs and chickens Feighner and Dashkevicz [11]. Disparity in mention result, value of TG and cholesterol in blood serum of birds that fed diet supplemented with VIR throughout study decreased (table6) as compared with control treatment (P>0.05), and lowest concentration observed at T3 (20 PPM).

Table 5. Effect of Virginiamycin on the sera parameters of broilers at 42 days

Treatments	N	Ca	P	Mg	TG (mg/dl)	Cholesterol (mg/dl)
T1	4	0.53	0.71	1.90	137.09	1.53
T2	4	0.62	0.87	2.03	133.87	1.54
T3	4	0.68	0.99	2.21	130.08	1.52
T4	4	0.62	0.90	2.06	132029	1.51
Mean ± SE		0.62±1.34	0.86±1.23	1.54±2.54	133.85±.01	1.52±2.51

CONCLUSION

the results current research indicated that:
A. Supplemented diet with 20 ppm virginiamycin caused improved of overall performance traits, and this level is the best measure for usage applied in broiler nutrition.
B. Improved immune system and function it due to effected VIR on the profile intestinal micro biota and thereby decrease count of pathogen and induced of optimum condition for proliferation of non pathogen bacteria, on the other hand by decreasing the production of growth depressing toxins or metabolites by intestinal microflora.
C. Because of positive effect mention above, we can formulated ration with low cost, especially in the countries that accessed to feedstuff source was limited

Acknowledgements
The Author wishes to thanks Prof. Dr. Javad pourreza for guides me as supervisor to do this research. The Author also acknowledges the staff of Lavark poultry farm in Isfahan Technology University for their assistance throughout this study.

REFERENCES

[1] Agudelo JH, MD Lindemann, GL Cromwell, RD Nimmo, *J. Anim. Sci*., **2003**, 81(Suppl. 2), 82. (Abstr.)
[2] Bauer E, BA Williams, H Smidt, MW Verstegen, R Mosenthin, *Curr, Microbiol*., **2006**,7, 35-51.
[3] Belay T, RG Teeter, *Poult. Sci*., **1996**, 75, 1383-1392.
[4] Buresh RE, RD Miles, RH Harms, *Poult. Sci*., **1985**, 64,757-758.
[5] Carlson M, TJ Fangman, http://muextension.missouri.edu/ xplor/agguides/ansci/g02353.htm, **2004**, Accessed Apr.
[6] Cebra J, *Am. J. Clin. Nutr*., **1999**, 69,1046- 1051.
[7] Cervantes H, K Bafundo, P Ewing, G Pesti, R Bakalli, *Poult. Sci.*, **2002**, 81 (Suppl. 1), 150. (Abstr.)
[8] Cummings TS, http://www. Cvm. Msstate. edu/ Poultry/Documents/Micro flora 20 Management.pdf., **2003**, Accessed Jan.
[9] Dibner JJ, JD Richards, *Poult. Sci*., **2005**, 84, 634-643.
[10] Elwinger K, E Berndton, OP Engstr, O Fossum, L Waldenstadt, *Acta. Vet. Scand*., **1998**, 39, 433-441.
[11] Feighner SD, MP Dashkevicz, *Appl. Environ. Microbiol*., **1987**, 53, 331-336.
[12] Hansen JL, PB Moore, TA Steitz, *J. Mol. Biol*., **2003** 330, 1061-1075.
[13] Henry PR, CB Ammerman, RD Miles, *Poult. Sci*., **1986**, 65, 321-324.
[14] Ives SE, EC Titgemeyer, TG Nagaraja, Adel Barrio, DJ Bindel, LC Hollis, *J. Anim. Sci*., **2002**, 80, 3005-3015.
[15] Joerger R D, *Poult. Sci*., **2003**, 82, 640-647.
[16] Lee C, M Minami, S Sakuda, T Nihira, Y Yamada, 1996, *Antimicrob. Agents Chemother*., **1996**, 40, 595-601.
[17] Miles, R D, DM. Janky, RH. Harms, *Poult. Sci*., **1984**, 63, 1218-1221.
[18] National Research Council (NRC), National Academy Press, **1994**, 9, 157.
[19] Niewold TA, *Poult. Sci.*, **2007**, 86, 605-609.

[20] Parks C W, J L Grimes, P R Ferket, A S Fairchild, *Poult. Sci.*, **2001**, 80, 718-723.
[21] Proudfoot FG, ED Jackson, HW Hulan, CDC. Salisbury, *Poult. Sci.*, **1990,** 69, 1713-1717.
[22] SAS, *SAS Institute Inc.*, **2001**, USA.
[23] Singh M, RK Srivastava, SS Chauhan, KS Singh, *Indian J. Poult. Sci.*, **2000,** 35, 272-275.
[24] Zhou H, J Gong, JT Brisbin, H Yu, B Sanei, P Sabour, S Sharif, *Poult. Sci.*, **2007,** 86: 2541–2549.