Kufa Journal for Veterinary Medical Sciences Vol.(5). No.(2) 2014



Kufa Journal for Veterinary Medical Sciences www.vet.kufauniv.com



# Effect of probiotic "Vetlactoflorum" on some physiological parameters of broiler chickens

Al-Aqaby Aamer Rassam Ali \*,\*\*

Glaskovich A.A.<sup>\*</sup>

\*Vitebsk State Academy of Veterinary medicine, Vitebsk, Republic of Belarus \*\* College of Veterinary Medicine, Alqadisiya University, Aldewaniya, Republic of

Iraq

E-mail: <u>Alaqaby2004@yahoo.com</u>

## Abstract:

The objective of the present study was to investigate the effect of supplemental probiotic preparation **"Vetlactoflorum"** on some hematological and biochemical constituents of blood in broiler chickens. One hundred fifty broiler chickens breed "Ross-308" used in one day old, divided into three groups of 50 chicks (control "un supplemented probiotic ", first treatment group received "Vetlactoflorum-C" and second treatment group received "Vetlactoflorum-M"). Samples were taken in 7, 14, 21, 28, 35 and 42 day-old. Experimental study conducted to determine whether supplementation of probiotic could improve the blood characteristics of broilers chickens, shown that these selected probiotic organisms are able to effect on hematological and biochemical aspects, and also some minerals in broiler chickens.

Key words: Vetlactoflorum, hematological aspects,

الخلاصة:

تهدف هذه الدراسة الى نقييم تأثير اعطاء المعزز الحيوي "Vetlactoflorum" على بعض المكونات الدمية و الكيموحيوية للدم في دجاج اللحم. اخذت 150 فروج لحم من سلالة "روس-308" في عمر يوم واحد ، وقسمت إلى ثلاث مجموعات كل مجموعة نتكون من 50 فروجة لحم (مجموعة السيطرة لم تعطى المعزز الحيوي) ومجموعة المعالجة الاولى التي اعطيت المعزز الحيوي "Vetlactoflorum-Vetactoflorus" ومجموعة المعالجة الثانية التي اعطيت المعزز الحيوي" Vetlactoflorum . تم أخذ عينات الدم في عمر 7 و

2014

14 و 21 و 28 و 35 و 42 يوما.اجريت التجربة لتحديد فيما لو كان اعطاء المعزز الحيوي يستطيع تحسين المواصفات الدمية والكيموحيوية لفروج اللحم حيث اظهرت النتائج ان المعزز الحيوي قادر على التأثير على الصفة الدمية وبعض العناصر المعدنية في دجاج اللحم.

## Introduction:

For poultry many years, been industry has looking for improvement of production indexes and broiler growth through breeding changes or through improving quality nutrients. Researchers worldwide are working on organic alternatives due to the ban of a wide range of drugs for animal production. Probiotics are alternatives for that. In Greek Probiotic means "for life" (1) and can be defined as a live microbial feed supplements, which beneficially affects the host animal by improving its intestinal balance (2).

Probiotics are one of the approaches that have a potential to reduce chances of infections in poultry and subsequent contamination of poultry products. Probiotic foods have been consumed centuries. for either as natural components of foods. A food can be said functional if it contains a component (which may or may not be a nutrient) that affects one or a limited number of functions in the body in a targeted way so as to have positive effects on health (3). Amongst the most promising targets for functional foods gastrointestinal functions, are the including those that control transit time, bowel habits, and mucosal motility as well as those that modulate epithelial cell proliferation. Promising targets are also gastrointestinal

functions that are associated with a balance colonic microflora, that are associated with control of nutrient bioavailability (ions in particular), that gastrointestinal modify immune activity, or that are mediated by the endocrine activity of the gastrointestinal system. Finally, some systemic functions such as lipid indirectly homeostasis that are influenced by nutrient digestion or fermentation represent promising targets (4, 5).

Republic of Belarus apply program for using of safety products, so probiotic preparation "Vetlactoflorum" made by limited liability company «Microbiotic», city of Vitebsk, Belarus. However, given the preparation has not been studied previously in the broiler chickens. We were the first in Belarus began the study of the preparation, which was approved by the result of scientific and technical papers (6, 7, 8, 9).

Vetlactoflorum-liquid preparation of live probiotic acidophilus bacteria strain *Lactobacillus acidophylus* EP 317/402 "Narine" containing 1 cm<sup>3</sup> of not less than 10<sup>7</sup> colony forming units of Lactobacillus.

The aim of this study was to extend our research and examine the effect of a defined probiotic on erythrocyte count,

leukocyte count, hemoglobin, some minerals, and liver functions enzymes in broilers chickens.

#### Materials and methods:

Experimental study was conducted by our clinic Epizootology Department, Department of Microbiology and Virology, Veterinary and Sanitary Inspection and laboratory in Scientific Research Institute of Veterinary Medicine and biotechnology/Vitebsk State Academy of Veterinary Medicine. For laboratory study,150 broiler chicks breed "Ross-308" were used, this chicks divided into three groups 50 broiler chickens in each, acquired by "Vitebsk broiler chickens farm".The broiler chickens were reared under same environmental conditions, they were kept in thermo-neutral hall (approximately from day one old 33°C until final 19°C).

According to the experience chicks one day-old were sorted by gender, 25 chicks' males and females in each group. Feed and water were provided *ad libitum* 

Throughout the experiment, broiler chickens given probiotic with drinking water according follow:

№ group	Diet and treatment
Group 1	Basic Diet.
(control)	
Group 2	Basic Diet + probiotic "Vetlactoflorum-M"
	daily drinking water at a dose of 0.1 ml / bird (1-27day)
	and 0.2 ml / bird (28-42 days).
Group 3	Basic Diet + probiotic "Vetlactoflorum-C"
	daily drinking water at a dose of 0.1 ml / bird (1-27day)
	and 0.2 ml / bird (28-42 days).

Collection of samples, data collection and analysis. At 7, 14, 21, 28, 35 and 42 days of age, 4 ml of blood for laboratory analysis were obtained by axillary vein (with aseptic and antiseptic) and decapitation, in two different sterile tubes (10, 11). In one of them stabilize blood anticoagulant Trilon B (disodium EDTA) of 0.1 - 0.2 ml of 10% solution in 10 ml blood, the other was without anticoagulant to obtain serum.

Serum obtained after coagulation at a temperature of (18-20)°C, then put tubes in centrifuge at 1500 RPM/min for 10-15 minutes to separate the serum. The serum was separated, and

No. (2)

2014

then stored at -20°C until assayed to measure erythrocyte count, leukocyte count, hemoglobin, magnesium, calcium and phosphor, and liver functions enzymes (AST and ALT) in broiler chickens.

Analysis of blood cells was used fresh blood sample. The amount of hemoglobin determined by standardized colorimetric method using photoelectrcometer. Count the number of red blood cells and white blood cells was performed in a chamber Goryaeva (12).Aminotransferases (ALT and AST) were performed by the clinic method IFCC. The enzyme activity was expressed as IU / l. Determination of serum calcium was performed by colorimetric method with 0crezolftaline. Calcium concentration expressed in mmol / l. Determination of serum magnesium was performed by colorimetric method with EGTA, the value expressed in mmol / l. Serum inorganic phosphorus was determined by colorimetric method with molybdate reagent and without deproteinization. Phosphorus content was expressed as mmol / l. (10). The results of study analyzed statistically to find the significant differences among the control and the treated groups by Excel programme.

## **Results and discussion:**

Results summarized in <u>Table 1</u> show both probiotic treatment groups had higher Hb values than the control although the difference was significant (P<0.05) in  $2^{nd}$  group of broiler

chickens in 14 and 35 day old (P<0.05) , while in 28 day old in 3<sup>rd</sup> group (P<0.05), at 42 day old also in 3<sup>rd</sup> < 0.01). group (P However, supplementation of *Enterococcus* faecium may improve health condition increasing by concentration of hemoglobin, hematocrit and red blood cell count in broilers (13, 14). values Increased Hb might be attributed to the probiotics which might have increased haematopoiesis.

Vol. (5)

Regarding the number of white blood cells on the contrary, their number increased to 42 day in the control group - 7.59%, in the 2nd group - by 11.84%, in the third group- by 9.41% (P <0.05). A similar increase in WBC count was reported in birds given probiotic-supplemented diets (15). While (16) found that Enterococcus faecium M-74 strain did not cause significant change in total leucocytic count in turkeys. Shareef and Al-Dabbagh observed there were no significant changes between treatment groups and control (17). Mannaa et al. noted that the addition of probiotic to broiler feed resulted in significant concerning hematoimprovement biochemical parameters (18).According to (19) supplementation of probiotic preparations with normal iron causes intensification of erythropoiesis, consequence probably as а of hypersynthesis of erythropoietin. Nolte claims that probiotic have a beneficial influence on erythrocyte indices (20). Increased WBC might be related to the production of more immune cells (21) that play important role an in defending against different diseases. Paryad and Mahmoudi also reported higher WBC in animals fed different levels of probiotics than those fed diets without probiotic (22).

AGE (DAY)	Group	Hemoglobin	erythrocyte,	leucocyte
		(G/L)	×1012/L	×109/L
7	1 Control	109,6±5,980	2,07±0,0350	25,12±0,732
	2 Experimental	113,2±3,121	2,14±0,052	26,19±1,023
	3 Experimental	108,4±3,586	2,11±0,040	27,48±0,598
14	1 Control	111±0,894	2,072±0,038	25,97±0,298
	2 Experimental	117,6±1,208*	2,162±0,020	27,03±0,895
	3 Experimental	111,8±1,36	2,096±0,027	28,61±0,657
	1 Control	208,8±14,667	2,28±0,084	25,81±0,826
21	2 Experimental	204,20±4,487	2,29±0,056	25,82±0,846
	3 Experimental	184,60±18,556	2,37±0,046	25,79±0,889
	1 Control	118,21±5,953	2,26±0,214	26,34±0,714
28	2 Experimental	131,82±7,067	1,90±0,110*	27,88±0,496
	3 Experimental	136,42±8,195*	2,15±0,159	27,28±0,537
	1 Control	126,60±15,781	1,75±0,082	27,22±0,984
35	2 Experimental	150,20±10,25*	1,95±0,163	29,66±0,718*
	3 Experimental	134,80±14,001	2,26±0,207*	28,34±0,638
42	1 Control	113,00±9,675	1,97±0,114	27,93±0,851
	2 Experimental	118,93±8,877	1,97±0,183	29,26±0,528
	3 Experimental	119,71±3,877**	1,59±0,006**	28,47±0,683*

Table 1: Blood parameters in broiler chickens (X±m)

Means of mark

\*Significat different compare with control(P<0.05)

<sup>\*\*</sup>Significat different compare with control(P<0.01)

<sup>\*\*\*</sup>Significat different compare with control (P<0.001) 2<sup>nd</sup> Group–diluted in milk. 3<sup>rd</sup> Group–diluted in whey.

No. (2)

2014

Vol. (5)

Table 2 show that there is variation in calcium, magnesium and phosphor levels in treatment groups supplemented probiotic where higher than in control group. Calcium levels compared with initial values increased in all experimental groups. Since the control group content of this indicator has increased by 62.24%, in the  $2^{nd}$ group - by 79.77% (P <0.01), in the third group - by 69.23% (P <0.001), this may be due to the increase of nutrient absorption by intestine this renders suggest that probiotic enhance nutrient absorption level the in intestine. Blood serum is frequently used for mineral assessment because they are significantly correlated to nutritional status of some mineral elements (23. 24. 25). Calcium regulates the contraction and relaxation of muscle and regulates the passage of substance into and out of the cells. So,

calcium is one of the most important nutrients for aquatic species (26). Phosphor levels were high in 14, 21, 28, 35, 42 old-day in  $2^{nd}$  group. Magnesium levels were high in 7, 14, 28, 42 old-day in 2<sup>nd</sup> group, while were high in 21, 35 old-day in 3<sup>rd</sup> group. Grigorieva and Topuriya reported positive effect of probiotic preparation Olin on the level of mineral metabolism in broiler chickens (27). Our results disagree with (28, 29) who reported unchanged serum minerals in lambs fed diets supplemented with or without probiotics. Usage of feed additive had positive effect on level of these minerals in blood. It has been reported that probiotics increase the calcium absorption from intestinal tract. Fermentation products as a result of probiotics' activity may increase the absorption surface by accelerating proliferation in enterocytes (30).

AGE	Group	Magnesium	Ca	Р
(DAY)		(Mmol/L)	(Mmol/L)	(Mmol/L)
7	1 Control	1,09±0,061	2,65±0,0605	3,84±0,3480
	2 Experimental	1,11±0,047	2,67±0,095	2,84±0,144*
	3 Experimental	1,02±0,054	2,44±0,093	3,06±0,104*
14	1 Control	1,09±0,068	2,71±0,157	0,97±0,099
	2 Experimental	1,11±0,031	$2,84\pm0,090$	1,046±0,050
	3 Experimental	1,06±0,042	2,81±0,096	1,022±0,021
21	1 Control	0,63±0,051	$1,31\pm0,100$	1,11±0,062
	2 Experimental	0,74±0,033	$0,89{\pm}0,079$	0,87±0,103
	3 Experimental	0,76±0,046	1,12±0,089	1,25±0,139
28	1 Control	$0,79\pm0,059$	1,28±0,0693	0,96±0,067
	2 Experimental	0,98±0,037	$1,47\pm0,138$	1,12±0,062*
	3 Experimental	0,91±0,031**	1,83±0,108**	1,08±0,083
35	1 Control	0,85±0,059	1,66±0,157	1,46±0,218
	2 Experimental	$0,85\pm0,047$	$1,59\pm0,278$	1,72±0,36
	3 Experimental	0,88±0,039	$1,55\pm0,200$	1,21±0,156
42	1 Control	0,86±0,0635	3,47±0,573	1,56±0,177
	2 Experimental	0,97±0,071	4,40±0,817	1,27±0,040*
	3 Experimental	0,90±0,050	3,64±0,179	1,38±0,152

<b>Fable 2: Show concentratior</b>	of minerals in serum	broiler chickens (	X±m)
------------------------------------	----------------------	--------------------	------

Means of mark

\*Significat different compare with control(P<0.05)

\*\*Significat different compare with control(P<0.01)

- \*\*\*Significat different compare with control (P<0.001)
- 2<sup>nd</sup> Group–diluted in milk.
- 3<sup>rd</sup> Group–diluted in whey.

#### **Conclusion:**

Concluded from our results, there was using improved probiotic by Vetlactoflorum-M" (dissolved in diluted milk) and "Vetlactoflorum-C" (dissolved in whey). From these results we can proved that probiotic did not induce any harmful effect on liver. Probiotic can be improving health condition by increasing concentration hemoglobin, of erythrocyte and leukocyte count in broilers.

#### **REFERENCES:**

- 1. Gibson,G.R. and Fuller,R. (2000). Aspects of in vitro and in vivo research approaches directed toward identifying probiotics and prebiotics for human use. J. Nutr., 130: 391-395.
- Fuller, R.(1989). Probiotics in man and animals. J. Appl. Bacteriol.66: 365-378.
- Bellisle,F. ; Diplock,A.T. and Hornstra,A.T. (1998). Functional food science in Europe. Br. J. Nutr., 80:3-4.
- 4. Clydesdale, F.A.(1997). Proposal for the establishment of scientific criteria for health claims for functional foods. Nutr. Rev., 55: 413-422.
- 5. Roberfroid, M.B. (1996). Functional effects of food components and the gastrointestinal chicory system: fructo-oligosaccharides. Butr. Rev.54:38-42.
- 6. Glaskovich, A.A.; Kapitonova,E.A.; Pritychenko, A. and Alaqaby, Aamer R.A.(2012).

The dynamics of the natural resistance of broiler chickens supplemented probiotic "Vetlactoflorum". Scientific notes: Sci. and prac.J./Vitebsk State Academy of Vet.Med.48(1):56-61.

- 7. Evshel, VA.; Kapitonova, E.A. and Glaskovich, A.A.(2012). Natural resistance of broiler chickens through supplementation of probiotics "Vetlactoflorum-C" and "Vetlactoflorum-M".Studentsscience and practice of agriculture: Proceedings the of 97th International Scientific Conference (Vitebsk on 22-23 May 2012) / Vitebsk State Academy of Vet.Med.179.
- 8. Ryzhik, Y.L.; Golubitskaya, A.V.; Glaskovich, A.A.; Kapitonova, E.A. and Alaqaby, Aamer R.A.(2012a). Technical conditions BY 391043609.008-2012. Feed additive biological active "Vetlactoflorum". State registration of Belarus No.034955 in 03.05.2012. The State Committee Standardization of the for Rep.Belarus.9p.
- Ryzhik,Y.L.; Golubitskaya,A.V.; Glaskovich,A.A.; Kapitonova,E.A. and Alaqaby, Aamer R.A.(2012b). Instruction for preparation use BY 391043609.008-2012.Feed additive biological active "Vetlactoflorum" State registration No. 034955 in 03.05.2012, the State Committee for Standardization of the Rep.Belarus.2p.
- 10. Dubina, I.N.; Kurdeko, A.P.; Fomchenko, I.V. and Smilgin,

I.I.(2008). Guidance on the biochemical blood animals using diagnostic kits: approved in Ministry of Agriculture and Food. Rep.Belarus in 27.11.2007. № 10-1-5/914. Vitebsk: Vitebsk state Academy of Vet.Med. 60p.

- Kurdeko,A.P.;Kovalenok,U.K.;De midovich,A.P.;Bratushkina,E.L.;Ve lecanov,V.V.; Ulyanov, A.G.; Kurilovich, A.M. and Makaruk, M.A. (2008).Take blood from animals: Manual of instructional method. Vitebsk: Vitebsk state Academy of Vet.Med. 36p.
- Gusakov, V.K., Kudryavtceva, E.N. and Ostrobskii,A.V. (2002). Counting blood cells in chickens (text), manual of teaching method. Vitebsk, Belarus.18p.
- Cetin, N.; Guclu, B.K. and Cetin, E.(2005). The effect of probiotic and mannanoligosaccharide on some haematological and immunological parameters in Turkeys. J. Vet. Med., 52: 263-267.
- Strompfova, V.;Marcinakova, M.;Si monova, M.;Gansarcikova, S.;Jonec ova, Z.;Scirankova, L.; Koscova, J.;Buleca, V.;Cobanova, K. and Laukova. (2006). *Enterococcus faecium* EK 13- an enterocin Aproducing strain with probiotic character and its effect in piglets. Anerobe, 12: 242-248.
- 15. Rahimi, S. and Khaksefidi, A.(2006). A comparison between the effect of a probiotic (Bioplus 2B) and an antibiotic (virginiamycin) on the performance of broiler chickens under heat stress condition. Iranian J. Vet. Res., 7: 23-28.
- 16. Marcela, C.; Kolesarova, A.; Massanyi, P. and Kovacik, J.(2008). Selected Blood Biochemical and Haematological Parameters in Turkeys after an

Experimental Probiotic Enterococcus faecium M-74 Strain Administration.

Inter.J.Poul.Sci.,12:1194-1199.

- 17. Shareef,A.M. and Al-Dabbagh,A.S. (2009). Effect of probiotic (*Saccharomyces cerevisiae*) on performance of broiler chicks. Iraqi J.Vet.Sci.,1: 23-29.
- Mannaa, F.; Ahmed, H.H.; Estefan,S.F.; Sharaf, H.A. and Eskander, E.F.(2005). Saccharomyces cerevisiae intervention for relieving flutamideinduced hepatoxicity in male rats. Pharm., 60, 689-695.
- 19. Sjaasted,Q.V.; Franstad,T. and Blom,A.K.(1996). Effect of iron on erythopoietin production in anemic piglets. Acta. Vet. Scand. 37:133-138.
- 20. Nolte,K.(1994). Comparative studies on the use of Streptococcus faecium M 74 and parenteral and oral iron supply in sucking piglets. Tierarztliche Hochschule Hanover, 109.
- 21. Lafleur-Brooks, M. (2008). Exploring Medical Language: A Student-Directed Approach, 7th Edition. St. Louis, Missouri, USA: Mosby Elsevier. p. 398.
- 22. Paryad, A. and Mahmoudi, M. (2008). Effect of different levels of supplemental yeast (Saccharomyces cerevisiae) on performance, blood carcass characteristics of broiler chicks, African J. Agricultural Res., 3(12): 835-842.
- 23. Claypool,D.W.; Adams,F.W.; Pendell,H.W.; Hartman,Jr.; Bone,J.F. (1975). Relationship between the level of copper in the blood plasma and liver of cattle. J. Anim. Sci. 41:911–914.

2014

- 24. Levander, O.A.(1986). In: Mertz, W. (Ed.), Trace elements in human and animal nutrition, vol. 2. Academic Press, New York:209– 279.
- 25. Mills, C.F. (1987). Biochemical and physiological indicators of mineral status in animals: copper, cobalt and zinc. J. Anim. Sci. 65:1702–1711.
- 26. Zimmermann, S.; Leboute, E.M. and de Souza, S.M.G.(1993). Effects of different calcium levels in feed and water on growth of freshwater prawn, Macrobrachium rosenbergii Man), postlarvae 19:183, (de Abstracts of contributions presented at the International Conference on World Aquaculture '93, Torremolinos, Spain, May. pp: 26-28.
- 27. Grigorieva, EV and Topuriya, LY .(2011). State of mineral metabolism in broiler chickens under the influence of probiotic Olin. Bulletin of veterinary medicine.4:128-129.

28. Antunovi,Z.; Peranda Marcela; Eri.V.: Liker. B.: Sen..I.: Domainovi M. and Peranda, T. (2005). Influence of feeding the probiotic pioneer pdfm® to growing lambs on performances composition. and blood Acta Veterinaria (Beograd), 55,4:287-300.

No. (2)

Vol. (5)

- 29. Masek, T.; Mikulec, Z.; Valpotic, H.; Antunac,N.; Mikulec,N.; Stojevic,Z.; Filipovic,N. and Pahovic S (2008). Influence of live veast culture (Saccharomyces cerevisiae) on milk production and composition, blood and biochemistry of grazing dairy ewes during the milking period. Acta Vet Brno.77:547-554.
- 30. Scholz-Ahrens, KE, Ade P, Marten B, Weber P, Timm W, Açil, Y, Glüer CC, Schrezenmeir J. (2007). Prebiotics, probiotics, and synbiotics affect mineral absorption, bone mineral content, and bone structure. J.Nut.137:838-846.

142