International Journal of Agricultural Science and Research Volume 3, Number 1, Winter 2012 (Serial #6)

A survey on *Chlorella vulgaris* effect's on performance and cellular immunity in broilers

M.Rezvani* morvarid.rezvani@gmail.com, M.Shivazad shivazad@ut.ac.ir, M. Zaghari mzaghari@ut.ac.ir, H. Moravej hmoraveg@ut.ac.ir

Department of Animal Science, Faculty of Agriculture, University of Tehran, Karaj, Iran

Received: February, 20, 2012 *Accepted: June, 13, 2012*

ABSTRACT

In order to evaluate the effects of chlorella on broiler performance and immune system, an experiment was performed by using 80 ROSS 308 male broiler chicks. The experiment was accomplished in completely randomized design including 5 treatments with 4 replication and 4 observations in each replicate. The basal diet was based on corn-soybean meal (control) and without any additives. A graded level of chlorella (0.07, 0.14 and 0.21%) was added to basal diet to formulating diets 2, 3 and 4 respectively. An extra diet (diet 5) was formulate by adding a commercial prebiotic to the basal diet. Results showed that feed conversion ratio significantly decreased by adding the chlorella and commercial prebiotic to the basal diet at 42 d of age ($p<0.05$). chlorella treatment had numerical increase in response to phitohemoglotenine-P at 35 d. Results of this study showed that chlorella could improve performance and may increase cell immune response.

Keywords: chlorella, broiler chicks, feed conversion, immune system,

INTRODUCTION

Over the last years, intense genetic selection has been performed to increase weight gain. Although this genetic selection has many advantages like more performance, lead to some qualities like immune becomes faint [5]. Thus, nowadays hypersensitivity to diseases is the most important problem in broilers farms. In this way annually, different kinds of diseases are occurred in flocks that are caused lots of mortality and losses.

Digestive system completes immune system in poultry thus each alteration in intestine environment which occurred by feed could affects mucosal immune system in poultry [9]. Therefore, using nutritional methods to support immune system could be a useful tool. Utilization of feed additives is the one of the most important methods for instance using prebiotic is profitable in broilers because prebiotics by adhesion to intestine lymphoid tissue could stimulate mucosal immune system.

In addition to prebiotic, using plants extract as a feed additive in poultry diet were studied in many researches and indicated that it causes many benefit such as improvement in performance and immune system [10].

Chlorella is a green microalgae that contain plentiful nutrients including essential amino acids and vitamins [3]. According to compound of chlorella is suppose that it may have some effects like prebiotic and plants extract effects. In this survey, effects of using chlorella on performance and immune system in broilers have been rewired.

Material and methods

Housing and experimental design

80 Ross 308 one-day-old male broiler chicks obtained from a commercial were used in this study. They are housed in cages (90 × 78 × 40 cm) individually. The experiment was performed in Poultry Research Laboratory Animal Science Department of University of Tehran. The chicks were divided to 5 groups with 4 replications and 4 observations in each replicate.

The 5 dietary treatments were fed Standard broiler basal diets (3-phase) based on corn-soybean meal without any additives as a control diet , a graded level of chlorella (0.07, 0.14 and 0.21%) was added to control diet as treatment 2, 3 and 4, respectively and an extra diet to comparing the effects of chlorella with commercial prebiotic as treatment 5. Treatment 5 was formulated by adding a commercial prebiotic (mananoligosacharid) to the basal diet. The diets compounds were presented in table 1. Spray dried chlorella was purchased from microalgae sina Company, Iran. The chlorella compound is presented in table 2. All clicks were provided the diets and water ad libitum and were received 24 h of light during the experiment. The experimental period lasted 42d.

Weight gain, feed intake and food conversation ratio was monitored weekly during the experiment.

Table 1. Composition of the basal diet fed to broilers (gr/100gr)

Ingredient and Composition	Starter (0-10d)	Grower (11-28d)	Finisher (29-42d)
Ingredient % as fed			
Corn	60.04	54.45	52.05
Soybean meal	29.27	34.21	36.28
Gluten	1.33	1.76	3.62
corn oil	5.42	5.53	3.48
Dicalcum phosphate	1.86	1.83	2.08
oyster	0.97	1.07	1.2
Salt (NaCl)	0.34	0.34	0.34
Vitamin Permix1	0.25	0.25	0.25
Mineral Permix2	0.25	0.25	0.25
DL-Methionine	0.16	0.17	0.16
L-Lysine	0.11	0.14	0.29
Nutritional content (calculated)			
ME, Kcal/kg	3200	3150	3010
CP	19	21	23

1-Vitamin premix provided per kilogram of diet: vitamin A, 9000 IU; vitamin D3, 2000 IU; vitamin E, 18 mg; vitamin B12, 0.015 mg; thiamine, 1.8 mg; riboflavin, 6.6 mg; pyridoxine, 2.9 mg; nicotinic acid, 30 mg; pantothenic acid, 25 mg; folic acid, 1 mg; cholin,500 mg.

2- Mineral premix provided per kilogram of diet: Fe, 50 mg; Cu,10 mg; Zn, 84 mg; Mn, 99 mg; Se, 0.2 mg; I, 0.99.

Evaluation of immune response

On 35 d of age, two chicks from each replication were randomly selected to testing their cell immune response. For this aid, at first the thickness of toe membrane in right food was measured and recorded then 100 micro liter phitohemoglotenine-P was injected into

there. After 24 h, the difference of membrane between toes thickness was calculated for each bird. Inflammation of membrane that is between the bird toes 24 h after injection 100 microgram phitohemoglotenine-P was considered as a lymphocyte proliferation response index [7].

Statistical Analyses

The experiment was accomplished in a completely randomized design with 5 treatments and 4 replication. Data was analyzed by using GLM procedure of SAS software. Duncan multiple range test compared means. The differences were considered significant at $P<0.05$ [8].

Reasults

The results of weight gain are presented in Table 3. Results showed that there were no significant differences in weight gain between the treatments in all ages. The results of feed conversion ratio are presented in Table 4. Feed conversion ratio significantly decreased by adding the chlorella and commercial prebiotic to the control diet at 42 d of age ($p<0.05$). There was no mortality during the experience period.

Table 3 . Effect of dietary treatment on weight gain of broiler chickens up to 42 d age (gr).

42 day	35 day	28 day	21 day	14 day	7 day	Treatment
2694.7	2036.9	1443.1	905.9	431.3	172.5	Control
2723.1	1988.4	1473.7	917.7	442.8	171.1	C[1]+0.07% chlorella
2755.0	2117.1	1483.3	949.7	475.7	170.7	C+0.14% chlorella
2850.8	2122.1	1490.4	904.0	438.2	176.2	C+0.07% chlorella
2740.6	2052.8	1437.8	883.0	433.9	172.2	C+ prebiotic
19.5	24.5	16.1	11.7	8.0	5.2	SE
0.13	0.43	0.82	0.55	0.90	0.98	P- value

C= control

Table 4. Effect of dietary treatment on FCR of broiler chickens up to 42 d age.

42 day	35 day	28 day	21 day	14 day	7 day	Treatment
1.60 [a]	1.45	1.30	1.11	1.07	0.96	Control
1.55 [b]	1.44	1.29	1.12	1.06	0.95	C[1]+0.07%chlorella
1.54 [b]	1.41	1.27	1.12	1.07	0.94	C+0.14%chlorella
1.54 [b]	1.44	1.26	1.11	1.06	0.97	C+0.07%chlorella
1.55 [b]	1.44	1.28	1.13	1.03	0.98	C+ prebiotic
0.007	0.006	0.008	0.004	0.007	0.02	SE
0.03	0.55	0.64	0.67	0.63	0.96	P- value

[a-b] Means with in columns with different superscript differ significantly ($P<0.05$) . 1C= Control

The results of phitohemoglotenine-P test were summarized in figure 1. Evaluation of cell immune response test showed a numerical increasing in lymphocyte proliferation response as a cell immune response index by increasing chlorella and commercial prebiotic to the control diet.

Table 5: Effect of adding chlorella and commercial prebiotic on cell immune response at 35 day.

phitohemoglotenine response	Treatment
1.36	Control
1.39	C[1]+0.07%chlorella
1.51	C+0.14%chlorella
1.54	C+0.07%chlorella
1.45	C+ prebiotic

0.043	SE
0.687	P- value

[a,b] Means within rows with different letters are significantly different ($P < 0.05$).

Discussion

In poultry industry, feed costs consist of 60-70% of the total costs. Thus, use of various feed additive caused reduction in feed conversion ratio could decrease the expenses. According to these results chlorella diets could decrease feed conversion ratio and could use as a beneficial additive in poultry nutrition.

The better feed conversion ratio in chlorella and prebiotic treated broilers could be related to better equilibrium intestine flora [1]. Chlorella has a specific cell wall that contain mananoligosacharid compound these compounds are found in lots of commercial prebitic that called MOS prebiotic.

These findings are in agreement with those of many previusing studies. Benites, V et al. (2008) reported that broiler chickens fed a diet supplemented with prebiotic (mananolisacharid) had a significant decrease in the feed conversion ratio in camper to control group [2].

The effects of feeding diet containing prebiotic on the immune response are related to enhancement of equilibrium intestine flora. The direct effect might be the lymphatic tissue stimulation [1]. The intestinal flora are in close contact with immune system cells.

The immune system is made up of a network of special cells, proteins, tissues, and organs that work together to defend broiler against germs and microorganisms. The cells include white blood cells, or leukocytes. The leukocytes circulate through the body and house in lymph nodes via lymphatic vessels and blood vessels. In this way, the immune system can monitor the body for germs or substances that might cause problems. The two basic types of leukocytes are phagocytes and lymphocytes. lymphocytes allow the body to remember and recognize previous invaders and help the body destroy them. The most common phagocytes type is the neutrophil, which primarily fights bacteria. The lymphocytes are two types; B lymphocytes and T lymphocytes. T Lymphocytes primary were formed in the bone marrow then they leave there to the thymus, where they mature into T cell [9].

B lymphocytes are triggered When antigens (foreign substances that invade the body) are detected. They produce antibodies, specialized proteins that lock onto specific antigens. Antibodies recognize an antigen and lock onto it but they are not able to destroying it solitarily.

T cells destroy antigens that have been tagged by antibodies or cells that have been infected or somehow changed. Thus, some T cells are actually called "killer cells." T cells also are helping other cells (like phagocytes) to do their functions. All of these specialized cells and parts of the immune system protect body against disease [6].

There are many interactions between immune responses for instance antibodies can activate a group of proteins called complement. Complement supports killing bacteria, viruses, or infected cells by T cell. Thus improvement in cell immune by using chlorella supplemented diets, could improve humeral

immune too. We suppose that humeral response will be examined in former researches.

Selenium is a vital trace mineral for poultry that lymphatic organs is the one the its target organs. According to formerly studies selenium increase burse and thymus weight. In addition, supplemented diet by selenium could increase immunity against Coccidiosis in broilers [4].

Chlorella is reach in selenium [3]. Results indicate numerical improvement in cell immune in our study so it could be related to high amounts of selenium in Chlorella.

Each kind of improvement in immune system could cause increase bioenergetic efficiency in body so increase body weight and decrease feed conversion ratio. Therefore it appears logical that performance increase in Chlorella and commercial treatments.

By consideration to these results, we suggest using higher levels of chlorella in former studies.

Acknowledgments

The authors thank Animal Science Department of Tehran University for providing Poultry Research Laboratory. to this experiment is performed. They thank microalgae sina Company for providing spray dried Chlorella

References

1-Bedford, M., 2000. Removal of antibiotic growth promoters from poultry diets: implications and strategies to minimize subsequent problems; World poultry science Journal; 56:347-365.

2-Benites, V., Gilharry, R., Gernat, A. G., and J. G. Murillo, 2008. Effect of Dietary Mannan Oligosaccharide from Bio-Mos or SAF-Mannan on Live Performance of Broiler Chickens. Poultry Science Association. 17:471–475

3-Halle, I. P.Janczyk , G.Freyer , and W. B. Souffrant, 2009.Effect of microalgae Chlorella vulgaris on lying hen performance ; Archiva Zootechnica 12:2, 5-13.

4-Hussain, M.I., Khan, S.A., Chaudhary, Z.I., Aslam, A., Ashraf, k., and M.F. Rae, 2004. Effect of organic and inorganic selenium with and without vitamin on immune system of broilers. Pakistan Vet. J., 24(1).

5-Manzoor, A. C., A. Qureshi, and G. B. Havenstein, 2003. Acomparison of the immiune profile of commercial broiler strains when raised on marginal and high protein diets. International Journal of Poultry Science. 2(5): 300-312.

6-Merlino, P. G., and Marsh, J. A. 2002. The enhancement of avian NK cell cytotoxicity by thymulin is not mediated by the regulation of IFN-gamma production. Developmental and Comparative Immunology, 26:103-110.

7-Rosa, P., Pesti, G. M., Edwards, Jr., and R. I. Bakalli, 2001. Threonine requirements of diffrerent broiler genotypes. Journal of Poultry Science, 80:1710-1717.

8-SAS Institute, 2000. The SAS System for windows 2000. Release 8.1. SAS Inst. Inc. Cary. NC.

9-Sharma, J.M. 1991. Overview of the avian immune system. Journal of Veterinary Immunology and Immunopathology. 30: 13-17.

10-Willis, W.L., Isikhuemhen, O. S., and S.A Ibrahim, 2007. Performance assessment of broiler chickens given mushroom extract alone or in combination with probiotics. Poultry Science. 86:1856-60.

11.Nicoleta, B. F. and I.M. Pop. 2010. Researches on the use of feed additive baby C4 in broiler chicken diets. Lucrări Ştiinţifice - vol. 53.