



Study Immune and productive treatise for broiler vaccinated with infectious bursal disease vaccine (IBD vaccine) after added boron powder to the feed

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Abstract

This study was performed to investigate changes in growth performance, blood Pictures, weights of internal organs and immune response to infectious bursal disease (IBD vaccine) of broilers fed different levels of Boron (100mg, 200mg and 300mg/kg) in feed from 1 week to 4 weeks. 150 broiler chicks (one day old) were randomly divided into five equal groups (each group contain 30), as following: first group (G1) was received 100 mg/kg of Boron, second group (G2) was received 200 mg/ kg of Boron, third group (G3) was received 300 mg/ kg of Boron and fourth group (G4) was not received Boron in feed Just take IBD vaccine and consider as control positive, (G5) was not received Boron in feed and consider as control negative, four groups only vaccinated with IBD vaccine at 10 and 18 days old, Growth performance of the chicks (Body weight, weight gain and feed intake) were weekly determined, at the end of experiment weights of internal organs (kidney, spleen and bursa of fabricius) were determined and Blood samples were collected to measure antibody titers (Ab) titers against IBD vaccine by ELISA test, total protein and determined blood pictures (RBCs, PCV, WBCs, H/L ratio), The results of this study showed that body weight, weight gain, feed intake, total protein, blood pictures and Ab titers were highly significant increase ($p < 0.01$) in G2 as compared to control group followed by G1 respectively in all weeks. Whereas the weights of internal organs (kidney, bursa of fabricius and spleen) were significant increase in G3, G2 and G1 respectively. In conclusion, the results indicated that 100mg/kg feed, 200mg/kg feed levels of Boron supplemented diet of broiler chicken caused improvement of blood parameters growth performance, there's no inhibition of the immunity response of chickens against IBD vaccine.

Keywords: immune response, Boron, growth performance, broiler, blood cells

دراسة مناعية وانتاجية لافراخ فروج اللحم الملقحة بلقاح كمبورو بعد اضافة عنصر البورون الى العليقة

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الخلاصة

أجريت هذه الدراسة لمعرفة التغيرات في أداء النمو، والصورة الدموية والاوزان النسبية للأعضاء الداخلية والاستجابة المناعية للافراخ الملقحة بلقاح الكمبورو حيث تم اضافة مستويات مختلفة من البورون للعلف (100 ملغم /كغم. علف ، 200 ملغم/كغم. علف و 300 ملغم / كغم. علف) من عمر 1 أسبوع إلى 4 اسابيع . تم تقسيم 150 الافراخ من عمر يوم واحد عشوائيا إلى خمس مجاميع متساوية (كل مجموعة تحتوي على 30 فرخ)، على النحو التالي: المجموعة الأولى (G1) (100 ملغم / كغم من البورون، المجموعة الثانية (G2) (200 ملغم / كغم. علف)، المجموعة الثالثة (G3) (300 ملغم / كغم. علف) والمجموعة الرابعة (G4) لم تعطى بورون وعدت مجموعة سيطرة موجبة (فقط لقاح) . المجموعة الخامسة (G5) عدت مجموعة سيطرة سالبة ولم تعطى اي شئى ، تم تلقح الافراخ بلقاح الكمبورو بعمر 10 و 18 يوما من التجربة ، وقد تم دراسة المعايير التالية (وزن الجسم وزيادة الوزن واستهلاك العلف) أسبوعيا على طول فترة التجربة ، وكذلك الاوزان النسبية للأعضاء الداخلية (الكلىة والطحال وجراب فابريشيا) وتم اجراء الفحوصات لبعض الصفات الدموية (حساب العدد الكلي لخلايا الدم الحمر RBC وخلايا الدم البيض WBC وحجم خلايا الدم المرصوصة PCV ونسب الخلايا المتغيرة للخلايا اللمفية H/L ratio وتركيز البروتين الكلي total protein) وكذلك قياس معيار الاضداد المناعية للقاح مرض الكمبورو بواسطة اختبار ELISA ، أظهرت نتائج وزن الجسم، الزيادة الوزنية، استهلاك العلف، و الصورة الدموية وكذلك الاستجابة المناعية تفوق مجموعة G2 بالمقارنة مع مجاميع السيطرة تليها G1 و G3 على التوالي في جميع الأسابيع. في حين اظهرت نتائج الاوزان النسبية الأعضاء الداخلية وجود زيادة معنوية واضحة في المجاميع G3 و G2 و G1 على التوالي حيث اظهرت نتائج هذه الدراسة انى التراكيز (100 ملغم/ و 200 ملغم) المضافة من مادة البورون الى علف الافراخ كانت ذات تاثير ايجابي على الصفات الدموية والاداء الانتاجي والاستجابة المناعية.

Introduction

For very long time period, it is well known that boron is an essential element for higher plants. Lately, it has been understood that boron also plays a role in the metabolism of minerals enzymes and steroid hormones in animals and humans (1, 2). Thus boron's distinctive chemical properties allow it to form complexes with organic molecules containing hydroxyl groups, and therefore to interact with various metabolites and enzymes to influence cellular activity (3). However, many questions still have to be answered in all segments of the poultry industry. In the field of animal nutrition, the role of some trace minerals is not completely elucidated and contradictory results are reported in the literature. In this context, some

studies focused on the micro-element boron, which is considered essential for superior plants but not for men or animals (4)

Although boron is not considered essential for poultry or most animals the addition of 2 ppm in poultry feed was recommended by (National Research Council, NRC) (5) Several studies have indicated that Boron is an important mineral for body weight, feed consumption, reduced mortality rate, normal cartilage, and bone formation in broilers; and for egg production, egg weight, and egg shell quality in laying hens (6-7) .

The objective of the present study was to determine the effects of boron diet supplementation in different levels on the performance of growth and, blood pictures, weight of internal organs and

immune response of broiler to infectious bursal disease vaccine.

Materials and Methods

- Chicks: The experiment was done at the Poultry house at the department of Veterinary Public Health, College of Veterinary Medicine-Baghdad University, after cleaning and disinfecting, Feeders and water utensils were cleaned and disinfected, newly hatched chicks of the Ross breed, were brought in good condition from Al-

Gamiyeh Hatchery-Baghdad-Abu-Ghraib. Chicks were vaccinated with IBD vaccine (vibro – USA) by manual oral drench at 10 days old, and 18 days. The basal diet was formulated for chicks according to National Research Council (8). Table (1) shows the ingredients and calculated chemical analysis of this diet, Boron was weighted individually and supplemented to diet in homogenous form.

Table (1) Ingredients of feed used in this study

Constituents	Percentages (%) of ingredients in D1
Wheat	10
Yellow corn	48
Soybean meal(44%protein)	25
Animal Protein (50% protein)	10
Vetivert	6
Calcium (CaCo ₃)	0.65
Salt (NaCl)	0.35
Total	100%

Design of Experiment: Boron powder was at the level (100,200, 300 mg/kg. feed) was tested at first day to the end experimental period (30 days). A total of 150 chicks at age of one day old were divided randomly into five groups each group have 30 chicks as follow:

- Group 1: broiler fed boron (100 mg /kg. feed) +IBD vaccine
- Group 2: broiler fed boron (200 mg/kg. feed) +IBD vaccine
- Group 3: broiler fed boron (300 mg/kg. feed) +IBD vaccine
- Group 4: controlpositive (IBD vaccine).
- Group 5: Control negative chicks fed the basal diet (no boron, no vaccine).

Parameters included in this study:

Live body weight: The weights of each group of chicks separately were taken at weekly intervals by weighting chicks individually at the end of every week and the total weights of all chicks were divided by the number to find the average weight of the chicks in gram (9).

Weekly body weight gain: The weight gains were calculated depending on the difference in body weight between the beginning of the week and the end of it, depending on the following equation:

Weekly body weight gain = Body weight at the end of the week - body weight at the beginning of the week (9).

Feed Intake (F.I.): Feed intake was calculated weekly depending on equation mentioned by Al-Zubaydi,(9) that depend on weighing the remained feed at the end of

each week period and subtracted from the total quantity offered at the beginning of the week taking in consideration the number of the dead chicks and the number of their feeding days according to this equation:

$$\text{Weekly feed intake (g. /broiler)} = \frac{W}{L + D} \times 7$$

In which:-

W = quantity of feed intake weekly for the whole group (g.).

L = number of alive chicks fed through a week x 7.

D = number of dead chicks x number of their feeding days.

Blood sampling for Serology and Hematological tests: Blood samples were randomly collected at the end of the experiment from (20) chicks from each group. The procedure of blood collection was carried out by slaughter of chicks followed and blood samples were taken to determine antibody titers against IBD vaccine in blood serum by using ELISA test (Synobiotics-USA), Total red blood cells counting RBC (cell/ mm³), Total White Blood cells counting WBC (cell/mm³), Packed cell volume PCV%, were measured by hemocytometer method described by (10), also measured of H/L ratio was measured (11) total protein count (12)

Estimation of relative weights of spleen, kidney, and bursa weight to the total body weight (relative weight): At the end of each experiment 20 birds were chosen from each group randomly and their live weight was taken then slaughtered, and samples from the spleen, kidney, bursa of Fabricius were collected after slaughtered the chicks. Carcass chicks were cleaned and weighted. The relative weight of each organ was measured according to the following equation:

organ was measured according to the following equation:-

Weight of organ

$$\text{Relative weight of the organ (\%)} = \frac{\text{Weight of organ}}{\text{Weight of live bird}} \times 100$$

Weight of live bird

Statistical analysis:-

Use the statistical program Statistical Analysis System - SAS (13) in the analysis of data to study the effect of various Groups in the traits according to a random design full (CRD), and compared the differences between the moral test averages Duncan (1955) polynomial

Mathematical model:

$$Y_{ij} = M + T_i + e_{ij}$$

Results

Results showed a significant increase (p <0.01) in Rate of the body weight, weight gain between groups Treated with Boron. Where It was highly In the group G2 and G1 and G3 On Respectively (Table no. 2, 3) There was no significant difference between the G2, and G1 In the third

week and fourth week. With regard to feed consumption, there were a significant increase on the level of probability (p <0.01) in the G2 compare with the G1 and G3. While did not Show and fourth week (Table no. 4). As well as the results showed the presence of a significant increase on the level probability (p <0.01) in

G2 compare with G4 and G5 for all Weeks. For blood pictures results (table no. 5). G2 appear highly significant level (P <0.01). In rate RBCs, WBCs, P.C.V compare With G1 and G3 On Respectively . As well as a significant decrease at the rate of the H / L ratio showed compare with the other groups. Where was the highest in the amount ratio H / L ratio the G4. When compare the G2 With Control groups (G4 and G5) of the blood pictures Observed Existence increase Level of total protein (P <0.01) on length Period Experience (Table no.6), for the measure of total protein G2 showed a significant increase on the level of a clear probability (p <0.01). Compared with the others groups , where the highest in the G2 , followed by G1, G3, G4 and G5 respectively . there was no significant difference for measuring total protein concentration when comparing between G3 and G5 (Table

no.6). The effect of boron on the relative weights of internal organs (spleen, bursa of fabricius, kidneys), these results showed the presence of a significant increase on the level of probability (p <0.01) G3 ,G2 and G1 compare with control groups. And also there was a significant difference on the level of probability (p <0.01) for the Relative weights bursa of fabricious where the highest in Group 4 compared with the G3, G2 and G1, respectively, (Table no.7). The effect of boron added to the diet. Its raising immune response of birds vaccinated with IBD vaccine where the results showed significant increase in the level probability (p <0.01). In (AB titer) where It was highly appear In the G2 Then G1 and G3 Respectively compare With the control groups (G4,G5) (table no. 8).

Table (2). The effect of Boron Supplemented formula &IBD vaccine on the rate of body weight for different weeks.

GROUPS	Average ± standard error (g/chicks)			
	first week	second week	third week	fourth week
Boron100 mg / kg feed	137.28 ± 1.85 b	364.07 ± 0.54 b	714.09 ± 1.42 c	1174.76 ± 0.97 b
Boron200 mg / kg feed	154.09 ± 0.74 a	406.08 ± 1.34 a	732.82 ± 13.66 b	1195.13 ± 1.89 a
Boron300 mg / kg feed	121.01 ± 1.56 d	314.09 ± 1.63 c	760.45 ± 0.71 a	1142.62 ± 2.46 c
control positive (vaccine IBD)	133.41 ± 0.53 c	299.41 ± 1.83 e	621.59 ± 1.16 d	977.95 ± 1.93 d
control negative (no vaccine)	120.34 ± 0.37 d	306.62 ± 1.14 d	621.03 ± 0.64 d	967.72 ± 2.79 e
level of significance	**	**	**	**

Averages that carry different letters within the same column significantly differ among themselves.** (P <0.01)

Table (3). Studied the effect of the Groups in the rate of weight gain of the Different weeks

Averages that carry different letters within the same column significantly differ among themselves.** (P <0.01).

Groups	Average ± standard error (g/chicks)			
	first week	second week	third week	fourth week
Boron100 mg/ kg feed	92.28 ± 1.84 b	226.78±1.96 b	350.02 ± 1.62 b	460.67 ± 2.16 a
Boron 200 mg / kg feed	109.09 ± 0.74 a	251.98±1.93 a	326.73±13.87 c	462.31 ± 13.47 a
Boron 300 mg / kg feed	76.01 ± 1.56 d	193.07±2.87 c	446.36 ± 1.76 a	382.17 ± 2.45 b
Control Positive (vaccine)	88.41 ± 0.53 c	166.01±2.16 e	322.17 ± 2.49 c	356.36 ± 2.14 c
control negative (no vaccine)	75.35 ± 0.37 d	186.27±1.09 d	314.41 ± 1.08 c	346.69 ± 3.134 c
Level of significance	**	**	**	**

Table (4).The effect of Boron Supplemented formula &IBD vaccine on feed intake of theChicks for different weeks .

Averages different letters within the same column significantly differ between

Groups	Average ± standard error (g / chicks)			
	first week	second week	third week	fourth week
Boron 100 mg / kg feed	72.78 ± 0.11 b	199.95 ± 0.09 b	381.46 ± 0.71 b	691.11 ± 0.09 b
Boron 200 mg / kg feed	77.18 ± 0.03 a	215.60 ± 0.06 a	400.71 ± 0.12 a	711.42 ± 1.55 a
Boron 300 mg / kg feed	71.97 ± 0.04 c	199.74 ± 0.02 b	380.79 ± 0.14 b	689.62 ± 0.04 b
control Positive	69.06 ± 0.03 e	198.68 ± 0.13 d	379.08 ± 0.03 c	681.05 ± 0.04 c
control negative	69.65 ± 0.02 d	199.15 ± 0.02 c	378.96 ± 0.96 c	690.01 ± 0.05 b
Level of significance	**	**	**	**

groups.** (P <0.01).

Table (5).The effect of Boron Supplemented formula &IBD vaccine on the blood pictures

Groups	Average ± standard error			
	(PCV%)	(RBCs)	(WBCs)	H / L ratio
Boron 100	27.95 ± 0.24 b	2.30 ± 0.008	18.94 ± 0.09	0.910 ± 0.02

mg / kg feed		c	c	c
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Treatment	Average ± standard error
Boron 100 mg / kg feed	2.95 ± 0.01 b
Boron 200 mg / kg feed	3.69 ± 0.02 a
Boron 300 mg / kg feed	2.85 ± 0.01 c
control positive (vaccine IBD)	2.53 ± 0.04 d
control negative (no vaccine)	2.81 ± 0.02 c
Level of significance	**

Boron 200 mg / kg feed	34.95 ± 0.04 a	3.44 ± 0.007 a	22.90 ± 0.04 a	0.808 ± 0.005 d
Boron 300 mg / kg feed	27.68 ± 0.04 b	2.52 ± 0.006 b	20.27 ± 0.12 b	0.989 ± 0.015 b
control positive	22.89 ± 0.02 d	2.07 ± 0.009 e	16.94 ± 0.05 c	1.09 ± 0.012 a
control negative	27.08 ± 0.02 c	2.26 ± 0.005 d	17.66 ± 0.05 d	0.903 ± 0.006 c
Level of significance	**	**	**	**

Averages that carry different letters within the same column significantly differ among themselves. ** (P <0.01).

Table (6).The effect of Boron Supplemented formula &IBD vaccine on the total protein

Concentration in the Blood serum rate (g / dl)

Averages that carry different letters within the same column significantly differ among themselves. ** (P <0.01).

Table (7).The effect of Boron Supplemented formula &IBD vaccine on the relative

weights of internal organ of the end of the experiment

Groups	Average ± standard error (%)		
	kidney	bursa of fabricius	Spleen
Boron 100 mg / kg feed	0.753 ± 0.001 c	1.008 ± 0.013 d	0.163 ± 0.005 c
Boron 200 mg / kg feed	0.849 ± 0.003 b	1.119 ± 0.01 c	0.230 ± 0.006 a
Boron 300 mg / kg feed	0.993 ± 0.001 a	1.431 ± 0.008 b	0.187 ± 0.004 b

control positive (vaccine IBD)	0.690 ± 0.002 d	1.643 ± 0.02 a	0.143 ± 0.005 d
control negative (no vaccine)	0.661 ± 0.007 e	0.853 ± 0.006 e	0.160 ± 0.006 c
Level of significance	**	**	**

Averages that carry different letters within the same column significantly differ among themselves. ** (P <0.01).

Table (8) The effect of Boron Supplemented formula &IBD vaccine on antibody titers against (IBD) in broilers (M±SE).

Groups	IBD titer (M±SE).
Boron 100mg/kg. feed	6842.42± 8.70 b
Boron 200mg/kg. feed	7112.30±1.94 a
Boron 300mg/kg. feed	6786.90±3.41 c
Control positive (IBD vaccine)	6628.60±6.09 d
Control negative	690.50± 1.36 e
Level of significance	**

Averages that carry different letters within the same column significantly differ among themselves. ** (P <0.01).

Discussion

In the present study, the results showed that three levels of boron (i.e. 100,200 and 300 mg/kg) induced improvement of the growth by significantly increase of BW , WG and feed intake especially the addition of 200 mg/kg, followed by 100 and 300mg/kg. Results are in agreement with other studies that found that boron supplementation had effect on growth performance (14, 15) who pointed out that different levels of boron added to diets caused a significant increase in

body weight compared to the diet which is not supplemented with boron,

Wilson J.H(16) stated that the body weight gains were not affected in broilers fed with boron diets. By contrast, (17) reported that the body weight significantly increased in broilers fed diet supplemented with boron, in birds supplemented with boron during the whole experimental period. The positive effect of boron on body weight and weight gain and feed intake may be depended to the role of

boron on certain metabolic processes of enzymes, minerals of chickens (18, 19),

The result of effect of boron supplementation on blood pictures showed that there was a positive effect on all parameters, it seems that also could affected blood cell counts and composition because blood cell formation and maturation are influenced by changes in cell membrane or kidney function or in calcium metabolism (20).

There were limited data about blood cell variables affected by boron supplementation in chicks or other animal spp, boron increase the response of cell membrane of blood cell to erythropoietin and this hormone synthesized in the kidney which plays a role in the maturation of red blood cell precursors in bone marrow (21)

In this study that the weights of internal organs (kidney, bursa of Fabricius and spleen) were significantly effect in birds supplemented with boron. This result also confirmed previous report from (22) who found dietary supplementation of boron significantly effects on visceral organ weights in broiler chickens. The dietary 60 mg/kg boron addition might help to enhance the immune system for the first 3 weeks age of broilers but further studies are required for exploring this point. The improvement of growth parameter for broiler that feed on boron which lead to enhance the immune response to (IBD vaccine) the increase in the antibody titer in this study significantly which may cause by addition of boron it will lead to enhance fc-receptor, interleukin, production of cytokines and interferon- γ (23,24).

As a conclusion, dietary 100,200 and 300 mg/kg boron addition cause positive effects on growth performance, blood pictures, total

protein concentrations and immune system in broiler chickens and don't inhibition the immune response of chickens against infectious bursal disease vaccine (IBD vaccine), dietary B addition may be useful for immune health of broiler chickens which is very important point in poultry industry.

Reference

- 1- Okuyan M.R (1997); Biochemistry of animal nutrition . okuyanm.r(ed), Ankara Universities ziraat fakultesi yayinlari, yayin no:1491, ders kitabi:450, Ankara, 350 pages.
- 2- world health organization WHO (1998).: international program on chemical safety, environmental health criteria 204, boron, ohio, USA., pp:1-20.
- 3- Park, M, Li, Q., Shcheynikov, N., Zeng, W., Muallem, S (2004)., NaBC1 is ubiquitous Electrogenic Na⁺- coupled borate transporter essential for cellular boron homeostasis and cell growth and proliferation. Mol Cell, 16, 331-341.
- 4- Underwood EJ, Suttle NF (2000). The mineral nutrition of livestock. 3rd ed. London: CABI Publishing. 602p.
5. NRC, National Research Council (1984). Nutrients requirements of poultry. 9th ed. Washington, D. C: National Academic press, P.71-
- 6- Qin, X., Klandorf, H. (1991): Effects of dietary boron supplementation on egg production, shell quality, and calcium metabolism in aged broiler breeder hens. Poult. Sci.; 70: 2131-2138.
- 7- Hunt, C.D.: (1989) Dietary boron modified the effects of magnesium and Molybdenum on mineral metabolism in the cholecalciferol deficient chicks. Biol.

- Trace Elem. Res; 22: 201-220.
- 8- N.R.C., National Research Council,(1994) . Nutrient Requirements of Poultry, 9th ed., National Acad. Press, Washington.D.C.: NAS, Pp. 155.
- 9- Al-Zubayedi, S.S.A (1986).Poultrysupplementation of drinking water with management. First ed., the College of Agriculture, Basra University .associated with the aflatoxin bio synthetic pathway, Bioorganic Chemistry, Vol.33, pp.426-438,
10. Campbell, T. W. (1988). Avian Hematology and Cytology. 1sted. Iowa State University.Press.Amess. Iowa, 1: 23-43.
- 11-Burton, R.R., andC.W.Guion(1978).the differential leucocyte blood count: its Precisionand individuality in chicks .poultry sci.471945-1949.
- 12- Wotton ,I.D.P.(1974).Micro – analysis in medical biochemistry 4thed.churchill Livingstone ,London .
13. SAS. (2012). Statistical Analysis System, Users Guide.Statistical. Version 9th ed. SAS. Inst. Inc. Cary.N.C.USA.
- 14- Kurtoglu F., Coskun B., Sekere.,Balevi T, EtinG-I.S.(2003)Effects of boron supplementation on performance and some serum biochemical parameters in laying hens, *Rev. Med. Vet.*, , 154, 828-839.
- 15-Rossi A.F., Miles R.D., Damro N B.L., Flunker L.K.(1993):Effects of dietary boron supplementation on broilers. *PoultSci.*, 72, 2124-2130.-
- 16-Wilson J.H., RuszlerP.L(1997):. Effects of boron on growing pullets Biol. Tr. Elem. Res, 56, 287-294.
- 17-ErenM., Guclu B.K., Uyanik F., Karabulut N.(2006): The effects of dietary boron supplementation on performance, carcass composition and serum lipids in Japanese quails. *J. Am. Vet. Assoc* , 5, 1105-1108.
- 18- Jin, E. &Gu, Y. (2014). Effectof Poultrysupplementation of drinking water with different levels of boron on performance and immune organ parameters of broilersItalian J. Ani. Sci.,13:205-213.
- 19-Mizrak C., Yenice E., Can M., Yildirim U., Atik Z(2010):.Effects of dietary boron on performance, egg production, egg quality and some bone parameters in laying hens, South Afr. J. Anim. Sci., 40, 3.
- 21-Nielsen, F. H.; Mullen, L. M. & Nielsen, E. J. (1991). Dietary Boron Affects Blood cell counts and Hemoglobin Concentrations in Humans. *J. Trace Elements in Exp. Med.*, 4:211-223.
- 22-YildizG., KoksalsB.H., AbacigluO.,(2009):. Effects of dietary boron addition on growth performance, , some carcass and tibia characteristics in different rearing periods in broiler chickens. *Revue Med.*, 164(4):219-224.
- 23-Armstrong, T. A.; Spears, J. W. & Lloyd, K. E. (2001). Inflammatory response, growth and thyroid hormone concentrations are affected by long-term boron supplementation in gilts. *J. Anim. Sci.*, 79:1549-1556.
- 24-Fassanie.J., Bertechina.G. Brito J.A.G., Kator.K, Fialho E.T.,(2004) Geraldo A.: Boron supplementation in broiler diets.*Rev. Bras. Cienc. Avic.*, , 6, 213-217

