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DIETARY EFFECTS OF ALGERIAN SODIUM BENTONITE ON GROWTH PERFORMANCE AND BIOCHEMICAL PARAMETERS IN BROILER CHICKENS

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ABSTRACT The present experiment was conducted to investigate the effect of supplementing poultry feed with graded levels of Algerian sodium bentonite (Na-B) on growth performance and the development of villus height in jejunum and some biochemical parameters during 50 days in broiler chickens. A number of 420 one-day old broiler chicks (Arbor Acres) were obtained from a commercial hatchery. The birds were randomly allocated into six groups (A, B, C, D, E and F). The treatments were 0 (control), 1%, 2%, 3%, 4% and 5% of Algerian Na-B levels. The results obtained indicate clearly that weight gain in the chickens fed treatments containing 4% Na-B had greater weight gain than the chickens fed different treatments (0, 1%, 2%, 3% and 5% Na-B). Feed conversion rate (FCR) was lower birds supplemented with Na-B 4% (2.45) than control group (3.06). Maximum feed consumption was observed in the birds' control (5,655.3 g), while the lowest was noted in the chickens with diet added 4% Na-B (5,009.5 g) ($p< 0.05$). The weight of duodenum, jejunum and ileum was decreased for the Algerian Na-B supplemented group, compared with the control group. The villus height was affected by dietary treatments (1%, 2%, 3% and 5%) on days 18 and 50 ($p< 0.05$). Feeding the supplemented graded levels Na-B resulted in an increase in plasma cholesterol, triglyceride and HDL concentrations at 50 days of age, compared with the control group. These results showed clearly that the Na-B from Algeria can improve the growth performance in broiler chickens. Thus,

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dietary inclusion of Na-B had positive effect on plasma triglyceride, cholesterol and HDL values in broiler chickens at the end experiment.

Keywords: Algeria; clay; growth performance; feed supplementary; poultry.

INTRODUCTION

Clay is a natural economic substance and a highly abundant product in the nature. Bentonite, one main constituent of clays, is white, light weight rock deposit, composed mostly of salts of hydrated aluminosilicates of sodium (Na), potassium (K), calcium (Ca) and occasionally iron, magnesium, zinc, nickel, etc. The special properties of bentonite, such as hydration, swelling, water adsorption and viscosity, made it a valuable material for wide range of applications in industrial and farming systems (Miazzo *et al.,* 2000). Bentonite as a feed additive has been used successfully in poultry without any harmful effects (Prvulović *et al.,* 2008; Safaeikatouli *et al.,* 2010). The use of clay supplements in animal and poultry feed manufacturing is not new.

Several studies showed that poultry feed supplemented with sodium bentonite (Na-B) can improve growth performance (Damiri *et al.,* 2010; Prvulović *et al.,* 2008; Safaeikatouli *et al.,* 2010; Salari *et al.,* 2006). Dietary clay supplements (bentonite and kaolinite) have been used as binding and lubricating agents in the production of pelleted feeds for chickens (Owen *et al.,* 2012).

Bentonite, as a toxin binder, decreased the adverse effect of aflatoxin (Shi *et al.,* 2009) improved the performance (Pasha *et al.,* 2007) and reduced mycotoxin concentration in the livers of affected birds (Bailey *et al.,* 2006). In natural breeding farms, the consumption of soil is a natural phenomenon observed in all animal species. It is know that hens consume the soil fauna voluntarily or by eating earthworms and insects.

There are two types of naturally occurring bentonites: calcium and Na-B (Wright, 1968); there are clays originating from the smectites and their physical properties are those of this mineral group. Variations in interstitial water and exchangeable cations in interlayer of the different space affect the properties of bentonite and, thus, the commercial uses of the different types of bentonite (Adamis *et al.,* 2005). Indeed, Khanedar *et al.* (2012) showed that the use of either kinds of bentonite in diets at 1% could improve the broiler performances; however, addition of 1.5% bentonite had not any significant effect in this study. Today there is a variety of commercial clay, especially bentonite, available in the market in the form of animal feed additives with different chemical characteristics (Magnoli *et al.,* 2008).

In Algeria, the Na-B is abundant in the North West area (Mostaganem, Maghnia) and it is the most important deposit with a production of 20,000 tons per year (Ministry of Energy and Mines, 2015). To our knowledge, there are very few works published

showing the effects of different levels of clay from Algeria on broiler performance. The present experiment was conducted to investigate the effect of supplementing poultry feed with graded levels of Algerian Na-B on growth performance and the development of villus height in jejunum and some biochemical parameters during 50 days in broiler chickens.

MATERIALS AND METHODS

Animals and dietary treatments

This study was conducted in Misserghin farm in Oran (western Algeria). The experimental protocol was approved by the Scientific Faculty Council of the University M. Istambouli (Mascara, Algeria).

A number of 420 one-day old broiler chicks (Arbor Acres) were obtained from a commercial hatchery.

The birds were randomly allocated into six groups (A, B, C, D, E and F; 70 birds/group) and housed in pens of identical size (2.35 × 6 m) in a deep litter system with a straw floor. The birds were fed a balanced commercial broiler ration *ad libitum* for a period of 7 weeks. Diets were formulated according to Smallix recommendations (*Table 1*). Chicks were managed according to the guidelines suggested by Cobb Broiler Commercial Management Guide. Birds were vaccinated against Gumboro (IBDL, Ceva), Newcastle (HB1, Ceva) according to laboratory recommendations. In order to prevent coccidiosis, the chicks were treated by anticoccidial during 36 days (Salinomycine 200). The chicks were reared for 7 weeks maintaining all the hygienic measures in a well-ventilated poultry house.

The treatments were 0 (control), 1%, 2%, 3%, 4% and 5% of Algerian Na-B levels for starter (0 to 14 days), grower (15 to 28 days) and finisher (29 to 50 days) periods. Clay used in this experiment was grayish abundant in the area of study (Maghnia, Algeria). The chemical composition of Algerian Na-B is presented in *Table 2* (Debieche and Kaoua, 2014).

Growth performances

All birds were weighted individually after their arrival from the hatchery to the experimental farm (initial weight) and on day 50. Feed intake, body weight gain and feed conversion ratio (FCR) were calculated every 5 days. The daily weight gain (DWG) was calculated as follows: DWG (g/d) = (Final weight – Initial weight) / 50. The FCR used the following formula: FCR = Total feed consumption / Total final weight. Mortality was recorded daily for each pen.

Histomorphological and blood samples

During the experiment, after weighting, five chicks per treatment were randomly selected and killed by cervical dislocation for histomorphological analysis of the intestinal mucosa. The gastrointestinal tract was weighted after removal of the content. The tissue samples for histology were taken from the jejunum at days 18 and 50. The samples were fixed in 4% buffered formalin for 48 hrs. The processing consisted of serial dehydration, clearing, and impregnation with paraffin. Tissues sections, 5 µm thick were cut by a microtome and were fixed on slides. A staining procedure was carried out using hematoxylin and eosin. The slides were examined on an optical microscope.

Table 1 - Composition (%) of the diets used in experiment

Ingredients	Starter period (0-14 d)						Growth period (15-28 d)						Finisher period (29-50 d)					
	A	B	C	D	E	F	A	B	C	D	E	F	A	B	C	D	E	F
Maize	57.1	56.1	55.1	54.1	53.1	52.1	62	61	60	59	58	57	64.3	63.3	62.3	61.3	60.3	59.3
Bicalcium phosphate	1.9	1.9	1.9	1.9	1.9	1.9	1.9	1.9	1.9	1.9	1.9	1.9	1.8	1.8	1.8	1.8	1.8	1.8
Calcium carbonate	0.5	0.5	0.5	0.5	0.5	0.5	1.1	1.1	1.1	1.1	1.1	1.1	1.2	1.2	1.2	1.2	1.2	1.2
Vegetal oil	0.5	0.5	0.5	0.5	0.5	0.5	1	1	1	1	1	1	1.5	1.5	1.5	1.5	1.5	1.5
Soybean	38.2	38.2	38.2	38.2	38.2	38.2	32.94	32.94	32.94	32.94	32.94	32.94	30.2	30.2	30.2	30.2	30.2	30.2
Methionine	0.24	0.24	0.24	0.24	0.24	0.24	0.2	0.2	0.2	0.2	0.2	0.2	0.15	0.15	0.15	0.15	0.15	0.15
Lysine	0.03	0.03	0.03	0.03	0.03	0.03	0.03	0.03	0.03	0.03	0.03	0.03	0.02	0.02	0.02	0.02	0.02	0.02
Salt	0.5	0.5	0.5	0.5	0.5	0.5	0.3	0.3	0.3	0.3	0.3	0.3	0.3	0.3	0.3	0.3	0.3	0.3
Cycostant	0.03	0.03	0.03	0.03	0.03	0.03	0.03	0.03	0.03	0.03	0.03	0.03	0.03	0.03	0.03	0.03	0.03	0.03
Sodium bentonite	-	1	2	3	4	5	-	1	2	3	4	5	-	1	2	3	4	5
Vitamin and mineral supplement	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1

Group A: 0% Na-B (control), group B: 1% Na-B, group C: 2%, group D: 3% Na-B, group E: 4%, group F: 5% Na-B.

Table 2 - Chemical composition (%) of sodium bentonite (Na-B) from Algeria (Maghnia, North West area)

Item	Content
SiO_2	64.98
Al_2O_3	16.08
Fe_2O_3	2.93
CaO	0.61
MgO	3.51
K_2O	2.02
Na_2O	3.88
TiO_2	0.2
Loss on ignition	6.07

The images were analyzed using Neubauer ruling. Villus heights were measured according to the method described by Ritz *et al.* (1995).

Blood samples were collected randomly from brachial vein in tubes containing heparin at days 18, 29 and 50. The plasma obtained was centrifuged (1500 × g for 15 min) and stored at -20°C until measurement for cholesterol, triglycerides and high density lipoprotein (HDL) concentrations using a spectrophotometer reader, according to the manufacturer recommendation (Spinreact, S.A./S.A.U. Sant Esteve de Bas, Spain).

Data analysis

Statistical analyses were carried out in Statview (Version 4.55). Statistical analysis was performed using *t*-test to compare between different groups. The data were expressed as mean ± SE, and $P < 0.05$ was considered significant.

RESULTS

Initial and final body weight (BW), weight gain, feed intake, FCR and mortality rate of the chickens fed different levels treatments are presented in *Table 3*. The initial body weight of chicks did not differ between the dietary treatments. At the end of the experiment (day 50), birds supplemented with graded levels of Algerian Na-B had a greater body weight, compared with control group (1,847.9 g). The birds supplemented with Na-B 4% had a higher body weight (2,044.1 g) than other groups. The average daily weight gain (from day 1 to 50) was higher for chicks supplemented with graded levels of Algerian Na-B (38.2 to 40.1 g) than for control group (36.4 g). The results obtained indicate clearly that weight gain in the chickens fed treatments containing 4% Na-B had greater weight gain than the chickens fed different treatments (1%, 2%, 3% and 5% Na-B).

Feed conversion rate (FCR) was lower in birds supplemented with Na-B 4% (2.45) than in control groups (3.06). In addition, Na-B supplemented birds (Group C) had a higher FCR than groups B, D, E and F (*Table 3*). Feed intake and increased weight gain in the chickens fed ration containing 4% Na-B recorded an im-

proved FCR, in comparison with the control and all other diets treatment. Maximum feed intake was observed in the birds' control (5,655.3 g), while the lowest was noted in the chickens with diet added 4% Na-B (5,009.5 g). There is a significantly difference between the control and the chicken fed different treatments ($p< 0.05$). The mortality rate was lower for the birds fed supplemented with Na-B 4% (1.4%) than other groups. The groups that received the Na-B 1%, 2% and 3% showed higher mortality rate (8.6%, 7.1% and 12.9%, respectively) than the control group (4.3 %).

The means of the weight of intestinal part for dietary treatments are shown in *Table 4*. The weight of duodenum, jejunum and ileum was decreased for the Algerian Na-B supplemented group, compared with the control group. *Fig. 1* shows the means of jejunum villus height from broilers fed different diets. The villus height was affected by dietary treatments (1%, 2%, 3% and 5%) on days 18 and 50. However, birds fed supplemented Na-B 4% decreased the villus height, compared with the control group on day 18 ($p< 0.05$).

The effects of different dietary treatments on plasma lipids on broiler chickens are presented in *Table 5*. When compared with control group, feeding the supplemented graded levels Na-B resulted in increase in plasma cholesterol, triglyceride and HDL concentrations at 50 days of age.

Table 3 - Effect of graded levels of Algerian Na-B on the performance of broiler chickens

Item	Diets treatments (Sodium bentonite %, n=70)					
	0	1	2	3	4	5
Initial body weight, g	41.1	41.1	40.3	40.3	40.3	40.9
Final body weight, g	1,847.9	1,948.9	2,035.8	2,025.1	2,044.1	1,995.9
Daily weight gain, g	36.1	38.2	39.9	39.7	40.1	39.1
Feed intake, g/bird	5,655.3[a]	5,074.7[b]	5,287.9[b]	5,200.1[b]	5,009.5[b]	5,161.0[b]
Feed conversion rate, %	3.06	2.60	2.59	2.56	2.45	2.58
Mortality rate, %	4.3	8.6	7.1	12.9	1.4	5.7

[a,b] A significant difference in mean feed intake (g/bird) between the control group (0 Na-B level) and the treated groups (1%, 2%, 3%, 4% and 5% Na-B level) is indicated by letters ($p < 0.05$).

DISCUSSION

The present study shows that inclusion of Algerian Na-B in diets of broilers chickens improves weight gain and decreases the values of FCR (*Table 3*). In addition, the final body weight (day 50) of broilers containing different levels Algerian Na-B was higher than that of control group. The reason for this improvement may be the action of silicate minerals enhancing the digestibility of certain nutrients (Safaei Katouli *et al.,* 2012).

Similar results have been described by Eser *et al.* (2011), who used the sepiolite in broiler diet.

Also, a higher growth response caused by bentonite supplemented diets has been reported in several animal species (Ibrahim *et al.,* 2000; Ivan *et al.,* 1992; Jacques *et al.,* 1986). However, the bentonite level above 4% could decrease the body weight gain in broiler chicken. A positive effect of bentonite on body gain seems to be inclusion dose on broiler dietary (Grosicki *et al.,* 2000). The feed efficiency of Algerian Na-B at different levels revealed non-significant difference statistically, but decreases the rate of FCR in broilers chicken, compared with control group. Bentonite at 4% level rather depressed the performance of chicks (Tauqir *et al.,* 2001). In the present experiment, we noticed that Algerian Na-B 5% in diet decrease the daily weight gain of broiler chicken, compared with all treated groups. Besides, additional Na-B level in feed higher 5% reduces the growth performance of birds (Mabbett, 2005). Damiri *et al.* (2012) reported that best results by addition low Na-B levels may be due to increased retention time of digesta in intestinal tract and more nutrients using. Therefore, the retention time increased had no negative effects on feed intake and increased performance.

Low weight gain and feed intake obtained in broiler with a Na-B level were in agreement with those reported by Tauqir and Nawaz (2001). It may be due to viscose nature of Algerian Na-B, which absorbs much water and decreased passage rate of digesta in lumen (Damiri *et al.,* 2012).

Table 4 - Effect of graded levels of Algerian Sodium bentonite (Na-B) on weight intestinal part of broiler chickens

Sodium bentonite (%)	Weight intestinal part (means ± SE), g		
	Duodenum (n=5)	Jejunum (n=5)	Ileum (n=5)
0	18.0 ± 4.0[a]	25.0 ± 6.8[a]	23.2 ± 5.3[a]
1	13.3 ± 1.5	21.1 ± 3.12	20.8 ± 6.1
2	12.2 ± 1.7	15.8 ± 2.9[b]	13.9 ± 2.6[b]
3	13.5 ± 2.1	19.9 ± 3.5	16.6 ± 3.0
4	11.5 ± 1.9[b]	18.0 ± 3.2	15.4 ± 1.3[b]
5	12.7 ± 2.5[b]	18.1 ± 5.9[b]	17.6 ± 2.5

Data presented in *Table 3* demonstrates that addition of the Algerian Na-B in feed had significant effect on the feed intake, compared with the control group ($p< 0.05$). Our results are different from of those obtained by Tauqir *et al.* (2001), who reported the use of level diet Na-B 2.5% improved feed consumption, compared with high Na-B level (>2.5%) that might have deleterious effect on the performance of birds.

However, Pasha *et al.* (2007) indicated that the indigenous Na-B 0.5% and 1% levels gave better results at the level of levels on birds' performance (Kubena *et al.,* 1993; Dale and Wyatt 1995).

Due to highly adhesive nature of the Na-B, it was suggested that Na-B absorbs moisture resist the flow of digesta through the gastrointestinal tract affecting negate-vely the feed intake (Van Olphen, 1963). This could be partially explained by the effect of clay on bile salts preservation (Prvulović *et al.,* 2007).The role of clinoptilolite, one of the clay, is involved in the adsorption and immobilization of the pathogenic flora and protection against their bile salt conjugation (Habold *et al.,* 2009).

Table 5 - Effect of different levels of Algerian Sodium bentonite (Na-B) on broiler plasma lipids

Diet treatment	Plasma lipids (n=5, means ± SE)		
	Cholesterol (mg/dL)	Triglyceride (mg/dL)	HDL (mg/dL)
0%			
Day 18	58.1 ± 32.0^{a}	48.7 ± 16.1	37.1 ± 19.5
Day 29	78.8 ± 33.0^{B}	63.8 ± 09.3	40.5 ± 09.3^{A,B}
Day 50	53.5 ± 28.7^{a}	37.8 ± 20.1	33.4 ± 11.4^{C,D}
1%			
Day 18	65.0 ± 12.4	42.7 ± 25.0^{a}	49.8 ± 09.2
Day 29	59.7 ± 14.8	41.0 ± 27.2	59.3 ± 22.6
Day 50	40.2 ± 22.5	55.1 ± 25.9	37.1 ± 08.9
2%			
Day 18	67.4 ± 20.5^{A}	96.4 ± 27.0^{a,b,A,B}	50.8 ± 24.2
Day 29	78.8 ± 33.0	57.3 ± 35.5^{A}	65.9 ± 16.8^{A}
Day 50	61.2 ± 26.6	76.7 ± 52.3^{B}	51.7 ± 10.7
3%			
Day 18	47.5 ± 09.8^{a,A}	67.58 ± 05.8	43.4 ± 07.7^{a}
Day 29	78.5 ± 10.8^{a,C}	49.21 ± 13.3	70.3 ± 06.7^{a,B}
Day 50	68.8 ± 03.2	43.26 ± 17.1	54.7 ± 10.7^{E}
4%			
Day 18	70.2 ± 29.9	50.6 ± 18.0	45.7 ± 14.0^{a}
Day 29	44.0 ± 23.4^{C}	14.9 ± 08.2	57.3 ± 24.6^{b}
Day 50	56.7 ± 39.6^{D}	45.6 ± 21.5	57.9 ± 14.9^{a,b,C,E}
5%			
Day 18	41.0 ± 29.5	45.7 ± 22.3^{b}	36.2 ± 19.6
Day 29	38.5 ± 08.2^{B}	30.8 ± 24.0	47.8 ± 06.3^{b}
Day 50	62.2 ± 26.5^{D}	53.1 ± 17.9	44.6 ± 11.2^{b,D}

^{a,b} Values with different superscripts in the same groups at different days that differ statically (p< 0.05).
^{A,B,C,D,E} Values with differences in the same day between different groups differ significantly (p< 0.05).

In this study, the results of FCR were lower in birds receiving graded Na-B diets than those of the control group. The different levels of Na-B had clearly an effect on FCR, but the FCR was improved by adding in diet Na-B 4%. This is in accordance with the results of some studies showing that the use of Na-B in broiler chickens diet would improve their weight gain (Prvulović *et al.,* 2008; Tauqir *et al.,* 2001). Moreover, some researchers found that animal diets containing kaolin and zeolite have been shown an improvement body weight gain and FCR in chicken (Cabuk *et al.,* 2004; Hesham *et al.,* 2004; Incharoen *et al.,* 2009; Safaeikatouli *et al.,* 2010).

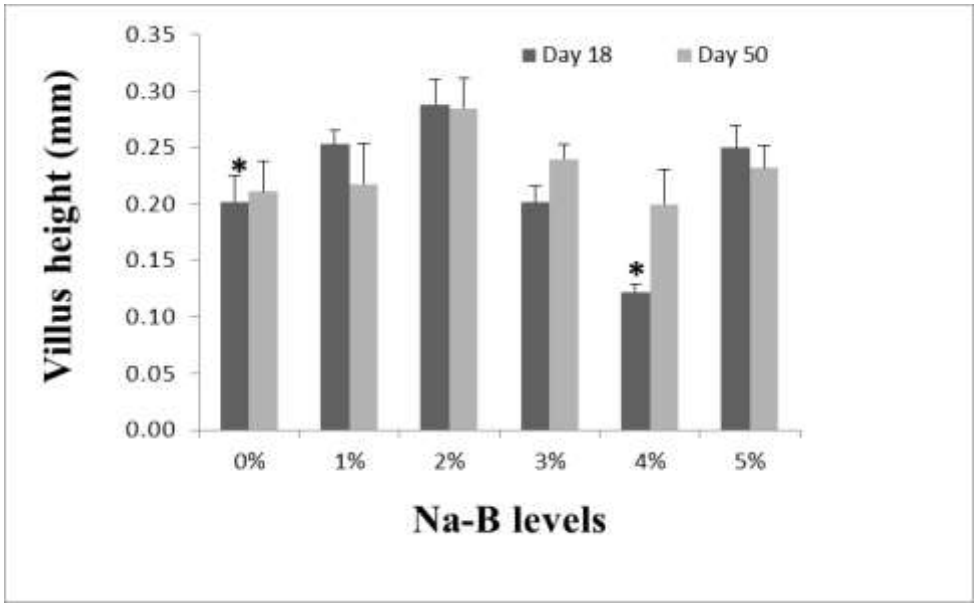

Figure 1 - Effect of different levels of Algeria Sodium bentonite (Na-B) on villus heights (mm) of broiler chickens (n=5). A significant difference in mean villus height between the Na-B level treated groups (0%, 1%, 2%, 3%, 4%and 5%) at Day 18 and Day 50 is indicated by asterisk (*)($P < 0.05$).

Our results agree with the data obtained by Safaeikatouli *et al.* (2010), where the FCR was increased in broiler diet Na-B 3%, compared with kaolin 3% and Na-B 1.5% treatments. Also, Damiri *et al.* (2012) observed that addition of Na-B 3.75% in diet of broilers fed decreased feed intake and weight gain.

Salari *et al.* (2006) indicated that chickens fed diets containing 1% and 2% of Na-B consumed more food. These latter had more weight gain and less FCR. Several studies have reported that weight gain in chicks given low energy diets is not affected by bentonite (Sellers *et al.,* 1980). Likewise, Tauqir *et al.* (2001)

indicated that the interaction between diet Na-B and energy was found in broiler chickens.

According to Xu *et al.* (2003), a shortening of the villus and deeper crypts may lead to poor nutrient absorption, increased secretion in the gastrointestinal tract and lower performance. It has been suggested that greater villi height is an indicator that the function of intestinal villi is activated (Langhout *et al.,* 1999; Shamoto and Yamauchi 2000). In the present experiment, the histological aspect of the jejunum of broiler chickens gives orientation concerning the potential for using Na-B in broiler feed. The result of the present study indicates that supplementation of broiler with Algerian Na-B increased the villi height of jejunum at the end of experiment (*Fig.1*). The feed supplemented with graded Algerian Na-B had no effect statistically, but decreases slightly on the weight intestine parts, compared with the control group.

The results of this study correspond with those reported by Xia *et al.* (2004), who noted that the diet supplemented with montmorillonite increased the villus height and decreased crypt depth. Similarly, other study has reported that higher villus height were observed when the diet was treated with prebiotics and probiotics (Pelicano *et al.,* 2005). This could be explained by an increased epithelial cell turnover due to feeding of fed microbial (Awad *et al.,* 2008). According to Cera *et al.* (1988), maximum absorption and digestion capacity is given by a large luminal area with high villi and mature enterocytes, and is essential to animal development. As regards the weight of intestine parts, the present data showed a decrease for birds fed with Algerian Na-B, compared with the control group what demonstrate that the feed supplemented with different Algerian Na-B levels had no effect on the weight intestine of broilers (*Table 4*).

When compared with the control group, diets supplemented with different levels Na-B resulted in a remarkable increase in the plasma triglyceride, cholesterol and HDL concentrations. Kececi *et al.* (1998) have shown that some serum biochemical changes could be ameliorated by bentonite administration to the diet in broiler chickens. Habold *et al.* (2009) reported that the presence of clay in the intestinal lumen promotes the hydrolysis of triglycerides and increases the concentration of free fatty acids.

Decreased plasma cholesterol in chicks given Na-B in diet is consistent with the general reduction of lipogenesis (Donaldson *et al.,* 1972) and impaired lipid transport (Tung *et al.,* 1972) in chicks and specific inhibition of hepatic cholesterol biosynthesis (Kato *et al.,* 1969).

CONCLUSIONS

The results obtained under the conditions of this experiment showed that the Algerian Na-B can improve the growth performance in broiler

chickens. The inclusion of 4% Algerian Na-B in diets is more effective on the weight gain and feed intake. Also, supplementation of grade Algerian Na-B at 4% level was found beneficial in ameliorating the feed conversion rate in broiler chickens. Thus, dietary inclusion of Na-B had positive effect on plasma triglyceride, cholesterol and HDL values in broiler chickens at the end experiment.

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