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SOME PHYSIOLOGICAL RESPONSES TO DIETARY BETAINE LEVELS IN BROILER CHICKENS

Z. S. H. Ismail[1] and Enas A. M.Ahmad[2]

[1]Anim. Prod. Dep., Fac. of Agric., South Valley Uni., Egypt.

[2]Anim. and Poult. Prod. Dep.t, Fac. of Agric. and Natural Reso., Aswan Uni., Egypt.

Corresponding author: zienhom shikhown Hassan E-mail: Zien7300@yahoo.com

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ABSTRAC:. This study was designed to evaluate some physiological responses to dietary betaine levels s in broiler chickens. One hundred and twenty, one-day-old unsexed chicks, of Cobb strain, were used in experiment period of 6 weeks.. The birds were allocated in four treatments,30 birds each (3 replicates of 10 birds). Treatments were:T1 as control group (C); T2,Diet C plus 0.01% betaine; T3, Diet C plus 0.02% betaine; T4, Diet C plus 0.03% betaine. Feed intake and body weight per replicate were recorded weekly, Body weight gain (BWG) and feed conversion ratios (FCR) were calculated. At the end of the experiments samples of chickens were randomly chosen and slaughtered to determine dressing percentage and blood samples were taken to measure serum Glucose, Cholesterol, Triglyceride, Total lipids, T3, T4 and Albumin.The results showed that using of 0.03 % betaine of broilers diet had significant differences on BWG, FI, and FCR compared with other groups at 3 and 6 wks of age. The carcass weight was significantly ($P<0.001$) improved in 0.02 % Betaine. There were a significant increase ($P<0.01$) in serum total protein, albumin, and globulin concentrations of birds fed 0.03 % betaine levels compared to other treatments. The results concluded that using of 0.03% dietary betaine showed beneficial of some physiological and productive responses of broilers chickens.

Key words: Betainelevels – broiler - Total Protein - triglycerides andtotal lipids

INTRODUCTION

Betaine is a common term for trimethylglycine, a substrate for betaine-homocysteine methyl transferase (BHMT) in the liver and kidney (Kettunen et al., 2001 and Attia et al., 2009). It is commonly occur at high levels in plant and herbsgrowing under drought conditions; where the betaine is distinguished byosmoregulatory properties (Kettunen et al., 2001 and Attia et al., 2009). Previous studies indicated that betaine may play important roles such as improving growth performance and fat distribution (Wang et al., 2004; Sun et al, 2008 and Attia et al., 2009). Betaine plays two main roles in metabolism: The first is to donate methyl groups, and the second is related to its ionic characteristics that help in maintaining cell water homeostasis (Klasing et al., 2002), without affecting cell metabolism. The betaine has a role in maintaining the proteins'stability, particularly in the presence of elevated uric acid levels and up-normal cell salinity.Moreover, several organ tissues depend on betaine as an effective osmolyte,such as the brain, kidneys, intestines, liver, andleukocytes (Klasing et al., 2002).The betaine osmo-protectance was reported to improve the resistance of birds exposed to heat stress and tolerate the changes in acid-base balance(Honarbakhsh et al., 2007).Furthermore, betaine was reported to lessen theharm effects of coccidiosis on birds; particularly throughdisrupting the coccidiosis proliferation andsustaining the intestinal function and structure (Augustine et al., 1997).

Betaine is the trimethyl derivative of the amino acid glycine. As a by-product of sugar beet processing, it is commercially available as a feed additive. Various osmolytic properties have been reviewed for betaine molecules in plants (Robinson and Jones, 1986), in animal body tissues (Law and Burg, 1991), in marine animals (Clarke et al. 1994), and in bacteria (Le Rudulier et al. 1984).In addition, the three methyl groups of betaine contribute in the transmethylation process which involved in the synthesis of various substances such as creatine and carnitine (Kidd et al. 1997). In addition to its methyl group content, betaine was reported to display similar properties of the amino acid glycine. Also, it is involved in the metabolism of protein and energy. In animal nutrition, betaine commonly considered as a growth promoting factor and as a carcass modifier;this was associated with its growth-promoting and lipotropic function. There is increasing evidence that it is a highly valuable feed additive in the diets of many farm animals (Hassan et al. 2011 and Rao et al. 2011). Furthermore, betaine has been shown to protect cells from osmotic stress and allow them to continue regular metabolic activities in conditions that would normally inhibit the cell function (Hamidi et al. 2010). Furthermore, betaine donates its labile methyl group which is used in transmethylation reactions for synthesis of carnitine and creatineand thus affects animal fat metabolism (Rao et al. 2011). However, most previous thermal environment studies were conducted at cyclic or constant-acute heat stress conditions in closed environments which did not explain the effects of betaine on broilers reared at constant-chronic high temperatures that may impose chronic heat stress (Khattak et al. 2012).

The objective of the present study is to evaluate the physiological responses to different dietary betaine levels inbroiler chicks.

MATERIALS AND METHODS

The present experiment was carried out at the Poultry Farm, Department of Animal and Poultry production, Faculty of Agriculture, University of South Valley, during the period from 1 February, to 15 March, 2016. It was designed to evaluatehow far the dietary betainelevels affect on the growth performance, carcass traits and some blood parameters in Cobb broiler chicks.

Birds and experiment design

One hundred and twenty, one-day-old Cobb broiler chicks were wing-banded, individually weighed, and randomly assigned to four experimental groups, (control and 3 treatments). Each group included three replicates, each of 10 chicks. Birds in the T1, which served as the control, were fed a commercial broiler diet, while those in T2, T3, and T4 received 0.1, 0.2, and 0.3 g betaine /kg diet, respectively.

Experimental treatments and management

The birds of each replicate (10 birds) were housed in a battery cage (97 x 50 x 45 cm), in a closed house and raised under adequate environmental and managerial conditions during the whole experimental period (6 weeks). They were exposed to 23 continuous lighting hours with an intensity of 15 Lux during the first three days of age, which decreased to reach only 5 Lux during the rest of the (39) days experimental period.

The birds were kept under 32 ºC during the first week, and the temperature then was reduced gradually (2 ºC / week) until 24 ºC by the fourth week of age, after which it was fixed till the experiment end. The relative humidity ranged between 55-60% during the experiment period. The birds received adlibitum starter and grower diets between 1 to 21 and 22 to 42 d of age, respectively. The chemical analyses of the tested diets are shown in Table 1.The studied parameters included body weight (BW),body weight gain (BWG), feed intake (FI), feed conversion ratio (FCR), and mortality rate.

Carcass traits:

At the end of theexperiment (42 days of age), out of 18 birds/ treatment were fasted for 8 hours (6/ replicate), slaughtered, scalded and plucked mechanically. The internal organs (liver, heart, spleen and empty gizzards) and the abdominal fat were weighed and expressed as percentages of the carcass weight. While, the dressing percentage was calculated by relating the carcass and giblets weights to the live BWs.

Blood parameters:

By the end of experiment period, blood samples were collected from 9 random birds from each treatment, centrifuged to get the sera,which were stored at -20°C until they were analyzed for determination of Total protein, Glucose, Cholesterol, Triglyceride, Total lipids, T3, T4 and Albumin

Statistical analyses:

Data were statistically analyzed by ANOVA, using the General Linear Model (GLM) Procedure of SAS software (SAS institute, version 9.1, 2005). Duncan's multiple range test (Duncan, 1955) was used to detect the differences among means of different groups.

Results and Discussion

Body weight (BW), body weight gain (BWG) and feed conversion ratio (FCR)

Data presented in Table 2,showed that, 0.02 % betainelevel increased broiler BW at 3 weeks of age compared with the other levels of Betaine. Whereas, there were no significant differences among treatments at 6 weeks of age. However, at three and sixweeks of age birds received 0.03% Betaine recorded the highest BWG compared with other groups. BWG recorded the highest values for birds fed betaine levels at 0.02 and 0.03%.Similarly, thedietary betaine supplementation improved broilers BWG by approximately 3-5 % (Hassan et al ., 2005).However, there were no significant differences among all treatments at 0-6weeks of age in BWG values.The obtained results concerned with BWG are in agreement with results of Zulkifli et al. (2004) but under heat stress. The results of feed intake showed that there were no significant differences among all treatments at 0-3 weeks of age. At 4-6 wks and 0-6 wks of age, birds fed 0.01% betaine had the highest feed intake compared to the other groups. The best values for FCR were achieved for birds fed 0.02 and 0.03% betaine at 0-3 and 4-6 wks of age compared to birds fed 0.01% betaine and control birds.Similarly, thedietary betaine supplementation improved broilers feed efficiency by approximately 3-5 % (Hassan et al ., 2005).No significant differences among all treatments at 0-6 weeks of age in FCR values.The obtained results are in agreement with results of Zulkifli et al. (2004) but under heat.Feed efficiency was improved in broiler fed betaine was reported byHassan et al. (2005) who mentioned that during periods of osmotic disturbance caused by heat stress in broiler chickens, betaine is involved in the protection and improvements in the morphological characteristics of intestinal epithelia، resulting in an improved growth and feed efficiency (Hamidi et al. 2010; Sakomura et al. 2013). Furthermore، the supplementation of betaine was also increased in particular, the fermentation of fiber, nitrogen retention and enhanced mineral absorption (Eklund et al. 2006).The dietary inclusion of betaine was reported to improve the poultry weight gain and feed conversion ratio(Attia et al., 2005).

Carcass characteristics

The results presented in Table 3 indicated the effect of treatments on dressed carcass, abdominal fat and relative weights of some edible organs such as gizzard, liver, heart, and spleen.There were no significant differences among the experimental groups in the relative weights (%)of dressed carcass, liver, heart, spleenand abdominal fat.On the other hand, carcass weight and gizzard% was increased by feeding broilers on 0.02% and 0.03% betaineas compared with other groups. Shaojun et al., (2015) reported considerable modifications in fat deposition in broilers due to dietary betaine supplementation. In another study, dietary betaine exerted positive effects on carcass characteristics of pigs by reducing carcass fat content and back fat thickness, without influencing performance (Sales 2011).Moreover, the improved carcass lean percentage was due to Betaine increases the levels of themethionine and cysteine in the organism for a protein synthesis (McDevitt, 2000). The improved utilizationof the amino acids from the diet for protein synthesiscould decrease the availability of amino acids fordesamination and subsequent fatty acid synthesis(Wallis, 1999).However, other authors reported that betaine supplementation was less effective in improving carcass characteristics of broiler chickens (Waldroup & Fritts 2005; Fernández-Fígares et al. 2008).

These differences may be related to the extent of heat stress, concentrations of betaine in diets and diet compositions.

Physiological changes in blood serumconstituents as affected by treatments.

Data of blood serum constituents throughout the experiment period are shown in Table 4. Some of the tested serum constituents (Total Protein, Glucose, Cholesterol,and Total lipids were achieved the highest levels with birds fed 0.03% betaine compared to the control birds.However, triglycerides and globulin were decreased with addition of 0.03% betaine as compared to control birds. No significant differences between all treatments in albumin concentration.T3 and T4 were decreased in the treated birds with betaine compared to vontrol birds. Potential positive effects of betaine under heat stress played a role in the present experiment and heat-stressed broilers retained more methionine than the normal diet could provide effectively, which may explain this (Chamruspollert et al. 2004). The involvement of betaine in lipid metabolism offers an interesting perspective in meat production to satisfy consumer needs for lean meat, hence, betaine is often referred to as the ‘carcass modifier’. The obtained results of the reduced triglycerides induced by betaine supplementation are in agreement with Ratriyanto et al., (2009), who found that betaine enhanced lipase activity and decreased the concentration of triglycerides. Addition of 0.02% betaine increased the albumin level in chicks compared to control group. The findings results are in agreement with that reported by Rao et al., (2011) who found that broiler chicks supplemented with betaine supplementation (800 mg/kg) significantly increased albumin concentration by about 5.1% compared to control group. Also, EL-Husseiny et al., (2007) found that albumin level significantly increased in broilers supplemented with betaine 0.05, 0.075 and 1 mg/kg diet by about 3.4, 6.2 and 2.3% respectively compared to the control group.The increased in total protein concentration as reported in the present study are in agreement with Rao et al., (2011) who found that the addition of 800 mg/kg betaine significantly increased serum protein by about 88% compared to the control group. The obtained results concerning globulin are in disagreement with RAO et al., (2011) found that the broilers supplemented with 800 mg/kg betaine significantly increased blood globulinby about 13.2% compared to the control group.

The results of this study concluded that the dietary supplementationof betaine havea beneficial effect on broilers performance including some parameters of growth performance and some blood constituents without any deleterious effect on broiler physiological responses.

Table (1): Composition and calculated analysis of experimental diets

Ingredients	Starter diet	Grower diet
Maize. Ground	27.59	30.00
Sorghum. Ground	27.59	30.00
Soybean meal (44%CP)	28.50	25.00
Corn gluten meal (60%CP)	9.50	6.00
Vit. & Min. premix	0.30	0.3
Sunflower oil	3.00	5.52
Di-calcium phosphate	2.00	1.8
Limestone	1.00	1.00
Salt	0.38	0.38
DL-methionine	0.04	---
L-lysine HCL	0.10	---
Total	100	100
Nutrient analysis		
ME (kcal/kg diet)	3000	3187
Crud protein (%)	23.67	20.46
Calcium (%)	1.00	1.00
Available phosphate (%)	0.50	0.50
Lysine (%)	1.16	1.16
Methionine (%)	0.52	0.52

[1]Premix provides by kg: Vit A, 5500 IU; Vit E, 11 IU; Vit D3, 1100 IU; riboflavin, 4.4 mg; Ca pantothenate, 12 mg; nicotinic acid, 44 mg; choline chloride, 191 mg; vitamin B_{12}, 12.1 ug; vitamin B_6, 2.2mg; thiamine (as thiamine mononitrate), 2.2 mg; folic acid, 0.55 mg and d- biotin, 0.11 mg. Trace mineral (mg /kg diet): Mn, 60; Zn, 50; Fe, 30; Cu, 5 and Se, 0.3.

Table (2): Productive performance of broiler chicks as affected by dietary betaine levels

Items	Treatments				SEM	P. value
	Control	0.01%Bet.	0.02%Bet.	0.03%Bet.		
BW(g/bird)						
3 Weeks of age	995.666[a]	930.000[d]	982.333[b]	942.666[c]	3.265	0.0001[**]
6 Weeks of age	2676.666	2658.000	2720.000	2672.666	19.379	0.2088[NS]
BWG (g/bird /day)						
0-3Weeks of age	83.286[c]	83.428[c]	87.285[b]	93.524[a]	1.544	0.004[*]
4–6Weeks of age	89.047[c]	93.523[b]	97.619[a]	96.571[a]	1.026	0.001[**]
0–6Weeks of age	80.380	82.285	82.746	82.381	0.938	0.343[NS]
FI(g /bird/day)						
0-3Weeks of age	134.952	139.809	135.714	137.57	1.415	0.149[NS]
4–6Weeks of age	153.857[d]	165.857[a]	162.809[b]	159.190[c]	1.626	0.004[*]
0–6Weeks of age	134.460[c]	141.635[a]	136.651[b]	134.127[c]	1.150	0.005[*]
FCR(g feed / g growth)						
0-3Weeks of age	1.636[a]	1.687[a]	1.553[b]	1.475[b]	0.030	0.005[*]
4–6Weeks of age	1.740[a]	1.771[a]	1.671[b]	1.652[b]	0.025	0.034[*]
0–6Weeks of age	1.687	1.720	1.654	1.632	0.025	0.158[NS]

a, d: Means in the same row having different superscripts are significantly different at (P<0.05).

Table (3): Slaughter traits of broiler chicks as affected by dietary betaine levels

Items	Control	0.01%Bet.	0.02%Bet.	0.03%Bet.	SEM	P. value
Carcass weight	2175.333^{b}	2169.000^{b}	2288.000^{a}	2028.000^{c}	29.878	0.009^{*}
Dressed carcass (including giblets) (%)	83.682	82.853	50.380	81.884	1.448	0.355^{NS}
Liver %	2.373	2.516	2.651	2.179	0.163	0.378^{NS}
Gizzard %	1.652^{a}	1.341^{b}	1.719^{a}	1.717^{a}	0.055	0.007^{*}
Heart %	0.558	0.782	0.610	0.660	0.118	0.556^{NS}
Spleen %	0.114	0.099	0.088	0.089	0.018	0.745^{NS}
Abdominal Fat %	1.155	1.091	1.050	0.952	0.307	0.962^{NS}

a, b: Means in the same row having different superscripts are significantly different at ($P<0.05$)

Table (4): Some serum biochemical parameters of broiler chicks as affected by dietary betaine levels

Items	Control	0.01%Bet.	0.02%Bet.	0.03%Bet.	SEM	P. value
Total Protein (g/dl)	5.76^{b}	5.51^{b}	5.99^{a}	6.60^{a}	0.130	0.002^{*}
Albumin (g/dl)	3.05	3.36	3.56	3.24	0.14	0.142^{NS}
Globulin (g/dl)	2.70^{a}	2.15^{b}	2.40^{b}	2.35^{b}	0.15	0.003^{*}
A/G ratio	1.13^{a}	1.56^{a}	1.54^{a}	0.97^{c}	0.12	0.024^{*}
Glucose (mg/dl)	160.00^{b}	111.66^{d}	117.33^{c}	193.33^{a}	8.261	0.011^{*}
Cholesterol (mg/dl)	92.00^{b}	101.66^{a}	84.66^{b}	98.33^{a}	4.123	0.018^{*}
Triglycerides(mg/dl)	135.00^{a}	98.66^{b}	86.00^{c}	91.33^{b}	2.99	0.001^{**}
Total Lipids (mg/dl)	725.26^{b}	701.32^{c}	604.91^{d}	766.22^{a}	9.18	0.00**
T3	2.08^{a}	1.79^{b}	1.40^{c}	1.32^{d}	0.17	0.050^{*}
T4	33.28^{a}	34.84^{a}	20.12^{b}	19.46^{b}	1.24	0.00**

a, b: Means in the same row having different superscripts are significantly different at ($P<0.05$)

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الملخص العربى

بعض الإستجابات الفسيولوجية لإضافة مستويات مختلفة من البيتايين لعلائق كتاكيت التسمين

زينهم شيخون حسن اسماعيل و إيناس أحمد محمد أحمد
قسم الإنتاج الحيوانى- كلية الزراعة - جامعة جنوب الوادى
قسم الإنتاج الحيواني والداجني - كلية الزراعه والموارد الطبيعيه-جامعه أسوان

أجريت هذة التجربة لدراسة الإستجابة الفسيولوجية لإضافة مستويات مختلفة من البيتايين لعلائق كتاكيت التسمين. تم استخدام عدد 120 كتكوت تسمين (كب) عمر يوم حيث تم تقسيمهم عشوائيا الى اربعه مجموعات كل مجموعة ثلاثة مكررات بكل من 10 طيور وكان تسكين الطيور فى اقفاص من السلك المجلفن وكان توزيع المجموعات كالتالى:-

1. المعاملة الأولى بدون اى اضافات واستخدمت ككنترول.
2. المعاملة الثانية العليقة الكنترول + 0.01 % بيتايين .
3. المعاملة الثالثة العليقة الكنترول + 0.02 % بيتايين ..
4. المعاملة الر ابعة العليقة الكنترول + 0.03 % بيتايين

تم قياس كل من الماكول اليومى بالجرام و وزن الجسم اسبوعيا وكذا تم حساب الزيادة فى الوزن و معدل التحويل الغذائى، كما تم عمل تجربة ذبح فى نهاية التجربة وكذا تم قياس تركيزات كل من البروتين الكلي للسيرم و الالبومين و الجلوبيولين و الجلوكوز و الكولستيرول و الجلسريدات الثلاثية و الدهون الكلية و الثيروكسين فى الدم.
و خلصت نتائج التجربة الى ان استخدام البيتافين بتركيز 0.03 % حسن من بعض صفات الآداء الفسيولوجى و الإنتاجى لكتاكيت التسمين حيث ان استخدامة بتركيز 0.03 % ادى الى زيادة وزن الجسم و الزيادة فى وزن الجسم مقارنة بالمجموعه الكنترول وكذا كانت هناك اختلافات معنوية بين المعاملات لتركيزات كل من البروتين الكلي للسيرم و الجلوبيولين و الجلوكوز و الكولستيرول و الجلسريدات الثلاثية و الدهون الكلية و الثيروكسين فى الدم مع اختلاف المعاملات. بينما وزن الذبيحة تحسن مع استخدام 02% بيتايين فى العليقة.
ونستنتج من هذة الدراسة ان استخدام البيتافين بتركيز 0.03 % حسن من بعض صفات الاداء الفسيولوجي و الإنتاجي لكتاكيت التسمين.