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Effect of partial substitution of crude glycerol as an alternative energy source to diets in productive performance and some blood parameters of broiler

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Abstract

This work was performed to investigate the effect of crude glycerol as an energy source in broiler on performance, blood biochemical constituents, Carcass yield and internal organ weights. A total of 240 Ross 308 broiler chicks (males) were distributed in a completely randomized design, with five treatments (0, 2.5 , 5 , 7.5 , and 10% crude glycerol from 1 to 42 days), with four replicates(12 chick/replicate). All diets were formulated to meet the same requirements. Broiler were fed with water and food *ad libitum*. The data of Body Weight Gain (BWG), Feed Consumption (FC) were recorded weekly and Feed Conversion Ratio (FCR) was also calculated. Birds fed diets with crude glycerol did not differ significantly in performance from those fed the control diet without crude glycerol. Birds receiving crude glycerol 5% for initially two weeks showed highest body weight gain and the better feed conversion ratio all over the experimental period , supplementation 10% crude glycerol recorded lowest values of body weight gain and feed conversion ratio. Glycerol inclusion at 5% of diet can be used as an effective source of energy in broilers, especially from 0 to 21 d of age. Glycerol supplementation had no adverse effects on the production performance , blood biochemical constituents , internal organ weights or carcass yield of broilers.

Keywords: Glycerol, production performance, biochemical constitutes, broiler.

Introduction

There is a production of crude glycerol above the market demand, which drives the researchers to seek new uses of this byproduct. Due to the lack of legislation to dispose the crude glycerol produced in excess, this by-product can become in this way, an environmental problem. Currently there is a lot of focus on the use of crude glycerol as a less expensive energy source in poultry diets. According to [1] , from each 1000 kg of biodiesel produced, about 100 kg of glycerin is obtained. Glycerin, known as glycerol or glycerine, is the principal co-product of biodiesel production, produced through a NaOH- or KOH- catalyzed transesterification of the triacylglycerols in oils or fats with an alcohol. Glycerol is known to be a valuable ingredient for producing food, soaps, cosmetics and pharmaceuticals. Currently, with plenty of glycerol available to the world market, more uses are expected to develop, especially as a potential energy source for poultry diets, with approximately 4,100 kcal/kg gross energy [2]. Glycerol is a sugar alcohol that exists naturally in foods and in living tissues. It is constantly being produced by the hydrolysis of lipids in the gastrointestinal tract and absorbed by the mucosa. Glycerol readily forms neutral fats, fatty acid esters, and phosphoglycerides that are widely distributed in living organisms [3]. Several researchers have reported that crude glycerol is an acceptable feed ingredient for broiler chicks up to 5% without adverse effect on performance, [4, 5, 6]. However, increasing dietary glycerol above 10% has been shown to adversely affect growth performance and meat yield of broiler chickens, although this may be due to feed flow ability and associated feed consumption [5]. Also, Simon, et.al 1996; and Simon, et.al 1997 [7 , 8] indicated that adding crude glycerol content up to 10% of broiler diet did not affect broiler performance. The purpose of the study was to investigate the effect of crude glycerol inclusion on the growth performance, blood biochemical constituents and internal organ weights of broiler chickens.

Materials And Methods

Experimental design and feeding

A total of 240 broiler chicks male (Ross 308) with an average body weight 38 gm, were obtained from a commercial hatchery. At hatch, broilers were weighed, wing-banded, and allotted to pens and dietary treatments so that the initial mean weight was similar among treatment groups and placed in 20 floor pens (12 birds/pen) and randomly assigned to five dietary treatments (4 four replicates/treatment) The experiment lasted for 42 days. Temperature was controlled and gradually decreased from 35°C on the first day to 22°C at 21 days of age. Broilers were fed a starter diet (0 to 14 d of age) , grower diet (15 to 28 d of age) and a finisher diet (29 to 42 d of age). Diets were formulated to be adequate in all nutrients [9]. The crude glycerol was obtained from a commercial facility (Turkey) that used plant oil as an primary substrate and its analyzed in a nutrition laboratory (commercial) to determine composition (Table 1).

Table 1: Characteristics of crude glycerol fed to broiler (as is basis)*.

Characteristics	%
Glycerol	86.95
Dry matter (%)	73.68
Crude protein (%)	0.41
PH	1.67
Moisture (%)	9.22
Ash (%)	3.19
Methanol (%)	0.028
Crude fat (%)	0.12
Sodium (%)	1.26
Sodium chloride (%)	3.9
Metabolizable energy kcal/kg	3625

* Chemical analysis of crude glycerin, were determined according to the procedures outlined by Association of Official Analytical Chemists [10].

Each pen was equipped with manual plastic feeder and an automatic nipple drinker, Water and experimental diets were provided *ad libitum*. The ingredients and chemical composition of the diets are presented in Table (2, 3 and 4). Crude glycerol used at the level 2.5, 5, 7.5 and 10% in diets of the second, third,fourth and fifth treatment groups, respectively. All chicks were individually weighed and FI was recorded at weekly intervals . Based on the recorded data, Feed Intake (FI), Feed Conversion Ratio (FCR), and Body Weight Gain (BWG) were subsequently calculated based on performance values. On day 42 of trial, eight birds representing treatment were randomly selected, allowed to fast for 12 hours, slaughtered. Chemical analyses of plasma were carried out for quantitative determination of blood parameters (plasma total Proteins, Albumin, Globulin, Triglycerides, Cholesterol, HDL-cholesterol, LDL-cholesterol, VLDL-cholesterol, AST, ALT, alkaline phosphatase were determined using commercial kits, following the same steps as described by manufactures.

Statistical analysis

Data were analyzed based on a completely randomized design by using the General Linear Models Procedure in SAS [11]. Pens were treated as the experimental unit. Significant differences among treatment groups were further analyzed using Duncan's multiple-range test. A significant level of P <0.05 was implemented.

Table 2: Ingredient and calculated analysis of the starter diets during 1-14 days.

Ingredient %	Glycerol supplementation (%)				
	0	**2.5**	**5**	**7.5**	**10**
Yellow corn	58.62	55.6	52.5	49.4	46.4
Soybean meal, (48%)	32.5	33	33.6	34.2	34.7
Protein concentrate *	5	5	5	5	5
Glycerol	–	2.5	5	7.5	10
Vegetable oil	1.5	1.5	1.5	1.5	1.5
Limestone	1.28	1.3	1.34	1.34	1.34
Dicalcium phosphate	0.3	0.3	0.3	0.3	0.3
DL-methionine	0.53	0.53	0.53	0.53	0.53
L-lysine	0.15	0.15	0.11	0.11	0.11
Salt	0.12	0.12	0.12	0.12	0.12
Total	100	100	100	100	100
Chemical composition, Calculated**					
ME, kcal/kg	2997	2997	2997	2997	2997
CP, %	22.6	22.6	22.6	22.6	22.6
Crude fat	4.3	4.2	4.1	4.0	3.9
Crude fibre	2.7	2.6	2.6	2.5	2.5
Met. + Cys.	1.03	1.03	0.98	0.98	0.97
Lysine	1.40	1.41	1.42	1.43	1.43
Ca	0.92	0.93	0.95	0.95	0.95
Available phosphorus	0.45	0.45	0.45	0.44	0.44

* Protein concentrate contains: 40% CP, 5% Ca,3.7% Methionine, 4.12% Methionine and Cystine, 3.85% Lysine , 4.68% AP, Metabolizable Energy 2107 Kcal kg^{-1}, 2.50 mg Sodium, 1.70 mg threonine, 0.42mg Tryptophan, 4.20 mg choline and each 1 kg of this concentrate contain: 100000 IU vitamin A, 33000; IU vitamin D3, 100 mg; vitamin E, 2.55 mg; vitamin K3, 25 mg; vitamin B1, 10 mg; B2, 50 mg; vitamin B6, 24 mg vitamin B12; 51 mg niacin; 1.5 mg folic acid; 15 mg; biotin,500 µg and 13.5 mg pantothenic acid.
**Calculated based on feed consumption Tables of [9].

Table 3: Ingredient and calculated analysis of the grower diets during 15-28 days.

Ingredient %	Glycerol supplementation (%)				
	0	**2.5**	**5**	**7.5**	**10**
Yellow corn	64.8	61.8	58.7	55.7	52.5
Soybean meal, (48%)	26	26.5	27.1	27.6	28.3
Protein concentrate *	5	5	5	5	5
Glycerol	–	2.5	5	7.5	10
Vegetable oil	2.1	2.1	2.1	2.1	2.1
Limestone	1.35	1.35	1.35	1.35	1.35
Dicalcium phosphate	0.1	0.1	0.1	0.1	0.1
DL-methionine	0.35	0.35	0.35	0.35	0.35
L-lysine	0.15	0.15	0.15	0.15	0.15
Salt	0.15	0.15	0.15	0.15	0.15
Total	100	100	100	100	100
Chemical composition, Calculated**					
ME, kcal/kg	3100	3100	3100	3100	3100
CP, %	20.0	20.0	20.0	20.0	20.0
Crude fat	5.1	5.0	4.9	4.7	4.6
Crude fibre	2.5	2.5	2.5	2.4	2.4
Met. + Cys.	0.96	0.96	0.96	0.95	0.95
Lysine	1.25	1.25	1.26	1.27	1.28
Ca	0.89	0.89	0.89	0.89	0.89
Available phosphorus	0.40	0.40	0.40	0.40	0.40

* Protein concentrate contains: 40% CP, 5% Ca,3.7% Methionine, 4.12% Methionine and Cystine, 3.85% Lysine , 4.68% AP, Metabolizable Energy 2107 Kcal kg^{-1}, 2.50 mg Sodium, 1.70 mg threonine, 0.42mg Tryptophan, 4.20 mg choline and each 1 kg of this concentrate contain: 100000 IU vitamin A, 33000; IU vitamin D3, 100 mg; vitamin E, 2.55 mg; vitamin K3, 25 mg; vitamin B1, 10 mg; B2, 50 mg; vitamin B6, 24 mg vitamin B12; 51 mg niacin; 1.5 mg folic acid; 15 mg; biotin,500 µg and 13.5 mg pantothenic acid.
**Calculated based on feed consumption Tables of [9].

Table 4: Ingredient and calculated analysis of the finisher diets during 29-42 days.

Ingredient %	Glycerol supplementation (%)				
	0	**2.5**	**5**	**7.5**	**10**
Yellow corn	65.6	62.6	59.5	56.4	53.39
Soybean meal, (48%)	23.8	24.3	24.9	25.5	26
Protein concentrate *	5	5	5	5	5
Glycerol	–	2.5	5	7.5	10
Vegetable oil	3.5	3.5	3.5	3.5	3.5
Limestone	1.25	1.25	1.25	1.25	1.25
Dicalcium phosphate	0.3	0.3	0.3	0.3	0.3
DL-methionine	0.35	0.35	0.35	0.35	0.35
L-lysine	0.11	0.11	0.11	0.11	0.12
Salt	0.09	0.09	0.09	0.09	0.09
Total	100	100	100	100	100
Chemical composition, Calculated**					
ME, kcal/kg	3199	3199	3199	3199	3199
CP, %	19.0	19.0	19.0	19.0	19.0
Crude fat	6.5	6.4	6.3	6.1	6.0
Crude fibre	2.5	2.4	2.4	2.3	2.3
Met. + Cys.	0.90	0.89	0.89	0.88	0.89
Lysine	1.14	1.14	1.15	1.16	1.17
Ca	0.85	0.85	0.85	0.85	0.85
Available phosphorus	0.40	0.40	0.40	0.40	0.40

* Protein concentrate contains: 40% CP, 5% Ca,3.7% Methionine, 4.12% Methionine and Cystine, 3.85% Lysine , 4.68% AP, Metabolizable Energy 2107 Kcal kg^{-1}, 2.50 mg Sodium, 1.70 mg threonine, 0.42mg Tryptophan, 4.20 mg choline and each 1 kg of this concentrate contain: 100000 IU vitamin A, 33000; IU vitamin D3, 100 mg; vitamin E, 2.55 mg; vitamin K3, 25 mg; vitamin B1, 10 mg; B2, 50 mg; vitamin B6, 24 mg vitamin B12; 51 mg niacin; 1.5 mg folic acid; 15 mg; biotin,500 µg and 13.5 mg pantothenic acid.
**Calculated based on feed consumption Tables of [9].

RESULTS

The effects of the dietary treatments on production performance are shown in Table 5. Results showed that at 14 days of age, the birds fed the diets with 5% glycerol has significantly increased in the body weight gain than birds fed the diets with 2.5, 7.5 and 10% glycerol. Also the results showed birds fed diets with 5, 7.5 and 10% glycerol, recorded the highest values of feed intake compared with the bird fed diet with 2.5% glycerol. However, at 14 days of age birds fed basal diets and birds fed diet with 5% glycerol showed best values in feed conversion ratio compared with birds fed diets 2.5 , 7.5 and 10% glycerol. No significant differences in average values of body weight gain or feed conversion ratio were noted at 28 days of age between birds fed diets with different levels of glycerol. Moreover, the inclusion of 5% crude glycerin in broiler dies recorded highest feed intake compared to control or the other treatments during 28 days of age. The high rates of feed consumption of rations containing glycerol may be due to the sweet taste and that improved absorption and utilization in birds gut [2]. On the other hand, Pure glycerol is the largest part of raw glycerol, can interfere with nutrients passage within the gastrointestinal tract , reducing utilization of feed energy. Therefore, the feed consumption rate will increases as a form of compensatory response [12]. Also, at 42 days of age It is noticed that body weight gain , feed intake and feed conversion ratio for treatments were almost comparable. The present findings are analogous with the findings of [13] also with [13] who found that the crude glycerin content of the rations did not affect body weight gain or feed conversion ratio. Also, they showed that using five or 10% crude glycerol in broiler diets instead of corn starch had no adverse effects. The economic evaluation of the use of glycerol is rather problematic due to the varying prices of individual feed components. The price of crude glycerol is likely to decrease with the overproduction of glycerol generated in the production of methylester. The use of crude glycerol in diets will therefore become very interesting from an economic point of view. It follows from our results data that the optimum level of crude glycerol in feed for broiler is 5%. If the diet programmed is designed properly, it is possible to use 10% of crude glycerol in a diet without a negative effect on the performance indicators of animals. However, there are still many issues associated with the use of glycerol in the nutrition of poultry that have to be addressed in future research studies.

The effect of different levels of crude glycerol on blood biochemical traits are shown in Table 6. It was observed in the present study that Crude glycerol supplementation had no significant ($P < 0.05$) effect on serum levels of total protein, Albumin, globulin, glucose, cholesterol, HDL , VLDL AST, ALT and ALP. Inclusion of 5 % glycerol resulted in a significant decrease in Triglycerides , Cholesterol and LDL compared to the diet of control treatment which recorded highest values. most serum biochemical parameters appeared to be did not affect by dietary glycerol supplementation in the present study. The circulating triglyceride concentrations were significantly reduced in birds receiving 5% glycerol, suggesting improved tissue storage or utilization of lipids. These results are in agreement with [14] who indicated that there was no significant effect due to the addition of 2, 4 ,6 and 8% glycerol on some blood biochemical traits. By contrast, [15] reported no signification variation of the tissue cholesterol concentrations in liver and in the semi membranous muscle in glycerol supplemented pigs. Moreover, Different responses to supplementary glycerol among the literatures might be due to the species, composition of glycerol used, composition of the diet, levels of glycerol in the diet and the duration of supplementation.

Table 5: Effect of partial substitution of crude glycerol as an alternative energy source on body weight gain, feed intake and feed conversion ratio of broiler chicken[1].

Items	Glycerol Supplementation (%)					SEM	*P*-value
	0	**2.5**	**5**	**7.5**	**10**		
Total Protein (gm/dl)	3.75	3.68	3.35	3.38	3.46	0.24	0.23
Albumin (gm/dl)	1.92	2.11	1.72	1.91	1.88	0.18	0.27
Globulin (gm/dl)	1.83	1.57	1.63	1.47	1.58	0.14	0.32
Glucose (mg/dl)	186.83	192.5	188.5	188.2	196.8	10.70	0.73
Triglycerides (mg/dl)	135.7 a	119.9 b	127.2 ab	141.4 a	129.5 ab	14.12	0.76
Cholesterol (mg/dl)	152.0 a	135.8 ab	128.1 b	144.2 a	146.8 a	6.75	0.18
HDL (mg/dl)	47.5	51.3	47.9	52.6	53.3	2.72	0.24
LDL (mg/dl)	77.3 a	60.5 b	54.6 b	63.3 ab	67.6 ab	7.61	0.13
VLDL (mg/dl)	27.14	23.98	25.44	28.28	25.9	3.44	0.22
ALP (U/L)	355.7	335.5	342.3	403.3	367.1	14.33	0.18
ALT (U/L)	6.67	6.33	7.00	7.33	7.67	4.58	0.72
AST (U/L)	161.00	172.67	163.33	160.00	170.67	0.67	0.81

1 Data represent mean values of 4 replicates per treatment.
a–b Means in the same row with different superscripts differ ($P < 0.05$).

Table 6: Effect of partial substitution of crude glycerol as an alternative energy source on Blood biochemical parameters of broiler chicken .[1]

Items	Glycerol Supplementation (%)					SEM	*P*-value
	0	**2.5**	**5**	**7.5**	**10**		
0-14 days							
BWG (g)	329 ab	316 b	337 a	325 b	324 b	8.41	0.4714
FI (g)	361 ab	359 b	371 a	374 a	370 a	6.92	0.0621
FCR	1.09 b	1.13 ab	1.10 b	1.15 a	1.14 a	0.03	0.4532
15-28 days							
BWG (g)	790	789	808	755	765	26.67	0.2417
FI (g)	1428 b	1450 ab	1468 a	1456 ab	1454 ab	17.88	0.6269
FCR	1.81	1.83	1.81	1.92	1.90	0.03	0.1634
29-42 days							
BWG (g)	1340	1367	1357	1380	1347	36.05	0.5217
FI (g)	2337	2370	2393	2341	2365	23.27	0.7858
FCR	1.74	1.73	1.76	1.70	1.75	0.04	0.5430
0-42 days							
BWG (g)	2459	2472	2502	2460	2436	36.31	0.0514
FI (g)	4126	4180	4232	4179	4190	35.72	0.7262
FCR	1.68	1.69	1.69	1.70	1.72	0.01	0.0172

1 Data represent mean values of 4 replicates per treatment.
a–b Means in the same row with different superscripts differ ($P < 0.05$).
HDL: high density lipoprotein, LDL: low density lipoprotein, VLDL: very low density lipoprotein, ALT: alanine aminotransferase, AST: aspartate aminotransferase, ALP: alkaline phosphatase.

Table 7: Effect of partial substitution of crude glycerol as an alternative energy source on Carcass yield and internal organ weights (%) of broiler chicken

Items	Glycerol Supplementation (%)					SEM	*P*-value
	0	**2.5**	**5**	**7.5**	**10**		
Hot carcass yield %	77.6	76.9	76.8	73.9	74.8	0.73	0.239
Liver %	1.75	1.64	1.59	1.71	1.78	0.13	0.828
Kidney %	0.13	0.12	0.12	0.11	0.11	0.03	0.462
Proventriculus %	0.425	0.413	0.420	0.413	0.453	0.06	0.907
Gizzard %	1.965	1.889	1.880	2.012	1.936	0.12	0.924
Heart%	0.426	0.488	0.456	0.412	0.433	0.03	0.998
Pancreas %	0.224	0.214	0.233	0.227	0.241	0.061	0.935
Abdominal fat %	2.10 a	2.02 ab	1.97 b	1.91 bc	1.88 c	0.29	0.36
Spleen %	0.11	0.13	0.11	0.13	0.12	0.05	0.68
Bursa %	0.14	0.11	0.13	0.10	0.11	0.05	0.97

1 Data represent mean values of 4 replicates per treatment.
a–c Means in the same row with different superscripts differ ($P < 0.05$).

The inclusion of crude glycerol in the diets did not affect the internal organ weights of broiler , except for the abdominal fat (Table 7). The results of our study showed that supplementation of 2.5, 5, 7.5 and 10% of crude glycerol in the diets had no negative effect on carcass yields or the liver, kidney, proventriculus , gizzard, heart , Pancreas , Spleen or bursa weights of birds. Also , the relative weights of some internal organs, such as the heart and liver, are known to be related to live body weight , therefore, we observed that feeding glycerol has no effects on difference of weights. Few studies have demonstrated the effect of diets containing glycerol on carcass yield were in agreement with the studies reported in the literature [14] observed broilers consuming feeds with different levels of glycerol did not differ significantly from birds fed the control diet for dressing percentage, the percentages of most internal organs, or weight of immune organs. Similar results were reported by [16] who observed that inclusion of glycerol 40 or 80 g of glycerol/kg in the diets did not affect the internal organ weights of broilers (females or males). Results in the present study were in contrast with the results of [17] , who observed that liver, gizzard, and heart relative weights of broilers fed a diet with 50 g of glycerol/kg were lower than those for the control group. Also, many authors reported that heart and liver weights of broilers fed 100 g of glycerol in the diets were higher than those for the cotrol group. However, in broilers dietary glycerol decreases the rate of fatty acid synthesis and lipogenic enzyme activities in the liver [18]. Moreover, in rats when glycerol and fat are fed together, they can reduce plasma cholesterol and liver lipids and testerification of fatty acids in rat fat tissue [19].

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