Crude Glycerin (Biodiesel By-product) as an alternative energy source in poultry diets

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Abstract

This study was aimed to study the possibility of using glycerin (biodiesel by product) as alternative energy source in broiler diets and effects on productive performance ,biochemical blood , Carcass traits and organ weights. Two-Hundred forty broiler chicks (Ross 308), one day old were divided to five groups (0, 2.5 , 5 , 7.5 , and 10% supplemented with crude glycerin from 1 to 42 days), with four replicates per group (12 chick/replicate). Diets were formulated Iso-nitrogenous and iso-caloric to meet the requirements of birds. Nutrition were *ad libitum*. Body weight gain, feed consumption were recorded weekly. Results showed no significant differences between groups in performance treatments from those fed the control diet without crude glycerin. Birds in group 3 which received crude glycerin 5% at 14 days recorded highest body weight with better feed conversion ratio during over all experimental period , while birds in group 5 (10% crude glycerin recorded lowest body weight with worst feed conversion ratio. Also, the results of biochemical traits indicated no significant differences between treatments in total protein , albumin , globulin, Glucose, HDL ,VLDL, ALP , ALT and AST Enzymes at 42 days. On the other hand, birds in group 2 (addition 2.5% crude glycerin) recorded lowest value in Triglycerides, While group 3 (addition 2.5% crude glycerin) recorded lowest value in total Cholesterol and low density lipoproteins. No significant differences between treatments in Relative weights of Hot carcass yield, liver, Kidney, proventiculus , gizzard , Heart , Pancreas, spleen and Bursa, while significant decrease were observed in Relative weights of abdominal fat of birds fed diets with crude glycerin as compared with Control Group .

We conclude from this work that diets supplementation with 5% crude glycerin can be used as an alternative source of energy, especially from 0 to 21 day .

Keywords: Crude glycerin, productive performance, biochemical, carcass, broiler.

Introduction

Energy sources represent the largest nutritional needs, they represent the largest proportion of feed costs, and accordingly, the biological value of the feed depends on the amount of energy it contains and also on the energy relationship with other food components in the feed such as the protein and energy relationship, which is considered one of the most important nutritional relationships and on the basis of which the feed is formed Which must contain the minimum amount of energy that is appropriate to the bird's age, type and production, due to its effect on the productive performance of birds (Summers and lesson, 2005; Plumstead, 2008).Raw glycerin is a by-product from oil extraction and biofuel production process, which has increased dramatically in recent years (ANP, 2015). glycerin is considered one from alternative energy sources because it has a good total energy content (4320 kcal/kg). The raw glycerin contains pure glycerol at (80-95%) and because of the high energy values of this compound, attention is drawn to the possibility of using it in animal feeding (Carvalho *etal*., 2012; Gomide *etal*., 2012; Chanjula *etal*., 2014; Guiomar *etal* l., 2017; Hyeok *etal*., 2017). The raw materials for production biofuels can be vegetable oils (cotton seeds, peanuts, canola, palms, sunflowers, soybeans, etc.) and animal fats (such as fish oil, animal grease and Tallow) or even fat waste from domestic, commercial or industrial processing (Hanna and Ma, 1999; Van Gerpen *etal*., 2005; Thompson and He, 2006; ANP, 2015). The type of raw materials and the biofuel production process affect the formation and quality of raw glycerin (Hansen *etal*., 2009). About 10% of the production of crude glycerin is associated with the production and manufacture of oils , for every three moles of ethyl esters, one mole of raw glycerin is produced (Karinen and Krause, 2006; Pagliaro and Rossi, 2008; Rahmat *etal*., 2010; Lenardão *etal*., 2017) and increase Biofuel production every year and thus increasing the production of raw glycerin opens up prospects for its potential in chemical and industrial production (ANP, 2015). Mandalawi *etal*. (2014) studied the effect of adding two sources of raw glycerin in broiler's diets with 2.5, 5, 7.5 and 10%, The results indicated that the levels of fat concentration in the liver were not significantly affected compared to the control treatment. Some studies also indicated that the value of glycerin determined by the energy level and not by method of manufacture that did not affect the productive characteristics which not effect on birds performance within 1-21 days of the bird's life. Henz *etal*. (2014) indicated that protein, lipid and dry matter were not significantly affected ($P \leq 0.05$) in broiler carcasses fed on diets supplemented with raw glycerin 3, 6, 9, 12 and 15% .Moreas *etal*. (2016) explained that adding pure glycerin 1 and 2% to drinking water did not significantly affect on characteristics and productive performance of birds compared to birds in control treatment during hot and temperate climates. The results of study Da Silva *etal*. (2017) did not indicate any significant differences between control treatment and treatments included adding pure glycerin in broiler (Cobb 500) diets on the productive performance (body weight, weight gain, food conversion ratio) and ratio of protein, fat, moisture in meat and carcass traits. Roll *etal*. (2018) did not notice any significant effect on deposition of saturated and unsaturated fatty acids in carcasses and cuts of broiler meat fed on diets based on palm oil supplemented with crude glycerin with 4, 8 and 16%. Mousa *etal*.,(2018) mentioned no negative effects by addition crude glycerol to broiler diets on productive performance and physiological traits.

Therefore, this study aimed to verify the possibility of adding raw glycerin in broiler diets as an alternative source of traditional energy sources and its effect on productive performance, some biochemical traits , internal organ weight and carcass yield.

Materials and Methods

This study was conducted in poultry field belong to Animal Production Department in College of Agriculture/University of Anbar. The aim of studying was studying the possibility of adding raw crude glycerin to broiler diets and its effect on productive, physiological performance and carcass yield. Two hundred –forty , One-day old unsexed Ross (308) chicks with average weight 38 gm. randomly distributed to five treatments with four replicates per treatment (12 chicks / replicate), the experimental treatments included control treatment, second , third, fourth and fifth treatments fed basal diet with glycerin at levels (2.5, 5, 7.5 and 10) % respectively. The birds fed diets formulated to meet birds requirements based with guide of breed . Diets were iso-caloric and iso-nitrogenous (NRC, 1994) . Broilers were fed a three phases feeding program starter, grower and a finisher diets. The crude glycerin was extracted from a soybean oil as an initial substrate in Laboratories of Ministry of Science and Technology (Baghdad) and its composition was analyzed and its composition was analyzed in a commercial laboratory (Table 1).

Table (1) . Chemical Composition of Crude Glycerin.*

Characteristics	**%**
Glycerol	86.88
Dry matter	72.61
Crude protein	0.44
PH	1.68
Moisture	9.21
Ash	3.18
Methanol	0.026
Crude fat	0.13
Sodium	1.27
Sodium chloride	3.8
ME kcal/kg	3624

* Chemical analysis of crude glycerin were determined According to (A.O.A.C, 2000).

Each replicate (pen) was supplemented with manual feeder and an automatic nipple drinker, feeds and water were *ad libitum*. The diets and chemical composition were presented in Tables (2)(3)(4). Crude glycerin was supplemented with levels 2.5, 5 , 7.5 and 10% in diets 2^{nd} , 3^{rd} ,4^{th} and 5^{th} treatments respectively. All birds were individually weighed and Feed consumption was recorded weekly. Based on data recorded, body weight gain (BWG) , feed consumption (FC) and feed conversion ratio (FCR) were calculated based on productive performance. On day 42 of age eight birds from each treatment randomly selected, blood samples were collected from brachial vein to determined biochemical traits which included (total Proteins, total Albumin, total Globulin, Triglycerides, Cholesterol, HDL, LDL, VLDL, AST, ALT, ALP) were determined. After blood test the birds allowed to fasting for 10 hours and then slaughtered , internal organs (liver , kidney, Proventriculus, gizzard, heart , pancreas, abdominal fat , spleen and bursa) were weighted .

Table (2). Ingredient and chemical analysis of starter diets.

Maize	58.62	55.6	52.5	49.4	46.4
Soybean (48%)	5.32	33	33.6	34.2	34.7
Animal Protein *	5	5	5	5	5
Crude Glycerin	–	2.5	5	7.5	10
Sunflower 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
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**					
M.E. (kcal/kg)	2998	2998	2998	2998	2998
Crude Protein %	22.5	22.5	22.5	22.5	22.5
Fat	4.3	4.2	4.1	4.0	3.9
Crude fibre	2.7	2.6	2.6	2.5	2.5
Meth. + Cyst.	1.03	1.03	0.98	0.98	0.97
Lys.	1.40	1.41	1.42	1.43	1.43
Calcium	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) : 40.5% C.P 5%, Ca3.6%, Methionine 4.11% ,Methionine and Cystine 3.84% , Lysine 4.67%, Metabolizable Energy 2104 Kcal $/kg$; 2.50 mg. Sodium, 1.70 mg. threonine 0.42mg, Tryptophan, 4.20 mg choline and each 1 kg of concentrate contains: 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 intake Tables of National Research Council (1994).

Table (3). Ingredient and chemical analysis of grower diets .

Maize	64.8	61.8	58.7	55.7	52.5
Soybean (48%)	26	26.5	27.1	27.6	28.3
Animal Protein *	5	5	5	5	5
Crude Glycerin	–	2.5	5	7.5	10
Sunflower 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
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**					
M.E. (kcal/kg)	3101	3101	3101	3101	3101
Crude Protein %	20.0	20.0	20.0	20.0	20.0
Fat	5.1	5.0	4.9	4.7	4.6
Crude fibre	2.5	2.5	2.5	2.4	2.4
Meth. + Cyst.	0.96	0.96	0.96	0.95	0.95
Lys.	1.25	1.25	1.26	1.27	1.28
Calcium	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) : 40.5% C.P 5%, Ca3.6%, Methionine 4.11% ,Methionine and Cystine 3.84% , Lysine 4.67%, Metabolizable Energy 2104 Kcal $/kg$; 2.50 mg. Sodium, 1.70 mg. threonine 0.42mg, Tryptophan, 4.20 mg choline and each 1 kg of concentrate contains: 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 intake Tables of National Research Council (1994).

Table (4). Ingredient and chemical analysis of finisher diets .

Maize	65.6	62.6	59.5	56.4	53.39
Soybean (48%)	23.8	24.3	24.9	25.5	26
Animal Protein *	5	5	5	5	5
Crude Glycerin	–	2.5	5	7.5	10
Sunflower 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
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**					
M.E. (kcal/kg)	3200	3200	3200	3200	3200
Crude Protein %	19.0	19.0	19.0	19.0	19.0
Fat	6.5	6.4	6.3	6.1	6.0
Crude fibre	2.5	2.4	2.4	2.3	2.3
Meth. + Cyst.	0.90	0.89	0.89	0.88	0.89
Lys.	1.14	1.14	1.15	1.16	1.17
Calcium	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) : 40.5% C.P 5%, Ca3.6%, Methionine 4.11% ,Methionine and Cystine 3.84% , Lysine 4.67%, Metabolizable Energy 2104 Kcal /*kg*; 2.50 mg. Sodium, 1.70 mg. threonine 0.42mg, Tryptophan, 4.20 mg choline and each 1 kg of concentrate contains: 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 intake Tables of National Research Council (1994).

Statistical Analysis :-

Data was analyzed by Statistical Analysis System (SAS) (2012) to study the effect of different factors on the characteristics studied according to a complete random design (CRD) , and the mean differences between the averages were compared by Duncan (1955) multiple-range test**.**

Results and discussion

Results in table 5. Showed the effect of substitute of crude glycerin on productive performance at 14 days of age, birds fed diets supplemented with 5% crude glycerin was increase significantly in live body weight than birds fed diets supplemented with 2.5 , 7.5 and 10% crude glycerin. Moreover, results in table (5) showed that birds fed on diets supplemented with 5, 7.5 and 10% crude glycerin recorded highest values in feed consumption as compared with birds fed diet with 2.5% crude glycerin which recorded lowest values. Also, birds fed control diet and T3 (5% crude glycerin) showed best values in feed conversion ratio as compared with birds fed diets supplemented with 2.5 , 7.5 and 10% crude glycerin at 14 days of birds age. Results of productive performance included not significant differences in the values of live body weight gain or feed conversion ratio at 28 days of age. Moreover, T3 (adding 5% crude glycerin) recorded highest values in feed consumption as compared with T1 or other treatments at 28 days of age. The increasing in feed consumption may be due to sweet taste of crude glycerin that improved consistency of feed, it can increase diets consumed and be more efficient absorbed in animals intestine (Min *etal*., 2010).On other hand, glycerol considered a large component of crude glycerin, and that could be decrease the passage of nutrient in digestive interact and increasing utilization in small intestine and that may be refer useful from energy uptake in animals (Barteczko and Kaminski 1999). However , statistical analysis showed at 42 days of age production performance characteristics which included (live body weight gain , feed consumption and feed conversion ratio) were not affected by addition of crude glycerin. The results of this study were in agreement with of Waldroup (2007) and (Abd-Elsamee *etal.* 2010) who mentioned that addition crude glycerin to feeds did not affect in productive performances by adding 5 or10% crude glycerol in broiler diets as an energy source replaced with yellow corn**.** Through results of this study it's clear that broiler feed supplemented with crude glycerin at 5-10% had no side effect on productive performance and that may be refer with economic evaluation due to high prices of feeds, and crude glycerin prices is decrease with overproduction in most the world. However, there are still many issues associated with using of crude glycerin in poultry nutrition that have to be studied in future .

Table (5). Inclusion Biodiesel by-product (crude glycerin) as an alternative energy source on body weight gain, feed consumption and feed conversion ratio of broiler.

Items	Glycerin Inclusion (%)					SEM	P-value
	0	2.5	5	7.5	10		
Starter							
BWG (g)	330 ab	317 b	338 a	326 b	325 b	8.42	0.4715
FC (g)	362 ab	360 b	372 a	375 a	371 a	6.93	0.0622
FCR	1.12 b	1.14 ab	1.11 b	1.16 a	1.15 a	0.04	0.4533
Grower							
BWG (g)	791	790	809	756	766	26.68	0.2418
FC (g)	1429 b	1451 ab	1469 a	1457 ab	1455 ab	17.89	0.6271
FCR	1.82	1.84	1.82	1.93	1.91	0.04	0.1635
Finisher							
BWG (g)	1341	1368	1357	1381	1348	36.06	0.5218
FC (g)	2339	2372	2395	2343	2367	23.29	0.7857
FCR	1.75	1.75	1.77	1.71	1.76	0.05	0.5431
0-42 day							
BWG (g)	2460	2473	2503	2461	2437	36.32	0.0515
FC (g)	4127	4181	4233	4180	4191	35.73	0.7263
FCR	69.1	1.70	1.70	1.71	1.73	0.02	0.0173

Means in same rows with different superscripts is differs at level (P< 0.05).

Effect of supplementation of crude glycerin with different levels on some blood biochemical tests were shown in Table (6). The addition of crude glycerin to diets had no significant effect in protein, globulin, Albumin, glucose, total cholesterol, high density lipoprotein , very low density lipoprotein, low density lipoprotein, GOT, GPT and alkaline phosphate. Results of table (6) shows birds fed diets include crude glycerin with 5% recorded significant decrease (P< 0.05) in Cholesterol , Triglycerides , low density lipoprotein as compared with T1 (control) which shows highest values and that decreasing may be due to crude glycerin improve lipogenesis in storage tissues by circulating of triglycerides in blood. By contrast, results of Mourot *etal.*(1994) indicated no significant differences cholesterol concentrations of liver tissues and semi membrane muscles by glycerin supplemented. Results in this study were in agreement with Abd-Elsamee *etal.*(2010) who declared that birds received diets with 2, 4 ,6 and 8% glycerol had not affect on some blood biochemical parameters. However , different views to glycerin supplementation in poultry diets might be due to glycerin species, glycerin used, composition of diets , levels of glycerin addition in diets and the duration of supplementation.

Table (6). Inclusion Biodiesel by-product (crude glycerin) as an alternative energy source on some blood biochemical parameters of broiler.

Item	Crude Glycerin (%)					SEM	P-value
	0	2.5	5	7.5	10		
Protein (*gm/dl*)	3.76	3.69	3.36	3.39	3.47	0.25	0.24
Albumin (*gm/dl*)	1.93	2.12	1.73	1.92	1.89	0.19	0.28
Globulin (*gm/dl*)	1.84	1.58	1.64	1.48	1.59	0.15	0.33
Glucose (*mg/dl*)	186.84	192.6	188.6	188.3	196.9	10.71	0.74
Triglycerides (*mg/dl*)	135.8 a	120.1 b	127.3 ab	141.5 a	129.6 ab	14.13	0.77
Cholesterol (*mg/dl*)	152.1 a	135.9 ab	128.2 b	144.3 a	146.9 a	6.76	0.19
HDL (*mg/dl*)	47.6	51.4	48.0	52.7	53.4	2.71	0.25
LDL (*mg/dl*)	77.5 a	60.7 b	54.8 b	63.5 ab	67.8 ab	7.63	0.15
VLDL (*mg/dl*)	27.15	23.99	25.45	28.29	26.0	3.45	0.23
ALP (*U/L*)	355.8	335.6	342.4	403.4	367.2	14.34	0.19
ALT (*U/L*)	6.68	6.34	7.01	7.34	7.68	4.59	0.73
AST (*U/L*)	162.00	173.68	164.34	161.00	171.68	0.68	0.82

Means in same rows with different superscripts is differs (P< 0.05).

Table **(7)** showed that inclusion of crude glycerin to diets did not affect on internal organ weights of birds , except for abdominal fat . We observed that levels 2.5 , 5 , 7.5 and 10% of crude glycerin had no negative effect on hot carcass yields , liver, kidney, Proventriculus , gizzard, heart , Pancreas , Spleen or bursa weights**.** The relative weights of some internal organs, such as heart and liver, are known to be related with body weight. Therefore, concluding from our study the differences were not based on feeding crude glycerin**.** A little of studies had demonstrated feeds containing crude glycerin on carcass yield in agreement with studies reported that Abd-Elsamee *etal.* (2010) who mentioned broilers consumed diets with different levels of glycerol did not differ with birds fed basal diet for dressing percentage, the relative weight of internal organs, weight of immune organs. Consistent results were noticed by Topal and Ozdogan (2013) who pointed out that addition of crude glycerin 40 or 80 g/kg feed did not affect on internal organ weights of broilers (unsexed). Some previous studies were in contrast with our results. Coşkun *etal.* (2007) , found that gizzard, heart, and liver percentage weights of broilers fed a diet with glycerol 50 g/kg were lower than control group. Jasim and Mousa (2018) reported that broiler fed diets supplemented with crude glycerol had no effect on relative weights of internal organs (liver , heart, gizzard, bursa and Proventriculus). Mousa *etal.*,(2018) noticed that significant decrease in abdominal fat of broiler fed crude glycerol with 0 , 2.5 , 5, 7.5 and 10%. Moreover , crude glycerin had the ability to decrease rate of fatty acid synthesis and lipogenesis enzymes activity in liver and tissues (Lin *etal.*, 1976). Narayan and Ross (1987) mentioned that rats fed glycerin with fat recorded decrease in plasma cholesterol , esterification of fatty acids in fat tissue and fat synthesis in liver .

Table (7). Inclusion Biodiesel by-product (crude glycerin) as an alternative energy source on Carcass yield and internal organ weights (%) of broiler.

Item	0	2.5	5	7.5	10	SEM	*P*-value
Hot carcass yield	77.8	77.0	76.9	74.0	74.9	0.74	0.240
Liver	1.76	1.65	1.60	1.72	1.79	0.14	0.829
Kidney	0.14	0.13	0.13	0.12	0.12	0.04	0.463
Proventriculus	0.426	0.414	0.421	0.414	0.454	0.07	0.908
Gizzard	1.966	1.890	1.881	2.013	1.937	0.13	0.925
Heart	0.427	0.489	0.457	0.413	0.434	0.04	0.999
Pancreas	0.225	0.215	0.234	0.228	0.242	0.062	0.936
Abdominal fat	2.11 a	2.03 ab	1.98 b	1.92 bc	1.89 c	0.30	0.37
Spleen	0.12	0.14	0.12	0.14	0.13	0.06	0.69
Bursa	0.15	0.12	0.14	0.11	0.12	0.06	0.98

Means in same rows with different superscripts is differs ($P < 0.05$).

References

Abd-Elsamee, M. O., Z. M. A. Abdo, M. A. F. El-Manylawi, and I. H. Salim. (2010). Use of crude glycerin in broiler diets. Egypt. Poult. Sci. 30:281–295.

ANP. 2015.(National Agency of Petroleum, Natural Gas and biofuels–Brazil). Biofuels.

AOAC, 2000. Official Methods of Analysis of AOAC International,17*ed*. Association of Official Analytical Chemists, AOAC International, Maryland.

Barteczko J., and J. Kaminski , (1999): The effect of glycerol and vegetable fat on some physiological indices of the blood and over fatness of broiler carcass. Annals of Warsaw Agricultural University – Animal Science, 36, 197–209.

Carvalho, P. L.O., Moreira I., Martins E. N., Piano L.M., Toledo J. B. and C.L. Costa Filho .2012. Crude glycerine in diets for piglets. Revista Brasileira de Zootecnia. 41:1654-1661.

Chanjula, P., P. Pakdeechanuan P. and S. Wattanasit. 2014. Effects of Dietary Crude Glycerin Supplementation on Nutrient Digestibility, Ruminal Fermentation, Blood Metabolites, and Nitrogen Balance of Goats. Asian Australas. J. Anim. Sci. Vol. 27, No. 3 : 365-374.

Coşkun, B., A. Şehu, S. Küçükersan, and H. K. Köksal. (2007). The use of glycerol as a byproduct of biodiesel in poultry rations. Pages 24–31 in IV Natl. Anim. Nutr. Congr., Uludağ University, Bursa, Turkey.

Da Silva, M.C., Marçal Vieira R.G., Rodrigues K.F., Da Silva G.F., Sousa L.F. , Fonseca F.L.R. , Campos C.F.A., Augusto W.F. , Parente I.P. and L. S. Bezerra.2017. Effects of purified glycerin in balanced diets of chicken broilers treated from 22 to 42 days of age. Semina: Ciências Agrárias, Londrina, v. 38, n. 4, p. 2083-2090.

Gomide, A.P.C., Brustolini P.C., Ferreira A.S., Paulino P.V.R., Lima A.L., Scottá B. A., Rodrigues V.V., Câmara L.R.A., Moita A.M.S., Oliveira Júnior G.M., Ferreira R.C., and Formigoni A.S. 2012. Substituição de milho por glicerina bruta em dietas para suínos em terminação. Arquivo Brasileiro de Medicina Veterinária e Zootecnia 64:1309-1316.

Guiomar, H. V., Anderson C., Douglas d. S.P., Ana P.S.T., Komiyama C.M. and Alexandre d. O. T.2017. Nutritional value of glycerin for pigs determined by different methodologies. R. Bras. Zootec., 46(7):584-590.

Hansen, C.F, Hernandez A., Mullan B.P, Moore K., Trezona-Murray M., King R.H, and J.R. Pluske. 2009. A chemical analysis of samples of crude glycerol from the production of biodiesel in Australia, and the effects of feeding crude glycerol to growing-finishing pigs on performance, plasma metabolites and meat quality at slaughter. Anim. Prod. Sci. 49:154-161.

Henz, J.R., Nunes R.V., Eyng C., Pozza P.C., Frank R., Schone R.A. and T.M.M. Oliveira. 2014. Effect of dietary glycerin supplementation in the starter diet on broiler performance. Czech J. Anim. Sci., (12): 557–563.

Hyeok, J. K., Min Y. P., Lee I.K., Hyun J. K., Min J. G., Cheol-Heui Y, Jagyeom S., and Myunggi B. 2017. Effects of ambient temperature and dietary glycerol addition on growth performance, blood parameters and immune cell populations of Korean cattle steers . Asian-Australas J. Anim. Sci. Vol. 30, No. 4:505-513 .

Jasim R.K. and Mousa B.H. 2018. Effect of adding crude glycerol to broiler diets on the performance and yield characteristics of carcass. Journal of Research in Ecology. Vol. 6(2), 1838-1845.

Karinen, R.S. and A.O.I. Krause. 2006. New biocomponent from glycerol. Applied Catalysis A: Gen., 306: 128-133.

Leeson, S. and J. D. Summers. 2005. Commercial Poultry Nutrition. (3rd Edition). Nottingham University Press, Nottingham, UK.

Lenardão, E.J., Borges E.L., Stach G., Soares L.K., Alves D., Schumacher R.F., Bagnoli L., Marini F., and G. Perin .2017. Glycerol as Precursor of Organoselanyl and Organotellanyl Alkynes. Molecules. 2 :22 (3).

Lin, M.H., D.R. Romsos and G.A. Leveille. (1976). Effect of glycerol on enzyme activities and on fatty acid synthesis in the rat and chicken, J. Nutr., 106: 1668-1677.

Ma, F., and M. A. Hanna. 1999. Biodiesel production: A review. Biores. Tech. 70:1-15.

Mandalawi, H. A., M. V. Kimiaeitalab , V. obregon , D. Menoyo , and G. G. Mateos.2014. Influence of source and level of glycerin in the diet on growth performance, liver characteristics, and nutrient digestibility in broilers from hatching to 21 days of age. Poultry Science 93 :2855–2863.

Min, Y., F. F. Yan, Z. Liu, C. Coto, and P. W. Waldroup. (2010). Glycerin—A new energy source for poultry. Int. J. Poult. Sci. 9:1–4.

Moraes, P.O., Ceron M.S., Borille R., Gianluppi R.D.F., Lovato G.D., Cardinal K.M., and A.M.L. Ribeiro.2016. Effects of the Addition of Pure Glycerin Supplementation in the Drinking Water on the Performance of Broilers Submitted to Heat Stress and Feed Restriction. Brazilian Journal of Poultry Science. Vol.18, 3 :413-418.

Mourot J., Aumaitre A., Mounier A., Peiniau P., and A.C. Fracois. (1994). Nutritional and physiological effects of dietary glycerol in the growing pig: Consequences on fatty tissues and post mortem muscular parameters. *Livest. Prod. Sci.*, 38, 237-244.

Mousa B.H., H.H. Nafa´a , Y.T. Al-Rawi and R.K.Al-Dulaimy.2018. Effect of partial substitution of crude glycerol as an alternative energy source to diets in productive performance and some blood parameters of broiler. Journal of Pharmaceutical Sciences and Research. Vol. 10(11), 2907-2911.

Narayan, K.A. and E.W. Ross. (1987). The interactive effect of glycerol and the type and level of fat on rat tissue lipids. Nutr. Rep. Int., 36: 335-343.

NRC.1994. Nutrient Requirements of Poultry. 9th rev. ed. Natl. Acad. Press, Washington, DC.

Pagliaro, M. and M. Rossi. 2008. The Future of Glycerol: New Usages for a Versatile Raw Material. 2ndEd., The Royal Society of Chemistry, Cambridge.

Plumstead, P., Leytem, A., Maguire, R., Spears, J., Kwanyuen, P. and J. Brake 2008. Interaction of calcium and phytate in broiler diets. 1. Effects on apparent prececal digestibility and retention of phosphorus. Poultry Science, 87: 449-458.

Rahmat, N. , Ahmad Z. A., and A. Mohamed.2010. Recent progress on innovative and potential technologies for glycerol transformation into fuel additives: A critical review. Renewable and Sustainable Energy Reviews. Volume 14, Issue 3, 987-1000.

Roll, A., Vilarrasa, E., Tres, A., and Barroeta, A. (2018). The different molecular structure and glycerol-to-fatty acid ratio of palm oils affect their nutritive value in broiler chicken diets. Animal, 1-9.

SAS. (2012). Statistical Analysis System, User's Guide. Statistical. Version 9.1th ed. SAS. Inst. Inc. Cary. N.C. USA.

Thompson, J.C., and He B.B. 2006 . Characterization of crude glycerol from biodiesel production from multiple feedstocks. Applied Engineering in Agriculture. V.22, :261-265.

Topal, E., and M. Ozdogan .(2013). Effects of glycerol on the growth performance, internal organ weights, and drumstick muscle of broilers. J. Appl. Poult. Res. 22 :146–151.

Van Gerpen, J. 2005. Biodiesel processing and production. J. Fuel Proc. 86:1097-1107.

Waldroup, Park W. (2007) Biofuels and Broilers-competitors or coopeterators, MANC proceeding. 31-40.