Revista Brasileira de Ciência Avícola
ISSN: 1516-635X
revista@facta.org.br
Fundação APINCO de Ciência e Tecnologia Avícolas
Brasil

Freitas, LW; Menten, JFM; Zavarize, KC; Pereira, R; Romano, GG; Lima, MB; Dias, CTS
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Revista Brasileira de Ciência Avícola, vol. 19, enero-marzo, 2017, pp. 91-96
Fundação APINCO de Ciência e Tecnologia Avícolas
Campinas, Brasil

Brazilian Journal of Poultry Science
Revista Brasileira de Ciência Avícola

ISSN 1516-635X Jan - Mar 2017

Special Issue Nutrition / 091-096

http://dx.doi.org/10.1590/1806-9061-2016-0226

Evaluation of Dietary Glycerin Inclusion during Different Broiler Rearing Phases

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■Keywords

Alternative feedstuff, biodiesel, broiler litter; nutrition, pododermatitis.

Submitted: April/2016
Approved: November/2016

ABSTRACT

The objective of this study was to evaluate the effect of the dietary addition of different levels of glycerin on the performance, litter moisture, pododermatitis incidence, and carcass and parts yield of broilers. In total, 1,610 broilers were reared in 35 pens with 46 birds each. A completely randomized experimental design, with five treatments with seven replicates was applied. The experimental treatments were: T1: control diet; T2: dietary inclusion of 5% glycerin from 1-42 days of age; T3: dietary inclusion of 10% glycerin from 1-42 days of age; T4: dietary inclusion of 5% glycerin from 7-42 days of age; T5: dietary inclusion of 10% glycerin from 7-42 days of age. The diets containing glycerin fed since the pre-starter period improved broiler weight gain and feed conversion ratio, but did not influence feed intake or livability. At the end of the experiment, the production efficiency index of the broilers fed 10% glycerin during the entire rearing period was significantly reduced compared with the other treatments. Litter moisture in the pens of broilers fed 10% glycerin during the entire rearing period was higher compared to the other treatments since day 21.Diets containing 10% glycerin, both for the entire rearing period (1-42 days) or only after the pre-starter phase (7-42 days), influenced broiler performance and incidence of severe pododermatitis, reducing the production efficiency indexes at 42 days. Glycerin may be added up to 5% in broiler´s diets with no effect on performance, litter moisture and carcass yield, indicating that this co-product of the biodiesel industry can be used as an alternative feedstuff for broilers.

INTRODUCTION

Corn is the main energy source used in poultry feeds. Approximately 80% of Brazilian corn production is used in the formulation of feeds. Corn accounts for 65-70% of poultry feed composition. It is a valuable grain also used in human foods, and its replacement in feeds by other energy sources may reduce animal feeding costs.

The use of alternative feedstuffs, particularly in poultry production, may reduce production costs and increase the availability of traditional ingredients for human foods. Among alternative feedstuffs, glycerin is an energy-rich and low-cost ingredient. The glycerin obtained from biodiesel production contains 70-80% glycerol. Literature reports a wide range of chemical composition and energy values for glycerin. This variation may be due to the lack of purity of the samples because of the methods used for biodiesel production (Zavarize *et al*., 2014).

The objective of this study was to evaluate the effect of the dietary addition of different levels of glycerin on the performance, litter moisture, pododermatitis incidence, carcass and parts yield of broilers relative to glycerin feeding period.

MATERIAL AND METHODS

In total, 1,610 Cobb 500 broilers were reared in 35 pens with 46 birds each. A completely randomized experimental design, with five treatments with seven replicates was applied. The experimental treatments were T1: control diet; T2: dietary inclusion of 5% glycerin from 1-42 days of age; T3: dietary inclusion of 10% glycerin from 1-42 days of age; T4: dietary inclusion of 5% glycerin from 7-42 days of age; T5: dietary inclusion of 10% glycerin from 7-42 days of age.

The glycerin product added to the diets contained 83% glycerol, 0.09% ether extract, 7% ashes, and 1.8% sodium. Glycerin gross energy value was determined in a bomb calorimeter as 3,620 kcal/kg, and the metabolizable energy value (3,258 kcal AMEn/kg) used for feed formulation was calculated as 90% of the GE value.

The rearing period was divided in four phases: pre-starter (1-7 days), starter (7-21 days), grower (21-35 days), and finisher (35-42 days), as the corresponding feeds were formulated to supply the broilers' nutritional requirements proposed by Rostagno *et al*. (2011), as shown in Tables 1 and 2.

Birds and feed residues were weekly weighed to calculate the live performance parameters (feed intake, weight gain, feed conversion ratio, live weight, livability, and production efficiency index). Production efficiency index was calculated according to the equation: PEI = [(daily weight gain x livability)/ feed conversion ratio] x 100.

On day 7, one bird per replicate, with the average body weight of the replicate, was selected, identified, sacrificed by neck dislocation, and immediately frozen at -18°C. Carcass samples were freeze-dried for 170 days for subsequent determination of their dry matter content.

Litter moisture percentage was determined on days 21, 28, 35, and 42. Litter samples were collected from three different locations in each pen, distant from the feeder and drinker Litter samples were weighed and

Table 1 – Ingredients and calculated nutritional composition of the pre-starter (1-7 days) and starter (8-21 days) diets.

Ingredients	Pre-starter			Starter		
	Glycerin inclusion (%)					
	0.0	5.0	10.0	0.0	5.0	10.0
Ground corn	48.91	42.97	37.04	53.78	47.85	41.91
Soybean meal	43.58	44.58	45.57	38.55	39.55	40.54
Corn oil	3.43	3.61	3.79	3.82	4.00	4.18
Glycerin	0.00	5.00	10.00	0.00	5.00	10.00
Dicalcium phosphate	1.86	1.87	1.88	1.53	1.54	1.55
Limestone	0.91	0.91	0.90	0.94	0.94	0.93
Salt	0.51	0.28	0.05	0.48	0.25	0.03
DL-methionine	0.33	0.33	0.34	0.39	0.40	0.40
L-lysine HCl	0.14	0.13	0.11	0.16	0.14	0.12
L-threonine	0.05	0.05	0.05	0.04	0.04	0.04
Vitamin supplement[1]	0.10	0.10	0.10	0.10	0.10	0.10
Mineral supplement[2]	0.05	0.05	0.05	0.05	0.05	0.05
Choline chloride 60%	0.08	0.08	0.08	0.08	0.08	0.08
Anticoccidial agent[3]	0.05	0.05	0.05	0.05	0.05	0.05
Growth promoter[4]	0.005	0.005	0.005	0.005	0.005	0.005
Calculated values						
ME, kcal/kg	2.960	2.960	2.960	3.050	3.050	3.050
CP, %	23.92	23.89	23.86	22.07	22.05	22.02
Ca, %	0.92	0.92	0.92	0.84	0.84	0.84
Available P, %	0.47	0.47	0.47	0.40	0.40	0.40
Na, %	0.22	0.22	0.22	0.21	0.21	0.21
Digestible methionine, %	0.63	0.63	0.64	0.67	0.67	0.68
Digestible Met + Cys, %	0.95	0.95	0.95	0.97	0.97	0.98
Digestible lysine, %	1.32	1.32	1.32	1.22	1.22	1.22
Digestible threonine, %	0.86	0.86	0.86	0.79	0.79	0.79
Electrolyte balance, mEq/kg	236	234	232	216	214	212

[1]Vitamin supplement (per kg feed): Vit. A: 9,000 IU; vit. D3: 2,500 IU;vit. E, 20 IU; vit. K_3, 2,5 mg; thiamine, 1.5mg; riboflavin, 6 mg; pyridoxine, 3 mg; cyanocobalamin 12 µg; nicotinic acid, 25 mg; pantothenic acid, 12 mg; biotin, 0.06 mg; folic acid, 0. 8 mg; selenium, 0.25 mg.[2]Mineral supplement (per kg feed):I, 1 mg; Fe, 50 mg; Cu, 10 mg; Co, 1 mg; Mn, 75 mg; Zn, 50 mg.[3]Coxistac:12% salinomycin.[4]Halquinol BP 80: 60% chlorohydroxyquinoline.

Table 2 – Ingredients and calculated nutritional composition of the grower (22-35 days) and finisher (36-42 days) diets.

Ingredients	Grower			Finisher		
	Glycerin inclusion (%)					
	0.0	5.0	10.0	0.0	5.0	10.0
Ground corn	56.53	50.60	44.66	60.46	54.53	48.57
Soybean meal	35.30	36.30	37.29	31.85	32.85	33.84
Corn oil	4.82	5.00	5.17	4.77	4.95	5.13
Glycerin	0.00	5.00	10.00	0.00	5.00	10.00
Dicalcium phosphate	1.31	1.32	1.33	1.10	1.10	1.11
Limestone	0.89	0.88	0.88	0.80	0.80	0.78
Salt	0.46	0.23	0.00	0.45	0.22	0.00
DL-methionine	0.27	0.27	0.28	0.24	0.25	0.26
L-lysine HCl	0.15	0.13	0.11	0.16	0.15	0.13
L-threonine	0.03	0.03	0.03	0.03	0.03	0.03
Vitamin supplement[1]	0.08	0.08	0.08	0.06	0.06	0.06
Mineral supplement[2]	0.05	0.05	0.05	0.05	0.05	0.05
Choline chloride 60%	0.06	0.06	0.06	0.04	0.04	0.04
Anticoccidial agent[3]	0.05	0.05	0.05			
Growth promoter[4]	0.005	0.005	0.005			
Calculated values						
ME, kcal/kg						
CP, %	3.150	3.150	3.150	3.200	3.200	3.200
Ca, %	20.74	20.71	20.68	19.49	19.46	19.42
Available P, %	0.76	0.76	0.76	0.66	0.66	0.66
Na, %	0.35	0.35	0.35	0.31	0.31	0.31
Digestible methionine, %	0.20	0.20	0.20	0.19	0.19	0.20
Digestible Met + Cys, %	0.54	0.54	0.54	0.51	0.51	0.51
Digestible lysine, %	0.83	0.83	0.83	0.77	0.77	0.77
Digestible threonine, %	1.13	1.13	1.13	1.06	1.06	1.06
Electrolyte balance, mEq/kg	0.74	0.74	0.74	0.69	0.69	0.69
	202	200	198	189	187	185

[1]Vitamin supplement (per kg feed):vit. A, 9,000 IU; vit. D_3, 2,500 IU; vit. E, 20 IU; vit. K_3, 2.5 mg; thiamine, 1. 5 mg; riboflavin, 6 mg; pyridoxine, 3 mg; cyanocobalamin, 12 µg; nicotinic acid, 25 mg; pantothenic acid, 12mg; biotin, 0.0.06 mg; folic acid, 0.8 mg; selenium, 0.25 mg.[2]Mineral supplement (per kg feed):I, 1 mg; Fe, 50 mg; Cu, 10 mg; Co, 1 mg; Mn, 75 mg; Zn, 50 mg.[3]Coxistac:12% salinomycin.[4]Halquinol BP 80: 60% chlorohydroxyquinoline.

dried in a forced-ventilation oven (55°C for 72 hours), and weighed again.

On day 42, four birds per replicate were scored for pododermatitis according to a 0-1 scale as: 0 (no lesions), 1 (lesion covering up to 50% of the footpad surface), and 2 (lesion covering more than 50% of the footpad surface).

On day 48, all birds were fasted for eight hours, weighed, and sacrificed to determine carcass, parts, and abdominal fat yields. Birds were sacrificed by electrical stunning and bled. After evisceration and removal of the abdominal fat, carcasses were not placed in the chiller. Carcasses with no feet, head, neck, and offal were weighed to calculate their yield as a percentage of live weight. Carcasses were then cut up according to industrial processing methods into breast, legs (thighs and drumsticks), back, and wings; and their yields were calculated as a percentage of carcass weight. Feet, head + neck, and abdominal fat (removed from the abdominal cavity and from the gizzard) yields were calculated as a percentage of live weight.

The results obtained for performance parameters, carcass dry matter content, and carcass traits were submitted to analysis of variance using the PROC GLM of SAS statistical package version 9.2. When significant effects ($p<0.05$) were detected, means were compared by the test of Tukey at 5% significance level.

Pododermatitis scores were analyzed by the PROC FREQ of SAS statistical package version 9.2, and treatment means were compared by the test of Kruskal-Wallis ($p<0.05$). When significant effects were determined, means were submitted to the Chi-square test (χ^2).

RESULTS AND DISCUSSION

Table 3 presents the performance results obtained. The statistical analyses showed that the diet with the inclusion of 5% glycerin promoted significantly better

Table 3 – Performance parameters measured in the periods of 1-7, 1-21, and 1-42 days of the rearing period of broilers fed with glycerin.

Parameters	Treatments[1]					p value	CV[8] (%)
	T1	T2	T3	T4	T5		
1-7 days							
BW[2], g	204[b]	212[a]	209[ab]	-	-	0.0014	2.03
WG[3], g	158[b]	166[a]	163[a]	-	-	0.0007	2.49
FI[4], g	167	170	169	-	-	0.5456	2.69
FCR[5]	1.059[b]	1.022[a]	1.035[ab]	-	-	0.0059	2.25
L[6], %	99.4	99.5	98.9	-	-	0.3752	1.18
Carcass DM, %	39.84	37.12	36.58	-	-	0.3058	10.87
1- 21 days							
BW[2], g	1116	1142	1106	1116	1126	0.0996	2.2
WG[3], g	1069	1096	1059	1069	1080	0.0935	2.28
FI[4], g	1484[ab]	1481[ab]	1513[a]	1448[b]	1438[b]	0.0034	2.42
FCR[5]	1.388[ab]	1.352[a]	1.428[b]	1.345[a]	1.341[a]	<0.0001	2.44
L[6], %	96.9[ab]	97.8[a]	91.6[b]	96.9[ab]	96.3[ab]	0.0184	3.63
1-42days							
BW[2], g	3218	3280	3211	3278	3277	0.4892	2.88
WG[3], g	3172	3234	3165	3232	3231	0.4857	2.92
FI[4], g	5177	5279	5272	5274	5299	0.6006	2.68
FCR[5]	1.634	1.632	1.666	1.632	1.64	0.349	2.02
L[6], %	89.9	87.6	79.7	90.10	86.0	0.058	7.52
PEI[7]	417[a]	413[a]	363[b]	425[a]	403[ab]	0.0111	7.45

[1]Treatments: T1 – control diet (no glycerin inclusion); T2 – diet with 5% glycerin inclusion from 1-42 days; T3 -diet with 10% glycerin inclusion from 1-42 days; T4 - diet with 5% glycerin inclusion from 7-42 days; and T5 - diet with 10% glycerin inclusion from 7-42 days. [2]BW:body weight on day 42. [3]WG: weight gain.[4]FI:feed intake.[5]FCR:feed conversion ratio.[6]L:Livability.[7]PEI:production efficiency index.[8]CV:coefficient of variation.[9]Carcass dry matter:Carcass dry matter after freeze-drying.[a,b] Means in the same row followed by different superscripts are different by the test of Tukey (p<0.05).

performance (p<0.05) until broilers were 21 days old, in agreement with the findings of Bernardino *et al*. (2014), Zavarize*et al*.(2012), Silva *et al*. (2012), and Dourado *et al*. (2010). In addition, consistent with studies evaluating the addition of increasing glycerin levels (up to 10%) in broiler diets (Zavarize *et al*., 2012; Menten *et al*., 2008; Fernandes *et al*., 2010), no negative effect of the inclusion of up to 8% of glycerin was detected on broiler performance, suggesting that glycerin can be included in broiler diets, provided its chemical composition. Mandalawi *et al.* 2014 showed that raw glycerin from the biodiesel industry can be used efficiently, up to 10% of the diet, as a source of energy for broilers from 1 to 21 days of age and that the energy content of well-processed raw glycerin depends primarily on its glycerol content. However, when high glycerin levels were added to the diet (20 and 25%), Simon *et al*. (1997) performance impairment was observed. Guerra *et al*. (2011) obtained worse feed conversion ratios due to high feed intake when broilers were fed a diet with 10% glycerin relative to lower levels of glycerin inclusion. On the other hand, Cerrate *et al*. (2006) obtained lower feed intake in broilers fed diets of 10% of glycerin compared with 5%, and attributed this result to the reduced flow of the feed in the feeders, which consequently limited feed intake.

Whole carcass dry matter content of the broilers sacrificed with seven days of age are shown in Table 3.There was no effect of dietary glycerin inclusion (p>0.05) on whole carcass dry matter content of broilers sacrificed with seven days of age. Previous studies indicated that the inclusion of high glycerin levels in starter broiler diets increased weight gain during the first days of rearing; however, when fed during the entire rearing period, performance losses were observed (Simon *et al*., 1997; Cerrate *et al*, 2006; Silva *et al*., 2012). This result suggests that the use of glycerin does not promote water retention in the carcass, but true weight gain.

Carcass trait results are shown in Table 4. The was no effect of the treatments (p>0.05) on carcass and parts yield. These results are consistent with the findings of Guerra *et al*. (2011), Silva *et al*. (2012), and Gianfelici (2009), using a maximum level of 10% of glycerin in broiler diets. This indicates that glycerin can be added up to this level in the diet with no adverse effects on carcass yield or commercial parts yields. However, Cerrate *et al*. (2006) observed that broilers fed diets containing 2.5 and 5% of glycerin, presented

Table 4 – Carcass and parts yields of 49-d-old broilers fed with different glycerin levels.

Treatments[2]	BW[2], g	CY[3], %	BrY[4], %	WY[5], %	LY[6], %	BY[7], %	AF[8], %
T1	3872	72.34	42.39	10.23	32.66	14.12	1.32
T2	3931	72.11	42.95	10.00	32.35	14.40	1.48
T3	3889	72.15	42.78	10.16	32.27	14.48	1.43
T4	4150	72.65	43.75	9.80	31.92	14.17	1.29
T5	4001	72.72	43.62	10.21	31.65	14.35	1.40
p value		0.4091	0.1875	0.1798	0.2873	0.5307	0.6896
CV[9] (%)		1.76	4.87	6.49	4.95	5.49	33.85

[1]Treatments: T1 – control diet (no glycerin inclusion); T2 – diet with 5% glycerin inclusion from 1-42 days; T3 -diet with 10% glycerin inclusion from 1-42 days; T4 - diet with 5% glycerin inclusion from 7-42 days; and T5 - diet with 10% glycerin inclusion from 7-42 days. [2]BW:body weight on day 48; [3]CY:carcass yield; [4]BrY:breast yield; [5]WY:wing yield; [6]LY:leg yield; [7]BY:back yield; [8]AF:abdominal fat; [9]CV:coefficient of variation.

higher carcass and breast yields compared with those fed a standard diet.

Litter moisture results are shown in Table 5. The litter of the broilers fed the diet with 10% of glycerin during the entire rearing period (1-42 days) presented higher moisture levels ($p<0.05$) after day 21, compared with the other treatments. Freitas *et al*. (2011) and Guerra *et al*. (2011), evaluating the inclusion of increasing glycerin level in broiler diets, also observed higher litter moisture after day 21, when the diets contained more than 10% of glycerin. Bernadino *et al.* (2014) found increased litter moisture when broilers were fed 7% of glycerin in the diet according to Robinson & Newsholme (1969), the digestion of glycerol, present in glycerin, is limited by the saturation of the enzyme glycerol kinase. In addition, glycerol is a highly hygroscopic molecule,

Table 5 – Litter moisture levels on days 21, 28, 35, and 42, according to experimental treatment.

Treatments[1]	21 days	28 days	35 days	42 days
T1	29.62^{b}	32.73^{b}	34.66^{bc}	40.90^{ab}
T2	34.17^{ab}	35.30^{b}	40.63^{ab}	46.26^{a}
T3	37.50^{a}	44.14^{a}	47.83^{a}	47.70^{a}
T4	29.43^{b}	30.90^{b}	30.29^{c}	40.82^{b}
T5	30.56^{b}	32.50^{b}	35.20^{bc}	37.10^{b}
Valor de P	<0.0001	<0.0001	<0.0001	0.0032
CV[2] (%)	9.69	11.15	13.19	12.06

[1]Treatments: T1 – control diet (no glycerin inclusion); T2 – diet with 5% glycerin inclusion from 1-42 days; T3 -diet with 10% glycerin inclusion from 1-42 days; T4 - diet with 5% glycerin inclusion from 7-42 days; and T5 - diet with 10% glycerin inclusion from 7-42 days.[2]CV: coefficient of variation.[a,b,c] Means in the same column followed by different superscripts are different by the test of Tukey ($p<0.05$).

carrying water during its excretion. Gianfelici (2009) and Romano *et al*. (2014) observed that levels higher than 7.5% of glycerin in broiler diets increased water consumption and excretion, which may have adverse effects on the field due to higher litter moisture. Glycerin is a low molecular weight hydrophilic compound, which is easily excreted by the kidneys. Silva *et al*. (2012) described a positive linear effect of dietary glycerin levels on litter moisture evaluated when broilers were 43 days old, and observed higher excreta moisture after the third week of rearing in broilers fed 10% of glycerin.

The incidence of pododermatitis observed in the present study is shown in Table 6. The incidence of

Table 6 – Incidence and severity (score) of pododermatitis in the right footpad (RF) and left footpad (LF) of broilers fed with different glycerin levels at 42 days.

		Treatments[1]				
	Score	T1	T2	T3	T4	T5
RF (%)	0	51.43 ^{a}	5.72 ^{b}	2.86 ^{b}	20.00 ^{ab}	31.42 ^{ab}
	1	31.43	28.57	17.14	34.29	14.29
	2	17.14 ^{b}	65.71 ^{a}	80.00 ^{a}	45.71 ^{ab}	54.29 ^{ab}
LF (%)	0	51.43 ^{a}	5.72 ^{ab}	0.00 ^{b}	17.14 ^{ab}	28.57 ^{ab}
	1	31.43	25.71	14.29	28.57	25.72
	2	17.14 ^{b}	68.57 ^{a}	85.71 ^{a}	54.29 ^{ab}	45.71 ^{ab}

[1]Treatments: T1 – control diet (no glycerin inclusion); T2 – diet with 5% glycerin inclusion from 1-42 days; T3 -diet with 10% glycerin inclusion from 1-42 days; T4 - diet with 5% glycerin inclusion from 7-42 days; and T5 - diet with 10% glycerin inclusion from 7-42 days. [a,b] Means in the same row followed by different superscripts are different by the test of Tukey ($p<0.05$).

pododermatitis was significantly higher ($p<0.05$) when birds were fed glycerin during the entire experimental period (1-42 days), and all birds fed 10% of glycerin were affected.

The incidence of severe pododermatitis (score 2) was only 17% in the broilers not fed glycerin, whereas more than 45% of those fed diets containing glycerin during the entire rearing period (1-42 days) presented score 2. This result may be attributed to the high litter moisture in the pens of the latter, which was higher than 25%, which is the highest recommended values (UBA, 2008).

Litter moisture may greatly influence the incidence and the severity of pododermatitis (Traldi *et al.*, 2007). Bernardi (2011) indicated that high excreta output increase litter moisture and nitrogen content, also increasing the incidence and severity of footpad lesions.

CONCLUSIONS

Diets containing 10% of glycerin, both for the entire rearing period (1-42 days) or only after the pre-starter phase (7-42 days), influence broiler performance and incidence of severe pododermatitis, reducing the production efficiency index at 42 days.

Glycerin may be added up to 5% in broiler´s diets with no effect on performance; litter moisture and carcass yield, indicating that this co-product of the biodiesel industry can be used as an alternative feedstuff for broilers.

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