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Purified glycerin in balanced diets of broiler chickens treated from 1 to 42 days of age

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ABSTRACT - We evaluated the technical feasibility of purified glycerin inclusion in balanced diets of broiler chickens treated from 1 to 42 days of age. A total of 240 broiler chickens were distributed in a completely randomized design into four treatments (0, 2, 4, and 6% purified glycerin inclusion), with six replicates of 10 broilers each. We evaluated productive performance (at 7, 21, and 42 days), edible viscera (heart, liver, and gizzard), carcass yield and cuts, color, chemical and physical composition, as well as protein and fat deposition in the breast muscle. Dietary inclusion of purified glycerin reduced feed conversion and increased feed intake, weight gain, and weight at seven days. From 1 to 21 days, there was a decrease in feed conversion and a linear increase in weight gain, with no effect on feed intake. Considering the total experimental period, increasing glycerin levels increased weight gain and weight at 42 days, with no effects on feed conversion or feed intake. Similarly, there was no effect on carcass and cut yields, liver and gizzard yields, and weight or length of the intestine, while heart yield was decreased. There was a linear increase in crude protein, ether extract, protein, and fat deposition and a reduction in moisture percentages, with no effect on ash content. Purified glycerin levels did not affect a*, b*, L*, pH, temperature, shear force, or weight loss by cooking in breast meat. The dietary inclusion of purified glycerin of up to 6% proved to be technically feasible in the diets of broilers from 1 to 42 days.

Keywords: biodiesel, energetic feedstuff, growth curve, productive performance

Introduction

Biodiesel production from animal or vegetable sources, through the reaction of saponification of fatty acids with sodium hydroxide or potassium hydroxide, generates crude glycerin as a co-product, which is of potential use in poultry diets, mainly due to its high energy efficiency (Dozier et al., 2008, 2011; Lammers et al., 2008).

Studies using glycerin in broiler diets have shown that its inclusion does not affect the main performance characteristics at different production stages (Cerrate et al., 2006; Sehu et al., 2013; Silva et al., 2012). In studies conducted to evaluate the effects of increasing levels of glycerin (0, 5, and 10%) in diets of broilers from 1 to 42 days of age, Sehu et al. (2012, 2013) observed that the inclusion of up to 5% glycerin did not affect performance traits, carcass yield, or body weight. Similarly, Romano et al.

(2014), using diets containing 2.5, 5.0, 7.5, and 10% glycerin, concluded that broilers can adequately metabolize up to 7.5% of dietary glycerol. However, the authors suggested that undesirable metabolic alterations, such as increased blood glucose concentrations, water intake, and fecal moisture, might occur at higher levels (Gianfelici et al., 2011; Romano et al., 2014).

There is conflicting evidence in the literature about the maximum glycerin inclusion level in poultry diets, mainly due to the lack of standardization during biodiesel production. As a result, glycerin can influence the composition and affect the final quality of the product (Gianfelici et al., 2011; Romano et al., 2014). Another reason reported by Bernardino et al. (2014) is that a high intake of glycerol may exceed the metabolizing capacity of broiler. Therefore, if the ingested glycerol is not phosphorylated by the enzyme glycerol kinase, it cannot be retained and used in the body, resulting in the excretion of the surplus through the kidneys.

So far, studies have focused on evaluating the effects of glycerin on the performance, and, consequently, our knowledge on the deposition of fat and body protein is rather scarce. Considering the above, the objective of this study was to evaluate the technical feasibility of purified glycerin inclusion in balanced diets of broiler chickens from 1 to 42 days of age.

Material and Methods

The trial was conducted from September 29 to November 10, 2015, in Araguaína, TO, Brazil (07°11'28"S and 48°12'26" W), according to the guidelines of the Ethics in Animal Use Committee, under case number 23101.000830/2014-16.

A total of 240 one-day-old male broiler chickens of the Cobb 500® lineage, with an average initial weight of 48±2.46 g, were used. Birds were assigned to four treatment groups using a completely randomized design (0, 2, 4, and 6% purified glycerin inclusion), with six replicates of ten birds per experimental unit. Broilers were housed in an experimental shed with 24 boxes of 2 m^2, supplied with tubular feeders and pendular drinkers. Feeders were replenished and drinkers were cleaned twice a day to ensure free access to water and feed throughout the experimental period.

Until the chicks were 14 days old, artificial heat was provided, using incandescent lamps (60 W) located inside the boxes. The environmental conditions inside the facility during the experimental period were monitored and recorded daily every 5 min, using HOBO Data Loggers OnSet® ware version 3.4.1. The devices were placed halfway up the boxes to measure temperature and relative humidity. Average air as well as maximum and minimum temperatures inside the facility during the experimental period were 29.2, 36.7, and 23.5°C, respectively, with a relative humidity of 64%.

The experimental diets were calculated considering the purified glycerin chemical composition (Table 1) and the nutritional requirements for medium-performance male broilers, following the

Table 1 - Composition of purified glycerin used in the formulation of experimental diets

Nutrient and energy	Purified glycerin[1]
Crude protein (g kg^{-1})[2]	2.3
Metabolizable energy (kcal kg^{-1})[3]	3560
Dry matter (g)[4]	899.80
Ether extract (g kg^{-1})[4]	11.9
Mineral matter (g kg^{-1})[4]	78.6
Methanol (g kg^{-1})[4]	<0.1
Glycerol (g kg^{-1})[4]	804.0
NaCl (g kg^{-1})[4]	74.7
Na (g kg^{-1})[4]	29.6

[1] Glycerin from the processing of soybeans sold in southern Brazil.
[2] Rostagno et al. (2011).
[3] Analysis conducted at the Animal Nutrition Laboratory from the School of Veterinary Medicine and Animal Science, Federal University of Tocantins.
[4] Approximate values supplied by manufacturer.

recommendations of Rostagno et al. (2011) during two stages: from 1 to 21 (Table 2) and from 22 to 42 days of age (Table 3).

The evaluated parameters were feed intake (FI), weight gain (WG), feed conversion (FC), final weight (FW; at 7, 21, and 42 days), edible viscera (heart, liver, gizzard), weight and/or length of the small intestine, carcass yield (CY), cut yield (thigh, drumstick, and breast), color of the breast muscle (L* = lightness, a* = redness, b* = yellowness), pH, crude protein, ether extract, ash content, moisture, protein deposition, and fat deposition.

Feed intake was calculated considering the amount of feed supplied and the leftovers in the feeders, using the ratio of FI and WG during the experimental period.

At 42 days of age, two broilers per repetition, with body weights ± 5% of the average, were fasted for 12 h and then slaughtered by cervical dislocation. Subsequently, they were subjected to bleeding, scalding, plucking, and evisceration. The relative weights of the whole carcasses (with legs, neck, and head) and special cuts (thigh, drumstick, and breast) were determined.

The edible viscera (gizzard, heart, and liver) and small intestines were collected during evisceration, cleaned, dried on paper towels, and weighed separately on a precision scale. From the gizzard, all the adhered fat, its contents, and the koilin membrane were removed. In addition to weight, the length of

Table 2 - Composition of experimental diets containing increasing levels of purified glycerin for 1 to 21-day-old broilers

Item	Level of purified glycerin (%)			
	0	2	4	6
Ingredient (g kg^{-1})				
Corn	570.52	548.56	526.60	504.64
Soybean meal (45%)	364.88	368.72	372.59	376.45
Purified glycerin	0.00	20.00	40.00	60.00
Dicalcium phosphate	17.08	17.11	17.13	17.15
Soybean oil	21.75	21.37	21.02	20.66
Limestone	9.190	9.170	9.140	9.120
Salt	4.930	3.470	1.980	0.500
DL-methionine	3.230	3.260	3.280	3.290
L-lysine HCl	2.530	2.450	2.380	2.310
L-threonine	0.890	0.890	0.880	0.880
Mineral and vitamin supplement[1]	5.00	5.00	5.00	5.00
Total	1000.00	1000.00	1000.00	1000.00
Calculated nutritional composition				
Metabolizable energy (kcal kg^{-1})	2975	2975	2975	2975
Crude protein (g kg^{-1})	215.0	215.0	215.0	215.0
Calcium (g kg^{-1})	8.69	8.69	8.69	8.69
Available phosphorus (g kg^{-1})	4.30	4.30	4.30	4.30
Digestible lysine (g kg^{-1})	12.42	12.42	12.42	12.42
Digestible methionine + cysteine (g kg^{-1})	8.95	8.95	8.95	8.95
Digestible methionine (g kg^{-1})	6.03	6.04	6.05	6.05
Digestible threonine (g kg^{-1})	8.07	8.07	8.07	8.07
Sodium (g kg^{-1})	21.5	21.5	21.5	21.5
Potassium (g kg^{-1})	8.34	8.34	8.35	8.36
Chlorine (g kg^{-1})	3.47	2.56	1.68	0.79
Electrolyte balance (mEq kg^{-1})[2]	208.9	234.6	259.7	285.0

[1] Composition per ton: folic acid, 150.00 mg; cobalt, 178.00 mg; copper, 2,675.00 mg; choline, 120.00 g; colistin, 2,000.00 mg; iron, 11.00 g; iodine, 535.00 mg; manganese, 31.00 g; mineral matter, 350.00 g; niacin, 7,200.00 mg; nicarbazin, 24.00 g; calcium pantothenate, 2,400.00 mg; selenium, 60.00 mg; vitamin A, 1,920,000.00 IU; vitamin B1, 300.00 mg; vitamin B12, 3,600.00 mg; vitamin B2, 1,200.00 mg; vitamin B6, 450.00 mg; vitamin D3, 360,000.00 IU; vitamin E, 3,600.00 IU; vitamin H, 18.00 mg; vitamin K, 480.00 mg; zinc, 22.00 g.
[2] Calculated according to Mongin (1981): electrolyte balance = (mg kg^{-1} of dietary Na^+/22.990) + (mg kg^{-1} of dietary K^+/39.102) – (mg kg^{-1} of dietary Cl^-/35.453).

the small intestine, from the beginning of the duodenum to the ileocecal junction, was measured. The relative weight of the plucked and eviscerated carcass was calculated in relation to the fasting weight. The relative weights of cuts, edible viscera, and small intestines were obtained in relation to that of the plucked and eviscerated carcass.

In the raw breast meat (without bones, skin, ligaments and fat), pH and meat color were evaluated by the CIELAB system (L* = lightness, a* = redness, b* = yellowness) with a colorimeter (Chroma meter®). Readings were performed at three different points of the musculature, and pH determination was performed by means of a penetration electrode inserted directly into the meat.

The breasts were cut in half and frozen in plastic bags. One of the separated halves was ground in an industrial meat grinder. The ground cuts were weighed, homogenized, and pre-dried in an oven at 55 °C for 72 h. Subsequently, they were ground in a knife-type mill and transferred to the laboratory for analysis (crude protein, ether extract, moisture, and ash), according to Silva and Queiroz (2002).

Protein and fat deposition rates in breast meat (g day^{-1}) were calculated based on the slaughter of an additional group of six one-day-old chicks, compared to birds slaughtered at 42 days of age, according to the formulas described by Scherer et al. (2011).

Table 3 - Composition of experimental diets containing increasing levels of purified glycerin for 22 to 42-day-old broilers

Item	Level of purified glycerin (%)			
	0	2	4	6
Ingredient (g kg^{-1})				
Corn	640.39	618.43	596.47	574.51
Soybean meal (45%)	294.34	298.21	302.06	305.92
Purified glycerin	0.00	20.00	40.00	60.00
Dicalcium phosphate	11.72	11.74	11.77	11.79
Soybean oil	30.78	30.40	30.05	29.68
Limestone	8.17	8.15	8.12	8.10
Salt	4.50	3.02	1.54	0.06
DL-methionine	2.48	2.50	2.53	2.55
L-lysine HCl	2.14	2.07	1.99	1.92
L-threonine	0.48	0.48	0.47	0.47
Mineral and vitamin supplement[1]	5.00	5.00	5.00	5.00
Total	1000.00	1000.00	1000.00	1000.00
Calculated nutritional composition				
Metabolizable energy (kcal kg^{-1})	3125	3125	3125	3125
Crude protein (g kg^{-1})	187.5	187.5	187.5	187.5
Calcium (g kg^{-1})	6.85	6.85	6.85	6.85
Available phosphorus (g kg^{-1})	3.20	3.20	3.20	3.20
Digestible lysine (g kg^{-1})	10.44	10.04	10.44	10.44
Digestible methionine + cysteine (g kg^{-1})	7.62	7.62	7.62	7.62
Digestible methionine (g kg^{-1})	5.01	5.02	5.03	5.04
Digestible threonine (g kg^{-1})	6.78	6.78	6.78	6.78
Sodium (g kg^{-1})	1.97	1.97	1.97	1.97
Potassium (g kg^{-1})	7.25	7.25	7.26	7.27
Chlorine (g kg^{-1})	3.22	2.32	1.43	0.53
Electrolyte balance (mEq kg^{-1})[2]	180.3	205.7	231.0	256.7

[1] Composition per ton: folic acid, 120.00 mg; cobalt, 179.00 mg; copper, 2,688.00 mg; choline, 108.00 g; iron, 11.00 g; iodine, 537.00 mg; lincomycin 800.00 mg; manganese, 31.00 g; mineral matter, 350.00 g; niacin, 6,000.00 mg; calcium pantothenate, 1,920.00 mg; salinomycin, 12.00 g; selenium, 54.00 mg; moisture 80.00 g; vitamin A, 1,500,000.00 IU; vitamin B1, 300.00 mg; vitamin B12, 2,800.00 mg; vitamin B2, 960.00 mg; vitamin B6, 450.00 mg; vitamin D3, 300,000.00 IU; vitamin E, 3,000.00 IU; vitamin H, 20.00 mg; vitamin K, 480.00 mg; zinc, 22.00 g.
[2] Calculated according to Mongin (1981): electrolyte balance = (mg kg^{-1} of dietary Na^+/22.990) + (mg kg^{-1} of dietary K^+/39.102) – (mg kg^{-1} of dietary Cl^-/35.453).

To determine weight loss by cooking, breast fillets were weighed and baked in an electric oven at 170 °C until reaching an internal temperature of 40 °C. Subsequently, they were turned once to reach an internal temperature of 70 °C. The samples were placed on absorbent paper for cooling to a temperature of 20 to 25 °C and then weighed again to determine weight loss by cooking. Finally, the samples were kept refrigerated at 4 °C for 24 h, according to the methodology adapted from Froning and Uijttenboogarte (1988).

To determine shear force, cylindrical samples (1.27 cm in diameter) were taken and placed with the fibers oriented perpendicular to the direction of plunger travel, using a Warner-Bratzler instrument.

The data were subjected to normality (Cramer Von Mises) and homoscedasticity (Levene) tests. As the data met these assumptions, the variables were subjected to analysis of variance according to the following statistical model:

$$Y_{ij} = \mu + NS_i + e_{ij}; \text{ with } i = 1, 2, 3, 4; j = 1, 2, 3, 4, 5,$$

in which Y_{ij} = observed value for the variable of interest of the *j*-th repetition receiving the *i*-th inclusion level of purified glycerin, μ = effect of general average, NS_i = effect of the *i*-th inclusion level of purified glycerin, and e_{ij} = experimental error.

The variables were subjected to regression analysis using polynomial models of the first or second order, considering the inclusion level of purified glycerin as an independent variable. To check the adjustment of the equation, we considered the following: the significance of the F test for models, the significance of the t test for parameters (β0, β1, and β2) of the models, and the coefficient of determination (R^2 = SS regression/SS total), considering the significance threshold of 5%. Statistical analyses were performed with the statistical software package SISVAR.

Results

Increasing levels of purified glycerin in diets linearly reduced (P<0.002) FC and linearly increased (P<0.001) FI, WG, and weight at seven days (W7d) (Table 4).

From the age of 1 to 21 days, the inclusion of purified glycerin in diets reduced FC (P<0.001) and linearly increased WG; weight at 21 days (W21d) was also increased (P<0.001). We observed no effect on FI (P>0.05) (Table 5).

Throughout the experimental period, there was a positive linear effect on WG and weight at 42 days (W42d) (P<0.034) with an increasing inclusion level of purified glycerin, with no effect on FC and FI (P>0.05) (Table 6).

Increasing levels of purified glycerin in diets did not affect the relative weights of carcass, thigh, drumstick, breast, gizzard, and liver or the relative weight and/or length of the small intestine (Table 7) (P>0.05). However, an effect was observed for relative heart weight, which linearly decreased with a greater inclusion of purified glycerin (P<0.05).

Table 4 - Average feed intake (FI), weight gain (WG), feed conversion (FC), and weight at seven days (W7d) of 1 to 7-day-old broilers according to the inclusion level of purified glycerin

Variable	Purified glycerin inclusion level (%)				Mean	P		CV (%)
	0	2	4	6		L	Q	
FI (g)	144.82	145.50	154.17	155.00	149.88	0.001	0.585	2.21
WG (g)	133.42	133.47	146.57	147.25	140.11	0.001	0.728	2.27
FC (g g^{-1})	1.09	1.08	1.06	1.05	1.07	0.002	0.379	2.12
W7d (g)	180.32	179.67	193.33	193.82	186.78	0.001	0.647	1.65

CV - coefficient of variation; L - linear effect; Q - quadratic effect; P - probability of type I error at 5% using F test.
Equations: WG (g) = 131.87 + 2.756 IL (P = 0.001; r^2 = 0.81); FI (g) = 144.25 + 1.875 IL (P = 0.001; r^2 = 0.79); FC (g g^{-1}) = 1.09 – 0.0075 IL (P = 0.002; r^2 = 0.79); W7d (g) = 178.67 + 2.708 IL (P = 0.001; r^2 = 0.79); in which IL = purified glycerin inclusion level (%).

The dietary inclusion levels of purified glycerin linearly affected the contents of crude protein, ether extract, moisture, protein deposition, and fat deposition (P<0.05), with no effect on the percentage of ash in the breast meat of broilers slaughtered at 42 days of age (Table 8).

The dietary inclusion of purified glycerin did not affect redness (a*), yellowness (b*), luminosity (L*), pH, temperature, shear force, or weight loss by cooking in the breast meat of broilers at 42 days of age (P>0.05) (Table 9).

Table 5 - Average feed intake (FI), weight gain (WG), feed conversion (FC), and weight at 21 days (W21d) of 1 to 21-day-old broilers according to the inclusion level of purified glycerin

Variable	Purified glycerin inclusion level (%)				Mean	P		CV (%)
	0	2	4	6		L	Q	
FI (g)[1]	1254.09	1249.64	1266.93	1291.07	1265.60	0.151	0.477	2.79
WG (g)	921.13	925.52	956.55	962.89	941.51	0.001	0.905	2.11
FC (g g^{-1})	1.35	1.34	1.33	1.33	1.35	0.001	0.553	3.01
W21d (g)	968.05	972.01	1003.29	1009.48	988.20	0.001	0.892	1.99

CV - coefficient of variation; L - linear effect; Q - quadratic effect; P - probability of type I error at 5% using F test.
[1] $\hat{Y}$ = NS.
Equations: WG (g) = 918.06 + 7.816 IL (P = 0.001; r^2 = 0.90); FC (g g^{-1}) = 1.39 – 0.017 IL (P = 0.001; r^2 = 0.82); W21d (g) = 964.87 + 7.779 IL (P = 0.001; r^2 = 0.89); in which IL = purified glycerin inclusion level (%).

Table 6 - Average feed intake (FI), weight gain (WG), feed conversion (FC), and weight at 42 days (W42d) of 1 to 42-day-old broilers according to the inclusion level of purified glycerin

Variable	Purified glycerin inclusion level (%)				Mean	P		CV (%)
	0	2	4	6		L	Q	
FI (g)[1]	4727.80	4670.00	4682.33	4761.83	4710.49	0.696	0.077	2.20
WG (g)	2846.02	2849.02	2929.31	2932.85	2889.30	0.034	0.993	2.84
FC (g g^{-1})	1.657	1.647	1.617	1.625	1.638	0.054	0.158	2.72
W42d (g)[1]	2892.95	2895.52	2976.06	2979.45	2935.98	0.034	0.990	2.79

CV - coefficient of variation; L - linear effect; Q - quadratic effect; P - probability of type I error at 5% using F test.
[1] $\hat{Y}$ = NS.
Equations: WG (g) = 2838.19 + 17.039 IL (P = 0.034; r^2 = 0.83); W42d (g) = 2884.99 + 17.002 IL (P = 0.034; r^2 = 0.83); in which IL = purified glycerin inclusion level (%).

Table 7 - Average values of carcass, special cuts, and organ yields of broilers slaughtered at 42 days according to the inclusion level of purified glycerin

Variable	Purified glycerin inclusion level (%)				Mean	P		CV (%)
	0	2	4	6		L	Q	
CY[1] (%)	85.08	84.51	84.45	84.65	84.67	0.616	0.533	1.75
TY[1] (%)	12.16	12.02	11.71	12.09	11.99	0.560	0.205	4.22
DSY[1] (%)	13.26	13.25	12.84	12.87	13.04	0.136	0.956	4.25
BY[1] (%)	35.69	35.56	36.35	35.54	35.79	0.908	0.583	4.25
HY (%)	0.46	0.45	0.44	0.41	0.44	0.011	0.147	5.83
GY[1] (%)	1.31	1.34	1.33	1.21	1.30	0.136	0.082	8.05
LY[1] (%)	1.83	1.87	1.94	1.88	1.89	0.274	0.317	5.37
SIY[1] (%)	3.01	3.00	2.97	2.95	2.98	0.586	0.998	6.62
LSI[1] (m)	1.73	1.77	1.78	1.68	1.73	0.701	0.261	7.40

CY - carcass yield; TY - thigh yield; DSY - drumstick yield; BY - breast yield; HY - heart yield; GY - gizzard yield; LY - liver yield; SIY - small intestine yield; LSI - length of small intestine; CV - coefficient of variation; L - linear effect; Q - quadratic effect; P - probability of type I error at 5% using F test.
[1] $\hat{Y}$ = NS.
Equation: HY (%) = 0.461 - 0.0065 IL (P = 0.011; r^2 = 0.80); in which IL = purified glycerin inclusion level (%).

Table 8 - Average values of crude protein (CP), ether extract (EE), moisture (M), ash, protein deposition (PD), and fat deposition (FD) in the breast meat of 42-day-old broilers

Variable	Purified glycerin inclusion level (%)				Mean	P		CV (%)
	0	2	4	6		L	Q	
CP (g)	21.82	22.27	22.77	23.10	22.48	0.001	0.805	2.51
EE (%)	5.27	5.51	5.54	5.71	5.51	0.001	0.680	2.84
M (%)	77.38	77.27	77.30	77.21	77.28	0.049	0.865	0.18
Ash[1] (%)	1.40	1.35	1.36	1.34	1.35	0.370	0.366	2.75
PD (g day^{-1})	4.31	4.32	4.48	4.64	4.44	0.045	0.585	6.54
FD (g day^{-1})	1.05	1.10	1.10	1.19	1.11	0.001	0.410	4.81

CV - coefficient of variation; L - linear effect; Q - quadratic effect; P - probability of type I error at 5% using F test.
[1] $\hat{Y}$ = NS.
Equations: CP (%) = 21.84 + 0.218 IL (P = 0.001; r^2 = 0.99); EE (%) = 5.299 + 0.0697 IL (P = 0.001; r^2 = 0.93); M (%) = 77.38 − 0.027 IL (P = 0.049; r^2 = 0.82); PD (g day^{-1}) = 4.27 + 0.056 IL (P = 0.045; r^2 = 0.93); FD (g day^{-1}) = 1.049 + 0.020 IL (P = 0.001; r^2 = 0.87); in which IL = purified glycerin inclusion level (%).

Table 9 - Average lightness (L*), redness (a*), yellowness (b*) pH, temperature, shear force (SF), and weight loss by cooking (CL) in the breast meat of 42-day-old broilers according to the inclusion level of purified glycerin

Variable	Purified glycerin inclusion level (%)				Mean	P		CV (%)
	0	2	4	6		L	Q	
L*[1]	59.86	62.10	61.36	61.20	61.14	0.185	0.035	2.13
a*[1]	9.45	8.85	9.17	9.94	9.36	0.234	0.054	8.58
b*[1]	10.04	9.58	9.96	10.22	9.95	0.736	0.556	14.83
pH[1]	6.23	6.13	6.14	6.07	6.13	0.153	0.843	2.81
Temperature	25.66	24.07	24.92	24.21	24.71	0.351	0.611	8.12
SF	1.36	1.39	1.34	1.36	1.36	0.563	0.943	9.33
CL	16.51	17.09	18.02	17.33	17.34	0.623	0.560	12.36

CV - coefficient of variation; L - linear effect; Q - quadratic effect; P - probability of type I error at 5% using F test.
[1] $\hat{Y}$ = NS.

Discussion

The increase in FI of broilers from 1 to 7 days may be related to the underdevelopment of the gastrointestinal organs of chicks during the pre-initial phase. Glycerol, the main component of glycerin, can interfere with the passage rate and impair energy absorption, leading to an increase in FI as a compensatory response (Henz et al., 2014).

However, the absence of an effect on FI in the subsequent phases can be attributed to experimental diets, which were balanced to contain the same nutritional levels. Therefore, it is possible that the nutritional requirements of birds were met in all evaluated treatments, regardless of the inclusion of purified glycerin.

Overall, better performance results were found in broilers as a function of purified glycerin levels. Similar results were reported by Cerrate et al. (2006) and Silva et al. (2012), evaluating different glycerin levels in broiler diets. The authors found that the inclusion of up to 5% glycerin had no effect on performance traits from 1 to 42 days of age.

Likewise, Abd-Elsamee et al. (2010) evaluated 0, 2, 4, 6, and 8% glycerin inclusion in broiler diets and concluded that 8% glycerin can be included without any effects on performance traits. However, it should be emphasized that in their study, the experimental diet with 8% glycerin exceeded the nutritional requirements for sodium (Na), unlike the present study, which met all nutritional requirements. Hence, the amount of dietary Na remained constant in all treatments (0, 2, 4, and 6%).

High concentrations of sodium and potassium, resulting from biodiesel catalysis, can cause dietary electrolytic imbalance (Cerrate et al., 2006). The electrolyte balance is directly related to water intake, and higher $Na^+ + K^+ - Cl^-$ ratios increase water intake (Mongin, 1981; Oliveira et al., 2010) and, consequently, bed moisture, as reported by Gianfelici et al. (2011) and Guerra et al. (2011).

Guerra et al. (2011), evaluating inclusion levels of 0, 2, 4, 6, 8, and 10% mixed crude glycerin in broiler diets at different production stages, concluded that it is possible to include up to 5% glycerin without influencing performance. However, additional care should be taken with bed management to reduce humidity.

The results for carcass yield and noble cuts are similar to those found by Cerrate et al. (2006), Guerra et al. (2011), and Silva et al. (2012), who evaluated increasing glycerin levels in broiler diets and found no effects on carcass and special cut yields.

Considering that the experimental diets were isoenergetic and that there was no difference in FI, the absence of any effects on carcass traits and special cuts can be considered an expected result, indicating that up to 6% glycerin can be included in broiler diets without affecting the relative weights of these variables.

Regarding edible viscera, Sehu et al. (2012) evaluated different glycerin levels in broiler diets and observed a reduction in relative heart weight of birds fed diets containing 5% glycerin. Contrary results were found by Topal and Ozdogan (2013), when using 0, 4, and 8% glycerin inclusion levels, with no effect on the relative weights of liver, gizzard, kidney, or proventriculus of male and female broilers. On the other hand, the authors verified that the relative heart weights of male broilers fed the diet containing 8% glycerin were higher compared with those of the males in other groups. According to the authors, the relative weights of some internal organs, such as heart and liver, may be associated with the WG of birds. Considering the above, it can be inferred that the reduction in relative heart weight was not based on WG of birds, which linearly increased with increasing levels of purified glycerin.

In a study conducted to evaluate the chemical composition of the meat of broilers fed diets containing glycerin, Topal and Ozdogan (2013) found no differences in moisture, ash, or crude protein contents. Nevertheless, they verified that the inclusion of 4 or 8% glycerin decreased the ether extract contents. The authors correlated this decline with the reduction in corn inclusion as glycerin was added, decreasing the ether extract content of diets, which may have favored the lower amount of fat in the meat.

Divergent results were found by Guerra et al. (2011), who evaluated the chemical composition of carcasses of broilers fed 0, 2, 4, 8, and 10% crude glycerin and reported the possibility of including up to 10% glycerin without any adverse effects on dry matter, crude protein, ether extract, or ash in the carcass.

Relative to protein and fat deposition, broilers fed diets containing glycerin had higher deposition values than those fed the control diet. These results are in agreement with reports of Silva et al. (2012), who stated that glycerol might increase protein deposition in breast meat by reducing gluconeogenesis from amino acids, caused by the inhibition of the enzyme phosphoenolpyruvate carboxykinase. On the other hand, Henz et al. (2014), evaluating 0, 3, 6, 9, 12, and 15% glycerin inclusion levels in broiler diets, did not observe effects on protein or fat deposition in carcasses of broilers slaughtered at 21 days of age.

In a study performed to evaluate the effects of different levels of dietary glycerin on the physical composition of broiler breast meat, Faria et al. (2013) observed an increase in redness and a change in tone angle. According to these authors, the meat showed an orange color, and increasing the levels of glycerin in the diets promoted a greater tendency towards the red color. For that reason, it was expected that the increase of purified glycerin in diets could influence the color of meat. However, this was not observed, indicating that the inclusion levels of purified glycerin were not high enough to influence these variables.

Conclusions

The dietary inclusion of up to 6% purified glycerin proved to be technically feasible in the diets of broilers from 1 to 42 days.

Conflict of Interest

The authors declare no conflict of interest.

Author Contributions

Data curation: M.C. Silva and L.F. Sousa. Formal analysis: M.C. Silva, L.F. Sousa and W.F. Augusto. Methodology: M.C. Silva, R.G.M.V. Vaz, K.F. Rodrigues, J.H. Stringhini, F.L.R. Fonseca and L.S. Bezerra. Project administration: R.G.M.V. Vaz. Supervision: M.C. Silva. Writing-review & editing: M.C. Silva, R.G.M.V. Vaz, K.F. Rodrigues and J.H. Stringhini.

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