

## Impact of dried brewers' grains supplementation on performance, metabolism and meat quality of broiler chickens

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### Abstract

The objective was to evaluate increasing levels of dried brewers' grains (DBG) in feed for broiler chickens from 1 to 21 days old and their effect on performance, blood parameters, intestinal morphometry, carcass characteristics and meat quality. The design was completely randomized with six treatments, which consisted of various levels of DBG inclusion, namely 0 (no inclusion), 20, 40, 60, 80, and 100 g/kg with seven replications and 17 animals per experimental unit, totalling 714 male broilers. Performance parameters, blood biochemical profile, morphology of the intestinal epithelium (duodenum), and carcass yield and composition were evaluated. Feed intake was not changed by DBG inclusion levels. Nor were weight gain and feed conversion ratio. Serum cholesterol levels were not influenced, and there were no effects on triglyceride, uric acid and creatinine levels. Enzyme aspartate aminotransferase showed a quadratic effect, as did alanine aminotransferase, with higher values at 79.5 and 63.9 g/kg DBG inclusion, respectively. No changes in carcass yield and relative organ weight were observed. The composition of the carcass in ether extract showed a quadratic effect, as fat deposition rate, with lowest values at 62.8 and 62.4 g/kg of DBG inclusion levels, respectively. Crude protein levels in carcass reduced linearly, as did fat deposition rate. The parameters of intestinal morphology and meat quality were not changed. Dried brewers' grains can be included in broiler diet from 1 to 21 days at levels up to 100 g/kg without influencing the metabolic parameters and broiler performance.

**Keywords:** alternative feed, blood, by-product, intestinal villi, performance

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### Introduction

Poultry production is an important source of animal protein, presenting strains with high productive indices, high efficiency in feed utilization and fast growth. Among the challenges are high feed costs and the availability of conventional feeds, which require a search for alternative sources with lower acquisition costs and good availability and that are not in conflict with human food (Bolu *et al.*, 2012; Ironkwe & Bangbose, 2011).

In this context, agro-industrial by-products are highlighted and can be economically viable alternatives (Abd El-Hack *et al.*, 2019; Swain *et al.*, 2012). Their utilization also allows a suitable destination for residues, preventing environmental problems (Brochier & Carvalho, 2009).

The brewing industry generates a considerable amount of residue, since every 100 L of beer that is produced results in 20 kg of brewers' grains being available for use in animal nutrition (Mussato *et al.*, 2006). This residue is of variable composition, since it may consist of a variety of grains, of which the most commonly used are barley, wheat, corn, rice and oats (Abd El-Hack *et al.*, 2019). After the fermentation process, the remaining insoluble material contains raw fibre fractions, ether extract, crude protein, amino acids, starch, minerals and vitamins (Ashour *et al.*, 2019; Alabi *et al.*, 2014), predominantly protein and fibre, owing to the removal of starch in the production process (Mussato *et al.*, 2006). Brewers' grains are obtained in wet form. However, because they deteriorate easily, they are normally dehydrated, resulting in dried brewers' grains (DBG) (Ashour *et al.*, 2019).

This by-product contains considerable protein and metabolizable energy. It can be used to reduce the amounts of corn and soybean meal in poultry diets. However, it has limiting characteristics, such as high fibre levels, which may reduce diet digestibility (Abd El-Hack *et al.*, 2019). Furthermore, DBG composition is variable, depending on plant maturity, processing, and the types of additive that are used by the industry (Santos *et al.*, 2003).

The results of the use of DBG are variable. Denstaldli *et al.* (2010) evaluated DBG in levels up to 400 g/kg in broilers from 12 to 33 days and concluded that its inclusion reduced growth and feed utilization. However, the performance in birds fed 10% to 20% DBG was similar to that of the control diets. On the contrary, Ashour *et al.* (2019) used DBG at levels up to 120 g/kg in broilers from 7 to 42 days old and there were no positive effects on their growth performance.

Thus, it is necessary to perform studies with levels of inclusion of DBG that do not compromise animal performance, enabling its use in poultry feeding. The hypothesis is that at a determined level, DBG can maintain poultry performance and prove to be a viable alternative to corn and soybean meal. The objective was to evaluate the performance parameters, blood parameters, carcass yield and intestinal morphology in broiler chickens fed with increasing DBG levels from 1 to 21 days.

## Material and Methods

This study was performed at the Poultry Education and Research Unit of the Federal University of Technology, Paraná, Campus Dois Vizinhos, PR, Brazil. All procedures were approved by the Ethical Committee and Animal Research under protocol number 2014-005.

A total of 714 one-day-old male Cobb 500 broiler chickens, weighing  $44.7 \pm 0.3$  g, were distributed in a completely randomized design, with six treatments, consisting of DBG levels of 0, 20, 40, 60, 80, and 100 g/kg, with seven replications and 17 broilers per experimental unit (EU). Broilers were kept in the thermal comfort zone recommended for each growth stage, with a water supply and feed ad libitum.

Brewers' grains were obtained in wet form (206.03 g/kg DM) from a brewery, then transported and distributed on a concrete floor, and exposed to the sun for three days, according to these periods of exposure (nine hours in the first day, 11 in the second, and 11 in the third day). The material was turned over every two hours and covered at night (Schone *et al.*, 2016). At the end of drying process, DBG were collected, packed in bags and stored. The nutritional composition of the dried brewers' grains is shown in Table 1.

**Table 1** Nutritional composition of dried brewers' grains

Dry matter, g/kg	915.80
Crude protein, g/kg	295.30
Fat, g/kg	49.60
Neutral detergent fibre, g/kg	650.00
Acid detergent fibre, g/kg	254.00
Gross energy, MJ/kg	18.95
Apparent metabolizable energy corrected for nitrogen balance, MJ/kg	8.06

The diets were formulated to be isocaloric and isoproteic (Table 2) according to the nutritional requirement recommendations (Rostagno *et al.*, 2011). The performance variables that were evaluated were weight gain (WG), feed intake (FI), and feed conversion ratio (FCR) and were corrected for mortality (Sakomura & Rostagno, 2016). Dried brewers' grains were included in the experimental broiler chick diets from 1 to 21 days. From 22 to 42 days, a basal ration was provided without DBG.

At 21 days old, after six hours of fasting, blood was collected by brachial puncture (ulnar vein) from two broilers per EU. Blood was allowed to coagulate and then was centrifuged at 1050 g for 10 min to obtain the serum, which was stored at  $-20$  °C (Nunes *et al.*, 2018). Blood parameters that were evaluated were cholesterol (CHOL), triglycerides (TAG), uric acid (UA), creatinine (CRE), aspartate aminotransferase (AST), alanine aminotransferase (ALT), and total protein (TP), using an auto biochemical analyser with automatic calibration (Flexor EL 200, Elitech) and commercial kits.

Evaluation of the intestinal morphometry was performed after a six-hour fast, when a broiler ( $\pm 5\%$  average weight EU) was slaughtered by cervical dislocation. A duodenal segment was collected, then fixed in formalin 10% and transferred to 70% alcohol solution. The tissue was dehydrated with alcohol solutions (80%, 90%, 95%, and 100%). The sample received a xylol bath before inclusion in histological paraffin.

Histological sections were made with a thickness of 5  $\mu\text{m}$ , fixed in blades, and subsequently stained with haematoxylin and eosin (Beçak & Paulete, 1976). For villi height and crypt depth reading, a light-coupled photomicroscope was used, which was connected to a computer with an image analysis program (Image Tool Version 3.0). The villi to crypt ratio was also determined.

**Table 2** Composition and specifications of experimental feeds for broiler chicks containing dried brewers' grains

Ingredients	Inclusion level of dried brewers' grains (g/kg)					
	0	20	40	60	80	100
Corn	597.00	575.70	554.40	533.00	511.7	490.40
Soybean meal	343.30	338.40	333.60	328.70	323.8	319.00
Dried brewers' grains	0.00	20.00	40.00	60.00	80.00	100.00
Soy oil	16.30	22.5	28.8	35.00	41.3	47.50
Dicalcium phosphate	16.90	16.80	16.7	16.60	16.50	16.40
Limestone	9.70	9.70	9.70	9.70	9.70	9.70
Biolysine	5.20	5.20	5.30	5.30	5.40	5.40
Sodium chloride	4.40	4.40	4.40	4.40	4.50	4.50
DL-Methionine (99%)	3.30	3.30	3.30	3.30	3.30	3.20
L-Threonine (98%)	1.30	1.30	1.30	1.30	1.30	1.30
Vitamin premix <sup>1</sup>	1.00	1.00	1.00	1.00	1.00	1.00
Mineral premix <sup>2</sup>	0.50	0.50	0.50	0.50	0.50	0.50
Antioxidant	0.05	0.05	0.05	0.05	0.05	0.05
<i>Nutrient specifications (g/kg)</i>						
Metabolizable energy (MJ/kg)	12.56	12.56	12.56	12.56	12.56	12.56
Crude protein	213	213	213	213	213	213
Calcium	8.8	8.8	8.8	8.8	8.8	8.8
Available phosphorus	4.3	4.3	4.3	4.3	4.3	4.3
Digestible lysine	12.7	12.7	12.7	12.7	12.7	12.7
Digestible methionine	6.1	6.1	6.1	6.1	6.1	6.1
Digestible methionine + cystine	9.1	9.1	9.1	9.1	9.1	9.1
Digestible tryptophan	2.3	2.3	2.3	2.3	2.3	2.3
Digestible threonine	8.2	8.2	8.2	8.2	8.2	8.2
Potassium	8.0	7.9	7.7	7.6	7.4	7.4
Sodium	2.0	2.0	2.0	2.0	2.0	2.0

<sup>1</sup>Vitamin A: 3000 mg; vitamin D3: 87.5 mg; vitamin E: 13.40 mg; vitamin K3: 2.5 mg; vitamin B1: 1.8 mg; vitamin B2: 6000 mg; vitamin B12: 16 mcg; vitamin B6: 2.6 mg; nicotinic acid: 40 mg; pantothenic acid: 12 mg; biotin: 65 mg; folic acid: 1 mg;

<sup>2</sup>Iron: 50 mg; copper: 9 mg; zinc: 60 mg; manganese: 70 mg; iodine: 1 mg; selenium: 300 mg; vehicle: 1000 g

Fat deposition rate (FDR) and protein deposition rate (PDR) were determined according to Scherer *et al.* (2011). On the first day, 10 chicks were slaughtered, plucked, weighed and frozen. At the end of the experimental period [repeat duration here for the reader], one bird per EU was slaughtered, plucked, weighed and frozen. The carcasses were crushed, homogenized and a sample was taken for pre-drying, grinding and subsequent analysis of dry matter, crude protein and ether extract. Determinations of FDR and PDR were performed according to Fraga *et al.* (2008).

At 21 days, after six hours of fasting, two broilers ( $\pm 5\%$  of the average weight per EU) were slaughtered by electronarcosis. Cut yield (breast, thigh, drumstick, and wing) was related to eviscerated

carcass weight (%). The relative weight of organs (liver, heart, gizzard, intestine) and the percentage of abdominal fat were calculated and related to broilers' live weight (percentage of live weight).

The pectoral muscle (*Pectoralis major*) was used to evaluate meat quality at 42 days old. Cooking loss (CL) was assessed in the right portion of the pectoral muscle without skin and surface fat (Osório *et al.*, 1998). After the sample was weighed, it was submitted to meat texture evaluation (shear force (SF)). For this, the fibres were first cut longitudinally, making five rectangles of 1.0 × 1.0 × 2.0 cm. The samples were inserted into the texturometer (Brookfield CT3 texture analyser) coupled to a probe (TA 3/100, fixture TA-SBA, calibration with 0.01 kg force; 20 mm deformation; 2.5 mm/s test speed) with fibre orientation perpendicular to the probe.

Water-holding capacity (WHC) was determined in two samples that were cut from the left portion of each breast and weighed on an analytical scale to determine the initial weight. They were then wrapped in filter paper and centrifuged for 4 min at 2000 rpm (Nakamura & Katok, 1985).

For statistical analysis, the PROC GLM of SAS<sup>®</sup> university edition (2017) statistical software (SAS Inst. Inc. Cary, NC, USA) was used. Data were submitted to polynomial regression ( $P < 0.05$ ). The mathematical model used was:

$$y_{ij} = \mu + t_i + e_{ij}$$

Where:  $y_{ij}$  = observation,  
 $\mu$  = overall mean,  
 $t_i$  = effect of diet, and  
 $e_{ij}$  = experimental random residual error.

## Results and Discussion

Inclusion of DBG levels did not influence ( $P > 0.05$ ) FI, WG or FCR of broilers (Table 3), which presented average values of 1.247, 0.893 and 1.396 kg, respectively. Dried brewers' grains contain non-starch polysaccharides, which reduce the utilization of nutrients. Replacing feeds such as corn with high-fibre sources could reduce the viscosity of the digestive tract content and interfere with nutrient digestion and absorption, consequently reducing bodyweight and carcass quality (Alabi *et al.*, 2014). However, this effect did not occur in the present study, demonstrating that these levels were adequate to maintain bird performance that was similar to those that received the treatment without DBG inclusion (0 g/kg).

**Table 3** Performance of broiler chickens fed with increasing levels of dried brewers' grains from 1 to 21 days old

Dried brewers' grains level	Feed intake (kg)	Weight gain (kg)	Feed conversion (kg/kg)
0	1.270	902	1.407
20 g/kg	1.241	877	1.415
40 g/kg	1.246	891	1.398
60 g/kg	1.246	898	1.387
80 g/kg	1.251	902	1.386
100 g/kg	1.228	889	1.381
Coefficient of variation %	3.290	4.410	4.210
SEM	0.006	0.006	0.009
P-value	0.580	0.827	0.872

Denstaldli *et al.* (2010) evaluated DBG inclusion levels of 0, 100, 200, 300 and 400 g/kg in the diet of broilers from 12 to 33 days old and concluded that their performance approached the control treatment at 100 and 200 g/kg inclusion levels. Inclusion of more than 200 g/kg resulted in a reduction in broiler performance. Onifade and Babatunde (1998) evaluated the inclusion of three levels of DBG (100, 200, and 300 g/kg) in broiler diets and observed an increase in FI, while feed efficiency was similar to that at 100 g/kg level, but decreased at higher DBG concentrations.

Blood levels of CHOL and TAG (Table 4) were not altered ( $P > 0.05$ ) by DBG inclusion. These metabolites are sensitive indicators of the intensity of lipid metabolism in poultry organisms (Bogusławska-Tryk *et al.*, 2016), so it could be inferred that lipid metabolism was not altered by the inclusion of DBG. Ashour *et al.* (2019) did not obtain effects of DBG inclusion on blood levels of total cholesterol and TAG at levels up to 120 g/kg.

Serum CRE was not changed ( $P > 0.05$ ) by DBG inclusion. Creatinine is an indicator of renal function and protein metabolism. Its concentration is related directly to muscle mass, age, physical activity and, like most of the chemical components of the blood, is influenced by diet (Piotrowska *et al.*, 2011). Uric acid level was not changed ( $P > 0.05$ ), being the main product of nitrogen and purine metabolism in broilers (Rezende *et al.*, 2017). The levels obtained in this study presented mean values of 4.34 mg/dl, which were considered within the acceptable range (under 15 mg/dl) (Schmidt *et al.*, 2007), although variations in critical values for this parameter occur in the literature.

**Table 4** Blood parameters of broiler chickens fed with increasing levels of dried brewers' grains at 21 days old

Dried brewers' grains level	CHOL (mg dL <sup>-1</sup> )	TAG (mg dL <sup>-1</sup> )	UA (mg dL <sup>-1</sup> )	CRE (mg dL <sup>-1</sup> )	AST (UI L <sup>-1</sup> )	ALT (UI L <sup>-1</sup> )	TP (g dL <sup>-1</sup> )
0	161.5	46.5	4.02	0.20	225.00	1.49	2.98
20 g/kg	162.5	45.5	4.32	0.20	234.42	1.51	3.42
40 g/kg	152.4	44.2	4.32	0.23	237.71	1.55	3.01
60 g/kg	150.4	46.1	4.22	0.20	264.57	1.61	3.15
80 g/kg	155.1	43.0	4.71	0.22	263.14	1.58	2.94
100 g/kg	143.4	41.4	4.47	0.21	250.57	1.53	2.69
CV	9.73	10.79	12.15	9.88	7.45	4.96	6.26
SEM	2.399	0.749	0.008	0.003	3.507	0.012	0.044
P-value	0.183	0.326	0.272	0.062	0.001	0.044	<0.001
Linear					<0.001	0.033	<0.001
Quadratic					0.050	0.026	<0.001

CHOL: cholesterol; TAG: triglycerides; UA: uric acid; CRE: creatinine; AST: aspartate aminotransferase; ALT: alanine aminotransferase; TP: total protein; g/dL: grams per decilitre; mg dl<sup>-1</sup>: milligrams per decilitre, IU: international units per litre; NS: not significant; CV: coefficient of variation; SEM: standard error of mean  
 $AST = 221.031 + 9.193 \cdot DBG - 0.575255 \cdot DBG^2$  ( $R^2 = 0.34$ );  $ALT = 1.473 + 0.035 \cdot DBG - 0.0027551 \cdot DBG^2$  ( $R^2 = 0.21$ );  $TP = 3.077 + 0.0788 \cdot DBG - 0.0118559 \cdot DBG^2$  ( $R^2 = 0.41$ )

Enzymes AST and ALT showed a quadratic effect ( $P < 0.05$ ) with higher values at the levels of 79.5 g/kg and 63.9 g/kg of DBG inclusion, respectively. These enzymes are commonly used to indicate liver function status (Rocha *et al.*, 2013). The critical limit of AST values that may be related to liver or muscle disorders is around 275 IU/L, which was not exceeded in the present study (Schmidt *et al.*, 2007). Contrary results were obtained by Ashour *et al.* (2019), who achieved reductions in blood AST and CRE levels in broilers fed up to 120 g/kg of DBG and related this result to a possible antioxidant effect of DBG components. Alanine aminotransferase is found in more significant amounts in the liver and is commonly used to identify liver problems, being released into the blood after hepatocellular damage (Senanayake *et al.*, 2015). The concentrations of serum TP had a quadratic effect ( $P < 0.01$ ), with higher levels at 32.0 g/kg DBG inclusion. However, it remained within the range considered physiological by Harr (2002), namely between 2.5 and 4.5 g/dl.

The variables carcass yield and proportions of breast, thigh, drumstick, and wing were unchanged ( $P > 0.05$ ) with DBG inclusion (Table 5), as were values for relative organ weight and abdominal fat percentage ( $P > 0.05$ ) (Table 6). The results for carcass yield corroborate those obtained by Kokol *et al.* (2012), who found no differences, even at higher levels of DBG inclusion (0, 150, 300, 450 and 600 g/kg) to replace corn in diets. The authors related this to the lower bodyweight that resulted at the inclusion levels of 60 to 120 g/kg.

**Table 5** Yield of carcass, breast, thigh, drumstick, and wing (%) at 42 days old of broiler chickens fed with dried brewers' grains from 1 to 21 days old

Dried brewers' grains level	Carcass	Breast	Thigh	Drumstick	Wing
0	78.15	34.73	11.91	13.89	9.22
20 g/kg	80.61	34.35	11.32	14.00	8.94
40 g/kg	81.56	35.00	11.50	13.79	9.09
60 g/kg	79.57	34.64	11.51	13.22	9.14
80 g/kg	82.46	35.87	11.53	13.52	9.11
100 g/kg	80.73	33.07	11.52	13.31	9.40
Coefficient of variation, %	4.18	3.93	5.15	7.19	8.19
SEM	5.32	2.25	0.90	1.48	1.10
<i>P</i> -value	0.244	0.066	0.589	0.600	0.915

Kokol *et al.* (2012) used DBG to replace corn in broiler diets at levels up to 600 g/kg and did not find effects in organ weight. As dietary fibre levels increased, higher gizzard development occurred, and consequently an increase in the weight of this organ (Braz *et al.*, 2011). However, even with the highest fibre content of DBG, this response was not evidenced in this study. Denstaldli *et al.* (2010) obtained an increase in broiler gizzard size at levels of 300 and 400 g/kg of DBG in diets. They highlighted that structural components and fibrous materials stimulate gizzard activity and can activate the secretion of pancreatic enzymes and bile acid. However, at higher levels, these fibrous components caused a reduction in feed utilization.

**Table 6** Relative organ weight (%) and abdominal fat percentage at 42 days old from broilers fed with increasing levels of dried brewers' grains from 1 to 21 days old

Dried brewers' grains level	Liver	Heart	Gizzard	Intestine	Abdominal fat
0	1.65	0.41	1.65	3.58	2.42
20	1.64	0.48	1.74	3.59	2.40
40	1.76	0.47	1.82	3.42	2.09
60	1.74	0.49	1.79	3.42	1.98
80	1.70	0.50	1.69	3.68	1.70
100	1.58	0.50	1.66	3.56	1.95
Coefficient of variation, %	11.70	17.31	13.23	5.29	23.84
SEM	0.019	0.128	0.034	0.030	0.082
<i>P</i> -value	0.560	0.360	0.643	0.087	0.079

Carcass ether extract composition and fat deposition rate (FDR) showed a quadratic effect ( $P < 0.05$ ), with the lowest values at 62.8 and 62.4 g/kg of DBG inclusion, respectively (Table 7). Carcass crude protein composition and PDR reduced linearly with increasing levels of DBG. One factor that can influence fat deposition is the balance of amino acids, since excess protein or its low digestibility can lead to an increase in FDR. Thus, the objective was to meet the requirements in amino acids, seeking maximum protein deposition and lower fat deposition (Trindade Neto *et al.*, 2009). Bolu *et al.* (2012) found no difference in the protein retention rate in broilers fed up to 100 g/kg dry distillers' grains to replace corn in diets. However, with levels of 300 and 400 g/kg, there were reductions in protein retention and fat retention rates.

**Table 7** Ether extract and crude protein content of carcasses at 21 days old from broilers fed increasing levels of dried brewers' grains

Dried brewers' grains level	Ether extract, %	Crude protein, %	FDR, g/day	PDR, g/day
0	16.31	17.50	6.72	7.10
20, g/kg	13.04	18.67	5.24	7.47
40, g/kg	12.26	17.13	5.04	6.99
60, g/kg	14.91	16.31	6.08	6.57
80, g/kg	12.94	16.22	5.33	6.64
100, g/kg	13.52	15.90	5.51	6.41
Coefficient of variation, %	12.54	5.75	13.87	6.63
SEM	0.331	0.203	0.144	0.086
<i>P</i> -value	0.001	<0.001	0.003	0.001
Linear	0.037	<0.001	0.064	<0.001
Quadratic	0.019	0.634	0.032	0.616

FDR: fat deposition rate; PDR: protein deposition rate;

EE = 15.565 – 0.826655\*DBG + 0.0657212\*DBG<sup>2</sup> (R<sup>2</sup> = 0.17); CP = 18.113 – 0.230486\*DBG (R<sup>2</sup> = 0.36); FDR = 6.35643 – 0.338872\*DBG + 0.0271199\*DBG<sup>2</sup> (R<sup>2</sup> = 0.15); PDR = 7.326446 – 0.091365\*DBG (R<sup>2</sup> = 0.31)

Villi height, crypt depth, and villi/crypt ratio (Table 8) were unchanged (*P* > 0.05), presenting mean values of 1192 µm, 224 µm and 5.35 µm, respectively. The maintenance of villus size, digestive capacity and absorption by the intestinal epithelium depends on the processes of cell renewal, performed by stem cells in the crypt and along the villi, and also on cell loss (extrusion), which usually occurs at the villus apex. If a higher rate of intestinal extrusion or a reduction in the proliferation rate occurs, the intestine responds with a reduction in villus height and a consequent decrease in the capacity to digest and absorb (Pelicano *et al.*, 2003; Pluske *et al.*, 1997). In the present study the authors observed the appearance of bifurcations in this region of intestinal tissue, which for Aleixo *et al.* (2011) is associated with the region's inefficiency in capturing nutrients, compensating the action of anti-nutritional factors, where the intestine epithelium forms bifurcations to increase the contact and absorption surface.

**Table 8** Morphology of duodenal mucosa at 21 days old of broilers fed with increasing levels of dried brewers' grains

Dried brewers' grains level	Villi height (µm)	Crypt depth (µm)	Villi: crypt ratio
0	1185	223	5.33
20, g/kg	1202	224	5.38
40, g/kg	1194	222	5.41
60, g/kg	1184	226	5.26
80, g/kg	1197	216	5.57
100, g/kg	1188	230	5.16
Coefficient of variation, %	2.97	7.43	8.65
SEM	0.005	0.002	0.069
<i>P</i> -value	0.926	0.762	0.674

Higher values for crypt depth indicate greater cell proliferative activity to ensure an adequate rate of epithelial renewal, compensating losses in villi heights (Pluske *et al.*, 1997). However, this effect did not occur in the present study, demonstrating that these levels of DBG did not have major effects on duodenal epithelium morphology. The villi to crypt ratio is a useful criterion for estimating the digestive capacity of the

small intestine. However, the effects of dietary fibre on epithelial morphology and cellular turnover in the gastrointestinal tract have not been studied in detail, with conflicting results between authors. Furthermore, factors such as type and level of fibre, broiler age, and the composition of the basal diet may influence the intestinal mucosa response (Mateos *et al.*, 2012).

There was no effect ( $P > 0.05$ ) on cooking loss (Table 9). This result differs from those obtained in the study by Ashour *et al.* (2019), in which lower values of CL occurred with the use of DBG in the broilers' diet compared with the control treatment, which affected the sensory properties of the meat.

**Table 9** Meat quality parameters in breast of broilers at 42 days old fed with increasing levels of dried brewers' grains (up to 21 days)

Dried brewers' grains level	Cooking loss (%)	Water-holding capacity (%)	Shear force (Kgf/cm <sup>2</sup> )
0	24.0	22.1	2.55
20, g/kg	23.3	23.4	2.62
40, g/kg	24.2	22.2	3.11
60, g/kg	23.8	21.9	3.46
80, g/kg	25.2	21.5	2.43
100, g/kg	24.5	21.3	2.47
Coefficient of variation	15.26	7.83	42.66
SEM	0.542	0.272	0.181
<i>P</i> -value	0.951	0.291	0.536

Water-holding capacity was not influenced ( $P > 0.05$ ) by DBG levels in the diets. Neither was the shear force. The results indicated extremely soft meat (Ramos & Gomide, 2007) with values below 3.62 kgf. Values between 6.62 and 9.60 are classified as slightly soft to slightly hard, and values above 12.60 kgf are rated as extremely hard.

## Conclusion

Inclusion of dried brewers' grains at up to 100 g/kg in broiler feed from 1 to 21 days old appears not to impair performance, blood parameters, carcass yield, body composition and meat quality. Future studies are necessary with diets which include dried brewers' grains at higher levels, and in combination with other ingredients for growing and finishing of broiler chickens.

## Authors' Contributions

RVN and PSC designed the study and were the supervisors. WP worked on the project, laboratory analysis and received his MSc. CE, JB, VDLS, ECS and ASA participated in management and discussion of the results, statistical analysis and writing, and corrected the manuscript.

## Conflict of Interest Declaration

The authors declare there is no conflict of interest.

## References

- Abd El-Hack, M.E.A., Alagawany, M., Patra, A., Abdel-Latif, M., Ashour E.A., Arif, M., Farag, M.R. & Dhama, K., 2019. Use of brewers dried grains as an unconventional feed ingredient in the diets of broiler chickens: A review. *Adv. Anim. Vet. Sci.* 7, 218-224. DOI: 10.17582/journal.aavs/2019/7.3.218.224
- Alabi, O.O., Atteh, J.O., Adejuno, I.O. & Ogundele, O.O., 2014. Effects of dietary levels of brewers' dried grains supplemented with commercial enzymes on performance, nutrient retention and gastro-intestinal tract characteristics of Arbor Acres broilers. *Int. J. Agric. Res. Innov. Technol.* 2, 818-823.
- Aleixo, V.M., Pressinoti, L.N., Campos, D.V.S., Menezes-Aleixo, R.C. & Ferraz, R.H.S., 2011. Histology, histochemistry and histometry of the captive-bred alligator gut. *Pesq. Vet. Bras.* 31, 1120-1128. (in Portuguese, English abstract).
- Ashour, E.A., Abd El-Hack, A.M.E., El-Hindawy, M.M., Attia, A.I., Osman, A.O., Swelum, A.A., Alowaimer, A.N., Saadeldin, I.M. & Laudadio, V., 2019. Impacts of dietary inclusion of dried brewers' grains on growth, carcass traits, meat quality, nutrient digestibility and blood biochemical indices of broilers. *S. Afr. J. Anim. Sci.* 49, 573-584. DOI: 10.4314/sajas.v49i3.18

- Beçak, W. & Paulete, J., 1976. Cytology and histology techniques. Technical and Scientific Books, Rio de Janeiro, BR, 1976. (in Portuguese)
- Boguslawska-Tryk, M., Piotrowska, A., Szymeczko, R., Burlikowska, K. & Głowińska, B., 2016. Lipid metabolism indices and fatty acids profile in the blood serum of broiler chickens fed a diet with lignocellulose. *Braz. J. Poultry Sci.* 18, 451-456. <http://dx.doi.org/10.1590/1806-9061-2015-0157>
- Bolu, S.A., Alli, O.I. & Esoula, P.O., 2012. Response of broilers to graded levels of distillers dried grains. *Sustain. Agric. Res.* 1, 147-150. DOI: 10.5539/sar.v1n1p147
- Braz, N.M., Freitas, E.R., Bezerra, R.M., Cruz, C.E.B., Farias, N.N.P., Silva, N.M., Sá, N.L. & Xavier, R.P.S., 2011. Fiber in growth ration and its effects on laying performance in the growing and laying phases. *R. Bras. Zootec.* 40, 2744-2753. (in Portuguese, English abstract).
- Brochier, M.A. & Carvalho, S., 2009. Environmental, productive and economic aspects of the use of wet brewery residue in feedlot lambs. *Ciênc. Agrotec.* 33, 1392-1399.
- Denstadli, V., balance, S., Knutsen, S.H., Westereng, B. & Svihus, B., 2010. Influence of graded levels of brewers dried grains on pellet quality and performance in broiler chickens. *Poult. Sci.* 89, 2640-2645. DOI: 10.3382/ps.2010-00724
- Fraga, A.L., Moreira, I., Furlan, A.C., Bastos, A.O., Oliveira, R.P. & Murakami, A.E., 2008. Lysine requirement of starting barrows from two genetic groups fed on low crude protein diets. *Braz. Arch. Biol. Technol.*, 51, 49-56.
- Harr, K.E., 2002. Clinical chemistry of companion avian species: A review. *Vet. Clin. Pathol.* 31, 140-151.
- Ironkwe, M.O. & Bamgbose, A.M., 2011. Effect of replacing maize with brewers' dried grains in broiler finisher diet. *Int. J. Poultry Sci.* 10, 710-712.
- Kokol, C., Zaklag, U., Antyev, M., Akade, F.T. & Bab, A.M.J., 2012. Response of broiler finisher fed graded levels of brewers dried grains on carcass and internal organ characteristics. *J. Agric. Vet. Sci.* 4, 70-76.
- Mateos, G.G., Jiménez-Moreno, E., Serrano, M.P. & Lázaro, R.P., 2012. Poultry response to high levels of dietary fiber sources varying in physical and chemical characteristics. *J. Appl. Poultry Res.* 21, 156-174. DOI: 10.3382/japr.2011-00477
- Mussato, S.I., Dragone, G. & Roberto, I.C., 2006. Brewers' spent grain: Generation, characteristics and potential applications. *J. Cereal Sci.* 43, 1-14. DOI: 10.1016/j.jcs.2005.06.001
- Nakamura, M. & Katok, K., 1985. Influence of thawing method on several properties of rabbit meat. *Bull. Ishika. Prefect. Coll. Ag.* 11, 45-49.
- Nunes, R.V., Broch, J., Wachholz, L., Souza, C., Damasceno, J.L., Oxford, J., Bloxham, H., Billard, D.J.L. & Pesti, G.M., 2018. Choosing sample sizes for various blood parameters of broiler chickens with normal and non-normal observations. *Poult. Sci.* 0, 1-9.
- Onifade, A.A. & Babatunde, G.M., 1998. Comparison of the utilisation of palm kernel meal, brewers' dried grains and maize offal by broiler chicks. *Br. Poult. Sci.* 39, 245-250. DOI: 10.1080/00071669889204
- Osório, J.C.S., Osório, M.T. & Jardim, P., 1998. Methods for evaluation of sheep meat production: in vivo, in carcass and meat. *Pelotas: UFPEL*, 107p.1998. (in Portuguese)
- Pelicano, E.R.L., Souza, P.A., Souza, H.B.A., Oba, A., Norkus, E.A., Kodawara, L.M. & Lima, T.M.A., 2003. Intestinal mucosa morphometry and ultrastructure of broilers fed diets containing different probiotics. *Rev. Port. Ciênc. Vet.* 98, 125-134. (in Portuguese, English abstract)
- Piotrowska, A., Burlikowska, K. & Szymeczko, R., 2011. Changes in blood chemistry in broiler chickens during the fattening period. *Folia biol-krakow* 59, 183-187. DOI: 10.3409/fb59\_3-4.183-187
- Pluske, J.R., Hampson, D.J. & Williams, I.H., 1997. Factors influencing the structure and function of the small intestine in the weaned pig: a review. *Livest. Prod. Sci.*, 51, 215-236. DOI: 10.1016/S0301-6226(97)00057-2
- Ramos, E.M. & Gomide, L.A.M., 2007. Meat quality evaluation: Fundamentals and methodologies. Viçosa, MG, BR. (in Portuguese).
- Rezende, M.S., Mundim, A.V., Fonseca, B.B., Miranda, R.L., Oliveira, Jr. W. & Lellis, C.G., 2017. Profile of serum metabolites and proteins of broiler breeders in rearing age. *Braz. J. Poultry Sci.* 19, 583-586. DOI: 10.1590/1806-9061-2016-0338
- Rocha, M.T., Andrade, A.M., Gonzales, E., Stringhini, J.H., Santana, E.S., Pôrto, R.N.G. & Rezende, C.S.M., 2013. Liver function and bacteriology of organs in broiler inoculated with nalidixic acid-resistant salmonella typhimurium and treated with organic acids. *Ital. J. Anim. Sci.* 12, 347-352. DOI:
- Rostagno, H.S., Albino, L.F.T., Donzele, J.L., Gomes, P.C., Oliveira, R.F., Lopes, D.C., Ferreira, A.S. & Barreto, S.L.T., 2011. Brazilian tables for poultry and swine: Food composition and nutritional requirements. 3rd edition. Viçosa, BR (in Portuguese).
- Sakomura, N.K. & Rostagno, H.S., 2016. Monogastric nutrition research methods. 2nd edition. FUNEP, Jaboticabal, SP, Brazil.
- Santos, M., Jiménez, J.J., Bartolomé, B., Gómez-Cordovés, C. & Del Nozal, M.J., 2003. Variability of brewer's spent grains within a brewery. *Food Chem.* 80, 17-21.
- Scherer, C., Furlan, A.C., Martins, E.N., Scarpinello, C. & Ton, A.P.S., 2011. Metabolizable energy requirement of broiler quails from 1 to 14 days old. *R. Bras. Zootec.*, 40, 2496-2501. (in Portuguese, English abstract)
- Schmidt, E. M.S., Locatelli-Dittrich, R., Santin, E., & Paulillo, A.C., 2007. Clinical pathology in poultry – A tool for monitoring poultry health. *Arch. Vet. Sci.* 12, 9-20. (in Portuguese, English abstract) doi: 10.5380/avsv12i3.10906
- Schone, R.A., Zambom, M.A., Nunes, R.V., Fernandes, T., Frank, R., Oliveira, T.M.M. & Castagnara, D.D. 2016. Brewers grains drying in layers with different thicknesses. *Sci. Agr. Parana.* 15, 127-131. (in Portuguese, English abstract)

- Senanayake, S.S.H.M.M.L., Ranasinghe, J.G.S., Waduge, R., Nizanantha, K. & Alexander, P.A.B.D., 2015. Changes in the serum enzyme levels and liver lesions of broiler birds reared under different management conditions. *Trop. Agric. Res.* 26, 584–595. DOI: 10.4038/tar.v26i4.8121
- Swain, B.K., Naik, P.K., Chakurkar, E.B. & Singh, N.P., 2012. Effect of feeding brewers' dried grains on the performance and carcass characteristics of Vanaraja chicks. *J. Appl. Anim. Res.* 40, 163-166. DOI: 10.1080/09712119.2011.645036
- Trindade Neto, M.A., Takeara, P., Toledo, A.L., Kobashigawa, E., Albuquerque, R. & Araújo, L.F., 2009. Digestible lysine levels for 37–49-day-old male broilers. *R. Bras. Zootec.* 38, 508-514. (in Portuguese, English abstract) DOI: 10.1590/S1516-35982009000300016