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Effects of guavira fruit *(Campomanesia adamantium)* peel extract on performance and meat quality of broilers

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Abstract

This article assessed the performance, carcass yield, and meat quality of finishing broilers fed increasing levels of hydroethanolic extract of guavira fruit peel (HEGP) were assessed. A total of 480, three-weeks-old male broilers were randomly allocated to dietary levels of HEGP (0, 100, 200, 300, 400, and 500 mg/kg), with five replicates and 16 birds each. There was a quadratic effect of HEGP inclusion on weight gain (WG) and feed conversion ratio (FCR), with the greatest WG and FCR being calculated at levels of 314 and 219 mg/kg HEGP, respectively. Broilers fed diets containing the extract had better performance than those fed an extract-free diet. There was no effect of extract inclusion on carcass yield and cuts. There was a quadratic effect of HEGP inclusion on *a** (redness) of thigh meat at 15 min post-mortem, and on water-holding capacity (WHC), with the lowest and highest values being calculated at 270 mg/kg and 263 mg/kg HEGP, respectively. There was a quadratic influence of HEGP inclusion on the malonaldehyde content at 30-day storage, with highest value being calculated at 218 mg/kg HEGP. Dietary inclusion of 219 mg/kg of HEGP resulted in better FCR for broilers in the finishing phase but without improvement in meat quality.

Keywords: Cerrado fruit, feed additives, flavonoids, meat quality.
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Introduction

For decades it has been common practice in the poultry industry to manipulate the intestinal health of broiler chickens with antibiotics in subtherapeutic concentrations. The dietary inclusion of these substances contributed to efficient nutrient absorption and assimilation, thereby favouring productive performance and reducing feed costs (Oleforuh-Okoleh *et al.*, 2015). Despite these benefits, the use of antibiotics to enhance broiler performance was correlated with the development of bacterial resistance in humans, which is a public health concern (Garcia-Migura *et al.*, 2014).

Mechanisms of action have been observed in natural extracts containing polyphenols and flavonoids that are similar to those of performance-enhancing antibiotics (Surai *et al.*, 2014). Plant extracts are natural non-toxic products that could be used as natural feed additives without inducing the resistance of microorganisms (Huyghebaert *et al.*, 2011). Guavira *(Campomanesia adamantium)* fruit extracts are among these potential natural additives. This fruit from the Cerrado biome has aroused interest among researchers because of its significant levels of proteins, vitamins, minerals, and fatty acids (Vallilo *et al.*, 2006).

In addition to its nutritional value, guavira contains bioactive substances such as ascorbic acid and phenolic compounds. Flavonoids, a class of polyphenolic compounds, are the major constituents, and include isoquercetin and quercetin (Pascoal *et al.*, 2011). These substances may have antioxidant, antimicrobial, or immunostimulant activity (Pereira *et al.*, 2012; Viecili *et al.*, 2014; Capeletto *et al.*, 2016). Phenolic compounds may inhibit the adhesion of pathogenic bacteria to the intestinal epithelia (Jamroz *et al.*, 2006; Windisch *et al.*, 2008), influence digestive processes (Ader *et al.*, 2000), and modulate the animal's immune system (Kamboh *et al.*, 2015), with consequent improvements in performance. Moreover, these compounds can improve meat quality by acting as antioxidant agents, reducing the lipid oxidation process by

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neutralizing reactive oxygen molecules and preventing toxin formation during meat processing and cooking (Jiang & Xiong, 2016).

Although guavira contains these compounds, the effects of using it in broiler nutrition have not yet been reported. Therefore, this study aimed to evaluate the effects of feeding increasing levels of HEGP on the performance, carcass yield, and meat quality of finishing broilers. The authors hypothesized, therefore, that the bioactive substances of guavira fruit peel extract will improve the performance and meat quality of broilers.

Material and Methods

The experiment was carried out at the Poultry Study Centre of Western Paraná State University, Unioeste, Marechal Cândido Rondon Campus, PR, Brazil. The birds were cared for according to ethical principles for animal experimentation established by the Brazilian Society of Laboratory Animal Science (SBCAL) and National Council for the Control of Animal Experimentation (CONCEA). The protocol of this experiment was approved by the Committee on Ethics in the Use of Animals of Western Paraná State University (Protocol #38/17). All efforts were made to minimize animal suffering.

A total of 600 one-day-old Cobb 500® male broiler chickens were housed in an experimental shed, divided into 1.95 m² pens (experimental unit), with a concrete floor and pine wood shavings reused five consecutive times. Each unit had access to a tubular feeder and a nipple-type drinker. The lighting programme followed the lineage manual recommendations. The temperature and relative humidity were measured daily by digital thermo-hygrometers (Instrutemp, ITHT 2250, São Paulo, Brazil) at bird height inside the facility. The temperature inside the shed was controlled with exhaust fans and evaporative cooling pads, set at maximum and minimum temperatures of 24.80 °C and 21.51 °C and maximum and minimum humidity of 74.99% and 63.82%.

For the first 21 days of rearing, all birds were kept under the same nutritional, management, and environmental conditions. The diets were formulated according to Rostagno *et al.* (2017) for intermediate-performance male broilers. On day 21, birds were weighed individually, divided into weight classes, and 480 birds, with an average weight of 1.007 ± 6.5 g, were redistributed in a manner that equalized pen weights and variances into a completely randomized design with six dietary treatments, with five replicates and 16 birds per unit. The treatments consisted of increasing levels of HEGP (0, 100, 200, 300, 400, and 500 mg/kg).

Guavira fruit was purchased in Ponta Porã - MS, Brazil and processed at the Faculty of Biological Sciences of the Federal University of Grande Dourados in Dourados - MS, Brazil. The fruits were washed and immersed in a sanitized solution of 0.66% sodium dichloroisocyanurate dehydrate (Diversey Chemical Industry, São Paulo, Brazil) for 15 min. Afterwards, the fruit was pulped, and the peel was dried in an oven at 40 °C with an airflow of 0.5 m^3/s for 36 hours. The dried material was packaged in low-density polyethylene package and stored at room temperature (25 °C) until the extract was produced. The extract was obtained by Soxhlet extraction using 150 g of the sample and 750 mL of 70% absolute ethyl alcohol (1:5 w/v ratio) (Synth, São Paulo, Brazil). The extraction was carried out at 80 °C for three hours, and then the solvent was removed under vacuum using a rotary evaporator (Fisatom model 810) to yield the aqueous extract, which was stored in amber glass vials at 7 ºC until analysis.

The bioactive compounds were extracted according to Singleton and Rossi (1965), with modifications to determine the total polyphenol and flavonoid content of the extract. Two millilitres of the extract were homogenized with 2 mL of methanol (Sigma Aldrich, São Paulo, Brazil) in Falcon tubes. Subsequently, the tubes were placed in a homogenizer (Criemaq C38, São Paulo, BR) for 10 min and then centrifuged (Centrifuge Kasvi K14-4000, Kasvi, São Paulo, BR) at 3000 rpm for 20 min. The supernatant was used directly for the measurements.

The total concentration of phenolic compounds in HEGP was determined according to Singleton and Rossi (1965), with modifications. An aliquot of the supernatant (125 µL) was homogenized with 125 µL of Folin–Ciocalteu reagent (Sigma Aldrich, São Paulo, Brazil) (1:1 v/v deionized water) and sodium carbonate (Sigma Aldrich, São Paulo, Brazil) (28 g/L) in a total volume of 2.25 mL. After incubation in the dark at room temperature (25 °C) for 30 min, absorbance was measured in a spectrophotometer (Evolution™ 300 e Thermo Scientific) at 750 nm. The results were expressed as milligrams of gallic acid equivalents (GAE) per litre of HEGP (mg GAE/L), using a standard curve constructed with gallic acid (Dinâmica Química Contemporânea LTDA, São Paulo, Brazil) at 0 - 300 mg/L. All analyses were performed in triplicate.

The flavonoid content was established with the aluminium chloride ($AlCl_3$) method (Buriol *et al.*, 2009) with modifications. Briefly, 300 µL of the supernatant was mixed with 150 µL $AlCl_3$ (5% w/v in methanol) (Sigma Aldrich, São Paulo, Brazil), and the volume was made up to 3000 µL with methanol (Sigma Aldrich, São Paulo, Brazil). After incubation in the dark at room temperature (25 °C) for 30 min, absorbance was measured in a spectrophotometer (Evolution™ 300 e Thermo Scientific) at 425 nm. A quercetin (Sigma

Aldrich, São Paulo, Brazil) standard curve was prepared, and the results were expressed as milligrams of quercetin equivalents (QE) per litre of HEGP (mg QE/L).

The experimental isoproteic and isocaloric diets were based on corn and soybean meal according to Rostagno *et al.* (2017) for intermediate-performance male broilers (Table 1). The HEGP was added to the diets by replacing kaolin and by mixing it into the soybean oil. The basal diet was formulated with Super Crac® 6.2 Premium software. The birds had ad libitum access to water and feed in the mash form in both periods (1 - 21 and 22 - 42 days).

Table 1 Feed ingredients and nutrient contents of basal diet

Ingredients	g/kg	Nutrients	
Corn	639.00	Metabolizable energy, MJ/kg	13.25
Soybean meal, 45% crude protein	279.98	Crude protein, g/kg	180.00
Soybean oil	41.59	Calcium, g/kg	7.17
Monocalcium phosphate	13.14	Available phosphorus, g/kg	3.49
Limestone	9.08	Sodium, g/kg	2.04
NaCl	4.84	Digestible lysine, g/kg	10.88
DL-methionine, 98%	3.09	Digestible methionine + cysteine, g/kg	8.05
L-lysine sulphate, 50.7%	5.03	Digestible threonine, g/kg	7.18
L-threonine, 98%	1.15	Digestible tryptophan, g/kg	1.98
Vitamin supplement[1]	1.50		
Mineral supplement[2]	0.50		
Kaolin[3]	0.50		
Choline chloride	0.60		

[1](per kg of the diet) vitamin A: 16.500 UI, vitamin D_3: 60.000 UI, vitamin E: 825 UI, vitamin K_3: 4.5 mg, vitamin B_1: 3.45 mg, vitamin B_2: 10.5 mg, vitamin B_6: 6 mg, vitamin B_{12}: 37.5 mcg, pantothenic acid: 18 mg, nicotinic acid: 90 mg, folic acid: 3 mg, biotin: 0.375 mg, selenium: 0.45 mg
[2]iron: 150 mg, copper: 30 mg, manganese: 195 mg, zinc: 195 mg, iodine: 3 mg
[3]The hydroethanolic extract of guavira peel replaced kaolin.

At 21 and 42 days old, the birds were weighed, as well as the leftover feed at 42 days to calculate average feed intake (AFI), weight gain (WG), and feed conversion ratio (FCR). Mortality was recorded daily for later correction of data on AFI and FCR (Sakomura & Rostagno, 2016). The mortality rate was considered normal for this facility, being less than 2% for all groups.

At 42 days old, two representative birds per experimental unit (mean weight ± 5%) were selected to determine yields of carcass and special cuts. The birds were weighed individually, slaughtered by electronarcosis (CONCEA normative resolution no. 37 of 15 February 2018) and then bled, plucked, and eviscerated. The carcass yield was calculated as the proportion of eviscerated carcass weight (without head, feet, neck, and abdominal fat) to live weight. The yields of cuts (breast, legs, and wings) were calculated relative to the eviscerated carcass weight. Abdominal fat was regarded as all adipose tissue around the cloaca and attached to the gizzard. Concomitantly, the right breast muscle and thigh were used for pH and meat colour evaluation. Afterwards, the cooking loss (CL), water-holding capacity (WHC), and shear force (SF) were determined using the breast muscle. The left thigh was used to determine lipid oxidation (TBARS).

Meat colour and pH were determined at 15 min and again at 24 hours post tmortem. pH determination was performed directly on the breast muscle *(Pectoralis major)* and thigh muscle, using a 250 Testo® pH meter (Testo Argentina S.A., Buenos Aires, Argentina). Meat colour was determined with a CR-400 Konica Minolta colorimeter. CIELab values of L^* (lightness - from 0 = black to 100 = white), a^* (redness = $+a^*$; greenness = $-a^*$), and b^* (yellowness = $+b^*$; blueness = $-b^*$) were evaluated in three regions of the upper, middle, and lower inner breast and inner thigh (Honikel, 1998).

Water-holding capacity was analysed according to Nakamura and Katok (1985). One-gram samples of raw breast muscle were wrapped in filter paper and centrifuged (Centrifuge Kasvi K14-4000, Kasvi, São Paulo, BR) at 1500 rpm for 4 min. The samples were then weighed and oven-dried at 70 °C for 12 hours.

Then the dried samples were weighed again to determine the WHC as a percentage (weight of the meat sample after centrifugation - weight of the sample after drying/initial weight of the raw sample × 100).

Breast fillets were weighed, wrapped in aluminium foil, and kept on an electrically heated plate at 180 ºC until the internal temperature of the breast reached 80 ºC to determine CL. The fillets were weighed after cooling to room temperature to obtain the post-cooking weight (Honikel, 1998).

The SF analysis was performed with the same steaks. The samples were trimmed, cut into three pieces (1.0 × 1.0 × 1.3 cm), and sheared perpendicular to the muscle fibre orientation with a TA.XT2i texturometer (Stable Micro Systems, Jarinu, Brazil) coupled to a calibrated 29 Warner–Bratzler SF mechanical probe, with 5-kg capacity and crosshead speed set at 20 cm/min (Fronning & Uijttenboogaart, 1988), providing the measurement of the SF in kilogramme force (kgf/cm^2).

The determination of TBARS in the thigh meat was performed immediately after slaughter (day 0) and after storage for 7, 30, and 60 days at -20 °C. The analyses were performed according Vince (1970) and Sorensen and Jorgensen (1996). The aldehydes were extracted by mixing 10 mL of trichloroacetic acid solution (7.5%) (Inlab, São Paulo, Brazil) and BHT (0.2%) (Sigma Aldrich, São Paulo, Brazil) with 2.5 g sample. The resultant supernatant was filtered through qualitative filter paper, and then 3 mL of thiobarbituric acid solution (0.02 M) (Sigma Aldrich, São Paulo, Brazil) was added to 3 mL of the solution (1:1 v/v). This solution was kept in a water bath at 80 °C for 40 min, and absorbance was measured using a spectrophotometer (Evolution™ 300 e Thermo Scientific) at 538 nm. A calibration curve was plotted with 1,1,3,3-tetraethoxypropane (Sigma Aldrich, São Paulo, Brazil), and the results were expressed as malonaldehyde (MDA)/mg meat.

The results were submitted to analysis of variance (ANOVA) and polynomial regression. Dunnett's test (P >0.05) was used to compare the control (0 mg/kg HEGP) with the treatment groups (100, 200, 300, 400, and 500 mg/kg HEGP) using SAS® university edition statistical software (2017) (SAS Inst., Inc., Cary, NC, USA). Data on TBARS were subjected to ANOVA using the PROC GLM procedure of SAS (2017) to test the individual effects of HEGP inclusion levels and storage time, and their interaction. Significant interactions (P <0.05) were revealed by applying polynomial regression equations to evaluate the HEGP inclusion levels at 0, 7, 30, and 60 days of storage.

Results and Discussion

The levels of total polyphenols and flavonoids in the HEGP were 47.80 mg GAE/L and 10.99 mg QE/L, respectively.

There was a quadratic effect of dietary HEGP inclusion (P <0.05) on WG and FCR of broilers from 21 to 42 days old. According to the adjusted equations, HEGP levels of 314 and 219 mg/kg resulted in greater WG and better FCR, respectively. The AFI increased (P <0.05) with rising extract levels in diets. Moreover, broilers fed diets containing 200 to 500 mg/kg of HEGP had greater WG compared with animals fed with extract-free diet, and animals fed diets containing 100, 200, 300, and 500 mg/kg of HEGP had greater AFI compared with control broilers (Table 2).

There was no effect of HEGP inclusion (P >0.05) on carcass yield, the yield of special cuts (wings, legs, and breast) and abdominal fat percentage. No significant difference (P >0.05) was observed for all yield traits when comparing each HEGP inclusion level with the control treatment (Table 3).

There was a quadratic effect of HEGP inclusion (P <0.05) on WHC, with the greatest WHC calculated at 263 mg/kg extract. The CL increased (P <0.05) with increasing extract levels in diets. No significant difference (P >0.05) was observed for WHC and CL when comparing each HEGP inclusion level with the control treatment. The SF was not affected (P >0.05) by the inclusion of HEGP in diets (Table 4).

There was a quadratic effect of HEGP inclusion (P <0.05) on a^* (redness/greenness) of thigh meat at 15 min post-mortem, with the lowest redness calculated at the level of 270 mg/kg HEGP. No significant difference (P >0.05) was observed for pH and meat colour traits when each HEGP inclusion level was compared with the control (Table 5).

Table 2 Performance of broiler chickens from 21 to 42 days old fed diets containing various levels of hydroethanolic extract of guavira peel

HEGP Inclusion, mg/kg	WG (g)	AFI (g)	FCR
0	1935	3511	1.817
100	2042	3702*	1.813
200	2119*	3712*	1.751
300	2124*	3789*	1.785
400	2085*	3675	1.762
500	2075*	3772*	1.818
Standard error	8.70	10.75	0.172
P-value	0.012	0.018	0.132
P-value: regression on HEGP level	0.003(Q)	0.015(L)	0.029(Q)
Equations	R^2	HEGP	Result
WG = 1942.10764 + 1.175963x - 0.0000187x^2	0.95	314 mg/kg	2126
FI = 3600,651067 + 0.37117x	0.49		
FCR = 1.8267 - 0.000438x + 0.000001x^2	0.61	219 mg/kg	1.769

HEGP: hydroethanolic extract of guavira peel, WG: weight gain, AFI: average feed intake, FCR: feed conversion ratio, L: linear; Q: quadratic
*significant difference (*P* =0.05) between control and experimental group

Table 3 Carcass, portion yield (%), and abdominal fat (%) of broiler chickens at 42 days old fed diets containing levels of hydroethanolic extract of guavira peel

HEGP Inclusion, mg/kg	Carcass	Wings	Thigh and leg	Breast	Abdominal fat
0	69.80	10.38	32.66	41.41	2.40
100	69.67	10.17	33.52	40.13	2.68
200	71.20	10.17	33.03	41.19	2.51
300	70.02	10.28	33.13	40.99	2.76
400	70.77	10.04	32.43	41.19	2.41
500	70.13	10.05	32.14	41.83	2.73
SEM	0.771	0.362	0.711	0.752	0.404
P-value	0.697	0.528	0.343	0.565	0.718
P-value regression on HEGP level	0.550	0.800	0.734	0.595	0.506

HEGP: hydroethanolic extract of guavira peel

Table 4 Water-holding capacity, cooking loss and shear force values of breast meat of broiler chickens at 42 days old fed diets containing levels of hydroetahnolic extract of guavira peel

HEGP inclusion, mg/kg	WHC (%)	CL (%)	SF (kgf/cm^2)
0	75.46	27.09	3.03
100	78.70	27.19	3.04
200	82.51	26.95	3.24
300	78.51	29.00	3.02
400	78.17	29.74	2.79
500	76.09	30.36	2.61
SEM	1.577	1.225	0.467
P-value	0.256	0.230	0.904
P regression	0.032(Q)	0.021(L)	0.318
Equation	R^2	HEGP	Result
WHC = 76,8458 + 0,0368x - 0,00007x^2	0.73	262.86 mg/kg	81.68
CL = 26,576 + 0,0066x	0.80		

HEGP: hydroethanolic extract of guavira peel, WHC: water-holding capacity, CL: cooking loss, SF: shear force

Table 5 Colour and pH values of breast and thigh meat of broilers, at 42 days old, fed diets containing levels of hydroethanolic extract of guavira peel excised at 15 min and 24 hours post-mortem

	pH				L*				a*			
	15 min		24 h		15 min		24 h		15 min		24 h	
HEGP inclusion, mg/kg	Breast	Thigh	Breast	Thigh	Breast	Thigh	Breast	Thigh	Breast	Thigh	Breast	Thigh
0	6.07	5.88	5.64	5.83	45.91	49.35	52.44	51.30	3.438	4.99	3.43	5.21
100	6.26	5.99	5.50	5.63	46.22	51.21	55.21	52.67	2.703	3.33	1.77	3.23
200	6.08	5.91	5.70	5.65	44.74	50.07	52.29	51.92	3.553	3.72	2.62	4.77
300	6.18	5.94	5.69	5.74	45.72	50.32	53.63	52.38	3.053	3.58	2.27	3.29
400	6.14	5.98	5.57	5.74	48.84	48.87	52.32	50.82	3.614	3.56	4.26	3.28
500	6.16	5.96	5.50	5.78	47.97	48.77	53.90	50.84	3.712	4.55	3.12	4.67
SEM	0.265	0.177	0.299	0.272	1.239	0.971	1.082	0.888	0.624	0.811	0.94	0.095
P-value	0.553	0.671	0.071	0.123	0.215	0.274	0.473	0.627	0.392	0.116	0.023	0.107
P-value regression on HEGP level	0.313	0.505	0.930	0.226	0.410	0.488	0.224	0.728	0.284	0.012(L)	0.281	0.129
Equation						R^2				HEGP		
a* Thigh 15 min= 4.7668-0.0108x+0.00002x²						0.76				270 mg/kg		

L*: Lightness, a*: redness, b*: yellowness, L: linear

An interaction (P <0.05) between inclusion levels of extract and storage period was observed for TBARS. Overall, the sliced data show that within each storage time there was only a quadratic influence (P <0.05) of the dietary inclusion levels of the extract on the malonaldehyde (MDA) contents at 30-day storage, with the highest MDA value being calculated at 218 mg/kg HEGP (Table 6).

Table 6 Values of thiobarbituric acid reactive substances (mg malonaldehyde/kg meat) of thigh meat of broiler chickens at 42 days old fed diets containing levels of hydroetahnolic extract of guavira peel and evaluated at various storage times (0, 7, 30 and 60 days)

HEGP inclusion, mg/kg	Storage times					
	0	7	30	60	Average	SE
0	0.264	0.468	0.225	0.110	0.267	0.0037
100	0.254	0.430	0.236	0.109	0.257	0.0038
200	0.262	0.433	0.237	0.109	0.260	0.0039
300	0.266	0.447	0.241	0.109	0.266	0.0037
400	0.269	0.444	0.234	0.108	0.264	0.0037
500	0.287	0.444	0.207	0.108	0.262	0.0038
Average	0.267	0.444	0.230	0.109		
SEM	0.0034	0.0029	0.0029	0.0029		
P-values	HEGP level = 0.432, Time < 0.001, Interaction = 0.006					
Regression of TBARS at 30 days (y) on HEGP level (x)				R^2	HEGP	Result
$y = 0.223 + 0.00172x - 0.000001x^2$				0.91	218 mg/kg	0.230

TBARS: thiobarbituric acid reactive substances, HEGP: hydroetahnolic extract of guavira peel

Bioactive compounds such as polyphenols and flavonoids are known for their antioxidant, anti-inflammatory, and antimicrobial effects (Viecili *et al.*, 2014; Capeletto *et al.*, 2016). Thus, the levels of total polyphenols and flavonoids in HEGP show the potential of this extract as a natural additive (Auharek *et al.*, 2013).

Bioactive compounds may stimulate the digestive process, thus providing higher feed digestibility and nutrient absorption (Ader *et al.*, 2000; Perić *et al.*, 2009). Recent studies identified correlations between natural substances and modulation of growth performance, intestinal microbiota and intestinal quality (Fascina *et al.*, 2017; Zhu *et al.*, 2019). The anti-inflammatory and immunomodulatory properties of these compounds must also be considered. Natural extracts could reduce the gene expression of cytokines involved in the inflammatory response at intestinal level (Herrero-Encinas *et al.*, 2020), leading to improved animal performance.

In terms of performance data, a dose-dependent effect was observed in this study, which was associated with the compounds remaining in the intestinal lumen, with local action, since the absorption rate of phenolic compounds is considered low in chickens (Brenes *et al.*, 2008). Phenolic compounds may reduce glucose absorption by interfering with the expression of the GLUT2 glucose transport protein (Kwon *et al.*, 2007), which would impair animal performance. In this context, Eyng *et al.* (2014) observed a linear reduction in saccharase activity in the duodenal mucosa of seven-day-old broilers fed diets containing up to 5000 mg/kg of ethanolic propolis extract. The authors reported a reduction in productive parameters, correlating the capability of phenolic compounds with alteration of carbohydrate metabolism and interference with monosaccharide absorption. Thus, attention should be paid to the dietary inclusion level of natural extracts in broiler diets.

In addition, the presence of these substances was correlated with improved meat quality owing to their antioxidant action (Starčević *et al.*, 2015), which influences sensory characteristics.

In this study a reduction on thigh muscle redness (a^*) at 15 min post-mortem was observed in response to inclusion of HEGP. Myoglobin is responsible for meat colour and contains an iron molecule in the ferrous state (Fe II). When exposed to oxygen, Fe II is transformed into Fe III, a state named metmyoglobin, which is accompanied by changes in meat colour and quality (Papuc *et al.*, 2017). Inai *et al.* (2014) demonstrated the in vitro ability of polyphenols to control the redox state of myoglobin. However, the presumably low bioavailability of these substances in humans and animals may have impaired their

antioxidant action. Thus, the protective action of polyphenols may be reduced when added to feeds because it is dependent on the processes of digestion, absorption, metabolism, and tissue deposition (Surai *et al.*, 2014). Despite the influence at 15 min post-mortem, the colour of meat samples at 24 hours post-mortem was considered normal (Barbut, 1997) as the dietary inclusion of HEGP did not affect the quality of the final product.

Meat tenderness is associated directly with WHC and CL, and consumers regard these attributes as fundamental traits. An increased muscle pH was correlated with enhanced WHC and meat tenderness, resulting in better quality meat (Selim *et al.*, 2013). Although no influence of extract levels on pH and SF was observed, there was an improvement in WHC. The active compounds in extracts can reduce protein oxidation and modulate the action of the enzymatic complex formed by calpain (Guttmann & Johnson, 1998; Huff-Lonergan & Lonergan, 2005) through their antioxidant action. These factors are related directly to WHC, affecting the quality of the final product.

Oxidation of meat lipid and protein components, which generates free radicals, is responsible for the quality deficiency owing to the loss of colour pigmentation, reduction in shelf life, the formation of toxic compounds, and deficit of nutrients (Contini *et al.*, 2014; Falowo *et al.*, 2014). Polyphenols are potent antioxidant agents that are capable of scavenging free radicals, largely by acting as metal ion-chelating molecules that catalyse oxidation reactions, delaying this process. In this context, phenolic groups donate hydrogen ions and prevent free radical generation, with the formation of stable end products (Zhang *et al.*, 2016).

Despite evidence for the antioxidant property of phenolic compounds and their ability to be incorporated in meat products (Saleh *et al.*, 2017), no reduction in meat lipid oxidation was observed when HEGP was included in the diets, rejecting the hypothesis that the inclusion of guavira extract in broiler diets could improve meat quality. The absorption of flavonoids depends on their physico-chemical properties, and the metabolization of these components involves processes of hydrolyzation, glucuronidation, sulfation, and methylation, which seems similar to mammal uptake (Rupasinghe *et al*., 2010; Fotakis *et al*., 2017). The mechanism of action of extracts may be related to the absorption and deposition of the phenolic compounds because, when absorbed, they are quickly metabolized, and a large part is eliminated by the kidneys (Lee *et al.* 2004). Kamboh *et al.* (2019) emphasized that the unabsorbed flavonoids from the small intestine, and some of the absorbed compounds, are secreted with bile, being after being degraded in the large intestine by the microbiota. Rupasinghe *et al.* (2010) observed that quercetin glucosides, which belong to a subgroup of flavonoids, can be absorbed and metabolized by broiler chickens. However, the absorption of the component was not sufficient to elevate the antioxidant capacity of the tissues (Rupasinghe *et al.*, 2010). In this sense, the direct use of these compounds in raw meat reduced lipid oxidation, increasing the shelf life of meat products (Krishnan *et al.*, 2014; Nikmaram *et al.*, 2018). Nunes *et al.* (2019) observed a reduction in lipid oxidation in eggs of laying quails fed diets containing dehydrated bocaiúva (*Acrocomia aculeata*) pulp, emphasizing that the antioxidants added to feed were preferentially incorporated in eggs rather than meat because the biological priority is to ensure the development of the embryo.

Conclusions

The dietary inclusion of 219 mg/kg of hydroethanolic extract of guavira fruit peel resulted in better FCR for broilers in the finishing phase but without changing the carcass characteristics. However, in terms of meat quality, it was not possible to verify the antioxidant capacity of this extract.

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Authors' Contributions

CE and RVN designed the study and were the supervisors. EJSA and CACC were in charge of extract production. MLL worked on the project, and laboratory analysis. TLK and APGCC participated in management and discussion of the results, statistical analysis and writing, and corrected the manuscript.

Conflict of Interest Declaration

The authors declare that they have no conflicts of interest relative to the content of this paper.

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