Partial corn replacement by glycerin and vegetable oils (cashew and castor) as alternative additive in the diets of crossbred bulls finished in a feedlot: Carcass characteristics and *Longissimus lumborum* muscle evaluation

Substituição parcial do milho por glicerina e óleos vegetais (caju e mamona) como aditivo alternativo em dietas de touros mestiços terminados em confinamento: características de carcaça e avaliação do músculo Longissimus lumborum

Sustitución parcial del maíz por glicerina y aceites vegetales (anacardo y ricino) como aditivo en dietas alternativas de toros mestizos terminados en confinamiento: características de la canal y evaluación del músculo Longissimus lumborum

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Abstract
This work was carried out to study the effects of corn grain replacement with glycerin (812 g of glycerol per kg/DM) and vegetable oils (cashew and castor) on the carcass characteristics and meat quality of Purunã bulls finished in a feedlot. A total of 32 Purunã bulls (¼ Aberdeen Angus + ¼ Caracu + ¼ Charolaise + ¼ Canchim) with a mean age of 12 ± 2.0 months and a mean body weight 206.1 ± 20.0 kg were distributed in a completely randomized design with four diets and eight replications per diet. The four experimental diets were as follows: CONT – basal diet; VOIL –basal diet and addition of vegetable oils (3 g/animal/day); GLYC – basal diet and addition of glycerin (20.1% glycerin on a DM basis); GLVO – basal diet and addition of glycerol (20.1% glycerin in DM basis) and vegetal oils (3 g/animal/day). The GLVO diet improved carcass conformation (+12.8%) in comparison with the CONT and GLYC diets. Likewise, fat thickness and proportion of fat in the carcass were higher in the GLVO group (+25.6% and +14.3%, respectively) versus the CONT group. Diets containing glycerin and vegetable oils increased hot (+5.0%) and cold (+5.1%) carcass weights, in comparison to the CONT diet. Diets containing vegetable oils (VOIL and GLVO) improved (+3.4%) carcass dressing relative to the CONT diet. Inclusion of glycerin and vegetable oils did not affect the *Longissimus* muscle area (68.0 cm^2), texture (4.24 points), marbling (6.68 points), or colour (3.51 points) at 24 h *post mortem*. Likewise, instrumental colour in terms of lightness (32.4), redness (13.9), yellowness (4.94), chroma (14.8), and angle hue (19.1) at 24 h *post mortem* were unaffected by diet. Finally, the diet did not affect moisture (26.6%), ash (1.1%), crude protein (21.6%), total lipids (2.2%), WBS (3.1 kgf), TBARS (0.28 mg of MDA per kg of meat) or calories (225.5 kcal/100 g of meat). Thus, up to 20% glycerin on a DM basis and vegetable oils from cashew and castor could be added to the diet of bulls finished in a feedlot for 250 days and fed with a high-density energy diet.
Keywords: *Anacardium* acid; Bio-fuels; Energy; Glycerol; Meat quality; Ricinoleic acid.

Resumo
Este trabalho foi realizado para estudar os efeitos da substituição do grão de milho pela glicerina (812 g de glicerol por kg/MS) e óleos vegetais (caju e mamona) sobre as características de carcaça e qualidade da carne de bovinos não castrados terminados em confinamento. Um total de 32 bovinos Purunã (¼ Aberdeen Angus + ¼ Caracu + ¼ Charolaise + ¼ Canchim) com idade média de 12 ± 2,0 meses e peso corporal médio de 206,1 ± 20,0 kg foram distribuídos em delineamento inteiramente casualizado com quatro dietas e oito repetições por dieta. As quatro dietas experimentais foram: CONT – dieta basal; VOIL – dieta basal e adição de óleos vegetais (3 g/animal/dia); GLIC – dieta basal e adição de glicerina (20,1% de glicerina na MS); GLVO – dieta basal e adição de glicerol (20,1% glicerina na base MS) e óleos vegetais (3 g/animal/dia). A dieta GLVO melhorou a conformação da carcaça (+12,8%) em comparação com as dietas CONT e GLYC. Da mesma forma, a espessura de gordura e a proporção de gordura na carcaça foram maiores no grupo GLVO (+25,6% e +14,3%, respectivamente) em relação ao grupo CONT. As dietas contendo glicerina e óleos vegetais aumentaram os pesos de carcaça quente (+5,0%) e fria (+5,1%), em comparação com a dieta CONT. As dietas contendo óleos vegetais (VOIL e GLVO) melhoraram (+3,4%) o rendimento de carcaça em relação à dieta CONT. A inclusão de glicerina e óleos vegetais não afetou a área do músculo Longissimus (68,0 cm2), textura (4,24 pontos), marmoreio (6,68 pontos) ou cor (3,51 pontos) 24 h post mortem. Da mesma forma, a cor instrumental em termos de luminosidade (32,4), vermelho (13,9), amarelo (4,94), croma (14,8) e tonalidade (19,1) 24 h post mortem não foi afetada pela dieta. Por fim, a dieta não afetou a umidade (26,6%), cinzas (1,1%), proteína bruta (21,6%), lipídios totais (2,2%), WBS (3,1 kgf), TBARS (0,28 mg de MDA por kg de carne) ou calorias (225,5 kcal/100 g) da carne. Assim, até 20% de glicerina na MS e óleos vegetais de caju e mamona poderiam ser adicionados à dieta de bovinos terminados em confinamento por 250 dias e alimentados com dieta de alta densidade energética.
Palavras-chave: Ácido Anacárdio; Biocombustíveis; Energia; Glicerol; Qualidade da carne; Ácido ricinoleico.

Resumen
Este trabajo se llevó a cabo para estudiar los efectos de la sustitución del grano de maíz por glicerina (812 g de glicerol por kg/MS) y aceites vegetales (anacardo y ricino) sobre las características de la canal y la calidad de la carne de ganado bovino de engorde no castrados. Un total de 32 bovinos Purunã (¼ Aberdeen Angus + ¼ Caracu + ¼ Charolaise + ¼ Canchim) con edad media de 12 ± 2,0 meses y peso corporal medio de 206,1 ± 20,0 kg fueron distribuidos en un diseño completamente al azar con cuatro dietas y ocho repeticiones por dieta. Las cuatro dietas experimentales fueron: CONT – dieta basal; VOIL – dieta basal y adición de aceites vegetales (3 g/animal/día); GLIC – dieta basal y adición de glicerina (20,1% glicerina en DM); GLVO – dieta basal y adición de glicerol (20,1% de glicerina en base a MS) y aceites vegetales (3 g/animal/día). La dieta GLVO mejoró la conformación de la canal (+12,8%) en comparación con las dietas CONT y GLYC. Asimismo, el espesor de grasa y la proporción de grasa en la canal fueron mayores en el grupo GLVO (+25,6% y +14,3%, respectivamente) en comparación con el grupo CONT. Las dietas que contenían glicerina y aceites vegetales aumentaron el peso de la canal caliente (+5,0%) y fría (+5,1%) en comparación con la dieta CONT. Las dietas que contenían aceites vegetales (VOIL y GLVO) mejoraron (+3,4%) el rendimiento de la canal en comparación con la dieta CONT. La inclusión de glicerina y aceites vegetales no afectó la área del músculo Longissimus (68,0 cm^2), la textura (4,24 puntos), el veteado (6,68 puntos) ni el color (3,51 puntos) 24 h post mortem. De manera similar, el color instrumental en términos de luminosidad (32,4), rojo (13,9), amarillo (4,94), croma (14,8) y tono de ángulo (19,1) a las 24 h post mortem no se vio afectado por la dieta. Finalmente, la dieta no afectó la humedad (26,6%), cenizas (1,1%), proteína cruda (21,6%), lípidos totales (2,2%), WBS (3,1 kgf), TBARS (0,28 mg MDA por kg de carne)

o calorías (225,5 kcal/100 g) de la carne. Así, hasta un 20% de glicerina en MS y aceites vegetales de marañón y ricino podría incorporarse a la dieta de bovinos terminados en cebaderos durante 250 días y alimentados con una dieta de alta densidad energética.
Palabras clave: Ácido de anacardium; Biocombustibles; Energía; Glicerol; Calidad de la carne; Ácido ricinoleico.

1. Introduction

Brazil is a significant producer and exporter of beef meat (FAPRI, 2021). However, in recent years beef production has been operating within a narrow profit margin due to high food costs. Thus, co-products from the bio-diesel industry, such as glycerin, have been studied as an alternative energy source to replace corn grains (Cruz et al., 2014; Eiras et al., 2014; Eiras et al., 2014; Françozo et al., 2013; Prado et al., 2015; Zawadzki et al., 2021). The biodiesel industry has expanded rapidly in recent years and increased the availability of glycerin (FAPRI, 2021). In 2018, the biodiesel industry produced more than 2.5 billion L of glycerin worldwide, and the Brazilian market produced about 3.0 million L (FAPRI, 2021). Glycerol contained in crude glycerin improves the synthesis of glucose in the liver (Chung et al., 2007), assists in the process of gluconeogenesis (Krehbiel, 2008) provides energy for cellular metabolism (Goff and Horst, 2001) and improves fat deposition (Cruz et al., 2014). In ruminal metabolism, glycerol is used by microorganisms for the synthesis of volatile fatty acids and increases propionic acid (Abo El-Nor et al., 2010). Likewise, which directly assists in gluconeogenesis, propionate from glycerol is interesting on animal performance, so that most propionate is used for gluconeogenesis in the liver (Bradford and Allen, 2007).

Compounds that can be used to replace the synthetic molecules currently used to improve ruminal modulation are being investigated worldwide (Monteschio et al., 2017; Rivaroli et al., 2016; Valero et al., 2011, 2014; Zawadzki et al., 2011a; Zawadzki et al., 2011b). Vegetable oils contain compounds that have antimicrobial activity against Gram-positive and Gram-negative bacteria (Himejima and Kubo, 1991; Zarai et al., 2012). This antimicrobial activity can be attributed to the interaction between such compounds and the bacterial cell membrane, affecting ion gradients, electron transport, protein translocation, phosphorylation and enzyme-dependent reactions (Benchaar et al., 2008; Ultee et al., 2000). Castor oil contains a high percentage of *ricinoleic* acid, which has antimicrobial properties (Shin et al., 2004) and their derivatives analogous (Narasimhan et al., 2004). Meanwhile cashew contains a high percentage of *anacardic* acid and a lower percentage of *cardol* and *cardanol* (Nagabhushana and Ravindranath, 1995). Compounds in cashew have antibacterial (Himejima and Kubo, 1991; Muroi et al., 1993), antioxidant and anti-parasitic properties (Kubo et al., 1999).

Synergisms between compounds extracted from different products may increase their antibacterial activity (Ultee et al., 2000). Thus, the synergism between compounds in cashew (*anacardic*, *cardol*, and *cardinal* acids) and castor oils (*ricinoleic* acid) may improve their antimicrobial effect on microorganisms. The association of the compounds extracted from cashew and castor oils, similar effects were observed in comparison the sodium monensin (Cruz et al., 2014; Valero et al., 2015, 2014, 2011; Zawadzki et al., 2021). However, the effects on carcass characteristics and meat quality remain unclear.

Thus, the aim of this study was to evaluate the effects of the replacement of corn grain with glycerin and vegetable oils from castor and cashew in the diet on the carcass characteristics and meat quality of Purunã bulls finished in a feedlot.

2. Material and Methods

2.1 Ethic committee, local, animals, diets and experimental design

The present experiment was approved by the Department of Animal Science of the State University of Maringá and performed at the Experimental Farm of the Agronomic Institute of Paraná, Ponta Grossa city, Paraná state, Brazil south, and followed the guidelines of biomedical research with animals (CIOMS/OMS, 1985) (Protocol – 550.891-2010-2/CNPq).

A total of 32 Purunã bulls (¼ Aberdeen Angus + ¼ Caracu + ¼ Charolaise + ¼ Canchim) with a mean age of 12 ± 2.0 months and a mean body weight 206.1 ± 20.0 kg were distributed in a completely randomized design with four diets and eight replications per diet. The bulls were selected at birth and kept in a pasture of *Hemarthria altissima* from birth until they were allocated into individual pens (8 m^2 for each animal) in a feedlot system.

The four experimental diets were as follows: CONT – basal diet; VOIL –basal diet and addition of vegetable oils (3 g/animal/day); GLYC – basal diet and addition of glycerin (20.1% glycerin on a DM basis); GLVO – basal diet and addition of glycerol (20.1% glycerin in DM basis) and vegetal oils (3 g/animal/day).

An adaptation period of at least 21 days was allowed before the start of the experiment, during which the bulls were fed corn silage and concentrate in separate troughs (40:60 ratio, respectively). The concentrate contained soybean meal, corn grain, and mineral salts. The animals had free access to the formulated diet to allow a weight gain of 1.0 kg/day and to meet the requirements for fattening beef cattle.

Half of the daily ration was offered in the morning at 08:00 h and the other half at 16:00 h. The concentrate intake was fixed at 1.4% BW and was adjusted every 28 days. Samples of corn silage and concentrate were collected twice weekly to estimate the DM%. The bulls were fed concentrate and corn silage in separate troughs, with *ad libitum* corn silage intake.

The glycerin used in this study was produced in a soy-diesel facility and its chemical composition was determined at the Institute of Technology of Paraná. It contained (g/kg, as-fed) 23.2 water, 4.76 ash, 812.0 glycerol, 11.63 Na and (mg/kg, as-fed) 3.3 methanol, 79.1 K, 35.8 Cl, 16.3 Mg, 239.8 P, and 3.65 Mcal/kg of crude energy. It was used as an energetic ingredient; therefore, to obtain four isoenergetic diets, the glycerol level was counterbalanced, mainly by reducing the corn grain content (Table 1).

Table 1. Ingredients and percentage of the basal diets.

Ingredients, %	Diets, g/kg of DM			
	CONT[1]	VOIL[2]	GLYC[3]	GLVO[4]
Corn silage	420	420	420	420
Soybean meal	103	103	150	150
Corn grain	477	447	227	197
Glycerin			203	203
Vegetable oils		30		30

[1]CONT – Without glycerin or vegetable oils. [2]VOIL – Vegetable oils. [3]GlLYC – Glycerin. [4]GLVO – Glycerin + vegetable oils.
Fonte: Authors.

The vegetable oils used in this study contain ricinoleic, anacardic, cardanol, and cardol acids. Ricinoleic acid was obtained from castor oil (extracted from castor seed) and anacardic, cardanol, and cardol acids were obtained from cashew oil. Vermiculite was used to solidify the vegetable oils. The vegetable oils were mixed by Oligo Basics Agroindustrial Ltda. It was used 3 g/animal/day. All diets were formulated to be isonitrogenous and isoenergetic (Table 2). Water and mineral salts were given *ad libitum*. The mineral salts contained (g/kg; as-fed) 150 Ca, 88 P, 0.08 Co, 1.45 Cu, 10 S, 1.0 Fe, 0.88 F, 0.06 I, 10 Mg, 1.1 Mn, 0.02 Se, 120 Na and 3.40 Zn.

Table 2. Chemical composition of the basal diets[1].

Ingredients	DM[2]	%/DM													
		OM[3]	Ash	CP[4]	EE[5]	TFC[6]	NFC[7]	NDF[8]	ADF[9]	CF[10]	NNE[11]	TDN[12]	CE[13*]	DE[14*]	ME[15*]
Corn silage	28.98	99.03	0.97	5.79	2.07	91.16	48.30	42.87	21.60	17.28	73.89	64.72	3.96	2.85	2.34
Corn grains	89.47	99.01	0.99	7.43	4.66	86.93	69.62	17.31	2.28	1.82	85.11	83.00	3.97	3.66	3.00
Soybean meal	91.62	99.06	0.94	45.04	2.63	51.38	33.31	18.07	8.23	6.58	44.80	86.46	4.29	3.81	3.13
Glycerol	94.22	95.24	4.76	0.07	0.12	95.05	95.05	-	-	-	95.05	80.61	3.65	3.55	2.91
Total diet															
CONT[16]	52.10	99.02	0.98	10.63	3.36	85.03	56.91	28.12	11.01	8.81	76.22	75.68	3.99	3.34	2.74
VOIL[17]	52.10	99.02	0.98	10.63	3.36	85.03	56.91	28.12	11.01	8.81	76.22	75.68	3.99	3.34	2.74
GLYC[18]	51.81	98.24	1.76	10.66	2.33	85.25	60.69	24.55	10.78	8.62	76.62	75.33	3.95	3.32	2.72
GLVO[19]	51.81	98.24	1.76	10.66	2.33	85.25	60.69	24.55	10.78	8.62	76.62	75.33	3.95	3.32	2.72

[1]Analyses conducted by Chemical Laboratory of State University of Maringá. [2]Dry matter. [3]Orgnic matter. [4]Crude protein. [5]Ether extract. [6]Total fiber carbohydrate. [7]Non fiber carbohydrate. [8]Neutral detergent fiber. [9]Acid detergent fiber. [10]Crude fiber. [11]Non Nitrogenous extract. [12]Total digestible nutrients. [13]Crude energy. [14]Digestible energy.[15]Metabolizable energy. *Mcal/kg. [16]Without glycerin or vegetal oils. [17]Vegetal oils. [18]Glycerin. [19]Glycerin and vegetal oils. Fonte: Authors.

2.2 Animal performance

The young bulls were weighed at the start of the experiment and then every 28 days after fasting from solid food for 16 hours, until the end of the experiment (252 days), to determine animal performance.

2.3 Slaughter

The bulls were slaughtered at a commercial slaughterhouse 10 km from Ponta Grossa Research Farm, according to industrial practices in Brazil, when the bulls reached a final BW of 468.2 $\pm$ 31.5 kg at around 19 months old.

2.4 Carcass characteristics

After slaughter, the carcasses were labeled and chilled for 24 h at 4 ºC. After chilling, the righthand side of the carcass was used to determine the following quantitative characteristics.

Hot carcass weight (HCW) was determined before chilling. The hot carcass dressing (HCD) of each animal was defined as the ratio of hot carcass weight and live weight multiplied by 100. Cold carcass weight (CCW) was determined after 24 h of carcass chilling. The cold carcass dressing (CCD) was defined as the ratio of cold carcass weight and live weight multiplied by 100. Carcass conformation (CAC) was evaluated using a point scale in which the highest value indicated the best conformation; muscle development was calculated after the exclusion of thickness fat. Carcass conformation was rated as superior, very good, good, regular, poor, or inferior; ratings were further qualified as a plus, average, or minus. Carcass length (CAL) was evaluated by measurements taken from the anterior board of bone of the pubis to the far side of the first rib, using a ribbon or a measuring tape. Cushion thickness (CUT) was evaluated by measurements taken using a wooden compass with metallic edges, being the distance between the lateral face and the median face in the upper part of the cushion. The cushion comprises flat muscle (*Biceps femoris* muscle). Fat thickness (FAT) was determined using a caliper at three points, between the 12th and 13th ribs on the *Longissimus dorsi* muscle. The percentage of muscle (MUS), fat (FAT), and bone (BON) in these muscles was calculated separately from the *Longissimus lumborum* section, which corresponded to the 9th, 10^{th,} and 11th ribs, and each section was individually weighed according to Hankins & Howe (1946). Data were regressed to equations, following Hankins, 1946, to find the percentages of muscle (MUS), fat (FAT), and bone (BON). Rates corresponding to the 9th, 10^{th,} and 11th ribs were regressed as follows:

MUS = (15.56 + 0.81) M%;

FAT = (3.06 + 0.82) F%; and

BON = (4.30 + 0.61) B%.

2.5 Longissimus muscle characteristics

After chilling (24 h at 4 ºC), the righthand side of the carcass was used to determine the qualitative characteristics of the *Longissimus* muscle (LM). LM samples were taken by complete cross-section between the 12th and 13th ribs and were immediately taken to the laboratory. The covering fat was discarded and the remaining muscle was frozen at -20º C for later chemical analysis. Temperature and pH were evaluated by measurements made on the LM between the 12th and 13th ribs at 0 h (about 40 min after the slaughter in hot carcass) and 24 h (after carcass chilling at 4° C). Temperature and pH values were measured using a portable PCE-228M pH-meter equipped with a penetrating metal electrode probe. *Longissimus* muscle area (LMA) in the right-hand side of the carcass was measured after a transverse cut was made between the 12th and 13th ribs, using a compensating planimeter, which measures the areas of irregularly-shaped objects. Marbling (MAR) in the LM was evaluated subjectively between the 12th and 13th ribs, following a point scale ranging from very fine (highest value), fine, slightly coarse, coarse to very coarse (lowest value); ratings were qualified as a plus, average, and minus. Texture (TEX) was determined based on the size of the fascicle (muscle "grain" size) and was evaluated subjectively using a point scale, in the same way as that of marbling in the LM between the 12th and 13th ribs. Colour (COR) was analyzed 24 h after carcass chilling according to a point scale, which ranged from cherry-red (highest value), red, slightly dark red, dark red to dark (lowest value), 30 min after a transverse cut was made in the LM between the 12th and 13th ribs.

Instrumental COR (I_{COL}) was analyzed 24 h *post mortem*, 30 min after a transverse cut had been made in the LM between the 12th and 13th ribs. COR was evaluated using a Minolta CR-400 spectrophotometer (illuminant D65, observer angle 10°, Konica Minolta Holdings, Inc., Osaka, Japan) in the CIELAB space. COR coordinates expressed as L*, a*, and b* were recorded, where L* is the lightness of COR, the value of which ranges from 0 for black to 100 for white; a* is redness, the value of which ranges from (+a*) for red to (–a*) for green; and b* is yellowness, the value of which ranges from (+b*) for yellow to (–b*) for blue. The Chroma (C*) and hue angle (H*) indexes were calculated as $C^* = (a^{*2} + b^{*2})^{0.5}$ and $H^* = \tan^{-1} (b^*/a^*)*[360° / (2*3.14)]$ expressed in degrees.

The mechanical properties of the meats were determined using a texture analyzer (Stable Micro Systems TAXT Plus, Texture Technologies Corp., UK) with a 5.00 kg load cell and a Warner-Bratzler (WB). The analysis was performed according to the methodology proposed by the Meat Animal Research Center of the USA (Wheeler et al., 1997).

Samples for lipid oxidation analysis were individually packaged and frozen (-20 ºC) for 6 months before analysis. Lipid oxidation was determined using the procedure described by (Botsoglou et al., 1994). Thiobarbituric Acid Reacting Substances (TBARS) rates were calculated from a standard curve and expressed as mg malondialdehyde (MDA) per kg of meat.

2.6 Chemical composition

The chemical composition of *Longissimus* muscle was analyzed 2 months after sampling. The samples were defrosted at 4º C, ground, homogenized, and analyzed in triplicate. Meat moisture and ash contents were determined according to ISO-R-1442 (1997), and crude protein content was obtained following ISO-R-937 (1978). The energy contained in the meat was determined by analyzing the fresh samples (in duplicate) in a Parr® 6200 bomb calorimeter.

2.7 Statistical analysis

The experimental design was completely randomized, with four diets and eight replicates. All variables were submitted to a normality test using the Univariate procedure of SAS (Version 9.1.2, SAS Institute; Cary, NC, USA). The variables were tested for conformity to normal distributions using the Shapiro–Wilk test at W > 0.90. Data were analyzed using the MIXED procedure of SAS (Version 9.1.2, SAS Institute). The model statement contained the fixed effect of diet. Data were analyzed using bull as the random variable. Results are reported as least-squares means and were separated using PDIFF. Significance was set at P < 0.05.

3. Results

3.1 Carcass characteristics

The partial replacement of corn grain by glycerin and vegetable oils (cashew and castor) inclusion in the diets did not affect (P > 0.05) final body weight (FBW) and average daily gain (ADG) of Purunã bulls finished in feelot (Table 3). However, diets containing glycerin and vegetable oils (VOIL, GLYC and GLVO diets) increased (P < 0.01) hot carcass weight (HCW, +5.3%) and cold carcass weight (CCW, +5.2%) in comparison to CONT diet (Table 3). Diets containing vegetable oils (VOIL and GLVO) improved (+3.4%) the dressing cold carcass (DCC) in comparison to CONT diet (Table 3). Diet had no effect (P > 0.05) on chilling loss, carcass length, cushion thickness andr muscle and bone percentages (Table 3). However, the inclusion of vegetable oils and glycerin (VOIL and GLVO diets) improved carcass conformation (+12.8%) in comparison to CONT and GLYC diets; the effects of diets containing vegetable oils were similar (P < 0.001, Table 3). Likewise, the fat thickness was higher (P < 0.01) in the bulls from GLVO group (+25.6%) than in the CONT diet; the VOIL, GLYC and GLVO groups had similar values (P > 0.05, Table 3). Similarly, the carcass fat percentage was increased in the GLVO diet (+14.3%) in comparison to CONT diets; the values in the VOIL, GLYC, and GLVO deits were similar (P > 0.05; Table 3).

Table 3. Glycerin level replacing corn as energy source and vegetal oils as additive on carcass and *Longissimus dorsi* characteristics of Purunã bulls finished in feedlot.

Itens	Diets				STD[5]	P[6]
	CONT[1]	VOIL[2]	GLYC[3]	GLVO[4]		
Final body weight, kg	460.2	467.8	471.1	473.7	6.86	0.49
Average daily gain	1.02	1.02	1.06	1.05	8.86	0.49
Hot carcass weight, kg	259.3b	272.1a	269.9a	277.0a	0.07	0.01
Cold carcass weight, kg	255.4b	267.9a	265.9a	272.5a	4.78	0.01
Dressing cold carcass, %	56.2b	58.0a	57.2ab	58.4a	0.02	0.07
Chilling loss, %	1.54	1.58	1.48	1.65	0.04	0.57
Conformation, points	13.0b	13.8ab	12.8b	14.8a	0.01	0.01
Carcass length, cm	128.8	130.3	130.3	132.7	0.02	0.32
Cushion thickness, cm	25.5	26.1	25.4	25.3	0.02	0.20
Fat thickness, cm	3.45b	4.00ab	3.81ab	4.64a	0.01	0.03
Muscle, %	64.99	64.35	64.31	62.92	1.11	0.88
Fat, %	21.31b	22.22ab	21.47ab	23.31a	0.57	0.01
Bone, %	13.72	13.43	14.23	13.78	0.26	0.60

[1]Without glycerin or vegetal oils.[2]Vegetal oils.[3]Glycerin. [4]Glycerin and vegetal oils.[5]Standard error.[6]Probability of *Tukey*-test for treatment, n = 8 per treatment. Fonte: Authors.

3.2 Longissimus muscle (LM) characteristics

Diet did not alter (P > 0.05) the mean temperature and pH of the LM at slaughter (37.3º C and 6.95, respectively) or at 24 h *post mortem* (5.93º C and 5.76, respectively) (Table 4). Similarly, there was no effect of the diets on LM area (68.0 cm^2), texture (4.24 points), marbling (6.68 points) or subjective colour (3.51 points) (Table 4). Likewise, instrumental colour did not differ at 24 h *post mortem*. Lightness (32.4 points), redness (13.9 points), yellowness (4.94 points), Chroma (14.80) and angle hue (19.11) were similar among treatments (P > 0.05) (Table 4).

Table 4. Glycerin partial replacing corn as energy source and vegetal oils as additive on chemical composition in *Longissimus dorsi*, fat thickness and perirenal of Purunã bulls finished in feedlot.

Itens	Diets				STD[5]	P[6]
	CONT[1]	VOIL[2]	GLYC[3]	GLVO[4]		
Characteristics mensured after slaughter						
Temperature, °C	37.01	37.61	37.34	37.30	0.14	0.53
Ph	6.97	6.94	7.01	6.88	0.04	0.74
Characteristics mensured[7] at 24 h						
Temperature, °C	5.95	5.93	5.98	5.88	0.14	0.98
pH at 24 h	5.78	5.74	5.77	5.78	0.06	0.99
Longissimus area, cm^2	63.3	68.5	69.9	70.5	0,09	0.43
Texture, points	4.00	4.37	4.15	4.47	0.10	0.41
Marbling, points	6.00	6.87	6.75	7.12	0.47	0.86
Color, points	3.62	3.25	3.40	3.78	0.03	0.44
Color charactreistics[7]						
L*	33.50	31.11	32.46	32.89	1.14	0.48
a*	13.80	14.01	14.09	13.70	0.69	0.97
b*	5.17	4.78	5.11	4.72	0.49	0.92
C*	14.78	14.82	15.04	14.56	0.80	0.97
H*	20.07	18.60	19.30	18.48	1.05	0.84

[1]Without glycerin or vegetal oils. [2]Vegetal oils. [3]Glycerin. [4]Glycerin and vegetal oils. [5]Standard error.[6]Probability of *Tukey*-test for treatment, n = 8 per treatment. [7]About 40 min. after slaughter. Fonte: Authors.

3.3 Longissimus muscle (LM) chemical composition

Diet had no effect on chemical composition of the LM (Table 5). Mean moisture (26.6%), ash (1.0%), crude protein (21.6%), total lipids (2.2%), WBS (3.07 kgf), TBARS (0.28 mg malondialdehyde (MDA) per kg of meat) and calories (225.5 kcal/100g of meat) were similar (P > 0.05) in the *Longissimus thoracis* of bulls finished four diets (Table 5).

Table 5. Glycerin replacing corn as energy source and vegetal oils as additive on chemical composition in *Longissimus dorsi* and fat thickness of Purunã bulls finished in feedlot.

Itens	Diets				STD[5]	P[6]
	CONT[1]	VOIL[2]	GLYC[3]	GLVO[4]		
Moisture, %	25.4	25.6	25.9	25.6	0.21	0.89
Ash, %	1.04	1.02	1.09	1.07	0.01	0.33
Crude protein, %	21.9	21.6	21.4	21.7	0.15	0.73
Total lipids, %	2.03	2.28	2.25	2.30	0.22	0.89
Braztler shear force, kgf[9]	3.32	2.80	2.98	3.21	0.12	0.49
TBARS[7]	0.21	0.33	0.31	0.28	0.03	0.51
Calories, kcal/100g of meat	227.9	221.5	217.0	235.9	4.76	0.54

[1]Without glycerin or vegetal oils. [2]Vegetal oils. [3]Glycerin. [4]Glycerin and vegetal oils. [5]Standard error. [6]Probability of *Tukey*-test for treatment, n = 8 per treatment. [7]2-Thiobarbituric acid reacting substances (mg malonaldehyde (MDA) per kg of meat). [7]Waner-Braztler shear measured by kilograms of force (kgf). Fonte: Authors.

4. Discussion

4.1 Carcass characteristics

The experimental design examined the effect of corn grain partial replacement by glycerin as an energy source and vegetable oils as natural compounds on performance, carcass characteristics, and meat evaluation of young bulls finished in feedlot for 252 days. Diet did not affect final body weight and average daily gain. The performance of bulls finished in a feedlot with glycerin and vegetable oils in the diet were similar to those observed by other researchers using the similar foods (Cruz et al., 2014; Eiras et al., 2014; Eiras et al., 2014; Zawadzki et al., 2021). However, Valero et al., (2014) and Valero et al. (2016) observed better performance of bulls fed glycerin and vegetables oils inclusion in the diet. The final body weight value observed in this study was consistent with industrial practices in Brazil (Rotta et al., 2009).

On the other hand, hot and cold carcass weights increased by partial substitution of corn grains by glycerin and vegetables oils inclusion in the diets. Previous studies reported similar values in young bulls finished in a feedlot (Eiras et al., 2014; Françozo et al., 2013; Fugita et al., 2018). In contrast to the results of the present study, Françozo et al. (2013) and Cruz et al. (2014) reported no difference in hot carcass weight of young bulls in response to corn grain replacement by glycerin levels ranging from 6 to 18%.

Diets containing glycerin and vegetable oils (VOIL, GLYC, and GLVO diets) improved cold carcass dressing, whereas the inclusion of vegetable oils in the diet (VOIL and GLVO diets) improved conformation, fat thickness, and percentage fat in the carcass. The increased dressing carcass results in the weight of hot carcass weight, which can be attributed to diet feeding treatments. Carcass conformation (13.6 points, on a scale from 0 to 18 points) was considered adequate to meet the standards of the Brazilian market (Rotta et al., 2009). However, young bulls finished fed CONT and GLYC diets showed a reduction in grade of carcass conformation, which was considered “good to very good”, in comparison with young bulls finished on the GLVO diet, in which carcass conformation was considered “very good”.

Françozo et al. (2013), Cruz et al. (2014) and Fugita et al. (2018) reported no effect of the glycerin inclusion in the diet on carcass conformation, which was considered very good (from 12.5 to 14.0 points). The higher carcass dressing and better conformation probably reflect higher fat deposition. The fat thickness and percentage fat were higher in carcasses from bulls fed with glycerin and vegetable oils. The inclusion of glycerin and vegetable oils in the diets of bulls finished in a feedlot improved apparent digestibility (Cruz et al., 2014; Eiras et al., 2014). Glycerol is the principal component of glycerin, and assists in the process of gluconeogenesis and triglyceride synthesis (Chung et al., 2007; Goff and Horst, 2001; Krehbiel, 2008). Likewise, glycerol increases volatile fatty acids during rumen fermentation (Abo El-Nor et al., 2010; Himejima and Kubo, 1991; Muroi et al., 1993; Shin et al., 2004). Vegetable oils improve ruminal fermentation via compounds that confer antibacterial properties (Abo El-Nor et al., 2010; Himejima and Kubo, 1991; Muroi et al., 1993; Shin et al., 2004). The higher propionate available in the rumen, as a result, improves gluconeogenesis in the liver (Bradford and Allen, 2007). The various mechanisms that glycerin can participate in, other nutrients or the same glycerol molecule can be targeted for fat synthesis and did not affect animal performance, which probably occurred in this study.

The *Longissimus* muscle temperature and pH were measured at 0 and 24 h *post mortem.* The mean temperature and pH at 0 h (37.3º C and 6.9, respectively) and 24 h (5.9º C and 5.7, respectively), LM area (68.5 cm^2), texture (4.2 points), and marbling (6.7 points) were similar to the values found by Françozo et al. (2013), Cruz et al. (2014), Eiras et al. (2014) and Fugita et al.(2018) in experiments with animals of similar age and body weight in response to the inclusion of glycerin and vegetable oils in the diet.

The texture is classified by granulation on the LM surface, and in the present study was rated as “thin” or “very thin” (4.2 points). Similar results were observed by Eiras et al. (2014), who reported no effect on texture or marbling in response to the inclusion of glycerin in the diet. Marbling was classified as “light” or “small” (6.7, points) in the current study. Medium marbling is well accepted within the domestic market; however, foreign markets require more accentuated marbling.

4.2 Longissimus muscle characteristics

Many factors influence meat quality (Ducatti et al., 2009; Rotta et al., 2009). The high *Longissimus thoracis* muscle pH observed in this study at 24 h *post mortem* is the result of glycogen depletion during pre-slaughter, which prevents the muscle from accumulating an adequate lactic acid concentration (Immonen et al., 2000). According to Page et al. (2001), a higher muscle pH is associated with beef that is more green (a*) and more blue (b*), whereas a lower muscle pH is associated with beef that is redder (a*) and more yellow (b*).

Colour is an important factor in meat quality and purchase decisions made by consumers of beef, contributing to a characteristic that is perceived by consumers as "food freshness". In the current study the mean subjective colour (3.51), instrumental colour for lightness (32.5), redness (13.9), yellowness (4.9), Chroma (14.8), and hue angle (19.1) were unaffected by feeding treatments; however, the observed values were considered high for lightness and redness (Page et al., 2001); whereas yellowness was lower.

The low value for yellowness (4.9) can be attributed to the long period in the feedlot (252 days) on diets with low pigment content (Dunne et al., 2009). Eiras et al. (2014) reported higher muscle pH (6.2) at 24 h *post mortem*, and better colour characteristics for L*, a*, and b* in the LM when glycerin was included at 0, 6, 12, or 18% with similar handling practices, experimental facilities, and place of slaughter. On the other hand, when we measured subjective colour in the LM at 24 h *post mortem*, meat that was considered to be good ranged from “red” to “slightly dark red”. According to Renerre & Labas (1987), nutrition and age affect meat colour.

4.3 Longissimus muscle chemical composition

Diets generally have little influence on chemical composition in the *Longissimus* mucle (LM) (Fugita et al., 2018; Monteschio et al., 2019; Rivaroli et al., 2020). The inclusion of glycerin and vegetable oils did not affect mean moisture (25.6%), ash (1.0%), crude protein (21.6%), total lipids (2.21%), shear force (3.07 kg) or calories (225.5 kcal/100 g of meat) in the LM. Prado et al. (2008) and Ito et al. (2012) reported similar values for the chemical composition of the LM from different genetic groups of bulls finished in a feedlot. Similarly, Ducatti et al. (2009) and Eiras et al. (2014) reported no difference in moisture (74.2%), ash (1.0%), crude protein (21.3%), or total lipids (2.0%) in the LM of similar genetic groups and age in response to glycerin inclusion. Another study reported no effect on total lipid content in the LM (3.8) in response to the inclusion of glycerin at 4, 8 or 12% (Mach et al., 2009). On the other hand, Françozo et al. (2013) observed a reduction in total lipids in the LM of bulls receiving glycerin (0, 5 or 12%). The total lipid levels observed in all groups were acceptable for the prevention of diseases related to the fat content in beef.

According to Jeleníková et al. (2008) tenderness is associated with the rate of glycolysis, post-mortem temperature decrease, and final pH in the LM. Peachey et al. (2002) associated tenderness with intramuscular fat content. In the present study total lipids (2.21%) and tenderness obtained by WBS (3.07 kg) in the LM were similar. Similarly, Eiras et al. (2014) reported no difference in total lipids and WBS in response to different glycerin levels in the diets of bulls finished in feedlot. In contrast, Mach et al. (2009) reported a reduction in total lipids but no change in LM tenderness following glycerin inclusion. Shackelford et al. (1999) reported that a WBSF result < 4.0 kg indicated tenderness that should result in high consumer acceptance.

Mean caloric values (225.5 kcal/100 g of meat in nature) in the LM were not affected by diet (Table 4). This may reflect the similar chemical composition of all treatments. One of the most important causes of meat deterioration is lipid oxidation, which is affected by the intramuscular fatty acid composition and particularly by polyunsaturated fatty acids (Faustman et al., 2010; Wood et al., 2008).

Table 5 shows the lipid oxidation values, as determined by TBARS (mg MDA/kg meat). Glycerin and vegetables oils did not affect ($P > 0.05$) TBARS values in the time evaluated (48 *post-mortems*). It has been reported that lipid oxidation may lead to drip losses, the development of an off-odor and off-flavor, and the production of potentially toxic compounds as well as inducing the oxidation of myoglobin (Faustman et al., 2010). The oxidation levels found in the present study (0.28 mg MDA/kg meat) were low and smaller the values found for animals fed silage and concentrate for 100 days before slaughter (Alexandre et al., 2020; Ornaghi et al., 2020). The values observed could be attributed to the duration of frozen storage until TBARS analysis (6 months), as observed by Muela et al. (2010) in meat from lambs. In any case, the MDA levels were above the recommended limit of 2 mg MDA/kg fresh tissue (Campo et al., 2006). The bulls in the current study were fed with corn silage and cereal, which contain vitamin E, making the samples oxidatively stable (Campo et al., 2006).

Red meat consumption contributes many minerals and vitamins to the diet (McAfee et al., 2010). The inclusion of glycerin and vegetables oils in the diet of bulls did not affect the calorie content of the meat (Table 5). The calorie levels found in the present study (225.2 Kcal in 100 g meat) contributed to adequate food. According to HMSO (1994), the recommended daily calorie intake is 1,940 Cal/day for women and 2,550 Cal/day for men, and fat should not exceed 35% of the total diet. Previous studies recommend consuming lean red meat as part of an energy-controlled diet (McAfee et al., 2010).

5. Conclusion

The partial replacement of corn by glycerin (81.2% glycerol) and vegetable oils (cashew and castor oils) in the diet of bulls in a feedlot did not negatively affect carcass characteristics, LM characteristics, or chemical composition. Diets containing glycerin and/or vegetable oils improved cold carcass weight. Diets containing vegetable oils improved dressing cold carcass. Glycerin associated with vegetable oils improved the conformation, fat thickness, and proportion of fat in the carcass. Thus, glycerin can be added up to a rate of 20% on a DM basis with vegetable oils from cashew and castor oils to the diets of bulls finished in a feedlot for 250 days.

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Declarions

Ethic statement: This experiment was approved by the ethics committee of Maringá State University, Paraná, Brazil (Protocol no. 3624120116).

Informed consent: All authors have given their consent that this work is valid and represent their views of the study, and all authors have given their consent for this wirk to be published.

Conflict of interest: We certify that there is no conflict of interest with any financial organization regarding the material discussed in the manuscript.

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