

Effect of oral glycerol on the carcass and beef quality

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Abstract

Glycerol can be used as a gluconeogenic substrate to increase glycogen reserves in steers subjected to stress before slaughter. This study aimed to evaluate the effect of oral glycerol 24 h before slaughtering on the pH of carcass and beef quality. Thirty-eight steers of 450 kg live weight were used. Of these, 19 did not receive glycerol (T₁), and 19 received 600 mL glycerol orally 24 h before slaughter (T₂), under a completely randomized design. During the 24-hour *postmortem*, the pH and temperature of the carcasses were determined. In the last reading, dry matter (DM), crude protein (CP), and ash were determined in the meat samples taken from the rib area, which were also used to determine pH, color, water holding capacity (WHC), texture, CP and ash at 7, 14, 21 and 28 d after slaughter. No differences ($P>0.05$) were found in pH, temperature, or DM and CP contents, in contrast ($P<0.05$) with ash at 24 h post-mortem. Oral administration of glycerol did not affect ($P>0.05$) pH, color, water retention capacity, and texture; however, these were affected ($P<0.05$) during shelf life. The interaction between glycerol and shelf life was not significant ($P>0.05$) for the indicated variables. Concentrations of CP and ash were similar ($P>0.05$) and with a positive interaction ($P<0.05$) for ash in T₂. The administration of 600 mL glycerol 24 h before slaughtering did not cause changes in the pH of carcass and beef quality; however, an effect was observed in color, WHC, and texture in the shelf-life of beef.

Key words: Bovines; Glycerol; Beef quality; pH carcass

Introduction

Transport and slaughter are the most stressful stages for animals because of handling, overcrowding with other animals, changes in temperature, lack of water, food, and noise, causing physiological and behavioral changes (Teke *et al.*, 2014.). Stress during transport and before slaughter decreases glycogen reserves in muscles, resulting in low production of post-mortem lactic acid and inadequate carcass pH (>5.8), impairing meat color, tenderness, water holding capacity, and dark meat that affect the appearance, juiciness, and shelf life; these aspects determine consumer acceptance (Liu *et al.*, 2022). Feeding different additives during fattening to the steers is gaining importance for its effect on carcass quality (Hamikoeva *et al.*, 2021; Kairov *et al.*, 2021; Khamikoeva *et al.*, 2021; Son *et al.*, 2025). Glycerol is also a suitable substrate for energy, as it can be converted to glucose through gluconeogenesis (Rotondo *et al.*, 2017). Studies have shown that administering glycerol in the form of glycerin to ruminants can promote metabolic changes and increase plasma glucose levels for short periods. However, although there are studies on this topic (Eiras *et al.*, 2014; Chanjula *et al.*, 2014), its effects on carcass and meat characteristics when it is administered before transport and slaughter are not entirely known (Orrico Junior *et al.*, 2015). Therefore, the objective of the study was to evaluate the effects of oral administering glycerol to steers, 24 h before slaughtering, on carcass temperature and pH, chemical composition of the meat at 24 h post-mortem, as well as water retention capacity, texture, crude protein, and ash content during the shelf life meat.

Materials and methods

Experimental site, animals and experimental design

The study was conducted in Tamuín, San Luis Potosí, México, located at 22° 00' 25" North and 98° 47' 19" West. Mean temperature 16°C and 53% relative humidity. This research was approved by the Animal Welfare Committee of the Colegio de Postgraduados. At the end of a 90-d fattening period, 38 Zebu (*Bos indicus*) crosses with Swiss or Simmental (*Bos taurus*) steers, with an average live weight of 450 ± 48 kg, the animals were weighed using a digital livestock scale (TORREY®, Model PG-2000, Monterrey, México) and were selected at random 24 h before slaughter. Steers were subjected to two treatments, in a completely randomized design (n=19); T₁: without oral administration glycerol (GLY), T₂: oral administration of 600 mL GLY (85% pure) per animal, 24 h before slaughter, using a Syrman graduated syringe and were manipulated with the support of a clamp to hold them.

Ante-mortem management

Steers were boarded for transport at 05:30 h (15°C and 90% relative humidity) into 20-steer capacity cages. The estimated time to slaughter in a commercial slaughterhouse with Federal Inspection Service was 20 min. During the entire process and at the reception offices before slaughter, observations were made to detect falls, disembarkation time, animal behavior, and handling. The total waiting time for the 38 steers up to slaughter was 75 min.

pH and temperature of steers carcass

Three h *post-mortem*, initial carcass temperature (T_{3h}) (DELTA TRAK, Model 11050, Waterproof Lollipop, USA) and initial pH (pH_{3h}) (HANNA pH meter Model-HI99163, Woonsocket, RI, USA) were measured, and consecutive readings were performed 6, 9, 12, 15, 18, 21 and 24 h *post-mortem*.

Physicochemical analysis of meat

Quartering and extraction of meat samples (four replications) in the rib area were done at the last reading (24 h). In these samples, immediately, dry matter (DM), crude protein (CP), and ash were determined following the AOAC (2005) methodology (9). Other meat samples were vacuum packed and kept at 5 °C for analysis at 7, 14, 21, and 28 d *post-mortem*, which correspond to meat shelf life in accord with the commercialization time of perishable products. pH was determined (PELICAN 1400 Case, Model SA 210, CA, USA), and color using Hunter (HUNTER QUEST 45/0, Hunter Associates Laboratory, Reston, VA, USA) values, which include L* (luminosity), a* (red to green tones), b* (yellow to blue tones). These were determined in a colorimeter (KONICA MINOLTA, Model CR400/410, NJ, USA) in meat samples at room temperature. The results are reported as the average of three consecutive measurements. WHC was determined by the methodology proposed by Guerrero *et al.* (2002). Five g of meat were placed in 160 mL tubes and homogenized with 8 mL 0.6 M NaCl. Tubes were then placed in an ice bath for 30 min and centrifuged 15 min at 1500 g (CLAY ADAMS, Model 420101, Sparks, USA). The supernatant was decanted and its volume was measured; this represented the non-retained water of the 8 mL NaCl that was added. Three meat segments were extracted with a punch perpendicular to the muscle fibers to determine each sample's texture profile. Then a Warner-Bratzler blade in a texture analyzer TA-XT2 (Texture Technologies Corp, Scarsdale, NY, USA) was used, with forward and backward movement speed of the blade of 5 mm/s (Guerrero *et al.* 2002). Crude protein and ash were also determined (AOAC, 2005).

Statistical analysis

The experimental design was completely randomized, and an analysis of variance was performed with Statistical Analysis System software (SAS, 2004). The chemical composition of the meat at 24 h were analyzed with PROC GLM; pH, temperature of carcass and pH, color, WHC, texture, protein and ash of the meat was

analyzed with measurements repeated through time with PROC MIXED, the model included principal effects and the interaction treatment*time. The means comparison was performed with the Tukey test.

Results and Discussion

pH and temperature of steers carcass

Administration of GLY to the steers 24 h before slaughter did not affect ($P>0.05$) carcass temperature (T) or pH at the different monitoring times (Figure 1). A descending trend occurred as the hours lapsed: initial T_{3h} of 33.05 and 32.91°C, intermediate (15 h *post-mortem*) of 13.4 and 13.8°C, ending at 27 h with 1.02°C and 1.31°C for GLY treatment and without GLY, respectively. Initial pH_{3h} was 6.5 and 6.9, decreasing after 9 h to 5.6 and 5.3, and both treatments stabilized and finalized at a pH_{24h} of 5.6 and 5.7 for GLY treatment and without GLY, respectively. The rate of cooling and the decrease in pH determine the final quality of the meat, so it is essential to know the conversion rates of the energetic substrates linked to changes in the pH and temperature of the muscles (Kufii *et al.*, 2017). pH is one of the most important meat quality parameters since it is related to muscle glycogen reserves and is a direct indicator of acid content in the muscle and affects cutting force, drip loss, and meat color (Carrasco-García *et al.*, 2020). Francozo *et al.* (2013) included 5 and 10% GLY in the diet of Nellore steers and shipped them 84 km at a density of one animal m^{-2} ; they found carcass pH_{24} of 6.35 and 6.11. Lage *et al.* (2014) obtained similar results when they included different GLY levels in the diet. However, Carvalho *et al.* (2014) reported similar values to those found in our study in which pH_{24h} was between 5.5 and 5.8. Chanjula *et al.* (2014) indicated that including 20% GLY in the diet reduces the acetate:propionate ratio because it benefits the population of *Selenomonas*, the primary GLY fermenters in the rumen, metabolizing approximately 80% after 24 h. In our study, pH was not affected by GLY inclusion 24 h before slaughter, possibly because the steers metabolized it rapidly or the amount administered was insufficient. Furthermore, GLY can be absorbed from the rumen in significant amounts, disappear through microbial digestion, and exit from the rumen through the omasal orifice (Werner *et al.*, 2015). The short transport time (20 min) and the rapid access to slaughter may not have produced enough stress in the animals to consume all the GLY as the pH values in the carcasses of the two treatments were similar. The pH values in the present study were in a normal pH range (5.4 to 5.7) of non-stressed animals (Piao *et al.*, 2021).

Chemical composition of the meat at 24 hours *post-mortem*

Administration of glycerol did not affect ($P>0.05$) DM or CP content 24 h *post-mortem*. However, it decreased ($P\leq 0.05$) the percentage of ash (Table I). Concentrations of CP are found within the 20.7 to 21.8% average in *Longissimus dorsi* of heifers reported by Almeida *et al.* (2021), indicating that administration of GLY (33.3, 66.6 and 99.9 g/kg DM of supplement) did not cause changes. However, Piao *et al.* (2020) report in thoracic *Longissimus thoracis* of Korean cattle steers, 19% CP when they included 3.57% GLY in the diet. The percentage of ash in the meat was high compared with the data of Piao *et al.* (2020), who obtained values of 1.3%. The difference between this study and those cited above can be attributed to the mineral content of the water consumed in the region where the feedlot is located, since the minerals are high (Vieyra-Alberto *et al.* 2013).

Physicochemical characteristics of beef at different days of refrigerated storage

Treatment with GLY did not affect ($P>0.05$) pH, luminosity (L^*), red tones (a^*), yellow tones (b^*), WHC, or meat texture. However, an effect of time was observed in all the variables ($P\leq 0.05$), even though the interaction treatment*time was not significant ($P>0.05$) (Table 2). Regarding crude protein and ash content, no differences were detected ($P>0.05$) due to treatment with GLY, and although differences were observed due to the effect of time ($P\leq 0.05$), the treatment*time interaction was not significant ($P>0.05$). The quality of meat storage depends on several factors, such as meat pH, growth and type of bacteria, oxidative stability, and enzymatic activity (Bostami *et al.*, 2023). The values during the shelf life were different, indicating a change in the use of GLY and the presence of lactic acid, modifying the pH levels over time (Strobel *et al.*, 2021), and quality meat is considered to have a final pH in the range of 5.4 – 5.6 (Hughes *et al.*, 2014a). The color, WHC, and tenderness are consumers' primary determinants of visual and sensorial attractiveness (Hughes *et al.*, 2014b). During meat shelf life, luminosity tended to decrease, which is affected by the quantity of water on the surface, a consequence of its WHC, which in turn affects pH (Francozo *et al.*, 2013). These results explain the values obtained of L^* at 28 days of shelf life. Animals under stress before slaughter more frequently produce dark, firm, and dry (DFD) meat (Hughes *et al.*, 2014b), however, this type of cut was not observed in this study, where L^* and b^* levels decreased, but a^* values increased, but not significantly. The results obtained from our study can be compared to those reported by Egea *et al.*, (2014) for Limousin bulls fed diets with 2 and 4% GLY; their carcasses 7 days *postmortem* had similar b^* , lower L^* and higher a^* . Eiras *et al.* (2014) obtained similar results for a^* and b^* , but higher values in L^* in carcasses of Puruná bulls fed diets with 0, 60, 120, and 178 g GLY/kg DM. Our results were higher than those of San Vito *et al.*, (2015) reported in grazed steers supplemented with GLY. Francozo *et al.* (2013) found that color coordinates were lower when 5 and 12% GLY was included in the diets of Nellore steers. However, Carvalho *et*

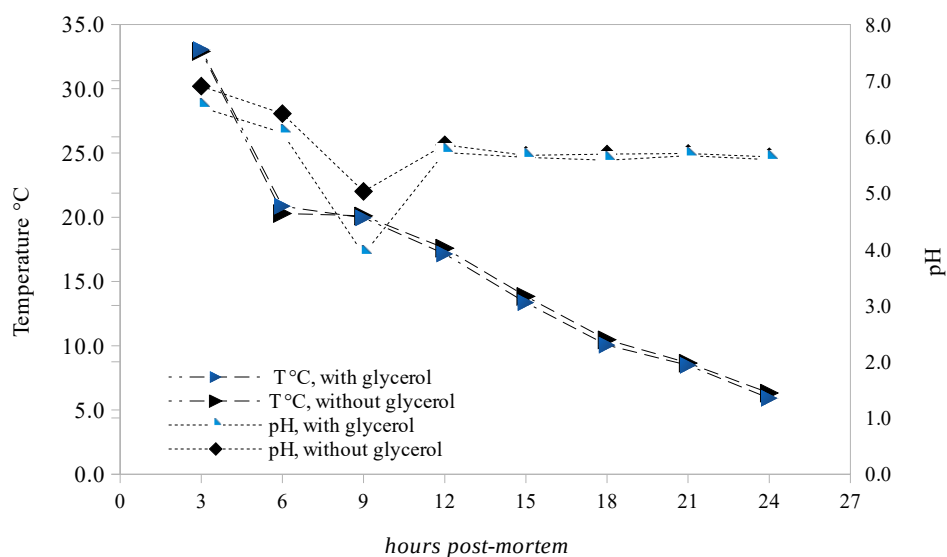


Fig 1. Temperature and pH of the carcass of steers administered with or without glycerol 24 h before slaughter

Table 1. Characteristic of meat of steers administered with and without glycerol 24 h before slaughter

Variable	T ₁	T ₂	SEM	p-value
Dry matter	28.01	26.82	0.18	0.884
As % dry matter				
Crude protein	23.40	23.72	0.15	0.745
Ash	4.00	3.66	0.04	0.032

T₁: Not glycerol administration; T₂: Oral administration of 600 mL glycerol; SEM: Standard error of the mean.

Table 2. Effect of treatments with and without glycerol on the characteristics of steer meat

	Refrigeration days (5°C)												
Items	7		14		21		28		SEM				
	T ₁	T ₂	T ₁	T ₂	T ₁	T ₂	T ₁	T ₂	T ₁	T ₂	Treat	Time	Treat*Time
Ph	5.6	5.6	5.5	5.5	5.5	5.5	5.6	5.6	0.30	0.27	0.651	0.005	0.075
L*	38.91	38.92	36.36	38.17	37.20	37.72	35.20	35.10	0.85	0.77	0.789	0.041	0.955
a*	14.15	14.36	17.09	17.52	15.21	15.32	16.33	16.77	0.50	0.46	0.058	0.025	0.087
b*	6.28	6.88	5.21	5.61	5.93	6.03	4.69	4.69	0.33	0.30	0.092	0.001	0.059
WHC	8.3	8.2	8.5	8.3	8.2	8.1	8.1	8.2	0.12	0.11	0.579	0.001	0.195
Texture	1.86	1.89	1.85	1.88	2.06	2.10	1.71	1.96	0.09	0.09	0.478	0.004	0.287
As % DM													
Protein	24.08	23.73	22.94	23.55	23.64	23.86	22.95	23.74	0.31	0.28	0.134	0.032	0.761
Ash	4.12	3.49	3.94	3.69	4.04	3.69	3.89	3.77	0.08	0.07	0.267	0.001	0.039

Color: L* = Luminosity; a* = red index; b* = yellow index. WHC = Water holding capacity of raw beef (mL 100⁻¹ g). Texture = resistance to cutting with a Warner-Bratzler blade (kg cm⁻²). DM: Dry matter. T₁: without glycerol administration T₂: Oral administration of 600 mL glycerol.

al. (2014) reported that the inclusion of 6, 12, and 18 % GLY in the diet affected steer carcass color, with higher values of *a** but lower values of *L** and *b**. Water retention may not be altered when GLY is administered in the diet, as has been demonstrated in studies by Francozo *et al.* (2013) and Egea *et al.* (2014). A similar situation occurs when administered directly, considering that GLY increases cell osmotic pressure, increasing intracellular water that can increase WHC (Egea *et al.*, 2014). The results obtained in this study indicate that GLY did not affect WHC or texture ($P > 0.05$). The differences in WHC over time can be attributed to protein deterioration and pH levels, which are affected by glycogen transformed to lactic acid, and reduced ATP (Abril *et al.*, 2023), however, in this study, the WHC levels in the beef were normal. These results are significant because they affect product quality, which is closely related to texture, color, and pH. According to Eiras *et al.* (2014), adding GLY does not

affect meat tenderness. In our study, the final pH values, as well as those of texture, were similar in the two treatments. According to Warner Bratzler's classification, this study's results are in the range of tender meat.

Conclusion

Oral administration of 600 ml of glycerol 24 h to steers before slaughter did not affect the pH of the carcass during the first 24 h post-mortem. Likewise, the dry matter and crude protein content (except ash) was not affected at 24 h post-mortem. The administration of glycerol before slaughter did not cause changes in the characteristics of the meat, however, it affected the pH, color, water retention capacity and texture on the different days of storage.

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Conflict of interest:

The authors declare no potential conflicts of interest.

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