



## Short Note [Nota corta]

## EFFECT OF PROPYLENE GLYCOL ON BLOOD METABOLITES, RUMINAL AND PRODUCTIVE PARAMETERS OF GROWING-FINISHING LAMBS †

## [EFECTO DEL PROPILENGLICOL SOBRE METABOLITOS SANGUÍNEOS Y PARÁMETROS RUMINALES Y PRODUCTIVOS DE CORDEROS EN CRECIMIENTO-FINALIZACIÓN]

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## SUMMARY

**Background.** Propylene glycol has been used successfully since the 1950's for the acute and prophylactic treatment of ketosis in dairy cows however; its use has been poorly evaluated in beef cattle and meat sheep. **Objective.** Evaluate the effects of different doses of propylene glycol on the productive parameters, blood metabolites and ruminal parameters in fattening lambs. **Methodology.** Twenty lambs 14.70 ± 0.57 kg of weight, 2 months old, males and Katahdin-Black Belly breed, were randomly assigned to one of four treatments, a control mixed ration and an experimental diet with three different levels of propylene glycol: 1.0, 1.5 and 2.0 g/20 kg live weight/day. The effects of propylene glycol on blood metabolites, ruminal and productive parameters were assessed. **Results.** Propylene glycol supplementation did not affect ( $P > 0.05$ ) glucose, total lipids, triglycerides and cholesterol concentration as well as increased ( $P < 0.05$ ) the concentration of beta hydroxybutyrate. In addition, its inclusion resulted in an increase in pH and protozoa population and in a decrease of reductive activity ( $P < 0.05$ ). No effect was observed ( $P > 0.05$ ) on feed intake, weight gain and feed conversion ratio of lambs but carcass yield was improved without increasing the feeding cost of production. **Implications.** Propylene glycol is a viable option as an energy source in fattening lambs. **Conclusion.** Inclusion of PPG increased the pH and the population of ruminal protozoa, reduced the formation of ketone bodies and improved the carcass yield of lambs without increasing the meat production cost.

**Key words:** Blood metabolites; feed additives; meat lambs; productive performance; ruminal parameters.

## RESUMEN

**Antecedentes.** El propilenglicol se ha utilizado con éxito desde la década de 1950 para el tratamiento agudo y profiláctico de la cetosis en vacas lecheras, sin embargo, su uso ha sido poco evaluado en ganado bovino y ovino productor de carne. **Objetivo.** Evaluar el efecto de diferentes dosis de propilenglicol sobre los parámetros productivos, metabolitos sanguíneos y parámetros ruminales en corderos de engorda. **Metodología.** Veinte corderos de 14.70 ± 0.57 kg de peso, 2 meses de edad, machos y raza Katahdin-Black Belly, fueron asignados aleatoriamente a uno de cuatro tratamientos, una ración mixta de control y una dieta experimental con tres niveles diferentes de propilenglicol: 1.0, 1.5 y 2.0 g/20 kg de peso vivo/día. Se evaluó el efecto del propilenglicol sobre la concentración de metabolitos sanguíneos, parámetros ruminales y productivos. **Resultados.** La suplementación con propilenglicol no afectó ( $P > 0.05$ ) la concentración de glucosa, lípidos totales, triglicéridos y colesterol, mientras que aumentó ( $P < 0.05$ ) la concentración de beta hidroxibutirato. Además, su inclusión dio como resultado un incremento del pH ruminal y la población de protozoos, así como una disminución de la actividad reductora

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( $P < 0.05$ ). No se observó efecto ( $P > 0.05$ ) sobre la ingesta de alimento, el aumento de peso y conversión alimenticia de los corderos, sin embargo, el rendimiento de la canal mejoró ( $P < 0.05$ ) sin aumentar el costo de producción por alimentación. **Implicaciones.** El propilenglicol es una opción viable como fuente de energía en la engorda de corderos. **Conclusión.** La inclusión de propilenglicol incrementó el pH y la población de protozoos ruminales, redujo la formación de cuerpos cetónicos y mejoró el rendimiento de la canal de los corderos sin aumentar el costo de producción de la carne.

**Palabras clave:** Metabolitos sanguíneos; aditivos alimenticios; corderos de carne; comportamiento productivo; parámetros ruminales.

## INTRODUCTION

Glucose is the main source of energy in animals, it is necessary for both maintenance and to comply with the production requirements. In the case of ruminants, about 90% of the total glucose required is contributed by the gluconeogenesis process from the volatile fatty acids absorbed after ruminal fermentation (Aschenbach *et al.* 2010). In dairy cows in particular, during the periparturient period, due to diminished dry matter intake, the demand for propionate and glucogenic amino acids from the rumen increases significantly (Gualdrón-Duarte and Allen, 2017).

Propylene glycol (PPG) is an organic compound with a brute energy content of 5.8 kcal/g that is characterized by its glycogenic activity (Ferraro *et al.* 2016) and has been shown to increase the concentration of propionate and decrease the ratio of acetate to propionate, thereby resulting in a ruminal volatile fatty acids pattern that is more glucogenic (Chung *et al.* 2009).

Propylene glycol has been used since the 1950s as an acute and prophylactic treatment of ketosis in dairy cows (Nielsen and Ingvarsen, 2004; Bjerre-Harpøth *et al.* 2015), as well as in different reproductive protocols in small ruminants and dairy cattle (Hackbart *et al.* 2017; Mikuła *et al.* 2020), however has been poorly evaluated in beef cattle and meat sheep. In this last, the effect of the inclusion of propylene glycol on intake, digestibility, and forage selection has been studied (Costa *et al.* 2019), however, there are no studies that evaluate the effect of its supplementation on productive parameters.

Therefore, the objective of this study was to evaluate the effects of different doses of propylene glycol on blood metabolites and ruminal and productive parameters in growing-fattening lambs.

## MATERIALS AND METHODS

The study was carried out in the Behavioral Testing Unit, Department of Zootechnics of the Autonomous University of Aguascalientes, located in Jesús María, Aguascalientes, México. Twenty Katahdin x Black Belly male lambs with an average weight of  $14.70 \pm 0.57$  kg of weigh, 2 months old, males and Katahdin-Black Belly breed, were randomly distributed into four experimental groups, with five replications per treatment ( $n=5$ ). Treatments consisted in a control mixed ration without PPG

(PPG 0) and an experimental diet in which 7 to 8% of grain (whole sorghum and rolled corn) was substituted with hay and three levels of propylene glycol (Nombre comercial del producto®, Laboratorio): 1.0, 1.5 and 2.0 g PPG/20 kg live body weight (PPG 1, PPG 1.5, and PPG 2, respectively), formulated according to the NRC (2007) specifications for fattening sheep (Table 1). Lambs were housed in individual pens, fed *ad libitum*, and had free access to water throughout the experiment which lasted 90 days.

**Table 1. Physical and chemical composition of control and experiental diets administered to growing lambs.**

Ingredient (g/100 g)	Control diet	Experimental diet
Whole sorghum	40.00	35.00
Soy meal	13.00	13.00
Alfalfa meal	7.00	7.00
Mineral nucleus	0.50	0.50
Calcium carbonate	1.00	1.00
Sodium bicarbonate	1.50	1.50
Corn, rolled	34.00	30.00
Hay	3.00	12.00
Analyzed composition		
Dry matter (%)	95.00	97.00
Crude protein (%)	15.00	15.00
Crude fiber (%)	4.00	10.00
Crude fat (%)	3.50	4.50
Ash (%)	3.50	9.00
Nitrogen Free Extract (%)	69.00	56.50
Acid Detergent Fiber (%)	17.00	28.00
Neutral Detergent Fiber (%)	42.00	44.00
Metabolic energy (Mcal/kg)	3.00	2.80

<sup>1</sup> Bromatological analysis was carried out in the nutrition laboratory of the Center for Agricultural Sciences of the Autonomous University of Aguascalientes.

Sampling and evaluation of production parameters was carried out on days 0, 30, 60 and 90 of the study. Blood samples were obtained by jugular venipuncture 2 h after feeding using tubes (i.e., without anticoagulant) and serum was separated by centrifugation at 2500 g for 10 min. Ruminal fluid

was taken 3 h after feeding by means of an oropharyngeal probe.

Blood glucose and  $\beta$ -hydroxybutyrate (BHBA) concentrations were determined using a glucometer with their respective test strips (Free Style Optium Neo, Abbott, Mexico City, Mex). The measurement of serum triglycerides and cholesterol were performed with the Triglycerides Reagent kit and the Cholesterol Reagent kit (Pointe Scientific Inc., Michigan, USA.) respectively, using a semi-automatic analyzer BTS-350 (BioSystems S.A., Barcelona, Spain). Total lipids concentration was also measured the Total Lipids kit (Cod. 8001602, MexLab Group, Guadalajara, Mex) in a spectrophotometer RA-50 (Bayer A.G., Leverkusen, Germany).

Immediately after ruminal fluid collection, the pH was determined using an electric potentiometer (Hanna Instruments, Mexico City, Mex). Bacterial reductive activity was evaluated by methylene blue reduction test and subsequently, 20  $\mu$ L of rumen fluid was placed in a Neubauer chamber and observed in an optical microscope at 40x magnification to determine protozoa population (Dirksen, 1969).

To determine the productive parameters feed offered and their respective refusals were recorded daily, and the lambs were weighed monthly in the morning before feeding. Feed intake was estimated as daily feed intake  $\times$  the days of confinement. Total weight gain (TWG) was determined for difference between the final and initial body weight, and the daily weight gain (DWG) was calculated as the quotient of the total weight gain and the confinement time (90 days). Feed conversion ratio (FCR) was calculated as the amount of feed consumed (kg) per body weight gain (kg), and the feed cost per kilogram of meat produced as the feed conversion ratio  $\times$  the cost of the diet. Lambs were slaughtered at a commercial slaughterhouse at the end of the trial and carcass yield was calculated as the quotient of the hot carcass weight and the live weight at slaughter  $\times$  100.

Data were analyzed using the statistical software SPSS (2017). The effect of PPG inclusion on blood metabolites and ruminal parameters was statistically analyzed using repeated-measurements ANOVA. Data from productive performance were subjected to one-way ANOVA including the initial weight of the lambs as a covariate. Post hoc comparisons were made using Tukey test and results were considered significant when  $P < 0.05$ .

## RESULTS

### Blood metabolites

Average serum glucose concentration was 67.85, 69.05, 66.70 and 71.25 mg/dL for PPG0, PPG1, PPG1.5 and PPG2 lambs respectively. Although

during the first 30 days the serum glucose concentration increased in the supplemented lambs to later show a gradual decrease, while that of the unsupplemented lambs decreased from the beginning of the test, this was similar ( $P > 0.05$ ) between the treatments and throughout the study period. Average serum BHBA concentration in PPG0, PPG1, PPG1.5 and PPG2 lambs was 0.51, 0.37, 0.35, 0.32 mg/dL respectively. BHBA concentration of the unsupplemented lambs increased significantly ( $P < 0.05$ ) at 60 days, also showing from that moment, a higher concentration ( $P < 0.05$ ) of BHBA than those lambs that received 1.5 and 2.0 g/20 kg BW of PPG. Average serum total lipids concentration was 271.00, 218.31, 201.34 and 232.87 mg/dL for PPG0, PPG1, PPG1.5 and PPG2 lambs respectively. Despite the fact that serum concentration of this metabolite in unsupplemented lambs showed a significant increase at 60 days, it did not present significant changes ( $P > 0.05$ ) throughout the study or between treatments. Average serum triglycerides concentration in PPG0, PPG1, PPG1.5 and PPG2 lambs was 37.85, 33.10, 31.05, 32.35 mg/dL respectively. In all treatments, the concentration of serum triglycerides increased during the first 30 days to later show a slight decrease, being statistically similar ( $P > 0.05$ ) throughout the study period. Average serum cholesterol concentration was 144.40, 127.05, 123.30 and 129.03 mg/dL for PPG0, PPG1, PPG1.5 and PPG2 lambs respectively. Serum cholesterol concentration was similar between all treatments ( $P > 0.05$ ), however it was significantly higher ( $P < 0.05$ ) at 60 days with respect to that observed at the beginning and at the end of the test (Figure. 1).

### Ruminal activity

Average ruminal pH in PPG0, PPG1, PPG1.5 and PPG2 lambs was 6.07, 6.63, 6.62 and 6.69 respectively. Ruminal pH of PPG0 lambs were lower ( $P < 0.05$ ) at days 30 and 60 than that of the rest of lambs. At day 90, pH of PPG1 and PPG2 lambs was higher ( $P < 0.05$ ) than that of the PPG0 lambs while pH of PPG 1.5 lambs showed intermediate values. Average time of reductive activity in PPG0, PPG1, PPG1.5 and PPG2 lambs was 3.83, 3.71, 3.55 and 3.46 minutes respectively. Time of reductive activity increased significantly ( $P < 0.05$ ) in all treatments on day 30 to later show values slightly lower to those found at the beginning of the trial. At day 30, reductive activity of PPG0 lambs were higher ( $P < 0.05$ ) than those observed in the rest of the lambs. Average protozoa population per mL of ruminal fluid was 341.50, 373.50, 380.00 and 393.00  $\times 10^3$  for PPG0, PPG1, PPG1.5 and PPG2 lambs respectively. Protozoa population of PPG1.5 and PPG2 lambs increased rapidly during the first 30 days of the trial to later remain stable. Protozoa population of PPG1 lambs showed an initial decrease to later increase rapidly and show values similar to those of the PPG1.5 and PPG2 lambs. Potozoa population of the PPG0 lambs showed

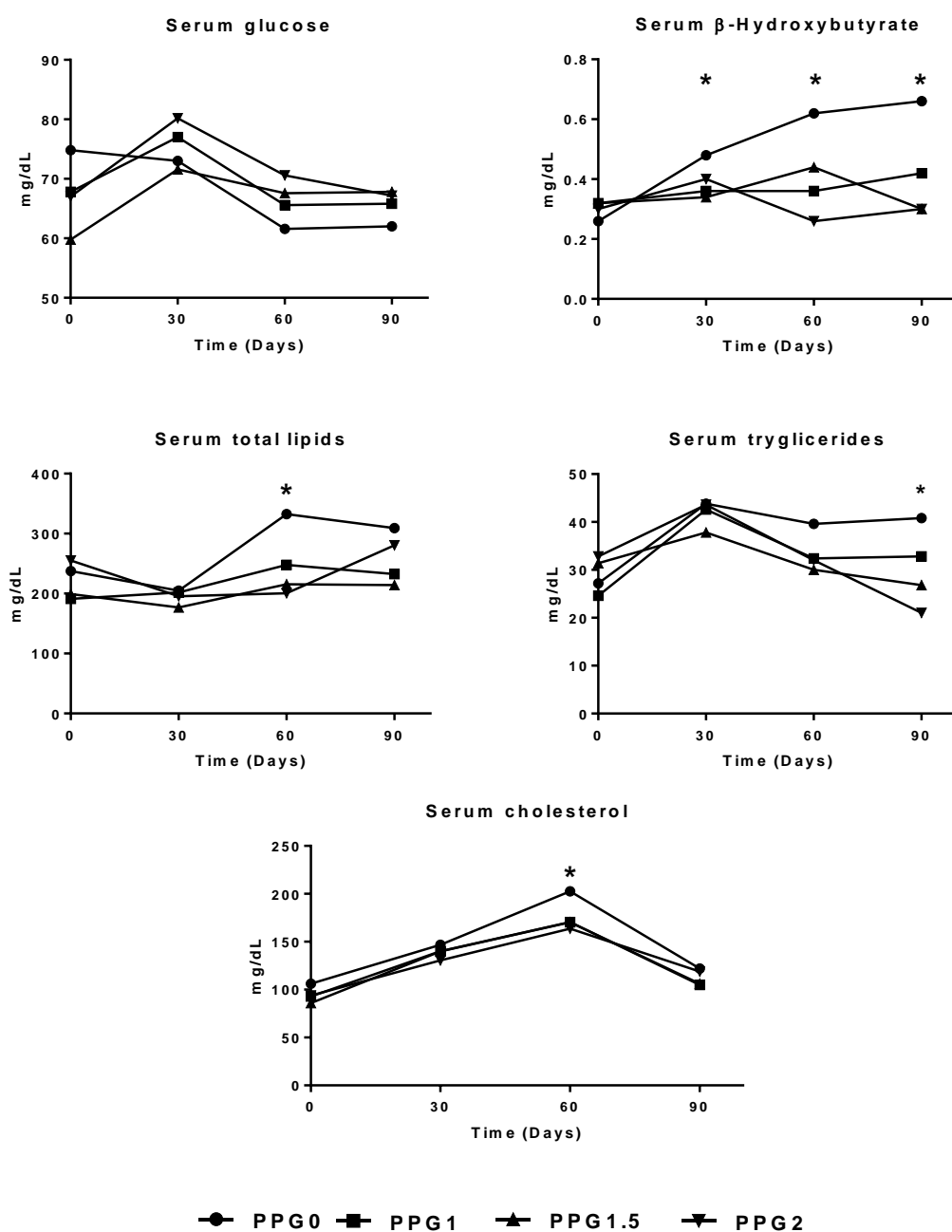
alternating increases and decreases to eventually, being significantly lower ( $P<0.05$ ) than that of the rest of the lambs (Figure. 2).

### Productive parameters

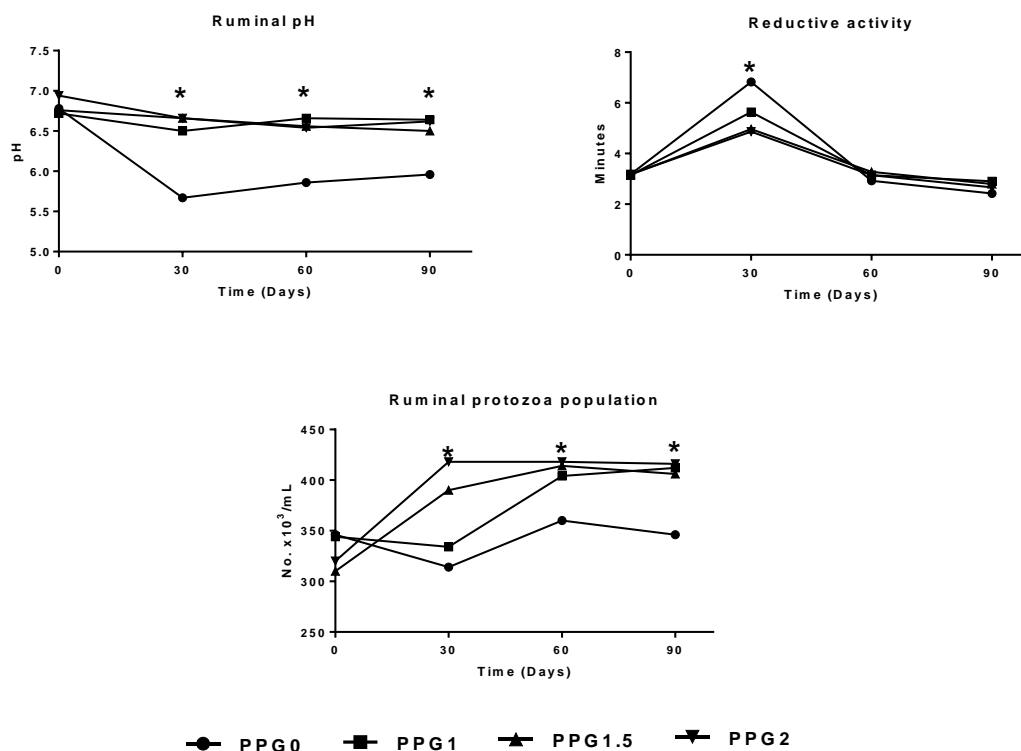
The inclusion of PPG in the diet did not affect ( $P>0.05$ ) feed intake, weight gain and feed conversion ratio of the lambs as it improved ( $P<0.05$ ) the carcass yield without increasing the feeding cost of production (Table 2).

### DISCUSSION

In the present study PPG supplementation with PPG did not increase the concentration of glucose which coincides with the studies carried out by Pickett *et al.* (2003) and Lomander *et al.* (2012) and, is contrary to that described by Nielsen and Ingvarsen (2004), Kristensen and Raund (2007) and Ferraro *et al.* (2016) who mention that increased plasma glucose concentration is indicative of the efficacious glucogenic effect of PPG. Ferraro *et al.* (2009) mention that PPG is rapidly metabolized to propionate in the rumen from where it escapes through the rumen wall and is converted to glucose by the liver increasing plasma concentrations of glucose and insuline.



**Figure. 1.** Effect of propylene glycol supplementation on serum metabolites of growing-finishing lambs.



**Figure 2.** Effect of propylene glycol supplementation on ruminal pH, reductive activity, and protozoa population of growing-finishing lambs.

**Table 2. Effect of time and propylene glycol supplementation on productive performance of growing lambs.**

Treatments <sup>1</sup>	Total Feed intake (kg)	Daily weight gain (g/d)	Total weight gain (kg)	Feed conversion ratio (kg)	Hot carcass yield (%)	Feeding cost/kg meat produced <sup>2</sup>
PPG 0	14.63±1.00	244.92±24.98	3.87±0.25	4.08±0.38	49.85±1.18 <sup>b</sup>	\$1.50±0.13
PPG 1	15.81±1.10	267.01±19.67	4.12±0.46	3.87±0.39	57.80±0.86 <sup>a</sup>	\$1.57±0.09
PPG 1.5	14.54±1.09	228.78±24.67	3.58±0.37	4.29±0.36	58.32±1.17 <sup>a</sup>	\$1.37±0.14
PPG 2	16.54±1.08	286.99±33.75	4.29±0.46	4.01±0.35	56.40±0.89 <sup>a</sup>	\$1.65±0.14
P value	0.20	0.149	0.46	0.632	0.05	

<sup>abc</sup> Means in the same column with different superscript are different ( $P < 0.05$ ); <sup>1</sup> PPG 0 control diet without propylene glycol, PPG 1 experimental diet + 1.0 g PPG/20 kg BW, PPG 1.5 experimental diet + 1.5 g PPG/20 kg BW, PPG 2 experimental diet + 2.0 g PPG/20 kg BW; <sup>2</sup> US Dollars.

Total lipids, tryglicerides and cholesterol concentration was similar between treatments which agrees with the findings of Toghdory *et al.* (2009) and Kabu and Civelek (2012) who found that the addition of PPG did not affect the concentration of total lipids, tryglicerides and cholesterol.

At the end of the test period which coincides with a higher energy demand of the lambs, a decrease in BHBA concentration together with a decrease in tryglicerides concentration was observed in lambs supplemented with the highest doses of PPG. Nielsen and Ingvarsten (2004), Kristensen and Raund (2007), Bjerre-Harpøth *et al.* (2015) and Chalmeh *et al.* (2020) stated that PPG caused an increase in the glucogenic status and a reduction of adipose tissue mobilization, which leads to decrease

NEFA in the liver and reduction in the formation of ketone bodies. In this way, Kalyesubula *et al.* (2019) concluded that treatment with PPG effectively reduced hyperketonemia and lipolysis. Contrary, non-supplemented lambs showed a higher concentration of serum BHBA which suggests that to support the energy requirement, the body fat and protein are mobilized for hepatic gluconeogenesis, which leads to the increase of NEFA, BHBA and ammonia in plasma (Moore and DeVries (2020).

Addition of PPG increased the ruminal pH, a situation contrary to that described by Nielsen and Ingvarsten (2004) since, according to their study, it could be expected that the inclusion of PPG in the diet would lead to a decrease in ruminal pH because of an increase in the concentration of ruminal

propionate. Chung *et al.* (2009) found a decrease in ruminal pH in cows receiving PPG as an oral drench, but not in cows that received it as a part of a total mixed ration or as a rumen drench. For their part, Kristensen and Raund (2007) found that ruminal pH was not affected by PPG supplementation.

The inclusion of PPG increased rumen protozoa which, as several studies pointed out, is closely related to ruminal pH. According to Dehority (2005) and Francisco *et al.* (2019), the abundance of total ciliates and entodinozoan protozoa was highly influenced and positively correlated with rumen pH, and values below 5.5 above 15 h/day generally causes protozoa death (Dehority, 2005; Franzolin and Dehority, 2010).

Time of reductive activity decreased with the inclusion of PPG observing values less than 4 minutes which was indicative of an adequate bacterial activity. Concurring to Karapinar *et al.* (2008) a methylene blue reduction time greater than 6 minutes is considered as evidence of acute ruminal lactic acidosis, a ruminal fermentative disorder that occurs in sheep feed with large amounts of seeds rich in ruminal fermentable carbohydrates such as grain whose inclusion level in the diets used in this study ranged between 65 and 74%.

Recapitulating, the addition of PPG increases the number of ruminal protozoa which would appear limit the accumulation of lactate, which leads to an increase in the pH and in the number and activity of bacteria with the consequent increasing of reductive activity (Brossard *et al.* 2004; Karapinar *et al.* 2008).

Notwithstanding that PPG has a high energy content and consequently the potential to increase food intake, this was not observed in the present study. Studies carried out in cows, bull calves and sheep coincide that food intake is generally not affected by the administration of PPG (Pickett *et al.* 2003; Kim *et al.* 2005; Chung *et al.* 2009; Yazdi *et al.*, 2015; Costa *et al.* 2019). The foregoing is attributed to the fact that the doses used are not sufficient to improve the energy density of the food and induce an increase in food intake. Also, PPG is considered unpalatable and generally does not stimulate food intake (Nielsen and Ingvarsen, 2004).

Addition of PPG did not increase TWG and DWG of the lambs, which is consistent with the study carried out by Kim *et al.* (2005), who did not observe differences in the initial and final weight in steers supplemented with propylene glycol. However, it should be noted that the lambs achieved the DWG for which the control diet was formulated (250–300 g/d), and the decrease in the energy content of the experimental diets did not affect the growth of the lambs or the feeding cost of production, which could be considered a positive effect of the PPG addition. This is very important if it is considered that in Mexico the energy of the diets used for the

fattening of lambs comes mainly from cereal and soybean meal, which are mostly imported and their price is subject to frequent fluctuations that could negatively affect the profitability of the farms (Reynoso *et al.* 2017).

FCR did not have a significant effect between treatments while the study of other gluconeogenic substrates showed that feed conversion efficiency of finishing cattle was improved by 21.9 and 33% when glycerine was used to replace 100 g/kg DM of the dry-rolled corn content and when it was included at 120 g/kg DM of the diet respectively (Pyatt *et al.* 2007; Eiras *et al.* 2014). However, FCR obtained in this study is within the reported range (4.11 – 4.99) for Katahdin lambs crossed with various meat breeds (Vázquez *et al.* 2011).

Carcass yield was significantly improved with the inclusion of PPG, effect not found by Kim *et al.* (2015) who reported that PPG did not affect carcass weight of Korean native steers.

## CONCLUSION

Inclusion of PPG increased the pH and the population of ruminal protozoa, reduced the formation of ketone bodies and improved the carcass yield of lambs without increasing the meat production cost.

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**Conflict of interests.** The authors hereby declare that they have no conflict of interest.

**Compliance with ethical standards.** This study was approved by the Ethics Committee for the Use of Animals in Teaching and Research (CEADI-UAA as stand in Spanish). The study was carried out in compliance with the provisions established in the Ethics regulations for the use of animals in teaching and research at the Autonomous University of Aguascalientes Code: DI-PL-NO-37.

**Data availability.** Data are available with the corresponding author ([teodulo.quezada@edu.uaa.mx](mailto:teodulo.quezada@edu.uaa.mx)) upon reasonable request.

**Author contribution statement (CRediT).** **Araceli López-Vargas** – Conceptualization, Methodology, Formal Analysis, Writing – original draft. **Teódulo Quezada Tristán** – Conceptualization, Methodology, Formal Analysis, Writing – original draft. **Carlos Haubi-Segura** - Formal Analysis, Writing – review & editing. **Rafael Macedo-Barragán** – Formal Analysis, Writing – review & editing. **Raúl Ortiz-Martínez** - Formal Analysis, Writing – review & editing. **Arturo Valdivia-Flores** - Formal Analysis, Writing – review & editing.

**Carlos Hernández-Millán** - Formal Analysis, Writing – review & editing.

## REFERENCES

- Aschenbach, J. R., Kristensen, N. B., Donkin, S. S., Hammon, H. M. and Penner, G. B., 2010. Gluconeogenesis in dairy cows: The secret of making sweet milk from sour dough. *IUBMB Life*, 62, pp. 869–877. DOI: <https://doi.org/10.1002/iub.400>
- Bjerre-Harpøth, V., Storm, A. C., Eslamizad, M., Kuhla, B. and Larsen, M., 2015. Effect of propylene glycol on adipose tissue mobilization in postpartum over-conditioned holstein cows. *Journal of Dairy Science*, 98, pp. 8581–8596. DOI: <https://doi.org/10.3168/jds.2014-8606>
- Brossard, L., Martin, C., Chaucheyras-Durand, F. and Michalet-Doreau, B., 2004. Protozoa involved in butyric rather than lactic fermentative pattern during latent acidosis in sheep. *Reproduction Nutrition Development*, 44, pp. 95–206. DOI: [10.1051/RND:2004023](https://doi.org/10.1051/RND:2004023)
- Chalmeh, A., Pourjafar, M., Badiei, K., Jalali, M and Sebdani, M. M., 2020. The comparative effects of dietary monensin and propylene glycol on insulin resistance of transition dairy cows. *Tropical Animal Health and Production*, 52, pp. 1573–1582. DOI: <https://doi.org/10.1007/s11250-019-02160-1>
- Chung, Y. H., Martinez, C. M., Brown, N. E., Cassidy, T. W. and Varga, G. A., 2009. Ruminal and blood responses to propylene glycol during frequent feeding. *Journal of Dairy Science*, 92, pp. 4555–4564. DOI: <https://doi.org/10.3168/jds.2009-2131>
- Costa, H., Saliba, E., Galvani, D., Bomfim, M., Lana, Â. M., Borges, A. L., Landim, A. and Faciola, A., 2019. Effects of zinc sulfate or propylene glycol on intake, digestibility, and forage selection by grazing sheep in a semi-arid region during the rainy season. *Animals*, 9, 867. DOI: <https://doi.org/10.3390/ani9110867>
- Dehority, B. A., 2005. Effect of pH on viability of *Entodinium caudatum*, *Entodinium exiguum*, *Epidinium caudatum*, and *Ophryoscolex purkynjei* in vitro. *The Journal of Eukaryotic Microbiology*, 52, pp. 339–342. DOI: <https://doi.org/10.1111/j.1550-7408.2005.00041.x>
- Dirksen, G., 1969. Ist die metylenblauprobe als schnelltest für die klinische pansensaftuntersuchung geeignet. *Deutsche Tierärztliche Wochenschrift*, 76, pp. 305–309. DOI: [10.1186/1751-0147-42-219](https://doi.org/10.1186/1751-0147-42-219)
- Eiras, C. E., Barbosa, L. P., Marques, J. A., Araújo, F. L., Lima, B. S., Zawadzki, F., Perotto, D. and Prado, I. N., 2014. Glycerine levels in the diets of crossbred bulls finished in feedlot: Apparent digestibility, feed intake and animal performance. *Animal Feed Science and Technology*, 197, pp. 222–226. DOI: [10.1016/j.anifeedsci.2014.07.004](https://doi.org/10.1016/j.anifeedsci.2014.07.004)
- Ferraro, S. M., Mendoza, G. D., Miranda, L. A. and Gutiérrez, C. G., 2016. In vitro ruminal fermentation of glycerol, propylene glycol and molasses combined with forages and their effect on glucose and insulin blood plasma concentrations after an oral drench in sheep. *Animal Feed Science and Technology*, 213, pp. 74–80. DOI: <https://doi.org/10.1016/j.anifeedsci.2016.01.010>
- Ferraro, S. M., Mendoza, G. D., Miranda, L. A. and Gutiérrez, C. G., 2009. In vitro gas production and ruminal fermentation of glycerol, propylene glycol and molasses. *Animal Feed Science and Technology*, 154, pp. 112–118. DOI: <https://doi.org/10.1016/j.anifeedsci.2009.07.009>
- Francisco, A. E., Santos-Silva, J. M., Portugal, A. P. V., Alves, S. P. and Bessa, R. J. B., 2019. Relationship between rumen ciliate protozoa and biohydrogenation fatty acid profile in rumen and meat of lambs. *PLoS ONE*, 14, e0221996. DOI: <https://doi.org/10.1371/journal.pone.0221996>
- Franzolin, R. and Dehority, B. A., 2010. The role of pH on the survival of rumen protozoa in steers. *Revista Brasileira de Zootecnia*, 39, pp. 2262–2267. DOI: <https://doi.org/10.1590/S1516-35982010001000023>
- Gualdrón-Duarte, L. B. and Allen, M. S., 2017. Increased anaplerosis of the tricarboxylic acid cycle decreased meal size and energy intake of cows in the postpartum period. *Journal of Dairy Science*, 100, 4425–4434. DOI: <https://doi.org/10.3168/jds.2015-9476>
- Hackbart, K. S., Bender, R. W., Carvalho, P. D., Vieira, L. M., Dresch, A. R., Guenther, J. N., Gencoglu, H., Nascimento, A. B., Shaver, R. D. and Wiltbank, M. C., 2017. Effects of propylene glycol or elevated luteinizing hormone during follicle development on ovulation, fertilization, and early embryo development. *Biology of Reproduction*, 97, pp. 550–563. DOI: <https://doi.org/10.1093/biolre/iox050>
- Kabu, M. and Civelek, T., 2012. Effects of propylene glycol, methionine and sodium borate on metabolic profile in dairy cattle during periparturient period. *Revue de Médecine Vétérinaire*, 163, pp. 419–430.

- Kalyesubula, M., Rosov, A., Alon, T., Moallem, U. and Dvir1, H., 2019. Intravenous infusions of glycerol versus propylene glycol for the regulation of negative energy balance in sheep: a randomized trial. *Animals*, 9, 731. DOI: <https://doi.org/10.3390/ani9100731>
- Karapinar, T., Dabak, M. and Kizil, O., 2008. Severe thiamine deficiency in sheep with acute ruminal lactic acidosis. *Journal of Veterinary Internal Medicine*, 22, pp. 662–665. DOI: 10.1111/j.1939-1676.2008.0094.x
- Kim, Y. K., Choi, H. and Myung, K. H., 2005. Effects of propylene glycol on carcass traits and its related gene expression in Korean native steers. *Journal of Animal Science*, 83, pp. 344–349. DOI: <https://doi.org/10.2527/2005.832344x>
- Kristensen, N. B. and Raun, B. M. L., 2007. Ruminal and intermediary metabolism of propylene glycol in lactating holstein cows. *Journal of Dairy Science*, 90, pp. 4707–4717. doi: <https://doi.org/10.3168/jds.2007-0295>
- Lomander, H., Frössling, J., Ingvarsen, K. L., Gustafsson, H. and Svensson, C., 2012. Supplemental feeding with glycerol or propylene glycol of dairy cows in early lactation—Effects on metabolic status, body condition, and milk yield. *Journal of Dairy Science*, 95, pp. 2397–2408. DOI: <https://doi.org/10.3168/jds.2011-4535>
- Mikuła, R., Pruszyńska-Oszmałek, E., Kołodziejcki, P. A. and Nowak, W., 2020. Propylene glycol and maize grain supplementation improve fertility parameters in dairy cows. *Animals*, 10, 2147. DOI: <https://doi.org/10.3390/ani10112147>
- Moore, S. M. and DeVries, T. J., 2020. Effect of diet-induced negative energy balance on the feeding behavior of dairy cows. *Journal of Dairy Science*, 103, pp. 7288–7301. DOI: <https://doi.org/10.3168/jds.2019-17705>
- Nielsen, N. and Ingvarsen, K., 2004. Propylene glycol for dairy cows: A review of the metabolism of propylene glycol and its effects on physiological parameters, feed intake, milk production and risk of ketosis. *Animal Feed Science and Technology*, 115, pp. 191–213. DOI: <https://doi.org/10.1016/j.anifeedsci.2004.03.008>
- NRC, 2007. National Research Council, *Nutrient Requirements of Small Ruminants: Sheep, Goats, Cervids, and New World Camelids*. Washington, DC: The National Academies Press. DOI: <https://doi.org/10.17226/11654>
- Pickett, M. M., Piepenbrink M, S. and Overton, T. R., 2003. Effects of propylene glycol or fat drench on plasma metabolites, liver composition, and production of dairy cows during the periparturient period. *Journal of Dairy Science*, 86, pp. 2113–2121. DOI: [https://doi.org/10.3168/jds.S0022-0302\(03\)73801-6](https://doi.org/10.3168/jds.S0022-0302(03)73801-6)
- Pyatt, A., Doane, P. H., and Cecava, M. J., 2007. Effect of crude glycerin in finishing cattle diets. *Journal of Animal Science*, 85(Suppl. 1), pp. 530(Abstr.).
- Reynoso, M.M.M., Oddone, N. and Flores, F.L., 2017. *Análisis de la cadena de valor del maíz y la soya y estrategias de intensificación sustentable para reducir las emisiones por deforestación y degradación del bosque en la Península de Yucatán*. Agencia de los Estados Unidos para el Desarrollo Internacional (USAID), Proyecto México para la Reducción de Emisiones por deforestación y degradación (M-REDD+), The Nature Conservancy, Rainforest Alliance, Woods Hole Research Center, Espacios Naturales y Desarrollo Sustentable. 146 p. [https://pdf.usaid.gov/pdf\\_docs/PA00W6ND.pdf](https://pdf.usaid.gov/pdf_docs/PA00W6ND.pdf)
- Toghdory, A., Torbatinejad, N., Kamali, R., Mohajer, M. and Chamani, M., 2009. Effects of propylene glycol powder on productive performance of lactating cows. *Pakistan Journal of Biological Sciences*, 12, pp. 924–928. DOI: 10.3923/pjbs.2009.924.928.
- Trabue, S., Scoggin, K., Tjandrakusuma, S., Rasmussen, M. A. and Reilly, P. J., 2007. Ruminal fermentation of propylene glycol and glycerol. *Journal of Agricultural and Food Chemistry*, 55, pp. 7043–7051. DOI: <https://doi.org/10.1021/jf071076i>
- UAA, 2019. *Regulations for the use of animals in teaching and research at the Autonomous University of Aguascalientes*. In: University legislation. 8th ed. Aguascalientes; 2019. p. 1–11. <https://www.uaa.mx/institucion/LEGISLACION%20UNIVERSITARIA.pdf>
- Vázquez, S. E. T., Partida, P. J. A., Rubio, L. M. S. and Méndez, M. D., 2011. Productive performance and carcass characteristics in lambs from crosses between Katahdin ewes and rams from four specialized meat breeds. *Revista Mexicana de Ciencias Pecuarias*, 2, pp. 247–258. <https://cienciaspecuarias.inifap.gob.mx/index.php/Pecuarias/article/view/1439/1434>
- Yazdi, M. H., Amanlou, H., Mirzaei-Alamouti, H. R., Harkinezhad, M. T., Nabipour, A., Mahjoubi, E., Aghaziarati, N., Noori, G. R. and Baumgard, L. H., 2015. Effects of a supplement containing multiple types of



gluconeogenic precursors on production and metabolism in Holstein bull calves during heat stress. *Livestock Science*, 178,

pp. 61–70. DOI:  
<https://doi.org/10.1016/j.livsci.2015.05.001>