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Technical note

DL-malic acid supplementation improves the carcass characteristics of finishing Pelibuey lambs

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Abstract:

The aim of the present study was to evaluate the effect of DL-malic acid addition in Pelibuey finishing diet on average daily gain, carcass characteristics and non-carcass components. Sixteen (16) male lambs with a mean body weight of 27 ± 1.92 kg, were used in a 48-d feeding experiment. Animals were fed a high-energy diet containing corn stover, as the only forage source, with and without the DL-malic acid (MA) addition. Animals were assigned randomly in two treatments with eight lambs each: 1) Addition of

4 g of DL-malic acid per kg of feed and 2) Control (diet 1 without MA). Four male lambs of each treatment were harvested after feeding experiment to measure carcass characteristics and non-carcass components. Lambs fed with MA presented a larger (P<0.05) *Longissimus lumborum* muscle area. Nevertheless, there were no effects (P>0.05) of MA on daily average weight gain and non-carcass components weight. In conclusion, addition of 4 g DL-malic acid to a high-energy feed enhances muscle accretion, which improves carcass quality of finishing lambs.

Key words: Malic acid, Animal performance, Carcass characteristics.

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Meat production is the main purpose[1] of ovine farms in many regions of the world. While ovine meat quality can be improved under an intensive feed system utilizing high grain diets[2], feed costs and ruminal disorders, mainly acidosis, increase. It is necessary to search for alternative feeding strategies to enhance the energy efficiency of rations and prevent metabolic disorders. Studies on malic acid (MA) have reported its ability to stimulate lactate utilization by *Selenomonas ruminantium*[3], which can account for up to 51 % of the total viable bacterial count in rumen[4,5]. MA has also been shown to cause increased pH[6], microbial protein[3] and total volatile fatty acid production[7]. Increases in pH, total volatile acids and propionic acid had been observed utilizing ground corn and soluble starch to feed microorganisms *in vitro*[8] since the hydrogen is utilized to convert malic acid to propionate[9]; decrease in hydrogen availability reduce the methane production[10].

Results obtained with MA supplementation are not constant[11]. The roughage and concentrate proportions in rations influence the success of this additive[12], with more favorable results being found with diets containing lower levels of forage[13] in which MA is naturally present[14,15]. Limited *in vivo* research has been conducted to evaluate the effects of MA on ruminant performance[9,16]. Therefore, researchers recommend further studies to test its effects on lamb performance[2].

Regarding with the influence of MA on carcass characteristics, there is evidence of an increase of hot carcass yield due to a greater average daily gain with malic acid inclusion in the concentrate of cross male lambs[17], but, some authors have not found effect on carcass yield of heifers[18]. The authors of this study are not aware of information relating to the effects of DL-malic acid on the productive performance and carcass characteristics of Pelibuey, a hair sheep breed (HSB).

The number of HSB has increased in several Latin American countries due to their ease of management and their resistance to parasites(19) and elevated environmental temperature and humidity. However, HSBs have presented lower daily gain and poorer meat quality than wool breeds(20). It should be noted that consumer demands for lean meats are increasing(21). The objective of this study was evaluate the effect of DL-malic acid addition in the concentrated finishing diet of Pelibuey male lambs on average daily gain, carcass characteristics and non-carcass components.

Animal management procedures were conducted within the guidelines of locally approved techniques for animal use and care (NOM-051-ZOO-1995; humanitarian care of animals during mobilization of animals; NOM-062-ZOO-1995: technical specifications for the care and use of laboratory animals. Livestock farms, farms, centers of production, reproduction and breeding, zoos and exhibition hall must meet the basic principles of animal welfare; NOM-024-ZOO-1995; animal health stipulations and characteristics during transportation of animals.

This experiment was conducted at a commercial Pelibuey farm called Los Limones, located in Nayarit, Mexico (21° 03' 48.11" N and 104° 31' 34.76" W). Sixteen (16) male lambs with a mean body weight of 27 ± 1.92 kg were used. Animals were divided randomly into two treatment groups of eight lambs each and placed in elevated pens with plastic slat flooring equipped with shade, feed and automatic waterers, with the two treatments comprising the addition of 4 g of DL-malic acid (MA) per kg of feed, and the control (the diet from the first treatment but without additive). The DL-Malic acid was purchased from the Sigma-Aldrich Chemical Company (St. Louis, MO, USA). Animals were fed *ad libitum* once daily at 0800 h. The first 7 d were used for adaptation to treatment followed by a 48 d trial. The composition and ingredients of the experimental diets are shown in Table 1. Basal diet was prepared weekly. Batch was divided in two parts, and MA was added to one part; mixing MA with 20 kg of feed followed by the incorporation with the rest of the feed.

Table 1: Ingredient and chemical composition of the experimental diets[1]

Ingredient, %	Control[2]	MA[2]
Cracked corn	12.48	12.43
Sorghum grain	53.05	52.84
Canola meal	9.82	9.78
Soybean meal	4.91|	4.89
Cane molasses	7.66	7.63
Corn straw	6.48	6.46
Minerals	2.95	2.94
Calcium	0.98	0.98
Sodium bicarbonate	0.69	0.68
Grease	0.98	0.98
DL-malic acid	-	0.39
Chemical composition, %		
Dry matter	88.70	89.09
Crude protein	14.01	13.95
NE g, Mcal/kg	1.14	1.13
NE m, Mcal/kg	1.77	1.76
Crude fiber	4.12	4.10
Neutral detergent fiber	16.44	16.37
Acid detergent fiber	5.76	5.74
Ether extract	3.88	3.87

[1]Expressed on a dry-matter basis.

NE g = Net energy of gain.

NE m = Net energy of maintenance.

The daily feed allotments for each pen were adjusted to allow for minimal (<5 %) feed refusal at the feed bunk. Daily adjustments were undertaken to either increase or decrease daily feed delivery and weekly intake was recorded. One sample per week of the feed that had been offered to the animals was collected in order to determine dry matter content (DM, method 930,15) in accordance with the AOAC(22).

Initial, weekly and final body weights were obtained after the animals' morning meal using an electronic scale (TOR REY TIL/S:107-2691, TORREY electronics Inc, Houston TX, USA). Body weight gains were calculated by subtracting the previous weight from the current weight, while the average daily body weight gains were calculated by dividing body weight gain by the number of days that had passed since the last weighing.

Four male lambs for each treatment were harvested in an ovine slaughterhouse (Asociación de Ovinocultores del Centro de Nayarit) after feeding period. Hot carcass weight was recorded immediately after animals were slaughtered. The carcasses were then covered with plastic to avoid cooling loss, and chilled for 24 h at -4 °C. Carcasses were ribbed between the 12th and 13th rib and back fat thickness measured with a metallic rule, with the *Longissimus lumborum* muscle then drawn on an acetate sheet in order to obtain the *Longissimus* muscle area (LMA) using a plastic grid.

Data was analyzed using a completely randomized design. The significance of the differences (P<0.05) between treatment means were determined using the Student t-test for independent samples utilizing the SPSS software[(23)].

The productive performance of finishing male lambs is shown in Table 2. In this study, lambs supplemented with DL-malic acid (MA) had similar (P>0.05) average daily gains, feed intake, feed efficiency and initial and final body weight compared to the non-supplemented ones.

Table 2: Productive performance of finishing Pelibuey male lambs

Variable	Control[1]	MA	SEM	*P*-value
Days of feed	48	48		
Male lambs, n	8	8		
Initial BW, kg	27.80	25.90	1.92	0.69
Final BW, kg	38.25	38.50	0.94	0.75
DMI, g/d	983	1022	64.75	0.69
ADG, g/d	218	263	58.05	0.87
FE, g/g	0.233	0.247	0.6	0.89

[1] Control = no malic acid supplementation.

MA= 4 g of malic acid supplementation/kg of feed; SEM= Standard error of the mean.

BW= Body weight, DMI= dry matter intake; ADG= average daily gain; FE= feed efficiency.

The influence of MA on the lambs' carcass characteristics is presented in Table 3. The addition of malic acid to male finishing diets did not alter (P>0.05) hot carcass weight, back fat thickness (FT), or dressing percentage. *Longissimus* muscle area (LMA) was significantly (P<0.01) larger in supplemented animals, while the MA did not affect non-carcass components (Table 4).

Table 3: Carcass characteristics of Pelibuey male finishing lambs

Variable	Control[1]	MA	SEM	*P*-value
HCW, kg	21.5	21.3	0.67	0.87
Dressing[2], %	56.2	55.3	0.81	0.23
LL area, cm^2	11.8	13.0	0.42	0.002
Fat thickness, cm	0.105	0.125	0.03	0.67

[1] Control = no malic acid supplementation.

MA= 4 g of malic acid supplementation/kg of feed; SEM= standard error of the mean.

HCW= hot carcass weight; LL= *Longissimus lumborum* muscle.

[2] Dressing = (HCW/Final weight)*100.

Table 4: Non-carcass components of Pelibuey male finishing lambs

Variable[1]	Control[2]	MA	SEM	*P*-value
Skin	11.04	12.16	0.52	0.08
Feet	2.15	2.19	0.66	0.77
Heart	0.48	0.51	0.05	0.60
Liver	2.25	2.26	0.15	0.92
Lungs	1.99	2.01	0.90	0.15
Spleen	0.30	0.38	0.01	0.06

[1] Weight of each non-carcass component is expressed as a percentage of final body weight.

[2] Control = no malic acid supplementation.

MA = 4 g of malic acid supplementation/kg of feed; SEM= Standard error of the mean.

Feed intake was similar for both groups 0.983 and 1.022 kg for the control and supplemented animals, respectively. Results of other authors show that malic acid does not alter the DMI of finishing lambs(2), lactating Pelibuey ewes(24), dairy goats(15), dairy cows(8,25,26), and feedlot cattle(27). Some authors mention that doses of MA lower than 2.6 % do not affect DMI(28). In this experiment, a 0.4 % MA level was evaluated because similar doses were used in previous research conducted with both the same(2,24) and different breeds of sheep(11).

Average daily gains (ADG) observed with MA corresponded to the mean weight for Pelibuey lambs fed high energy diets(29,30). The null effect of MA on ADG in this experiment is in agreement with other reports on the MA supplementation of lambs(11;31) and bull calves(8) fed with a high concentrate diet. However, some studies have reported the positive effect of MA on the daily live weight gain of cattle fed with a high concentrate diet(16). Malate content varies with plant age (mature < early) and plant type (gramineous < legumes)(32). Therefore, a significant ADG increase was expected since lambs were fed with a high concentrate diet comprising only corn straw (which is low in malate content) as source of forage. Incorporation of DL-malate into soluble starch and cracked corn fermentations with mixed ruminal microorganisms have modify final pH, CH_4, and volatile fatty acids (VFA) in a manner analogous to ionophore effects[33]. However, according to this experiment, as other factors such as days on feed may influence the effects of MA, not only the concentrate forage ratio or type of grain should be criteria for the positive effects of this additive on ADG. The degree to which MA effects ADG may depends on diet composition[11], in terms of grain type, dosage, chemical form (salt or free acid) or the productive stage of the animal[8]. MA levels ranging from 0.6 to 1.1 % have enhanced the ADG of steers fed with high energy corn-based diets[34]. Also, steer ADG have increased linearly as more DL-malate was added to high energy diets based on rolled corn[16]. This study used a lower level of MA than 0.06 %[34], due to the favorable results obtained in a previous experiment on lactating Pelibuey ewes fed with a similar diet[24]. It is likely that significant differences could be found by prolonging the feeding experiment because ADG differences among treatments tended to be higher the more feeding days passed. However, in the region of Mexico where this study was conducted, retail customers, looking for leaner carcasses, prefer animals with a final body weight (FBW) of between 35 and 40 kg.

Hot carcass weight (HCW) and dressing percentage were not influenced by MA supplementation. Similarly, some researchers reported null effect of malate on HCW, cold carcass weight, and dressing percentage when it was added to a Merino lamb diet at 0.4 and 0.8 %[11].

The MA supplementation of finishing male lambs increased (P=0.002) *L. lumborum* muscle area (LMA). Greater LMA is associated with higher yield and wholesale cuts of the carcass[35]. Malate supplementation have increased nitrogen retention in sheep and steers[34]. Higher muscle growth may be attributed to an increase of microbial protein production[3,36], or to the high availability of propionate converted from the added MA as it 'sinks' H_2 when reducing methanogenesis in rumen[7,37]. Both high nitrogen and propionate levels in rumen could increase muscle size firstly by depositing more nitrogen directly into the tissue, and secondly through the higher level of alanine bioavailability produced by the propionate metabolism through gluconeogenesis[38]. Moreover, higher amounts of propionate lead to the hypertrophy of intramuscular adipocites[39] and bovine muscle[40]. The use of propionate, a primary precursor for gluconeogenesis as energy for production has been documented mainly for milk synthesis. A significant effect of MA has been found in the milk protein yield from early lactation Pelibuey ewes[24], early lactation dairy cows[28], and mid lactation dairy cows[25]. This effect is attributed to an increase in microbial efficiency resulting from increased carbohydrate use for microbial N production[25]. However, propionate could also be used for higher muscle gain, as proposed in this study. Furthermore, percentage corresponding to each tissue may vary considerably among carcasses of similar weight, depending on the breed and type of feed[41].

Subcutaneous fat thickness (FT) was not different between treatments. It is likely that MA did not affect subcutaneous fat in Pelibuey sheep because hair sheep breeds deposit a small amount of subcutaneous fat[42] and, most importantly, because the animals were harvested young when the fat deposition in pre-formed subcutaneous adiposities is not complete. In this case, higher amounts of ruminal propionate and, subsequently, glucose in the tissue does not result in the formation of higher levels of subcutaneous adiposities, as occurs with intramuscular adiposities(39). Typically, body fat increases as harvest weights increase(43,44). Thicker subcutaneous fat in Pelibuey lambs and a heavier final weight (>43 kg) have been reported(45).

Smooth muscle growth was not affected by MA supplementation, with non-carcass components, expressed as a percentage of final body weight, similar for both treatments. These research results are in agreement with others authors, who found similar increases in total splanchnic tissue across the finishing phase that were consistent with similar rates of live and carcass gain observed in other studies(46). There is a negative relationship between carcass residues (organs and offals) and carcass yield(47). The weight of organs such as gastrointestinal tract and liver decrease during subnutrition periods(48). Therefore, bigger differences in nutrients intake would be necessary to influence the weight of organs.

DL-malic acid supplementation (4 g per kilogram of feed) in finishing male Pelibuey lambs does not significantly improve average daily gain. While DL-malic acid supplementation does improve the *Longissimus* muscle area, it does not affect non-carcass components. Larger *Longissimus* muscle area could have a positive economic impact since it implies higher muscle proportion.

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