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## Effect of adding *Salix babylonica* Extracts and Exogenous Enzymes to Basal Diets on the Meat Quality of Growing Suffolk Lambs<sup>#</sup>

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### ABSTRACT

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It was evaluated the effect of adding *Salix babylonica* L. extracts and exogenous enzymes in combination or individually on meat quality in growing Suffolk lambs. Animals were divided into 4 groups of 4 animals each in a completely randomized design (CRD). Treatments were: (i) Control: basal diet of concentrate (30%) mixture and corn silage roughage (70%); (ii) EZE (exogenous enzymes): basal diet plus 10 g of enzyme (Zado<sup>®</sup>); (iii) SB (*Salix babylonica*): basal diet plus 30 ml of *S. babylonica* extracts, and (iv) EZESB (exogenous enzymes + *Salix babylonica*): basal diet plus 10 g enzyme and 30 ml of *S. Babylonica* extracts. Lambs were housed in individual cages for 60 days. Extracts were dosed orally while EZE was mixed with concentrate. At the end of the trial, lambs were slaughtered and *Longissimus dorsi* samples were analysed. Samples were analysed for CP, CF, ash and DM. Meat quality parameters included color, pH, carcass temperature and kidney fat. No significant differences for live weight, chemical composition, as well as hot carcass weight and cold, initial and final temperature and kidney fat. Meat lightness (variable L \*) and pH<sub>f</sub> were improved (P<0.05) with EZESB treatment compared to the other three treatments, and the most optimal pH<sub>f</sub> four treatments being the most acidic. Lambs fed SB or EZE were not different from the control. In conclusion, a combined administration of EZESB to the basal diet improves meat quality by reducing the pH and increasing its lightness when compared to either EZE or SB, individually.

**Key words:** Exogenous enzymes, Lambs, Meat quality characteristics, *Salix babylonica* extracts

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## INTRODUCTION

Globally, it is recognized that the cost of feeding livestock accounts for over 50% of total production costs, thus there is a constant search for alternative feeding methods that will allow for optimum growth and performance with minimum economic investment. Some of the alternative production methods developed to date include the use of pre and probiotics, ionophores, exogenous enzymes and even local fodder shrubs and trees. Use of foliage from local resources has helped in some areas to reduce costs of production considering that most of these trees grow naturally and there are virtually no management costs associated with their production. In Mexico, the search for these alternatives production/feeding methods has become relevant. Studies performed in Egypt have reported some profitable alternative methods preparing animal feeds (Salem *et al.*, 2006, 2007, 2010). Research conducted with local trees and shrubs and extracts from the same species as additives indicated they represent an important potential in feeding ruminants in arid and semi-arid regions of Northern Egypt.

Addition of fodder tree and shrub extracts to the basal diet of sheep have resulted in positive effects in performance, nonetheless, the addition of exogenous enzymes might prove further improvements in sheep nutrition practices. Utilization of tree/shrub extracts have promoted fiber digestion (non-starch polysaccharides), reduced viscosity of the bolus, increased the bioavailability of nutrients, inhibited the growth of pathogens bacteria and enhanced the growth of probiotic intestinal flora by the hydrolysis of non-starch which allow a better use of the energy value of cereals. Moreover, plant extracts tend to reduce losses and excretion of certain compounds such as phosphorus and nitrogen, reducing negative environmental impacts. Furthermore, they prevent diarrhea and digestive disorders and as a result improves growth performance, reproductive, and animal immune system. Gado *et al.* (2007) indicated that supplementing dairy and beef cattle with exogenous improved fiber degradation, such information was further supported in lambs through *in vitro* and *in vivo* studies (Gado and Salem, 2008). Meat quality evaluation in sheep supplemented with extracts and exogenous enzymes ought to be performed. Bianchi (2007) mentioned that an important attribute for the consumer while buying meat was color, however, other features such as pH, water holding capacity, flavour, smell and fatty acid concentration are also considered, the later being an important issue in human health due to cardiovascular disease concerns. Previous work have indicated positive impacts while using *Salix babylonica* extracts on productive parameters, whereas favourable properties have also been determined by addition of exogenous enzymes (Zado®), thus, this study was conducted to evaluate the meat quality response of these two additives added individually or in combination hoping for positive effects on meat quality of Suffolk lambs.

## MATERIALS AND METHODS

The research was conducted at the premises of the zoo technical post at the Faculty of Veterinary Medicine of the Autonomous University of the State of Mexico.

### *Salix babylonica L. extracts and exogenous enzymes in growing lambs*

Extract preparations and quality meat evaluations conducted at the Food Science Laboratory of Animal Nutrition of the Faculty.

#### *Preparation of extracts*

The extraction of tree/shrub (SB) was conducted according to Salem (2006), where leaves of *S. babylonica* were collected randomly from several young and mature trees during Summer. The fresh leaves were chopped into 1-2 cm lengths and immediately extracted with a prepared solvent mixture (1 g leaf/8 ml). The extraction mixture contained 10 ml methanol (99.8%, analytical grade, Fermont®, Monterrey, Mexico), 10 ml ethanol (99% analytical grade, Fermont®, Monterrey, Mexico) and 80ml distilled water. The chopped leaf materials were individually soaked and incubated in the solvent mixture at laboratory room temperature at 25-30°C for 48 to 72 hrs in closed black/dark 500 ml bottles. After incubation, jars were heated at 39°C for 1 h, and then immediately filtered after which the filtrates were collected and stored at 4°C for further use.

#### *Animals and treatments*

Sixteen growing male lambs (4 animals in each treatment) (28 kg BW) and from 7 to 8 months were used. Animals were fed a basal diet consisting of 70% corn silage and 30% commercial concentrate (Purina®) for sixty days. Lambs were provided throughout the study with feed and drinking water *ad libitum*.

Treatments were: (i) Control: basal diet of concentrate (30%) mixture and corn silage roughage (70%); (ii) EZE (exogenous enzymes): basal diet plus 10 g of enzyme (Zado®; commercial product from El Cairo, Egypt). The enzyme treatment was prepared by mixing 10 g of enzyme complex with 200 g concentrate mixture fed to each animal; (iii) SB (*Salix babylonica*): basal diet plus 30 ml of *S. babylonica* extracts; lambs were drenched with the extract directly in the mouth, and (iv) EZESB (exogenous enzymes + *Salix babylonica*): basal diet plus 10 g enzyme and 30 ml of *S. Babylonica* extracts. The experiment was performed during 60 days.

#### *Measurements and instrumentation*

Temperature of hot carcass and pH was recorded on carcass immediately after slaughter and at 24 hours post-mortem. Samples from the *Longissimus dorsi* muscle were collected from 12, 13, 14, 15<sup>th</sup> ribs at 24 hours post-mortem and stored in a freezer for later analysis.

pH Measurements: The determination of pH and temperature of carcass were performed using a pH meter that was equipped with a penetration electrode (HANNA HI99163). Measurements were taken on the *Longissimus dorsi* muscle (at the last rib). pH was measured by electrode penetration at 45 min after slaughter (time taken to reach carcass) and at 24 h.

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#### Chemical composition analysis

Samples stored in the freezer, which were taken from the ribeye area of the 12<sup>th</sup> and 13<sup>th</sup> rib, were thawed at 4°C for 24 hours and immediately analysed for moisture content, crude protein, ether extract and ash, according to the AOAC (1990).

#### Kidney fat scoring

The amount of fat covering the kidneys were scored on a scale of 1 to 4, where 1 was for Kidneys that were completely uncovered 2: Kidneys with large a uncovered window, 3: Kidneys with a small uncovered window and 4 for kidneys that were completely covered in fat. The methodology used for scoring renal fat coverage was according to Delfa *et al.* (1995).

#### Colour scoring

To measure the colour of meat from the various treatments, frozen meat chops were first thawed at 3°C, measurements were performed using a Minolta Chroma Meter CR-measuring head 400, calibration plate CR.A43, viewing angle 20°, according to Brewer (1999) and Honikel (1998).

#### Statistical analysis

Data were analysed according to a completely randomized design, with 4 treatments and 4 replications for each treatment. The statistical model used was:  $Y_{ijk} = m + t_i T_i + E_{ijk}$ , where  $m$  is the mean,  $T_i$  as the treatment effect and  $E_{ijk}$  as the residual error effect. Data was analysed using the GLM procedure of SAS (2002). Tukey's test was used for mean comparisons (Steel and Torrie, 1980).

## RESULTS

In Table 1, initial pH values (pHi) were not affected by dietary treatments ( $P > 0.05$ ). However, for the final pH (pHf), animals offered the SB had lower values ( $P < 0.05$ ) compared to those registered in lambs fed the control and the EZE treatments. Initial and final meat temperature values were not different among treatments ( $P > 0.05$ ).

Table 1. The effect of adding *Salix babylonica* extracts (SB), exogenous enzymes (EZE) or the mixture of the two (EZE-SB) on pH and the carcass temperature in sheep

Variables	Treatments				LSD	P
	Control	EZE	SB	EZE-SB		
pHi	6.87	6.83	6.80	6.73	0.300	0.572
pHf	6.51 <sup>a</sup>	6.53 <sup>a</sup>	6.46 <sup>ab</sup>	6.25 <sup>b</sup>	0.226	0.010
Ti	20.27	21.36	20.55	20.82	2.226	0.556
Tf	8.82	8.23	8.00	8.02	1.206	0.174

pHi: pH initial, pHf: pH final, Ti: initial temperature, Tf: final temperature, EZE: exogenous enzymes, SB: *Salix babylonica*, EZE-SB: exogenous enzymes + *Salix babylonica*

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The meat production parameters are shown in Table 2. No significant differences were registered among treatments in all the studied variables. Nonetheless, some numerical trends in BLW, HCW, CCW and KF were observed. Highest record variables regarding weight were registered in animals fed EZE treatment, followed by EZE-SB, SB and Control. Kidney fat was slightly increased in lambs fed EZE than other treatments. This being one of the parameters that give the conformation pattern of sheep body fat, thus supporting the fact that EZE-SB can be considered an suitable treatment.

Table 2. The effect of adding *Salix babylonica* extracts (SB), exogenous enzymes (EZE) or the mixture of the two (EZE-SB) on some production parameters in sheep

Variables	Treatments				LSD	P value
	Control	EZE	SB	EZE-SB		
BLW	34.47	40.13	36.85	37.37	15.656	0.775
HCW	16.30	18.96	16.00	17.47	9.145	0.783
CCW	15.92	18.50	15.50	17.17	9.389	0.791
KF	3.50	3.66	3.50	3.25	1.234	0.797

BLW: Body live weight, HCW: hot carcass weight, CCW: cold carcass weight, KF: kidney fat, EZE: exogenous enzymes, SB: *Salix babylonica*, EZE-SB: exogenous enzymes + *Salix babylonica*.

Meat chemical analysis are shown in Table 3. There were no significant effects of dietary treatments on the studied variables. Average CP values were 17%, while lambs fed EZE had a slightly higher value (17.8%), whereas the control group registered the lowest (17.2%). Regarding fat content, administration of ENZ in lambs resulted in decreased ( $P < 0.05$ ) fat content by 3.9% than other treatments.

Table 3. The effect of adding *Salix babylonica* extracts (SB), exogenous enzymes (EZE) or the mixture of the two (EZE-SB) on parameters of chemical composition of sheep meat

Variables	Treatments				LSD	P value
	Control	EZE	SB	EZE-SB		
CP	17.20	17.81	17.47	17.26	1.491	0.654
Fat	7.15	3.96	5.78	5.60	3.735	0.164
Ash	0.58	1.21	0.86	0.98	0.978	0.345
DM	25.32	25.45	24.80	24.93	2.699	0.871

EZE: exogenous enzymes, SB: *Salix babylonica*, EZE-SB: exogenous enzymes + *Salix babylonica*.

Meat color was significantly different ( $P < 0.05$ ) for lightness among treatments ( $L^*$ ), with lambs fed EZE-SB having the highest value when compared to SB, Control and EZE treatments. For the red ( $a^*$ ) and yellow ( $b^*$ ) colours, there were no significant differences among treatments. However, a numerical trend was recorded for the various variables of  $L^*$ ,  $a^*$  and  $b^*$ , the lamb fed EZE-SB, which was relatively higher when compared with that of other treatments.

Table 4. The effect of adding *Salix babylonica* extracts (SB), exogenous enzymes (EZE) or a mixture of the two (EZE-SB) on color of sheep meat

Variables <sup>†</sup>	Treatments				LSD	P
	Control	EZE	SB	EZE-SB		
L	34.84 <sup>bc</sup>	34.34 <sup>c</sup>	41.42 <sup>ab</sup>	44.36 <sup>a</sup>	6.708	0.0019
a	12.49	13.58	14.05	15.67	3.481	0.0888
b	4.08	4.80	4.62	5.59	2.0723	0.2072

<sup>†</sup>L: lightness, a: red colors, b: yellow colors

EZE: exogenous enzymes, SB: *Salix babylonica*, EZE-SB: exogenous enzymes + *Salix babylonica*.

## DISCUSSION

Values obtained in this study regarding pH are not in accordance with Hernández *et al.* (2009) and Torrescano *et al.* (2009) who reported initial (6.1 and 6.3) and final (5.8 and 5.5) values, respectively; in Pelibuey sheep. Lawrie (1998) reported that sheep meat has a slightly higher pH than other species. However, results herein are slightly elevated compared to other data in sheep. Accordingly, Ramírez *et al.* (2007) indicated a normal range of 5.8-6.2 for pH values in sheep. In addition, it is recognized that sheep are less susceptible to stress ante-mortem factors which might have contributed to these final pH values. Moderate acidic pH values are known to prevent microbial contamination which favours the preservation of meat. Thus lambs fed EZE-SB with mildly acidic conditions compared to the other treatments are to prove a more favourable condition. It is well known that the final pH as it approaches the protein's isoelectric point (pH 5.0-5.5) will produce gradual decrease in water retention. López (1987), reported that the maximum amount of juice is expelled with higher values of pH in muscles.

Similar results in terms of CP content in Suffolk lamb meat were reported by Pérez *et al.* (2002), with average values being 17% and 18% CP for males and females, respectively.

Results obtained herein regarding meat color are not in agreement to the values reported by Germano *et al.* (2009) who mentioned values for L\* of 27.73, which is low compared to our results. Similarly, data from this study is not in accordance to Costa *et al.* (2008) who while performing studies with Nova, Santa Ines and Dorpper x Santa Ines sheep breeds reported values for L\* (26.46, 28.99 and 28.00 respectively), for a\* (average =12.22) and for b\* (average=18.00). The pH has a close relationship with meat color, high pH in meat results in low luminosity (L\*). It has been indicated (Hopkins *et al.*, 1995) that there is a significant negative correlation between L\* and b\* values and muscular pH, which is in accordance to our study, where treatments with high pH yielded low degrees of lightness (L\*) such as in groups fed Control and EZE, whereas low pH treatments yielded higher degrees of brightness, as SB and EZE-SB. Accordingly, (EZE-SB). had low pH against higher lightness (L\*). In addition, Sañudo

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*et al.* (1998) indicated differences in the variable L\* for sheep meat with females and males 39.80 and 41.26, respectively.

## CONCLUSION

The combination of SB extract enzymes as additives improved meat pH and color compared to the control or individual additives which resulted in a more favourable color and might contribute to a better conservation profile.

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