



## Influence of Zilpaterol and Mineral-Yeast Mixture on Ruminal Fermentation and Growth Performance in Finishing Steers

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### Abstract

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*To evaluate the effects of zilpaterol hydrochloride (ZH) and a mineral-yeast (MY) mixture two experiments were conducted on finishing steers. Four treatments were: 1) control; 2) MY (chromium, selenium and zinc yeast); 3) ZH; 4) YM+ZH. 125 and 800 g/ton of Zilmax<sup>®</sup> (ZH) and Bioplex<sup>®</sup> (MY), respectively, were supplemented to the total mixed ration (TMR). Four beef steers (420±32 kg) fitted with ruminal cannulae and 20 BrahmanxSwiss beef steers (375±6 kg BW) divided in four equal groups were used in the two experiments, respectively. Ruminal pH, volatile fatty acids, ammonia N and DM degradability were not affected ( $P > 0.05$ ) by treatments. After 40 d of feeding ZH alone or with MY improved average daily gain (ADG) and feed conversion (F:G). Dressing percentage (hot and cold) were higher ( $P < 0.05$ ) in supplements fed steers and the highest values were observed with MY+ZH. Our results suggested a beneficial synergetic effect of zilpaterol hydrochloride (ZH) and mineral-yeast (MY) mixture on growth, feed efficiency and carcass characteristics of finishing steers.*

Key words: Zilpaterol, mineral-yeast, growth, ruminal fermentation, steers, carcass characteristics, total mixed ration.

### Introduction

Various attempts are being made to reduce fat content and increase protein content in red

meat using  $\beta$ -agonists (e.g. Zilpaterol hydrochloride, Bell *et al.*, 1998) or some mineral supplements such as high-chromium yeast, chromium picolinate, high-selenium yeast, zinc proteinate, zinc methionine and high-zinc yeast in finishing steer (Boleman *et al.*, 1995; Spears and Kegley, 2002; Lawler *et*

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*al.*, 2004; Juniper *et al.*, 2006). Because supplementation of yeast have shown beneficial effects on dry matter intake and nutrient digestibility in ruminants (Wohlt *et al.*, 1991), trace minerals in the form of mineral yeast may have an impact on feed intake, ruminal fermentation and its digestibility. A synergetic effect of zilpaterol hydrochloride (ZH) with mineral-yeast mixture (MY) has been speculated to improve growth performance and lean carcass in finishing steers. Therefore, this study was conducted to evaluate the effects of ZH and a MY mixture of chromium, selenium and zinc supplementation on rumen fermentation and degradability, as well as on growth and carcass characteristics in finishing steers.

### **Materials and Methods**

Two experiments were conducted under the supervision and approval of the Academic Committee of Livestock Science Department of "Colegio de Postgraduados, Campus Montecillo, and the Instituto de Investigación de Zonas Desérticas of the Universidad Autónoma de San Luis Potosí", according to regulations established by the Animal Protection Law, enacted by the Estado de México and San Luis Potosi state in México. In experiment 1, four cross bred steers (ZebuxSwiss, 420±32 kg) fitted with ruminal cannulae were randomly assigned to the following four treatments: 1) control basal diet consisting of corn grain (320), sorghum grain (240), barley straw (175), rice hulls (120), canola meal (40), molasses cane liquid (75), mineral premix (15), urea (10) and salt (5 part per kg); 13% CP, 25% NDF, 15% ADF; 2) basal diet supplemented with 800g mineral-yeast (MY, chromium-yeast, 233 mg/kg, selenium-yeast, 117 mg/kg, zinc methionine, 23 g/kg; bioplex, quality meat, Alltech Inc., Nicholasville, KY, USA) mixture per ton of feed; 3) basal diet supplemented with 125g zilpaterol hydrochloride per ton of feed (ZH, Zilmax, Intervet México S.A. de C.V., México) and 4) basal diet supplemented with MY+ZH. Experimental periods were 15 d for adaptation and 5 d for sample collection and

ruminal *in situ* incubation. *In situ* DM disappearance (Vanzant *et al.*, 1998), volatile fatty acid (VFA) concentration (Erwin *et al.*, 1961) and ammonia N (NH<sub>3</sub>N; McCullough, 1967) were determined. Analysis of ruminal DM kinetic was estimated using the Gompertz model of Susmel *et al.* (1999).

In experiment 2, twenty BrahamanxSwiss steers (375±6 kg) were housed in dry lot pens and randomly assigned to the four treatments described in experiment 1. Experimental period was 100 d, but Zilmax was added from 40 to 80 d. Dry matter, CP (ID: 954.01), ash were determined according to AOAC (1990) and NDF, ADF (Van Soest *et al.*, 1991). Animals were weighed after every 20 d while feed intake and refusal were recorded daily. Post-slaughter hot and chilled (0C,24 h) carcass weight were recorded in an authorized slaughter house (Los Reyes, Edo. México).

Data were analyzed using the 'GLM'/ 'MIXED' option of SAS (1999) and 'LSMEANS' (SAS, 1999) were used to compare means. A repeated measures analysis with at 'PROC MIXED' (Littell *et al.*, 1998) in SAS (1999) was used for continuous data collected over time.

### **Results and Discussion**

All ruminal fermentation activities, e.g. pH, VFA, NH<sub>3</sub>-N and ruminal DM degradability were not affected ( $P > 0.05$ ) by supplementation of MY, ZH or MY+ZH as compared to control diet (Table 1). There are no previous reports about MY effects on ruminal fermentation and DM degradability, but there are evidences that high levels of Zn supplementation may (Arelovich *et al.*, 2000) or may not alter (Mandal *et al.*, 2007) ruminal pH, NH<sub>3</sub>-N, nutrient digestibility and the population of specific ruminal microbes and production of different VFAs. Probably inorganic minerals are more readily reduced within the rumen than mineral-yeast (Koenig *et al.*, 1997) and, therefore, no impact on ruminal variables was observed. These results agree to those of Mwenya *et al.* (2005) and Cooke *et al.* (2007), who also did not find any positive effect of yeast

Table 1  
Ruminal pH, ammonia N (mg/dl), volatile fatty acids (VFA) and ruminal DM degradability of diets with zilpaterol hydrochloride (ZH) and mineral-yeast mixture (MY) in finishing steers

	Treatments				SEM
	Control	MY	ZH	MY + ZH	
pH	5.5	5.8	5.6	5.7	0.06
Ammonia N, mg/dl	15.3	15.3	15.4	15.3	0.04
Acetate, mol/100 mol	64.2	64.3	64.5	64.1	0.04
Propionate, mol/100 mol	21.6	21.5	21.2	21.6	0.05
Butyrate, mol/100 mol	14.2	14.2	14.3	14.3	0.05
Total VFA, mmol/l	74.6	74.8	74.4	75.9	0.06
DM <i>in situ</i> kinetics					
Soluble fraction (a), %	35.3	33.4	35.4	34.1	3.42
Potentially degradable fraction (b), %	50.1	51.4	49.8	52.1	5.91
a + b	85.4	84.8	85.2	86.2	6.12
Potentially degradation rate (k), /h	4.4	4.2	4.6	4.0	0.49

Table 2  
Growth performance of finishing steers supplemented with zilpaterol (ZH) and mineral-yeast mixture (MY)

	Treatments <sup>1</sup>				SEM
	Control	MY	ZH	MY + ZH	
Initial BW, kg	369	380	379	372	4.20
Total LWG, kg	158 <sup>c</sup>	160 <sup>c</sup>	170 <sup>b</sup>	180 <sup>a</sup>	2.83
d 0 to 40					
Average daily gain, kg	1.50	1.52	1.54	1.53	0.09
Dry matter intake, kg	8.3	8.5	8.4	8.6	0.44
Feed:Gain	5.5	5.6	5.5	5.6	0.32
d 40 to 80					
Average daily gain, kg	1.59 <sup>c</sup>	1.61 <sup>c</sup>	1.77 <sup>b</sup>	1.94 <sup>a</sup>	0.03
Dry matter intake, kg	11.1	11.5	11.3	12.0	0.36
Feed:Gain	7.0 <sup>a</sup>	7.1 <sup>a</sup>	6.4 <sup>b</sup>	6.2 <sup>b</sup>	0.17
d 80 to 100					
Average daily gain, kg	1.64 <sup>c</sup>	1.67 <sup>c</sup>	1.80 <sup>b</sup>	1.92 <sup>a</sup>	0.02
Dry matter intake, kg	11.3 <sup>b</sup>	11.6 <sup>ab</sup>	11.4 <sup>ab</sup>	12.2 <sup>a</sup>	0.24
Feed:Gain	6.9 <sup>a</sup>	6.9 <sup>a</sup>	6.3 <sup>b</sup>	6.4 <sup>b</sup>	0.12
Overall					
Average daily gain, kg	1.58 <sup>c</sup>	1.60 <sup>c</sup>	1.70 <sup>b</sup>	1.80 <sup>a</sup>	0.10
Dry matter intake, kg	10.2	10.5	10.3	10.9	0.33
Feed:Gain	6.5 <sup>a</sup>	6.6 <sup>a</sup>	6.1 <sup>b</sup>	6.1 <sup>b</sup>	0.34

<sup>a-c</sup> means with different superscript in the same row are different (P<0.05).

Table 3  
Carcass characteristics of finishing steers supplemented with zilpaterol (ZH)  
and mineral-yeast mixture (MY)

	Treatments <sup>1</sup>				SEM
	Control	MY	ZH	MY + ZH	
Hot carcass dressing, %	54.4 <sup>b</sup>	57.9 <sup>a</sup>	58.6 <sup>a</sup>	59.6 <sup>a</sup>	0.44
Cold carcass dressing, %	53.5 <sup>b</sup>	57.0 <sup>a</sup>	57.7 <sup>a</sup>	58.6 <sup>a</sup>	0.43
Marbling score <sup>1</sup>	5.4	5.7	5.0	5.3	0.99

<sup>a-c</sup>Means with different superscript in the same row are different (P<0.05).

<sup>1</sup>4 = Small; 5 = Modest; 6 = Moderate.

culture supplementation on apparent digestion of DM or potential degradation rate of total mixed rations.

Average daily gain (ADG), dry matter intake (DMI) and feed:gain ratio (F:G) were not affected (P>0.05) by the treatments during the first 40 d (Table 2). During d 40 to 80, ADG and F:G were significantly increased (P<0.05) in steers fed MY+ZH supplemented diet than those fed MY and control steers, while the DMI was not affected by both supplementations. Our observation can probably be explained by the synergetic effect between ZH and MY on improving TMR utilization, which appeared on ZH supplementation from d 40 of experiment. In the third period (e.g. 80 to 100 d), ADG, F:G and also DMI were improved (P<0.05) with MY+ZH supplemented diet than other treatments. This effect probably reflexes the disappearance of ZH addition to TMR during the last 20 d of the experiment before slaughtering animals. However, O'Neill (2001) reported an increase of 10.4% in ADG and 15.1% in the efficiency of gain in steers consuming 6 mg/d of ZH. Contrary to above studies evaluating chromium-yeast (Swanson *et al.*, 2000), selenium yeast (Richards *et al.*, 2004), and zinc proteinate (Malcolm-Callis *et al.*, 2000) have not reported any positive effect of these forms of minerals on growth and carcass weight in beef steers.

Hot and cold dressing percentage were improved (P<0.05) with MY, ZH and MY+ZH supplementation and the highest improvement

was observed with MY+ZH diet (Table 3). Similar to above Avendaño-Reyes *et al.* (2006) reported an increase of 22 and 14 kg in hot carcass weight when steers were fed ZH or ractopamine hydrochloride compared to control steers.

Based on this study, it may be concluded that ZH and MY have a beneficial synergistic effect so far as growth, feed utilization and carcass yield are concerned. But these have no effect on rumen parameters.

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- जिल्पैटेराल हाइड्रोक्लोराइड (जेडएच) और खनिज-खमीर (एमवाई) के एक मिश्रण के प्रभाव के मूल्यांकन हेतु समापनी बधिया गौ पर दो परीक्षण किए गये। इस के चार उपचार क्रमशः 1) नियामक, 2) एमवाई (क्रोमियम, सेलेनियम और जिंक खमीर), 3) जेडएच, 4) एमवाई + जेडएच थे। संपूर्ण मिश्रित आहार में जिल्मैक्स (जेडएच) 125 ग्राम और बायोप्लेक्स (एमवाई) 200 ग्राम प्रति टन की दर से मिलाए गये। रूमेनी छिद्रित 4 और 20 ब्राह्मन x स्वीस मांसी बधिया गौ (375 ± 6 किग्रा) को 4 सम वर्गों में बांट कर दो परीक्षणों में उपयोग किया गया। उपचारों का रूमेनी पीएच, वाष्पशील वसा अम्ल, अमोनिया-नाइट्रोजन और शुष्क पदार्थ अपक्षरण पर सार्थक प्रभाव नहीं पाया गया। जेडएच या एमवाई मिश्रित जेड युक्त आहारों के प्राशन के 40 दिन पश्चात् औसत दैनिक भार वृद्धि और आहार परिवर्तन दक्षता पर सार्थक प्रभाव पड़ा। पूरक प्रदत्त वर्गों के गरम और ठंडे वधज का प्रतिशत नियामक से अधिक था एवं एमवाई-जेडएच वर्ग में सर्वाधिक था। निष्कर्ष यह निकला कि समापनी बधिया गौ के वर्धन, आहार उपयोग दक्षता और वधज गुणों पर दोनों पूरकों का योगवाही प्रभाव था।





## Effect of a Yeast Culture on Nitrogen Balance and Digestion in Lambs Fed Early and Mature Orchard Grass

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### **Abstract**

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*To evaluate the effect of feeding a yeast culture (0 or 5 g/d per sheep) on digestion and N balance in lambs fed orchard grass hay cut at 5 or 16-wk of re-growth, 4 male Criollo lambs (45 ± 4 kg) fitted with ruminal cannulae in a 4x4 Latin square with a factorial 2x2 arrangement of treatments were used. Addition of a yeast culture did not affect (P>0.05) intake, rumen pH and ammonia N, N balance and total tract digestion of nutrients. Ruminal total protozoa changed only according to orchard grass maturity (P<0.05); however, treatments did not affect total and cellulolytic bacteria, rumen pH and rumen ammonia N concentration. It is concluded that addition of a yeast culture to lambs fed early and mature orchard grass did alter ruminal protozoa without affecting feed intake, total tract digestion and N balance.*

Key words: Yeast culture, orchard grass, digestion, rumen fermentation, N balance.

### **Introduction**

Results about the effects of yeast cultures (YC) have been variable. Addition of YC did not

stimulate intake, digestibility of nutrients and fiber and N retention in lambs fed total mixed rations (Corona *et al.*, 1999). However, YC has increased total tract digestion of organic matter, N, NDF and ADF in steers (Lehloenya

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*et al.*, 2008), weight gain, feed conversion (Haddad and Goussous, 2005) and carcass traits of fattening lambs (Titi *et al.*, 2008). Orchard grass is one of the most important and economical grass sources to feed ruminants, but nutritional quality is affected by maturity due to an increase in cell-wall concentration and a decrease in cell-wall digestibility and crude protein content. Therefore, the objective of this trial is to evaluate the effects of YC supplementation to lambs fed orchard grass cut at 5 and 16-wk.

### Materials and Methods

This experiment was conducted under the supervision and approval of the Academic Committee of the Animal Science Department of Colegio de Posgraduados-Campus Montecillo in Mexico according to regulations established by the Animal Protection Law enacted by the Estado de México.

Four male Criollo lambs (45 kg  $\pm$  4) fitted with ruminal cannulae were used in a 4x4 Latin square with a 2x2 factorial arrangement of treatments: orchard grass hay (5 or 16-wk maturity) and yeast culture (0 or 5 g/d per sheep). Yeast culture (*Saccharomyces cerevisiae*<sup>1026</sup>; Yea-Sacc, Alltech Inc., Nicholasville, KY, USA) was placed daily through the ruminal cannula with the morning feeding. Orchard grass (*Dactylis glomerata* var. Potomac) grown at Campus Montecillo, Colegio de Postgraduados, México, was cut at 5 and 16-wk, sun cured and chopped.

Lambs were housed in metabolic pens in a naturally ventilated barn. There were four 15 d periods in the experiment: 10 d of adaptation, 5 d for fecal and urine sample collection and ruminal fluid samples. Lambs had free access to orchard grass hay, water and a mineral premix.

Forages, orts and feces were collected daily (d 11 to 14), sampled and analyzed for dry matter (65C; 72 h) and ash (550C; 8 h). Crude protein (CP) and acid detergent fibre (ADF) were determined according to the AOAC

(1997). Neutral detergent fibre (NDF) was determined according to Van Soest *et al.* (1991).

On day 15, ruminal fluid samples (250 ml) were taken at 0, 3, 6, 9, 12, 24, 48 and 72 h after the morning feeding, pH was measured immediately and samples were frozen; later, ammonia N concentrations (NH<sub>3</sub>-N; McCullough, 1967) were determined. To evaluate ruminal cellulolytic bacteria and protozoa concentration, 150 ml of the original ruminal fluid samples (at 3 h) were utilized. In a 10 ml tube, 1 ml rumen fluid was mixed with 1 ml trypan blue (1%) and diluted 1:10 (with 9 ml distilled water); then, rumen ciliate protozoa were counted using a Newbaver chamber and a microscope (40X magnification). For cellulolytic bacteria, a mixture of 1 ml of rumen fluid plus 9 ml of an anaerobic medium (Cobos *et al.*, 2002) was prepared; then, bacteria concentration was determined according to the most probable number technique (Harrigan and McCance, 1976).

Data were statistically analyzed by the 'MIXED' procedure of SAS (1999) as:  $Y_{ijkl} = \mu + P_i + A_j + B_k + AB_{jk} + D_l + \epsilon_{ijkl}$ , where  $Y_{ijkl}$  = the response;  $\mu$  = overall mean;  $P_i$  = effect of period ( $i = 4$ );  $A_j$  = effect of orchard grass maturity ( $j = 5$  or 16-wk);  $B_k$  = effect of yeast culture ( $k = 0$  or 5 g);  $AB_{jk}$  = interaction maturity <sub>$j$</sub>  x yeast culture <sub>$k$</sub> ;  $D_l$  = effect of sheep ( $l = 4$ );  $\epsilon_{ijkl}$  = experimental error <sub>$ijkl$</sub> .

### Results and Discussion

No substantial differences of chemical composition were found between 5 and 16-wk orchard grass (Table 1). Daily DM intake was not affected by maturity of orchard grass or yeast culture (Table 2). Ruminal pH values, ammonia N concentrations, total bacteria and cellulolytic bacteria were not affected by maturity of orchard grass and YC (Table 2).

Lambs fed mature orchard grass had a higher total protozoa concentration than those fed early orchard grass (Table 2). An interaction maturityxYC was detected suggesting that YC increased total protozoa in

Table 1  
Morphological and chemical composition of orchard grass hay cut at 5 (early) and 16-wk (mature)

	Orchard grass hay	
	Early	Mature
Morphological composition, % as DM		
Leaf	31.8	31.4
Stem	3.1	1.3
Weeds	2.1	11.4
Others	1.5	11.6
Senescent biomass	61.5	44.3
Chemical, % DM		
Dry matter	91.3	91.2
Crude protein	10.8	9.6
Neutral detergent fiber	66.6	71.4
Acid detergent fiber	44.8	47.6
Ash	24.6	22.0

the rumen of lambs fed early orchard grass, as compared to those fed mature orchard grass.

Total tract digestions of DM, NDF, ADF and N balance in lambs fed early or mature orchard grass, with and without YC, were similar (Table 2).

The main effect of yeast culture on rumen is improving rumen pH when ruminants are fed high grain diets (Bach *et al.*, 2007). Because in the current study lambs were fed orchard grass and ruminal pH was not below 6.7, yeast culture benefits could be limited.

Results from this experiment indicate that addition of a yeast culture to lambs fed early and mature orchard grass did alter ruminal protozoa without affecting feed intake, total tract digestion and N balance. Thus, fiber content of mature orchard grass may not have

Table 2  
Effect of a yeast culture on intake, digestibility and ruminal variables in lambs fed orchard grass hay cut at 5 (early) and 16-wk (mature)

	Orchard grass hay				SEM <sup>b</sup>
	Early		Mature		
	-YC <sup>a</sup>	+YC	-YC	+YC	
Dry matter intake (kg)	1.15	1.10	1.30	1.34	0.129
Total tract digestion, %					
Dry matter	50.7	52.2	49.8	53.6	2.94
Organic matter	54.5	60.5	55.9	56.6	4.60
Neutral detergent fiber	50.4	52.8	54.1	57.8	3.59
Acid detergent fiber	42.3	45.0	41.2	47.2	4.33
N balance, g/d					
Intake	15.2	14.9	15.5	16.0	1.58
Feces	5.9	6.2	5.0	6.2	0.87
Urine	3.4	4.0	4.0	3.8	0.55
Retained	5.9	4.7	6.5	6.0	1.09
Ruminal variables					
pH	6.76	6.80	6.79	6.72	0.112
Ammonia N, mg/dl	11.4	13.3	12.4	12.3	1.90
Total protozoa, x10 <sup>4</sup> d	9.0 <sup>z</sup>	9.7 <sup>y</sup>	15.1 <sup>w</sup>	14.1 <sup>x</sup>	1.68
Total bacteria, x10 <sup>9</sup> d	0.6	10.0	4.5	4.6	1.00
Cellulolytic bacteria x 10 <sup>7</sup> d	4.6	0.5	0.1	2.1	0.57

<sup>a</sup>-YC, 0 g yeast culture; +YC, 5 g yeast culture/lamb. <sup>b</sup>SEM, standard error of means.

<sup>d</sup>P and SEM values are from algorithm transformations.

<sup>w,x,y,z</sup>Means in a row with different superscripts differ (P<0.05).



been enough for the yeast culture to have an effect on digestion and ruminal variables.

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- पांच सप्ताह अथवा 16 सप्ताह की पुनर्वधित आर्चर्ड घास की हे खिलाए जाने वाले मेमनों के आहार में एक खमीर प्रवर्ध (0 या 5 ग्रा/दिन/भेड़) के पाचन और नाइट्रोजन लब्धि पर प्रभाव के मूल्यांकन के लिए रूमेन छिद्रित 4 नर क्रायोल्लो भेड़ों (45±4 किग्रा) को 2×2 फैक्टोरियली व्यवस्था के साथ 4×4 लैटिन स्क्वायर डिजाइन में उपयोग किया गया। खमीर प्रवर्ध का आहार ग्रहण, रूमेन पीएच, अमोनिया-नाइट्रोजन, नाइट्रोजन लब्धि और पोषकों के पाचन पर कोई प्रभाव नहीं पड़ा, केवल आर्चर्ड घास की प्रौढ़ता से रूमेन के सकल प्रोटोजोआ में परिवर्तन हुए, परन्तु उपचार का सेलुलोजभंजक और सकल सूक्ष्माणु, रूमेन पीएच, रूमेन-आमोनिया सांद्रता पर प्रभाव नहीं पड़ा। निष्कर्ष यह निकला कि खमीर प्रवर्ध देने पर आर्चर्ड घास की प्रौढ़ता से सकल प्रोटोजोआ की संख्या के अतिरिक्त आहार ग्रहण, पाचन और नाइट्रोजन लब्धि पर प्रभाव नहीं पड़ा।