THE EFFECT OF A MICROBIAL PRODUCT ON PRODUCTION PERFORMANCES OF FATTENING STEERS: PRELIMINARY RESULTS

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Abstract

The effect of a bacteria-based product (Bovilact), on ruminants production performances was studied on 32 fattening steers (150-250 kg weight category), randomly distributed in two groups (control/experimental). Both groups were fed ad libitum a diet composed of 20% corn silage and 80% compound feed (based on wheat and sunflower meal). For the experimental group, 10% of the compound feed was enriched with Bovilact (inoculated with a polyculture of acilolactic bacterias and yeasts and incubated until desired density of bacterial cells). Similar diets were fed to two fistulated cows, organized in a Latin Square design, in order to assess the effect on postprandial ruminal pH dynamics: average pH, minimum pH, maximum pH, duration of pH decrease under 6.2 threshold (t<6.2; hours), intensity of pH decrease under 6.2 threshold (a<6.2; hours x pH units), area under pH curve (auc; hours x pH units). pH was measured 10 hours after the morning meal, with a frequency of sampling of two hours. After 36 days of experiment, inclusion of Bovilact tended to slightly improve the daily weight gain: + 4.9% in experimental group, versus control. Bovilact had no clear effect on the level of ruminal pH level but changed the shape of pH curves, suggesting an influence on microbial populations involved in carbohydrates metabolism. Further studies on the end-products of carbohydrate (VFA) are needed in order to clearly assess the effects of Bovilact at ruminal level.

Key words: steers, performance, probiotics, rumen, pH

INTRODUCTION

The term "probiotic" has been defined as "a live microbial feed supplement, which beneficially affects the host animal by improving its intestinal microbial balance" [4] and has been used to describe viable microbial cultures, culture extracts, enzyme preparations, or various combinations of the above [20]. The original concept of feeding bacterial supplements to man and livestock was based primarily on the potential for beneficial intestinal effects, including the establishment of a desirable gut microflora and/or prevention of the establishment of pathogenic organisms. Supplementing diets on a daily basis with lactate-producing and/or lactate-utilizing bacteria has been shown to improve feed efficiency and daily gain of feedlot cattle [17], [5], [13], enhance milk production in dairy cows, and improve health and performance of young calves. In several experiments, supplementing feedlot cattle with lactate-utilizing and/or lactate-producing bacteria has been shown to improve daily gain (approximately 2.5%), with little change in dry matter intake (DMI). Ware et al. [19] was one of the first to report that *L. acidophilus* BT1386 increased daily gain and improved feed efficiency in yearling steers fed a high-concentrate diet compared with controls.

The objective of this study was to evaluate the effects of microbial feed supplement on production performances of fattening steers.

MATERIAL AND METHOD

A number of 32 Romanian Spotted fattening steers were used, weighing in average 220 kg, with an average daily gain of 1200 g, assigned randomly to two groups (control and experimental). Both groups were fed on a diet consisting of 20% corn silage and 80% ground compound feed (based on wheat and sunflower meal) formulated according to Burlacu [1].

In the experimental group, 10% of the compound feed was enriched with Bovilact feed additive. This is a polyculture of selected strains of lactic acid bacteria and yeasts (*Lactobacillus acidophilus, Lactobacillus casei, Lactobacillus plantarum, Enterococcus faecium, Saccharomyces boulardi* and *Saccharomyces cerevisiae*), which, by numeric superiority, production of lactic acid and some bacteriocins, might prevent and control enterocolitis, might improve feed conversion and animal performance. The product has 70% DM, acidity 83° T, colony density $3.0x10^9$ CFU/g, sweet/sourish taste, specific smell of lactic fermentation.

The diets were fed in one meal / day (8:30 a.m.), the compound feed preceding the silage and the animals had free access to the water. The daily amounts of given feed and the leftovers were recorded, to give the real feed intake. Tabel 1 shows the diets structure.

Table 1
Structure of diets

Diet	Diet C	Diet E
Corn silage (kg/day)	6	6
Compound feed (kg/day):	6.5	5.85
- wheat	65.5%	65.5%
- sunflower meal	20%	20%
- corn	10%	10%
- $CaCO_3$	2%	2%
- salt	1%	1%
- vitamin/mineral mix T3	1.5%	1.5%
Compound feed with Bovilact (kg/day)	-	0.65

C = control; E = experimental

Two fistulized Romanian Black Spotted cows, with an average weight of 550 kg, set up in a Latin square design, received similar diets in order to determine the effect of the studied product on postprandial ruminal pH dynamics. The animals were kept in individual digestibility stands and feed in the same manner as the fattening steers.

Ruminal fluid samples were collected in the morning, just before feeding, and then at 2, 4, 6, 8 and 10 hours after the animals have consumed the entire amount of compound feed. The samples were collected through the ruminal cannula using a vacuum pump. The ruminal content (about 100 ml) was filtered through 4 layers of gauze and the pH was read with a Beckman pH meter immediately after sampling (2 readings/sample). A Visual Basic script was used to calculate the following parameters: average pH, minimal pH, maximum pH, duration of pH decrease below 6.2 ($t<6.2$), intensity of pH decrease below 6.2 ($a<6.2$), area under the pH curve (auc).

The experimental data were recorded and processed statistically, making an evaluation of the dispersion parameters (average, variance, standard deviation) to show the following aspects:

- representativeness of the average;
- comparison in time and space of two or more samples to determine the level of homogeneity;
- characterisation of sample variation;
- quantification of the influence of different factors on the variation of the specific characteristics.

We used a null hypothesis and an alternative hypothesis for the significance tests. Sample and variance homogeneity were tested by ANOVA (for the bifactorial experiments, to determine not just the significance of the difference, but also of the interaction between the factors of influence), Student and Fisher.

RESULTS AND DISCUSSION

The daily recording of the feed administered and leftover by the steers allowed determining the average daily intake, shown in Table 2. The actual intake was lower than the planned one in both groups. This may be due to the acidic structure of the diet given by the 8:2 ratio of concentrate to bulk forages, susceptible to affect diet ingestibility.

Table 2
Average daily feed intake

	Control Group	Experimental Group
Administered corn silage (kg/steer/day)	6	6
Consumed corn silage (kg/steer/day)	5.82	5.66
Administered CF (kg/steer/day)	6.5	6.5
Consumed CF (kg/steer/day)	5.71	5.68
Dietary nutrient supply:		
DM (g/day)	6614.60	6544.78
mFU/ day	8.19	8.11
IDPN (g/ day)	654.03	648.95
IDPE (g/ day)	554.77	549.86
Ca (g/ day)	51.49	51.09
P (g/ day)	30.30	30.04

DM = dry mater; mFU = meat feed units; IDPN = intestinally digestible protein allowed by nitrogen supply; IDPE = intestinally digestible protein allowed by energy supply

The diets were balanced in terms of protein and energy, the differences between groups being very small. Table 3 shows the evolution of the body weight, average daily gain and feed conversion ratio.

Table 3
Animal performance and intakes

	Control Group	Experimental Group
Initial weight (kg)	218.8 ± 39.2	226.7 ± 40.6
Final weight (kg)	264.5 ± 43.3	274.6 ± 39.8
Average daily weight gain (g)	1269.1 ± 269.0	1331.3 ± 214.7
Feeding efficiency:		
Kg DM/kg weight gain	5.21	4.92
mFU / kg weight gain	6.45	6.09
g IDPN / kg weight gain	515.35	487.46
g IDPE / kg weight gain	437.14	413.02

According to the data shown in Table 3, on the background of a high energy/protein diet, the inclusion of Bovilact produced a slight increase of the average daily gain (+4.9%) in the experimental group compared to the control group. The difference was not significant and must be interpreted as trend, further tests on a larger number of animals being required. The experimental group used 5-6% less nutrients for one kg of gain compared to the control group.

Because the diet had a high concentrate to bulk forage ratio, with a high risk to induce ruminal acidosis, we also investigated the effects on the postprandial ruminal pH dynamics, using two fistulized cows, set up in a Latin square design (2x2).

Figures 1, 2 and 3 show the evolution of the ruminal pH using the animal individuality as constant factor and the diet as controllable factor, while quantifying this influence on the variation of the specific characteristic.

Figure 1 (showing pH postprandial evolution independently on the factors of influence, diet, period, environment) shows that the pH displays a clear individual variability (0.22 pH units difference between the two cows). In both animals pH peaked in the morning before feed administration (7.01 and 7.41), decreased fast during the first two hours after the feed was consumed (to 6.38 and 6.37), reaching a minimum of 6.04 and 6.16 after 4 hours, increasing thereafter between 6 and 10 hours, to 6.26 and 6.62, for cow 1 and 2, respectively.

The change in the shape of pH curve (Figure 2), determined by the addition of 10% Bovilact to the diet, suggest a positive influence on the ruminal microbial population, favouring improvement of production performances, as found in the trial on steers. However, no major influence on the parameters describing ruminal pH was detected (Table 4). Nocek et al. [10] reported daily low ruminal pH was higher, and area under pH 5.5 was reduced, for dairy cows receiving *Enterococcus* and *Lactobacillus*, compared to cows fed a control diet.

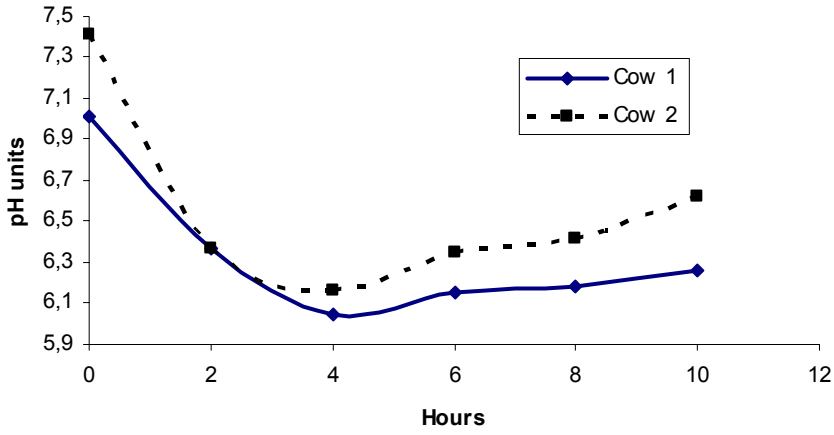

Figure 1. Individual variability of ruminal pH, postprandial evolution

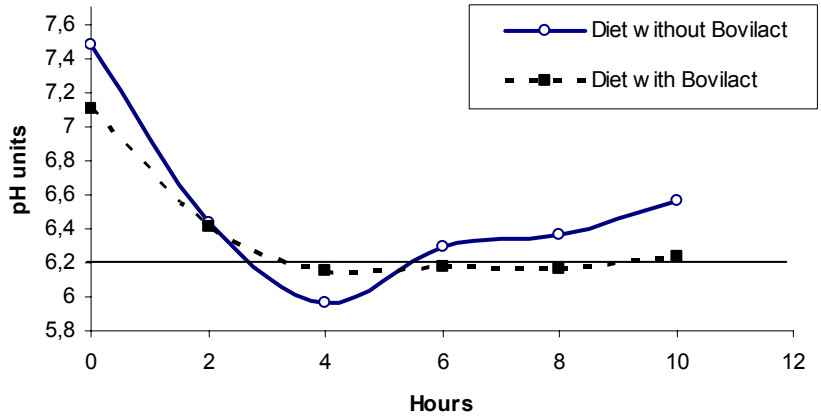

Figure 2. Ruminal pH postprandial evolution without and with Bovilact

Although Bovilact generates lactic acid, the decrease of pH was not too large. Figure 2 shows that the Bovilact supplementation to the diet modified pH shape, maintaining it for a longer period in the plateau stage around 6.2 threshold. In the absence of Bovilact, a clear decrease of pH below the 6.2 threshold (down to 5.95) occurred between 3-5 hours after feeding, suggesting a potential onset of the subacute rumninal acidosis, but the restoration of pH to normal values was sharper.

Figure 3 shows the interaction animal-diet on the ruminal pH. Cow 1 had an average initial 6.0 pH (before feeding) during the control day. After feeding of the Bovilact diet, pH increased by 0.48 units and after Bovilact was removed from the diet, pH decreased by 0.08 units. Cow 2 had an average initial pH of 6.90. It decreased by 0.33 units after feeding the diet without Bovilact, and continued to decrease by 0.4 units after Bovilact was included in the diet. The phenomenon can be put on the account of the individual pH variation (Figure 1). We need to analyse the final products of carbohydrates metabolism (VFA) in order to fully assess the effects of the product at rumen level.

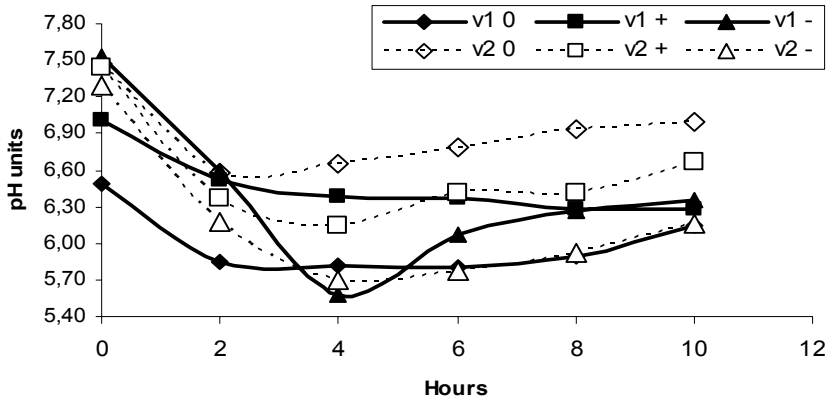

Figure 3. Ruminal pH postprandial evolution function of the animal – diet interaction
(v1 0, v2 0 = initial pH; v1 +, v2 + = pH due to dietary Bovilact; v1 -, v2 - = pH due to exclusion of the dietary Bovilact)

Table 4 shows the effect of the individuals and of the two diets on the synthetic parameters (t<6.2, a<6.2) which express the postprandial evolution of ruminal pH. The duration of pH decrease below a level (6.2 in this case) is a more reliable parameter, having a biological signification, confirmed by in vitro [15], [16], [12] and in vivo [9], [8] studies. This is explained by the fact that pH values higher than the threshold and, consequently, with no relevant effect at the ruminal level, are not taken into consideration. For example, a pH value of 6.4 or 6.8 has the same effect at the ruminal level.

As concerning the shape of pH curve, a flattened pH curve may lead to a lower average than a normal one, even if the period and intensity of pH decrease below a relevant level for the ruminal metabolism are identical.

Table 4
Effect of the individual and of the diets on ruminal pH parameters

	Average pH	Minimum pH	Maximum pH	t< 6.2	a< 6.2	auc
Effect of the animal (individual variability):						
Cow 1	6.34	5.95	7.01	3.81	1.05	62.76
Cow 2	6.56	6.09	7.41	2.96	0.69	64.63
Effect of Bovilact:						
Diet with BVL	6.37	5.97	7.11	3.19	0.89	63.14
Diet without BVL	6.52	5.90	7.47	2.81	0.50	64.15

t<6.2 = duration of pH decrease below 6.2; a<6.2 = intensity of pH decrease below 6.2; auc = area under the pH curve

The synthetic parameters shown in Table 4 confirm the individual variability of the postprandial evolution of the ruminal pH (between 2.96 – 3.81 hours for t<6.2, and between 0.69 – 1.05 hours x pH units for a<6.2). The addition of Bovilact tends to increase the duration and intensity of pH decrease below 6.2. However, these decreases are in the range of individual variability (animal effect) and their magnitude is not able to affect the ruminal metabolism (the general level of pH remaining thus high).

At our knowledge, no studies thus far have evaluated the influence of different types of direct-fed microbials on diurnal ruminal pH variation on ruminal digestibility but there are specific strains of microbials that, when selected and combined, could strategically manipulate and regulate ruminal metabolism. It is possible that certain direct-fed microbials combinations that synthesize lactic acid, when incorporated into the diet, may sustain a tonic level of lactic acid in the rumen that would be higher and less variable. This would stimulate

lactic acid utilizing bacteria, which would reduce total lactic acid available in the rumen and total ruminal acidity.

CONCLUSIONS

On the background of a high energy/protein diet, the inclusion of Bovilact in fattening steers diet produced a slight increase of the average daily gain (+4.9%) in the experimental group (1331.3 g) compared to the control group (1269.1 g) and a 5 – 6% improvement of feeding efficiency.

No clear effects of Bovilact on the level of ruminal pH level could be detected, but it changed the shape of pH curves, maintaining it for a longer period in the plateau stage, suggesting an influence on microbial populations involved in ruminal metabolism.

REFERENCES

Journal articles

[3] Dragomir C. şi col.,: The effect of replacing dietary barley with dry corn gluten feed on the dynamics of ruminal pH, Arhiva Zootehnică, 2008, 11:1, 16 – 23.

[4] Fuller R.,: A review: Probiotics in man and animals, J. Appl. Bacteriol, 1989, 66:365–378.

[5] Galyean M. L., Nunnery G. A., Defoor P. J., Salyer G. B., and Parsons C. H.,: Effects of live cultures of *Lactobacillus acidophilus* (Strains 45 and 51) and *Propionibacterium freudenreichii* PF-24 on performance and carcass characteristics of finishing beef steers, 2000.

[6] Ghorbani G. R., Morgavi D. P., Beauchemin K. A., and Leedle J. A. Z.,: Effects of bacterial direct-fed microbials on ruminal fermentation, blood variables, and the microbial populations of feedlot cattle, J. Anim. Sci., 2002, 80:1977–1986.

[7] Krehbiel C. R., Rust S. R., Zhang G., and Gilliland S. E.,: Bacterial direct-fed microbials in ruminant diets: Performance respponse and mode of action, J. Anim. Sci., 2003, 81 (E. Suppl. 2): E120 – E132.

[8] Martin C. and Michalet-Doreau B.,: Variations in Mass and Enzyme Activity of Rumen Microorganisms: Effect of Barley and Buffer Supplements, Journal of the Science of Food and Agriculture, 1995, 67 (3): 407 – 413.

[9] Mould F.L., Orskov E. R. and Mann S. O.,: Associative effects of mixed feeds. 1. Effects of type and level of supplementation and the influence of rumen pH on cellulolysis in vivo and dry matter digestion of various roughages, Animal Feed Science and Technology, 1983,10 (1): 15 – 30.

[10] Nocek J. E., Kautz W. P., Leedle J. A. Z., and Allman J. G.,: Altering diurnal pH and in situ digestion in dairy cows with ruminal supplementation of direct-fed microbials (DFM) and yeast, J. Dairy Sci., 2000, 83 (Suppl. 1): 1242 (Absr.).

[11] Nocek J. E., Kautz W. P., Leedle J. A. Z., and Allman J. G.,: Ruminal supplementation of direct-fed microbials on diurnal pH variation and in situ digestion in dairy cattle, J. Dairy Sci., 2002, 85: 429–433.

[12] Russell J. B. and Wilson D. B.,: Why are ruminal cellulolytic bacteria unable to digest cellulose at low pH?, J. Dairy Sci., 1996, 79: 1503 – 1509.

[13] Rust S. R., Metz K., and Ware D. R.,: Effects of Bovamine rumen culture on the performance and carcass characteristics of feedlot steers, Michigan Agric. Exp. Stn. Beef Cattle, Sheep and Forage Sys. Res. Dem. Rep., 2000a, 569:22–26.

[15] Shriver B. J., Hoover W. H., Sargent J. P., Crawford R. J., Thayne W. V.,: Fermentation of a high concentrate diet as affected by ruminal pH and digesta flow, J. Dairy Sci., 1986, 69: 413 – 419.

[16] Slyter I. L., Rumsey T. S.,: Effect of coliform bacteria, feed deprivation and pH on ruminal D-lactic acid production by steer or continuous culture microbial populations changed from forage to concentrates, J. Anim. Sci., 1991, 69: 3055 – 3066.

[17] Swinney-Floyd D., Gardner B. A., Owens F. N., Rehberger T., and Parrott T.,: Effect of inoculation with either strain P-63 alone or in combination with *Lactobacillus acidophilus* LA53545 on performance of feedlot cattle, J. Anim. Sci., 1999, 77(Suppl. 1):77.

[18] Ware D. R., Read P. L., and Manfredi E. T.,: Lactation performance of two large dairy herds fed *Lactobacillus acidophilus* strain BT138 in a switchback experiment, J. Dairy. Sci., 1988a, 71(Suppl. 1):219.

[19] Ware D. R., Read P. L., and Manfredi E. T.,: Pooled summary of eight feedlot trials evaluating performance and carcass characteristics of steers fed *Lactobacillus acidophilus* strain BT138, J. Anim. Sci. 1988b, 66(Suppl. 1):436.

[20] Yoon I. K., and Stern M. D.,: Influence of direct-fed microbials on ruminal microbial fermentation and performance of ruminants: A review, Asian-Australas, J. Anim. Sci. 1995, 8:533–555.

Books

[1] Burlacu Gh., Cavache A., Burlacu R.,: The new system of nutritive value appreciation and animals feed requirements, Ed. Ceres, Bucharest, 1996.

[2] Burlacu Gh., Cavache A., Burlacu R.,: Productive potential of feeds and their use, Ed. Ceres, Bucharest, 2002.

[14] Rust S. R., Metz K., and Ware D. R.,: Evaluation of several formulations of Bovamine rumen culture on the performance and carcass characteristics of feedlot steers, Michigan St. Univ. Beef Cattle Res. & Ext., 2000b, Available: http://beef.ans.msu.edu/ MSU Beef Research and Extension 1999-2000.pdf. Accessed April 17, 2003.