

Performance of Calves Submitted to Protocols Using Extruded or Ground Starter

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ABSTRACT

Feed processing can affect rumen development in new born calves, and potentially define animal performance. Two feed management systems, extruded starter (Ruter) with possible early weaning and ground starter (control), were evaluated in thirty-two Holstein calves (16 females and 16 males). Animals were randomly assigned to the treatments using a randomized block design with birth weight as a covariate. They were weaned when starter intake reached 800 g for two consecutive days. Twenty-one days after the weaning, males were slaughtered and the stomach compartments were isolated. Rumen and omasum fragments were processed for morphological evaluation. Animal performance, clinical condition and stomach compartment weight did not differ between the treatments ($P > 0.05$), despite weaning weight of animals receiving extruded starter being 5.68% higher than the control animals. Extruded starter stimulated cell proliferation of the ruminal epithelium ($P < 0.05$), but did not affect the dimensions of the papillary rumen and omasum mitotic index (MI). The Ruter feeding system was potentially beneficial for weight gain and morphofunctional rumen development in lactating animals; however, this treatment did not allow early weaning as proposed by the feeding system.

Key words: Milk-feeding period, grain processing, reticulorumen

INTRODUCTION

Early transition from simple gastric digestion to functional ruminant digestion in dairy calves is essential for animal health and growth. The type of diet and management system defines the speed of functional stomach development (Lucci 1989; Coelho et al. 1999; Nussio et al. 2003). The development of rumen papillae is determined by the fermentation of carbohydrates in the diet, with consequent production of short-chain fatty acids (SCFA). These changes and consequent ruminant development in pre-weaned calves are essentially

affected by the intake and composition of solid feed (Coelho et al. 1999).

Most conventional milk production systems in Brazil use a liquid diet as the basic food supply for 90 days. However, these systems may hinder calf rumen development, since the ruminal fermentation pattern necessary for its morphofunctional development decreases. Use of a solid diet to accelerate the consumption and early rumen development has been proposed for better economic returns. The Association of Cooperatives in Argentina (ACA) has suggested early weaning of calves and use of a starter containing starch extruded with higher protein

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content than the conventional formula. This weaning method leads to a brief transition from lactation to ruminant digestion, based on the functional development of the rumen, with the possibility of weaning at 21 days.

Performance data of lactating calves fed a starter with extruded grains and possible early weaning, are still limited. Thus, this study was conducted to evaluate two feed protocols on calf performance and stomach morphology: extruded starter (Ruter®, ACA, Buenos Aires, Argentina) following the manufacturer's recommendations or ground starter in a conventional layout.

MATERIALS AND METHODS

The experiment was conducted at a commercial farm in the region of Lavras, Minas Gerais, Brazil. Sample consisted of 16 female and 16 male Holstein calves with average birth weight of 38.4 ± 6.2 kg. After birth, the animals were weighed and housed in individual pens, where they received four liters of colostrum and had the navel treated with 10% iodine solution. Animals were then paired, primarily by the gender and secondarily by date of birth, and randomly assigned to the treatments (Table 1). One group received extruded starter (Ruter®, ACA, Buenos Aires, Argentina) following the manufacturer's recommendations, and the other group (control) received ground starter following feed management recommended by the experts working in the region of Lavras, MG. All the animals regardless of treatment received four liters of milk in two daily meals, at 7:00 am and 2:00 pm. The daily milk volume was reduced to two liters in the treatment with extruded starter when the consumption of extruded starter reached 400 g for two consecutive days, and to three liters of milk in the treatment with ground starter, from the 29th day of life onwards. All the animals regardless of treatment were weaned when starter consumption reached 800 g for two consecutive days. The experimental period continued another 21 days after the weaning. Animals fed ground starter continued to receive ground starter *ad libitum* during this time. Animals fed with extruded starter were gradually adapted to the ground starter utilized in the control animals during this post-weaning period. Daily consumption of the extruded starter was restricted to 1,000 g during the first week and 500 g during the second, while

the ground starter was fed *ad libitum*. During the third week, animals were offered only the ground starter *ad libitum*. All the animals had free access to water and starter concentrate. The starter intake was measured daily by weighing the remains. Samples of the starter supplied were collected weekly to determine the dry matter (DM), crude protein (CP), ether extract (EE) and ash content, according to AOAC (1990), and neutral detergent fiber (NDF) was determined as described by Van Soest (1991). Animals were weighed at birth and every seven days before the afternoon meal. Daily weight gain was defined as the slope of the linear regression of the weekly weight gain.

Table 1 - Composition and nutrients of ground and extruded starter concentrates.

Ingredients	Ground starter,	
	% DM	
Finely ground mature corn	57.0	
Soybean meal	18.5	
Wheat meal	18.5	
Minerals and vitamins ^A	6.0	
Nutrients	Ruter	Ground
	% DM	
DMI	93.5	92.0
Crude protein	21.6	16.7
Neutral detergent fiber	13.5	18.5
Ether extract	4.1	2.3
Ashes	4.1	6.2

^AMineral Milk Young MCassab: 17.0% of Ca; 7.3% of P; 5.0% of Mg; 6.9% of Na; 1.6% of K; 2.2% of S; 1131 ppm of Cu; 2105 ppm of Mn; 4078 ppm of Zn; 18.1 ppm of Co; 37 ppm of I; 20.4 ppm of Se; 5602276 UI of Vit. A; 55732 UI of Vit. D; 1120 UI of Vit. E; 1500 ppm of Monensin.

Each animal was clinically evaluated over the experimental period by measuring the rectal temperature, feces consistency and behavior (adapted from Magalhães et al. 2008). Rectal temperature was measured daily around 2:00 pm, and temperatures above 39.5°C were considered to be febrile. Animal behavior was assessed during the afternoon milk feeding using the following scoring system: 1) alert and responsive; 2) non-active; 3) depressed, and 4) moribund. Fecal consistency was scored at the same time as rectal temperature measurement, in which the following classification was adopted: 1) firm, 2) soft or moderate consistency, 3) runny or mild diarrhea, and 4) watery and profuse diarrhea. Calves with fecal score >2 were used for the analysis of incidence of diarrhea. Animals with diarrhea were treated with antibiotics and oral rehydration with electrolyte solution. Other clinical occurrences of

respiratory origin or hemoparasites were diagnosed and treated, and medications were recorded, considering the dose and duration of the treatment.

At the end of the trial period, in the afternoon, the males were slaughtered and gutted, and the stomach compartments were isolated: reticulorumen, omasum and abomasum. After cleaning and removing the excess tissue, the organs were opened, emptied, washed with running water and weighed on a precision scale. For the histological evaluation, samples were collected from the wall fragments of the cranial rumen sac, the ventral rumen sac (rumen recess) and the omasal laminae. Samples were then fixed for 12 h in Bouin solution and processed routinely for paraffin embedding. Slides were stained with Hematoxylin-Eosin (Luna 1968). Papillary length and area were measured in rumen samples using CellB image analysis software (Imaging Software for Life Science Microscopy, Olympus Optical do Brasil Ltda., São Paulo, SP). The mitotic index (MI) was determined in both the rumen and omasum samples, using an optical microscope at 400 magnification. The mitotic index was determined by counting all the mitotic nuclei and expressed as a percentage of total visible nuclei. Two independent reviewers performed the counts. Animal MIs were calculated as the average of these individual ratings.

For data interpretation, the experimental period was divided into three phases: 1 - in which the animals received four liters of milk per day; 2 - in which the animals consumed two liters of milk per day in extruded starter treatment, or three liters in ground starter treatment, and Period 3 - 21-day post-weaning period. Data were analyzed using the GLM procedure of SAS (1985) as a randomized complete block design, with birth weight as a covariate. Blocks were based primarily on gender and secondarily on the date of birth with 16 blocks for performance data and eight blocks for morphological data. The latter was based solely on male specimens, since female animals remained in the herd, following the farm routine. The two groups were compared by the analysis of variance and Tukey test. Significance was determined at $P < 0.05$ unless otherwise noted. The following

model was used: $Y_{ijk} = \mu + T_i + B_j + \beta(X_k - \bar{X}) + e_{ijk}$, where Y_{ijk} = dependent variable, μ = overall mean, T_i = treatment i (extruded or ground starter), B_j = block effect (1-16) or (1-8), β = slope of the linear regression between X and Y ; \bar{X} = overall average of the covariate, X_k = covariate (continuous effect of birth weight), e_{ijk} = residual.

RESULTS AND DISCUSSION

There was no significant difference ($P > 0.05$) in weaning weight between the treatments, although weaning weight of the animals receiving extruded starter was 5.68% higher than the control animals ($P = 0.07$) (Table 2). Consequently, animals fed extruded starter (Ruter) gained higher body weight 21 days after weaning than the control animals ($P = 0.06$), given that the animals in both the treatments had the same starter intake ($P > 0.05$) and were weaned, on average, at 49 days of age (SEM=1.91, $P = 0.72$). The protocol used by the Ruter manufacturer predicted lower milk consumption ($P < 0.01$) in the Periods 1 and 2, which suggested that the animals compensated for the unavailability of nutrients with higher feed efficiency in the extruded starter during the starter phase. Zhang et al. (2010) using the starter and extruded corn and soybeans, steam-flaked corn and soybeans or ground corn and soybeans found no differences in body weight, calf starter intake, milk intake, total dry matter intake and body structural growth during the study; however, feed efficiency was significantly improved by the steam-flaked treatment.

Milk intake in the Periods 1 and 2 was different for the two treatments ($P < 0.01$); however, due to the different lengths of these periods, total intake was not affected by grain processing ($P > 0.05$) (Table 2). Other authors also found no benefits of grain processing regarding starter intake and performance in livestock (Nussio et al. 2003b). Grain processing may not be as important to an animal with a developing rumen as it is for a functional ruminant, since digestion is still predominantly chemical and enzymatic in the abomasum and small intestine (Nussio et al. 2003b).

Table 2 - Measurements of body weight, daily weight gain (DWG), dry matter intake (DMI) and test duration of calves in Ruter and Control treatments (n = 32).

Variables	Ruter	Control	SEM ^A	P-value
Body weight		kg		
Weaning	56.30	53.10	1.17	0.07
Total	66.94	60.06	2.21	0.06
DWG		kg/d		
Period 1 ^B	0.29	0.26	0.03	0.43
Period 2 ^B	0.48	0.34	0.07	0.19
Period 3 ^B	0.66	0.68	0.05	0.82
Total ²	0.44	0.39	0.02	0.09
Milk intake		l		
Period 1	154.40	111.80	7.28	<0.01
Period 2	22.10	62.10	5.49	<0.01
Total	176.50	173.90	5.95	0.76
DMI		kg		
Period 1	4.86	2.60	0.48	<0.01
Period 2	6.54	9.38	1.03	0.07
Period 3	31.20	32.30	0.99	0.44
Total	42.60	44.30	1.29	0.36
Duration		days		
Period 1	38.6	28.0	1.82	<0.01
Period 2	11.1	20.7	2.08	<0.01
Period 3	21.0	21.0	0.00	-
Total	70.7	69.7	1.91	0.72

^ASEM = Standard error of the means

^BPeriod 1 – in which animals consumed four liters of milk per day. Period 2 – in which animals consumed two liters of milk per day in the Ruter or three liters in the Control treatment. Period 3 – post-weaning period of 21 days. Total – Period from birth to 21 days after weaning.

Grain processing in the Ruter treatment not only improved starch digestibility in the gastrointestinal tract, but also increased short chain fatty acids (SCFA) production and absorption in the rumen, providing higher performance in preweaned dairy calves (Suarez et al. 2006a). This probably did not occur in the present study, since total CP intake was 9.2 kg for the animals fed extruded starter compared to 7.4 kg for the animals fed ground starter (SEM=0.25; $P<0.01$), which was 19.5% less CP in the animals fed ground starter.

Animals in the Ruter treatment required less time to reach 400 and 800 g/d of the starter intake ($P<0.01$, 0.007, respectively), which was represented by the duration of Period 1 and 2, respectively (Table 2). The higher intake of solid food in Ruter study group animals during the Period 1 ($P<0.01$) could be associated with reduced availability of the nutrients. These animals began to take less milk and consequently, needed to consume concentrate earlier than those who received three liters of milk per day until 29 days of life. Early initiation of solid feed intake and possible ruminant activity in the calves can mitigate the negative effects of lower milk intake

on the growth and performance of dairy calves during pre-and post-weaning periods (Bittar et al. 2009; Azevedo et al. 2014).

Animals showed alert and responsive behaviour ($P>0.05$) throughout the experimental period. Feces consistency was soft or moderate for both the treatments. This was consistent with the report of Zhang et al. (2010) in which no significant treatment differences for the calf feces were observed. Most cases of diarrhea were observed in both the treatments during the first and second weeks of life. Cases of diarrhea are typical for this period of animal life and consistent with data from other authors (Franklin et al. 2003). At this stage, roughly 80% of calves had at least two days of fecal score >2 , ranging from two to seven days. Rectal temperature averaged 38.9°C, which was considered to be within the physiological limits of the species. No differences were observed ($P>0.05$) in the days that calves were treated with medications, since the number of diseases observed and the medicine administered were similar for all the animals ($P>0.05$).

There was no relationship between the weight of calf stomach compartments and treatment

($P>0.05$) (Table 3). Nussio et al. (2003a) found a trend effect ($P=0.11$) for grain type only on the weight of reticulorumen expressed as percentage of total stomach in the calves fed steam-flaked grain. However, other authors had no significant effect on the weight of reticulorumen of calves fed with different levels of processed grain (Bittar et al. 2009).

Table 3 - Weight of stomach compartments in Ruter and Control animals (n=16).

	Ruter	Control	SEM ^A	P-value
	weight (grams)			
Reticulorumen	1,672.5	1,647.5	100.89	0.87
Omasum	246.3	304.4	23.87	0.13
Abomasum	296.3	320.6	13.30	0.24
Total stomach	2,235.0	2,268.8	124.17	0.85
	% body weight			
Reticulorumen	2.6	2.7	1.14	0.64
Omasum	0.3	0.5	0.36	0.08
Abomasum	0.5	0.5	0.22	0.07
Total stomach	3.4	3.7	1.22	0.23
	% total stomach			
Reticulorumen	75.0	72.2	0.99	0.12
Omasum	11.0	13.5	1.01	0.15
Abomasum	13.3	14.3	0.70	0.39

^ASEM = Standard error of the means

There was no difference between the treatments on papillae length ($P>0.05$) (Table 4). These were similar to that of the calves fed corn grain pellets and slaughtered at 70 days (Khan et al. 2008). The higher MI in the animals in the Ruter treatment ($P<0.05$) might have occurred due to higher production and absorption of SCFA in the rumen, increased insulin secretion or actions of other epithelial growth regulators. However, Goodlad (1981) noted that MI transition in the dietary forage to concentrate increased and peaked five to six days after the dietary change, followed by a sharp decline back to the original level. In the present study, even if MI declined after a possible peak in cell proliferation, which was not measured at the time, MI values were higher in the animals fed extruded starter, suggesting a prolonged effect of the extruded starter on the morphological and functional development of the reticulorumen.

There are reports describing that physical stimulation of the rumen with feed could account for the measurable increases in rumen weight and muscle development (Nussio et al. 2003; Suarez et al. 2006b). Thus, for the development of the

ruminal epithelium to progress normally, available ruminal fermentation must be established, suggesting that there is a requirement for the presence of SCFA, especially propionate and butyrate (Costa et al. 2008) in the ruminal lumen to promote normal papillary development. Several studies have been conducted to evaluate the effect of physical form and particle size of grain on the development of rumen mucosa (Franklin et al. 2003; Bittar et al. 2009), but results are quite contradictory.

There was no effect of the treatment on MI in the omasum ($P>0.05$) (Table 4); however, MI was higher in the omasum than in rumen recess. Daniel et al. (2006) found MI to be 0.52% in the omasum and 0.28% in the rumen in adult crossbred animals. According to these authors, higher MI in the omasum indicated faster cell proliferation in the basal layer of the omasum in relation to the rumen. Morphological and functional aspects of the omasum are little understood, in spite of the importance of this organ in the digestive process of ruminants.

Table 4 - Papillae length, area papillary, and mitotic index (MI) in the rumen recess and MI in the omasum of Ruter and Control animals (n=16).

Stomach	Ruter	Control	SEM ^A	P-value
Rumen recess				
Papillae length (μm)	1,955	1,661	105	0.13
Area papillary (μm^2)	1,592,200	935,400	411,100	0.34
MI (%)	1.11	0.59	0.13	0.04
Omasum				
MI (%)	1.52	1.24	0.16	0.34

^ASEM = Standard error of the means

CONCLUSIONS

Ruter feed system, using extruded starter, with the possibility of early weaning, was potentially beneficial for weight gain and morphofunctional rumen development in lactating animals, suggesting a prolonged effect of extruded starter on the morphological and functional development of the reticulorumen in calves. Since the animals in the Ruter treatment were weaned at the same time as those in the control group, early weaning was not possible as suggested by the Ruter feed system.

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