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# Grazing cows are more efficient than zero-grazed and grass silage-fed cows in milk rumenic acid production

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#### **ABSTRACT**

Six rumen-cannulated Holstein cows in early lactation were assigned to 3 treatments: grazing (G), zerograzing (ZG), and grass silage (GS) harvested from the same perennial rye grass sward in a  $3 \times 3$  Latin square design with three 21-d periods. The objectives of this study were to investigate the underlying mechanisms for the reported elevation in milk rumenic acid (RA) concentration associated with G compared with ZG and GS, and to identify the important variables contributing to the milk RA response. Grazing animals were offered 20 kg of dry matter/cow per day; indoor animals were offered ad libitum grass or silage. A concentrate at a rate of 3 kg/d was also offered to all cows. Rumen, plasma, and milk samples were collected in the third week of each period. Data were analyzed by the MIXED procedure of SAS. Dry matter intakes were less for GS with no difference between G and ZG. Milk yield was greater for G than for ZG or GS. Milk fat and protein contents were less for GS with no difference between G and ZG. The combined intake (g/d) of linoleic and linolenic (18:3n-3) acids was different across the treatments (G: 433; ZG: 327; and GS: 164). Rumen pH was less for G with no difference between ZG and GS. Concentrations of volatile fatty acids and ammonia nitrogen in rumens were not different across the treatments. Wet rumen fill was less for G with no difference between ZG and GS. Vaccenic acid concentrations were different across the treatments in rumen (G: 12.30%, ZG: 9.31%, and GS: 4.21%); plasma (G: 2.18%, ZG: 1.47%, and GS: 0.66%) and milk (G: 4.73%, ZG: 3.49%, and GS: 0.99%). Milk RA concentrations were greater for G (2.07%) than for ZG (1.38%) and GS (0.54%). Milk desaturase index based on the ratio cis-9-14:1/14:0 was not different across the treatments. Milk RA vield per 100 g of linoleic acid and linolenic

acid intake (efficiency) was 2.23, 1.50, and 0.62 g in G, ZG, and GS, respectively, suggesting that G cows were more efficient than ZG and GS cows in milk RA production. Stepwise regression analysis of a group of variables revealed that plasma vaccenic acid accounted for 95% of the variation in milk RA production. Milk desaturase index did not enter into the model. Overall findings suggest that substrate intake influenced milk RA production but it was not the only factor involved. There were differences in efficiency of milk RA production, which appears to depend on the factors regulating ruminal vaccenic acid production and its supply to the mammary tissue.

**Key words:** rumenic acid, vaccenic acid, conjugated linoleic acid, grazing

#### INTRODUCTION

Rumenic acid (**RA**) is a component of milk fat with demonstrated health benefits. Biosynthesis of milk RA requires a source of polyunsaturated fatty acid (PUFA) substrate such as linoleic acid (LA) and linolenic acid (LNA) in the feed that is biohydrogenated in the rumen supplying a combination of precursor, vaccenic acid (VA), and small amounts of product (RA) to the mammary gland (Bauman et al., 1999). The extent of ruminal biohydrogenation of PUFA in turn has been reported to be influenced by the initial concentration of dietary LA (Harfoot et al., 1973), passage rate, and rumen pH (Martin and Jenkins, 2002; Troegeler-Meynadier et al., 2003; Qiu et al., 2004). In the mammary tissue, most of the RA is formed from VA by the action of  $\Delta^9$ -desaturase (Griinari et al., 2000). Thus biosynthesis of milk RA is dependent on the amount of substrate in feed, the extent of biohydrogenation in the rumen, and the desaturase activity of the mammary tissue. Although the significance of the above 3 factors on milk RA concentrations is well known, the relative contribution of each has not been established.

Fresh grass is a rich source of LNA. However, its concentration varies with growth stage and the form

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in which it is fed. Chilliard and Ferlay (2004) reported that milk RA response varies among forages, with fresh grass > hay > maize silage > grass silage. Moreover, pasture (grazing) and barn (zero-grazing) feeding, which is feeding fresh grass outdoors and indoors, respectively, has been reported to influence milk RA response differently (Leiber et al., 2005). Offer (2002) and Elgersma et al. (2003) found that beneficial fatty acids (FA) including RA were greater in milk for grazed animals than for zero-grazed grass or grass silage.

The present study was designed to investigate possible mechanisms underpinning the differences in milk RA concentrations and yields among cows grazing (G), zero-grazing (ZG), or fed grass silage (GS) with the following objectives: 1) to determine the supply of substrate from the different diets; 2) to investigate if offered and consumed grass by grazing cows differed in substrate content; 3) to determine the effect of the diets on rumen environment; 4) to gain an understanding of the biosynthesis of milk RA by evaluating the FA composition of feed, rumen ingesta, plasma and milk with specific interest in VA and RA; 5) to investigate if there were treatment differences in the efficiency of milk RA production; and 6) to identify the most important variables contributing to the differences in milk RA.

#### **MATERIALS AND METHODS**

#### **Experimental Design and Treatments**

All procedures were carried out under license in accordance with the European Community Directive 86-609-EC. Six spring-calving, rumen-cannulated, multiparous (lactation number 2 to 5) Holstein-Friesian dairy cows in early lactation (average milk yield 21.9  $\pm$  4.8 L/d; average DIM 76  $\pm$  18 d; average BW of 542  $\pm$  34 kg) were balanced for milk yield and DIM and assigned to 2 squares. Within a square, the animals were randomly assigned to 3 treatments: grazed grass, zero-grazed grass, and grass silage in a  $3 \times 3$  Latin square design with three 21-d periods. Cows received 3 kg/d per cow of concentrate supplement. The experiment was undertaken between June and August on a perennial ryegrass sward at the Dairy Production Research Centre, Moorepark, Ireland (55°10′ N, 8°16′ W). All the experimental animals were grazing a perennial ryegrass sward before the feeding experiment. Silage was harvested from the same perennial ryegrass sward, field-wilted for 2 h, and baled in bags in May.

### Management of Grazing Cows

A grazing area of 1.7 ha consisted of 2 main grazing blocks. These were subdivided into 2 equal halves to

offer grass for G or ZG animals. Paddock area utilized for the study was divided width-wise in such a way that the G and ZG cows received the same quality of grass throughout the experiment. Grass cuts (GC) were taken twice weekly (at 4 cm from the ground) to determine the herbage mass by cutting 2 strips  $(1.2 \times 10 \text{ m})$  with a motor Agria (Etesia UK Ltd., Warwick, UK). Ten grass height measurements were recorded before and after harvesting on each cut strip using an electronic plate meter (Urban and Caudal, 1990) with a plastic plate (30  $\times$  30 cm and 4.5 kg/ m<sup>2</sup>; Agro Systemes, Choiselle, France). This allowed us to determine the sampled height precisely and allowed the calculation of the sward density [DM per hectare divided by the precutting height minus the postcutting height (kg of DM/cm per ha)]. All mown herbage from each strip was collected and weighed, and a representative sample (300 g) was retained. A 100-g subsample of the herbage was dried overnight at 90°C in an oven for DM determination. The remaining 200 g of the herbage collected from the 2 strips was bulked, subsampled ( $\sim 100 \text{ g}$ ), and stored at  $-20^{\circ}\text{C}$ . Pregrazing and postgrazing sward heights were recorded daily by taking measurements across the diagonals of the grazed strip. The measured pregrazing sward height multiplied by the mean sward density was used to calculate the daily herbage allowance of the grazing animals. Grazing cows were strip-grazed through each half of the 2 blocks to offer 20 kg of DM/cow per day. Electric fences were used for strip-grazing and were moved once daily with a back fence so that the cows did not have access to the area already grazed. A concentrate containing rolled barley (300 g/kg), citrus pulp (460 g/kg), soybean meal (180 g/kg), soybean oil (30 g/kg), and mineral/vitamin mix (30 g/kg) was offered to all cows at a rate of 3 kg/d before the evening milking.

Individual intakes for the grazing cows were measured using a controlled release n-alkane marker [Alkane CRC, Captec (NZ) Ltd., Auckland, New Zealand] in each period. For each animal, one alkane bullet was introduced into the rumen through the cannula on d 7. Herbage snip (**HS**) samples (approximately 500 g in polythene bags), which closely represented the herbage that the cows were selecting, were collected daily from d 7 following the introduction of the bullet for a period of 6 d and stored at  $-20^{\circ}$ C. Fecal grab samples (approximately 250 g) were collected twice daily for 7 d from d 8 following the introduction of the bullet and stored at  $-20^{\circ}$ C until analysis. The HS were freezedried, milled, and analyzed individually. The fecal grab samples of each grazing cow from the total collection period were bulked to obtain one sample per cow per period. This was dried for 48 h at 40°C, milled, and analyzed. The n-alkane concentration in HS and feces

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