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## **BLOAT IN RUMINANTS: THE CELL RUPTURE HYPOTHESIS**

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### **Abstract**

An alfalfa (*Medicago sativa* L.) cultivar with a low initial rate of digestion (LIRD), AC Grazeland B , was developed to test the hypothesis that the rate of cell rupture was a cause of frothy bloat in cattle or sheep. The amount of cell wall increases and digestibility declines as plants mature. Thus the cell rupture theory also predicts that the bloat potential of alfalfa is related to maturity. This study compared the digestibility of cell walls (NDF) in LIRD cultivars with that of a standard, Beaver, in relation to maturity and bloat incidence. Fresh alfalfa was fed to wethers during two experiments. NDF was greater (P#.05) in Beaver (43.9%) than the less mature LIRD cultivar (41.1%) during Experiment 1. The digestibility of NDF was greater (P#.05) in the LIRD cultivar (48.4%; Beaver = 44.5%) during Experiment 2, when both cultivars were in vegetative stages of development but NDF digestibility was lower (P#.05) when the cultivars had matured to the bud stage (LIRD=37.1%; Beaver=48.4%). Bloat incidence did not differ between cultivars at the same stage of development. The number of bloats declined as the alfalfa matured, an effect which was attributed

to the barrier to microbial invasion created by increasing amounts of cell wall and reduced cell wall digestibility. The cell rupture hypothesis of frothy bloat was supported by these results demonstrating that alfalfa maturity plays a key role in bloat etiology.

**Keywords:** Bloat, alfalfa, lucerne, ruminant, cattle, sheep, cell wall, digestibility

## **Introduction**

The cell rupture theory considers the development of bloat in ruminants to be a consequence of the rate of lysis of plant cell walls. Proposed by Howarth et al. (1982), the hypothesis was an advanced theory of forage digestion in the ruminant forestomach. Underlying the theory was the supposition that the surface area and thickness of the plant cell wall limited the initial rate of digestion.

The cell rupture theory implies that equivalent rates of cell lysis will generate similar limits to digestibility and equivalent bloat potentials in the ruminant. As the cell walls are broken apart in the rumen, many forages create a froth of cell constituents, a result of the mixing of gas and fluid during digestion. If the rate of cell lysis is limited then the volume, stability and rate of foam formation will also be constrained, limiting the bloating potential of these forages.

Cell wall thickness, digestibility and therefore cell lysis, are well correlated with age and stage of development in alfalfa (Buxton and Casler 1993). Thus as a corollary, the cell rupture theory predicts that bloat incidence will be a function of alfalfa maturity. A breeding program, undertaken by Agriculture and Agri-Food Canada, has produced AC Grazeland B<sup>f</sup>, a cultivar with a thicker cell wall and a low initial rate of ruminal disappearance (LIRD) (Goplen et al. 1993). The objective of this study was to evaluate maturity, amount and digestibility of cell wall and bloat incidence in

wethers fed diets of fresh cut AC Grazeland, LIRD-3, a progenitor of AC Grazeland, or Beaver alfalfa, a parental standard.

### **Material and Methods**

All trials were conducted at the Agriculture and Agri-Food Canada Research Center near Lethbridge, AB, Canada (49°42' N; 110°47' W). Alfalfa plots were seeded at a rate of 11.25 kg/ha, fertilized with 225 kg/ha of 12-51-0 (N-P-K) and sprinkler irrigated as required.

Two feeding experiments were conducted. Twelve Suffolk wethers (wt 81.6  $\pm$  9 kg) were blocked by weight and randomly assigned to feedings of either LIRD-3 or Beaver alfalfa during Experiment 1. Eight Romanov x Suffolk wethers (wt 48.6  $\pm$  2 kg) received feedings of either AC Grazeland or Beaver alfalfa during Experiment 2. All animals were ruminally cannulated and handled according to the guidelines of the Canadian Council for Animal Care (CCAC 1993). Fresh feed was harvested daily before 0700 am, and all animals received a daily dry matter allowance equivalent to 1.7% of their individual body weight by 0900 am. Wethers were carefully monitored for signs of distension and distress. Ruminal tympanites were diagnosed and bloat was confirmed by palpating the left flank (Garry 1990). Distensions resulting from bloat were relieved by removing the cannula plug. Bloat incidence was recorded as the number of distensions in 24 h since the previous feeding.

After a 10 d adaptation period, samples of feed and feces were collected daily from each wether for the remainder of each experiment. These samples were dried, ground and analyzed for dry matter and neutral detergent fibre (NDF) (Van Soest and Robertson 1980). Each experiment was divided into two or more feeding periods based on the maturity of the LIRD cultivar. In Experiment 1 there were two feeding periods and in Experiment 2, four feeding periods. A standard maturity score was calculated by using the equations of Kalu and Fick (1983) that relate NDF to

morphological stage of development in alfalfa. The frequency of bloats occurring on each cultivar was evaluated against a Poisson distribution of random events (Sokal and Rohlf 1995) to compare differences in the pattern of bloat between cultivars.

### **Results and Discussion**

During feeding Period 1 in Experiment 1, LIRD-3 was at an early bud stage of development while Beaver was at the late bud stage. During Period 2 the two cultivars were in similar ( $P>.05$ ) late bud stages of development. During Experiment 2, maturity of both cultivars increased from the early vegetative stage in Period 1 to the late vegetative stage in Period 4 but did not differ between cultivars ( $P>.05$ ) within each feeding period. The cultivars in Experiment 2 were not as mature as those in Experiment 1.

When both cultivars were in the same stage of development, the amount of cell wall (NDF) was also similar ( $P>.05$ ) (Table 1). The only difference supported the original hypothesis, the more mature cultivar had the greater NDF. Digestibility of the cell wall did not differ ( $P>.05$ ) within Experiment 1, except overall between the cultivars and the LIRD cultivar had a reduced digestibility. Cell wall digestibility declined during Experiment 2 from Period 1 to Period 4 as the two cultivars matured. AC Grazeland cell wall was more digestible at the early vegetative stages of development encountered in Experiment 2. No bloats were recorded during Experiment 1. However, wethers bloated 23 times during Experiment 2. The number of bloats between animals fed each cultivar did not differ ( $P>.05$ ) (12 bloats on AC Grazeland, 11 on Beaver) but the distribution of bloats was considerably more frequent ( $P\#.05$ ) in Period 1 (12 bloats) than in Period 2 (5 bloats), Period 3 (2 bloats) or Period 4 (4 bloats). The frequency distribution of bloats was not random (Table 2). Overall incidents of one bloat per day occurred less frequently than expected, while incidents of 2 or more

bloats per day occurred with greater frequency. The frequency of bloat in animals fed the standard cultivar, Beaver, was clustered. In other words, if one animal bloated there was a greater than expected probability that other animals would bloat at the same time. Bloat frequency in the AC Grazeland fed animals was randomly distributed.

The cell rupture hypothesis as a causal theory of bloat is supported by these results. Increasing amounts of cell wall and declining cell wall digestibility, associated with maturity of the feed, were accompanied by a drop in bloat incidents. The results suggest that barriers to microbial degradation help to maintain a stable digestive environment.

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**Table 1** - Amount and digestibility of cell wall (NDF) and maturity of two alfalfa cultivars, LIRDS-3 and AC Grazeland B<sup>f</sup> compared to a standard, Beaver when fed fresh cut to wethers during consecutive feeding periods

Feeding Period	Experiment 1			Experiment 2		
	Maturity <sup>y</sup>	NDF% <sup>z</sup>		Maturity	NDF%	
		Total	Digestibility		Total	Digestibility
<i>Period 1<sup>x</sup></i>						
LIRD Alfalfa	3.0 <sup>a</sup>	39.4 <sup>a</sup>	36.8	1.2	29.3	53.6
Beaver	4.1 <sup>b</sup>	43.6 <sup>ab</sup>	47.4	1.0	27.8	45.6
<i>Period 2</i>						
LIRD Alfalfa	3.9	42.8 <sup>ab</sup>	37.3	1.1	28.5	50.4
Beaver	4.3	44.1 <sup>b</sup>	49.4	1.0	28.2	48.5
<i>Period 3</i>						
LIRD Alfalfa		-	-	1.6	32.1	46.2
Beaver		-	-	1.7	32.2	44.9
<i>Period 4</i>						
LIRD Alfalfa		-	-	1.7	32.3	43.3
Beaver		-	-	1.4	30.8	39.0
<i>Between Periods</i>						
Period 1	3.5 <sup>a</sup>	41.5 <sup>a</sup>	42.1	1.1	28.6 <sup>a</sup>	49.6 <sup>a</sup>
Period 2	4.1 <sup>b</sup>	43.5 <sup>b</sup>	43.3	1.0	28.3 <sup>a</sup>	49.4 <sup>ab</sup>
Period 3		-	-	1.6	32.1 <sup>b</sup>	45.5 <sup>abc</sup>
Period 4		-	-	1.5	31.5 <sup>b</sup>	41.1 <sup>c</sup>
<i>Between Cultivars</i>						
LIRD Alfalfa	3.4 <sup>a</sup>	41.1 <sup>a</sup>	37.1 <sup>a</sup>	1.4	30.5	48.4 <sup>a</sup>
Beaver	4.2 <sup>b</sup>	43.9 <sup>b</sup>	48.4 <sup>b</sup>	1.3	29.8	44.5 <sup>b</sup>

<sup>z</sup> NDF= Neutral Detergent Fibre as a percentage of dry matter.

<sup>y</sup> Maturity index: Mean Stem Stage by Weight after Kalu and Fick (1983);

1.0= early vegetative, 2.0= late vegetative, 3.0= early bud, 4.0= late bud.

<sup>x</sup> LIRD alfalfa cultivar for Experiment 1= LIRD-3; Experiment 2 = AC Grazeland B<sup>f</sup>.

<sup>a,b</sup> Means in the same column under the same heading, followed by the same letter or no letter are not different (P>0.05).



**Table 2** – Observed and expected (Poisson) frequency distributions of bloat in sheep fed two alfalfa cultivars (cv. AC Grazeland and Beaver)

Frequency <sup>z</sup>	AC Grazeland		Beaver		Overall	
	Observed <sup>y</sup>	Expected	Observed	Expected	Observed	Expected
0	133	132.49	136	133.41	270	265.89
1	10	11.04	5	10.19	15	21.23
2	1	0.46	3	0.39	2	0.85
3	0	0.00	0	0.00	0	0.02
4	0	0.00	0	0.00	1	0.00
5	0	0.00	0	0.00	0	0.00
<i>Statistic</i>						
Mean	0.083	0.083	0.076	0.076	0.080	0.080
SE <sup>x</sup>	0.025	0.024	0.028	0.023	0.021	0.017
CD <sup>w</sup>	1.090	1.007	1.479	1.006	1.621	1.003
X <sup>2</sup>	NS		**		***	

<sup>z</sup> Bloats / day of feeding

<sup>y</sup> Cumulative days of feeding

<sup>x</sup> Standard error of the mean

<sup>w</sup> Coefficient of dispersion =  $s^2/\text{mean}$ . CD=1 indicates random distribution of incidents, CD>1 indicates clumping, CD<1 indicates dispersion (Sokal and Rohlf 1995).

X<sup>2</sup> Goodness of Fit test, \*\* significant at 0.01 probability,\*\*\* = 0.001 probability level