THE EFFECT OF FIBROLYTIC ENZYME SUPPLEMENTATION ON PROTOZOAL POPULATIONS IN BEEF CATTLE FED HIGH GRAIN DIETS

D.R. MILLER[A], *B.W. NORTON*[A] *and R. ELLIOTT*[B]

[A] Schools of Veterinary Science and Animal Studies, University of Queensland, St Lucia, Qld 4072
[B] DSM Nutritional Products, Princeton Court 3, Suite 6, 13 Princeton Court, Kenmore, Qld 4069

The pre-feeding addition of fibrolytic enzyme products to high grain diets fed to dairy cows has been shown to modify microbial populations in the rumen (Nsereko *et al*. 2002). Four *Bos indicus* steers (mean 314 kg, s.d. 26 kg) were allocated per treatment to either barley or sorghum grain-based diets (approx. 60% dry-rolled grain, 27% Pangola grass chaff, 10% cottonseed meal + supplements) with 1 of 2 levels (0 or 13200 IU Xylanase/kg diet DM) of a *Trichoderma longibractiatum*-derived enzyme complex, Roxazyme® G2 Liquid (RG2; F.Hoffmann-La Roche AG, Basel, Switzerland). The RG2 was applied to the grain portion of the concentrate mix at least 24 h before feeding. The steers were fed *ad libitum* such that all the chaff and half of the daily concentrate allocation was offered at 0800 h and the other half of the concentrate mix was fed at 1400 h.

Two weeks after RG2 treatment commenced, a stomach tube was used to collect 5 mL rumen fluid samples both before, and 4 h after, the morning feed. Samples were strained through nylon stockings and stored in 5 mL of 10% formal saline. The preserved rumen fluid (0.25 mL) was diluted with 0.5 mL of 30% glycerol solution and stained with 0.25 mL of Lugol's Iodine Solution. Liquid-associated ciliate protozoa (LAP) were counted (2 chambers/sample) in an Improved Neubauer Bright-Line® Hemacytometer (Hausser Scientific, Horsham, PA) under 100 times magnification. Protozoa were differentiated into the family *Ophryoscolecidae* (subfamily *Entodiniinae*, subfamily *Diplodiniinae* and subfamily *Ophryoscolecinae* as *Epidinium* spp.) or family *Isotrichidae* (as genus *Isotricha* or genus *Dasytricha*) according to Dehority (1993).

Table 1. Liquid-associated ciliate protozoa in rumen fluid after 2 weeks of RG2 supplementation (see the text for details).

Count ($x10^4$/mL)	Barley control	Barley + RG2	Sorghum control	Sorghum + RG2	s.e.m.
Total LAP	191 ± 23 (6.23)[a]	101 ± 17 (5.90)[a]	22 ± 3 (5.22)[b]	29 ± 5 (5.18)[b]	(0.11)
Entodiniinae	179 ± 23 (6.18)[a]	91 ± 16 (5.84)[a]	18 ± 2 (5.13)[b]	28 ± 5 (5.16)[b]	(0.12)
Diplodiniinae	4.2 ± 1.3	2.9 ± 0.8	2.5 ± 0.9	0.7 ± 0.3	
Ophryoscolecinae	N.F.	N.F.	N.F.	N.F.	
Isotricha	5.6 ± 0.9	5.6 ± 0.8	1.5 ± 0.5	0.6 ± 0.2	
Dasytricha	2.2 ± 1.1	2.6 ± 0.8	0.4 ± 0.2	0.1 ± 0.1	
Composition (%)					
Entodiniinae	93.3[a]	87.3[ab]	81.5[b]	96.4[a]	3.5
Diplodiniinae	3.3	3.4	10.6	1.9	2.9
Isotrichidae	3.4[ab]	9.3[b]	7.9[ab]	1.8[a]	2.0

Actual means ($x10^4$/mL ± s.e.) reported with Log_{10} transformed counts (as Least Square Means) in brackets. Population composition data shown as Least Square Means. Means in rows with different superscripts are significantly different ($P<0.05$). N.F. = Not Found.

The barley-based diet recorded higher total LAP and *Entodiniinae* counts than the sorghum-based diet (Table 1). Before feeding, total LAP and *Entodiniinae* counts were higher (P=0.004) than the counts 4 h post-feeding, which probably related to nutrient availability. There was a significant diet/enzyme interaction with respect to the composition of the total LAP population, as a result of RG2-induced changes in the contribution of the *Entodiniinae* population (Table 1). This may reflect differences in the physical structure of the 2 grains, and the resulting mode of enzymatic action, for example an increase in the availability of starch from sorghum grain.

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Email: d.miller2@mailbox.uq.edu.au