Digestion kinetics of New Zealand native flax

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ABSTRACT

This experiment compares the nutritive value of New Zealand native flax, *Phormium tenax,* by-products i.e. leaves (L), butts (B), thrash-stripped fibre (F) and green matter (G), with white clover (WC) via digestion kinetics derived from *in vitro* and *in sacco* incubations. The WC had a high nutritive quality (11.1 MJME/kg DM, CP 28% DM), G a moderate (10.1 MJME/kg DM, CP 9% DM), and L, B, F were of very low nutritive quality (< 8 MJME/kg DM, CP < 6 % DM). After 24 hours, 96, 76, 47, 47, and 30 % of the DM was degraded for WC, G, L, B and F, respectively ($P < 0.001$). The degradation rates (%/h) of digestible DM were faster ($P < 0.001$) for WC (13) and G (11) than B (3.8), L (4.6) and F (2.7). Flax by-products had a net loss of ammonia after 2 (B), 4 (F and L) and 8 hours (G). The proportion of DM converted to VFA by 8 hours was 21 %, and 14 % for WC and G respectively, but only 9-10 % for the other flax by-products ($P < 0.001$). The feeding model BestFeed™ predicted that G could grow a 500 kg bull at 0.8 kg/day and had potential as a supplement for pasture-fed animals.

Keywords: flax; *Phormium tenax; in sacco; in vitro;* digestion kinetics; nutritive value; forage ration, BestFeed.

INTRODUCTION

Recently there has been resurgence in research into the potential of New Zealand native flax, *Phormium tenax,* for the production of bioactives, high fashion fibres, and eco-composites for building (Harvey & Waring, 1987; Duchemin *et al.*, 2003; Sims, 2003; Carr *et al.*, 2005). The harvesting and processing of flax will generate by-products such as leaf blades and flax leaf bases or butts. In addition during processing for fibre, flax is separated into thrash-stripped fibre and green strippings and all these processing by-products may have potential as feeds for ruminants.

The nutritive value for ruminants and digestion kinetics of flax by-products can be described using *in sacco* and *in vitro* techniques (Barrell *et al*., 2000; Burke *et al*., 2000; Chaves, 2003) . The *in sacco* incubation enables digestion to be evaluated in terms of the rate of disappearance of dry matter, crude protein and fibre. The *in vitro* incubation method measures nutritive value in terms of the products of fermentation such as ammonia and volatile fatty acids.

The BestFeed™ (Pacheco, 2002) model was developed to assist farmers with the selection and use of supplementary feeds. In addition to the nutritive value and digestion kinetics of the feed, inputs such as environment (terrain, legume content, feed availability) and animal type (species, age, weight, sex, breed, fleece weight) are used to predict live-weight gain (LWG). Animals can be fed to achieve target LWG and/or fed combinations of feeds to optimise cost or predict maximum intake. BestFeed is based on the Australian and British energy and protein standards (Feeding standards for Australian livestock, 1994; AFRC, 1995) and intake prediction algorithms developed for New Zealand stock by Marc Ulyatt (Pacheco, 2002).

The objective of this study was to evaluate the nutritive value of flax via *in sacco* and *in vitro* incubations, and then utilising these results, to model the nutritive potential of flax by-products as feed for cattle.

METHOD

Sample preparation

Thirty mature flax leaves were harvested by hand from 5 flax bushes growing at a Foxton swamp site in April 2004. Twenty of these leaves were then randomly selected and thrashed at a flax fibre mill (Foxton, Manawatu, New Zealand) and both the stripped green matter and remaining fibre were collected. All fibrous samples were chopped by guillotine into cubes (10 mm^2), then all samples were freeze-dried, and sieve-ground (< 1 mm) and a subsample was analysed for all nutrients by wet chemistry.

Flax samples along with the laboratory standard of freeze-dried white clover (WC, *Trifolium repens*) were digested by *in sacco* and *in vitro* incubation. Flax was analysed as components of whole flax leaf or blade (L), flax leaf butt or base (B), flax green matter stripped by thrashing (G), and flax fibre remaining after thrashing (F).

In sacco

Freeze-dried and sieve-ground (< 1 mm) sample was weighed (6g) and placed into 100 x 100 mm Dacron bags (mean pore size 25 μm) (Burke *et al.,* 2000). In April 2004, three non-lactating dairy cows, two Friesian-Jersey cross and one Jersey, grazing high quality perennial ryegrass and white clover mixed pasture, were used for the *in sacco* digestions. Six bags of each sample type were placed in the rumen of each of

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the three cows, and one bag of each sample type was removed from each cow after 2, 6, 12, 24 and 72 hours. Bags were hand-rinsed in cold water until the wash water ran clean, and oven-dried (60°C, 48 hours). Residues were ground and analysed by Near Infrared Reflectance Spectroscopy (NIRS) to estimate crude protein (CP) and fibre contents (neutral detergent fibre, NDF; and acid detergent fibre, ADF) (Corson *et al.,* 1999). Ten samples were also analysed by wet chemistry to assist in the establishment of a NIRS calibration standard curve.

In vitro

Freeze-dried and sieve-ground (< 1 mm) sample was weighed (0.6 g) into 50ml Schott® bottles and warmed to 39°C in an oscillating incubator (90 oscillations minute^{-1}). Twelve ml of artificial saliva (McDougall, 1948), reducing agent (0.5 ml), and strained bovine rumen liquor (3 ml) (sourced from 1 cow) were added (Burke *et al.,* 2000). Bottles were then gassed with CO_2, returned to the incubator and removed after 1, 2, 4, 8, and 24 hours of incubation. One ml sub-samples were taken and acidified (conc. HCl) for determination of ammonia (Bergemyer and Beutler, 1985). Further 1 ml sub-samples were taken at 0 and 8 hours for determination of volatile fatty acids (VFA) by gas liquid chromatography (Attwood *et al.,* 1998). All sub-samples were centrifuged at 4000 rpm for 15 minutes and the supernatant collected and frozen (-20 °C). Rumen liquor pH was also measured at each sampling point. The *in vitro* digestion was performed three times over three days.

Statistical analysis, digestion kinetics and ration formulation

Disappearance rate of DM and other fractions were analysed using a non-linear model described by Burke *et al*. (2000). Fractional disappearance rate (k, % h^{-1}), lag time (L, h) and potential degradation (P %) were determined according to:

$$P = A + B\,(1 - e^{-k\,(t - L)})$$

where A is soluble DM (% of DM at t = 0 h), B is degradable insoluble DM, and t is time in hours.

Kinetic parameters were analysed using the general linear model (SAS, 1999) and analysed for effects of feed type. Flax fibre exhibited unusual digestive behaviour relative to that of common pasture and forages; therefore certain limits were imposed on some of the kinetic parameters to achieve fitted curves.

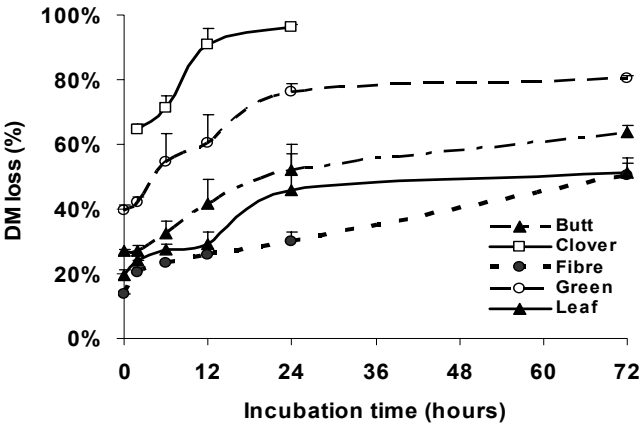
BestFeed™ (Pacheco, 2002) simulations were conducted using 500 kg Friesian bulls grazing typical autumn (9.8 MJME/kg DM, 20% CP, 44% NDF) and winter (11 MJ ME/kg DM, 26% CP, 40% NDF), flat North Island beef farm pasture (Litherland *et al*., 2002). Bull LWG was modelled using sole diets of pasture or flax by-products, and then flax G was supplemented to autumn pastures for growing bulls. The minimum amount of pasture required to be fed with flax by-products, in order to maintain bull live weight (LW), was also determined.

RESULTS

In sacco

The chemical composition of the WC and flax by-products as determined by wet chemistry are presented in Table 1. White clover had a typically high nutritive quality, G a moderate quality, and L, B and F were of very low nutritive quality (Table 1). Following 24 hours incubation in the rumen, 96% DM of the WC had been digested, compared with 76, 52 and 46% DM of the G, L, and B, respectively (Figure 1). Only 30 % DM of the F had been digested after 24 hours incubation ($P < 0.001$). The potential degradability of the F was 51%, however this required retention in the rumen for 72 hours. The fractional DM degradation rates of the fibrous flax by-products, i.e. L, B and F, were approximately half that of the fractional DM degradation rates for G and WC (Table 1).

FIGURE 1: *In sacco* dry matter degradation curves of white clover and New Zealand native flax: 'leaf', stripped 'green' matter, 'fibre' and 'butt'. Error bars are standard deviations.



While flax fibre was poorly degradable, flax CP was readily degradable. After 24 hours in the rumen, ADF degradability was 93, 45, 33, 24 and 3% DM; NDF degradability was 95, 57, 33, 30 and 9 % DM; and CP degradability was 98, 88, 88, 87, and 81% DM for WC, G, B, L and F, respectively.

In vitro

In vitro incubations showed that up to 39% of WC plant nitrogen was released as ammonia over 24 hours. However, the flax by-products had a net loss of ammonia after 2 (B), 4 (F, L) and 8 hours (G) incubation (Figure 2).

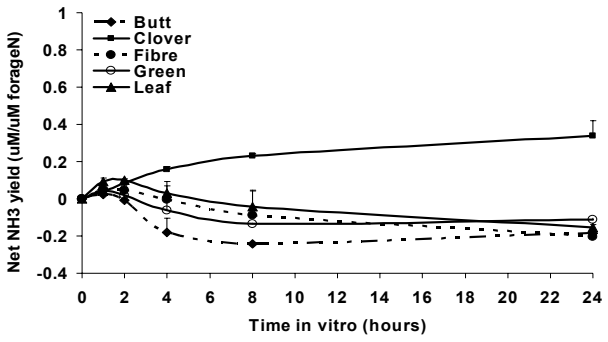
TABLE 1: White clover (WC) and New Zealand native flax; stripped green matter (G), leaf (L), butt (B), and stripped fibre (F): DM percentage; acid detergent fibre (ADF), neutral detergent fibre (NDF) crude protein (CP); soluble carbohydrates (CHO), lipid, starch and organic matter digestibility (OMD); kinetic data (% DM) defined as soluble DM (A), degradable insoluble DM (B), potential degradable DM (P), fractional degradation rate (k, % h^{-1}), lag time (L, h) and effective degradability (E).

Forage	ADF[1]	NDF[1]	CP[1]	CHO[1]	Lipid[1]	Starch[1]	OMD[2]	A	B	P	k	L	E[3]
WC	16	21	28	8	2.9	1.4	78	53 [a]	43	96 [a]	13	0.0	82 [a]
G	30	39	9	14	1.9	3.6	70	40 [b]	41	81 [b]	11	2.1	63 [b]
B	40	58	4	11	0.8	2.1	48	27 [c]	44	71 [b]	6	2.5	43 [c]
L	49	73	5	6	0.9	0.33	41	21 [d]	36	56 [c]	6	1.9	35 [d]
F	49	69	6	6	1.1	0.76	43	16 [e]	34	51 [c]	3	0.1	27 [e]
SEM								1	4	3	3	1.0	2
P								0.001	0.28	0.001	0.12	0.46	0.0001

[1] Measured by wet chemistry
[2] Estimated by NIR
[3] Calculated using an assumed fractional passage rate of 0.06 h^{-1}
Different superscripts in the same column differ significantly ($P < 0.05$)

For WC, 21 % DM was used for the production of VFA by 8 hours, which was higher ($P < 0.001$) than the 14% produced from G. The fibrous flax by-products converted 9-10% of the DM to VFA (Table 2). Acetate represented a greater proportion of the VFA produced by L and F digestion than the other flax components.

FIGURE 2: Net ammonia (NH_3) release from *in vitro* incubations for white clover and Zealand native flax: 'leaf', stripped 'green' matter, 'butt' and stripped 'fibre'. Error bars are standard deviations**.**



Overall the fibrous flax by-products maintained an *in vitro* pH at higher levels (all pH 7.0, $P < 0.0001$) than WC (pH 6.7) or G (pH 6.5). The rumen pH was similar in all forages with up to 2 hours of digestion but at 4 hours WC and G diverged progressively (Forage*time $P < 0.01$). Following 24 hours of incubation, the pH of the G had dropped more than that of the WC (L, F, B 7.4; WC 6.5, G 5.9). However, *in vitro* pH never declined below 5.6, which is considered to be the threshold of impaired digestion (de Veth and Kolver, 2001).

Ration formulation

Growing bulls. Simulations using BestFeed suggested that sole diets of typical winter or autumn pasture or G could support 2.3, 0.4 or 0.8 kg/day of bull LWG respectively. BestFeed™ predicted a 2 kg DM/day increase in feed intake compared with sole diets of autumn pasture or G and an increase in LWG to over 1 kg/day. This was driven by an empirical substitution algorithm in BestFeed™ derived largely from concentrate plus forage feeding data, whereas our scenario was more akin to forage plus forage supplementation.

Bulls at maintenance. BestFeed™ calculated that 7.2, 7.9 or 8.5 kg DM of winter pasture, G or autumn pasture, respectively, were required to achieve LW maintenance. The sole feeding of fibrous by-products resulted in substantial LW loss (more than 1 kg/day LWG).

On both autumn and winter pastures, if DM intake of pasture was halved, maintenance LW was achievable by supplementing with 3.9 kg DM of G. The amount of fibrous by-products that could be substituted for either winter or autumn pastures, while still achieving maintenance feeding level were determined (Table 3). Maintenance feeding could still be achieved following replacement of 24% of winter pastures for L, however, for all other combinations of fibrous flax by-products and pasture types the savings in pasture following feeding of flax by-products were less than 10%.

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TABLE 2: Volatile fatty acid (VFA) production from *in vitro* incubations after 8 hours for white clover (WC), Zealand native flax; leaf (L), stripped green matter (G), butt (B) and stripped fibre (F).

	WC	G	L	B	F	SEM	P <
Yield of VFA (mg/gDM)	210 [a]	141 [b]	101 [c]	101 [c]	90 [c]	9	0.001
Acetate (%)	40 [b]	44 [b]	51 [a]	43 [b]	49 [a]	1	0.001
Propionate (%)	38 [a]	33 [a]	29 [ab]	33 [a]	28 [b]	2	0.01
Butyrate (%)	15 [b]	21 [a]	16 [b]	22 [a]	19 [ab]	1	0.05
Minor VFA (%)	7 [a]	3 [c]	5 [b]	2 [c]	4 [bc]	0.5	0.001
Acetate:Propionate	1.0 [b]	1.35 [b]	1.8 [a]	1.3 [b]	1.8 [a]	0.1	0.01

Different superscripts in the same column differ significantly (P < 0.05)

TABLE 3: BestFeed™ simulations for feed intake required of minimum pasture and flax leaf (L), butt (B) or fibre (F) rations to achieve maintenance of a 500 kg Friesian bull.

	Intake (kg DM/hd/day)			
	Winter pasture	**Flax**	**Autumn pasture**	**Flax**
L	5.5	5.8	7.8	2.3
B	6.7	4.1	8.3	1.8
F	8.5	1.5	8.5	1.5

On both autumn and winter pastures, if DM intake of pasture was halved, maintenance LW was achievable by supplementing with 3.9 kg DM of G. The amount of fibrous by-products that could be substituted for either winter or autumn pastures, while still achieving maintenance feeding level were determined (Table 3). Maintenance feeding could still be achieved following replacement of 24% of winter pastures for L, however, for all other combinations of fibrous flax by-products and pasture types the savings in pasture following feeding of flax by-products were less than 10%.

DISCUSSION

In vitro and *in sacco* digestions revealed that flax G possessed similar nutritive and digestive characteristics to that of mature perennial ryegrass leaf (Burke *et al*., 2000; Chaves, 2003). BestFeed™ predicted that the energy and protein levels contained in G were sufficient, even as a sole diet, to support moderate LWG older cattle. However in young cattle the low protein content of G would limit its use as a sole diet. Green strippings could be used to supplement pasture-fed cattle of all ages both to maintain LW during feed deficits, or to boost LWG on poor quality pasture. This experiment shows that field studies with G are warranted and should examine aspects such as palatability, anti-nutritional factors, and the development of practical storage and feeding systems.

The degradation rates of digestible DM for B and L were equivalent to that of grass stem (Chaves, 2003) with F being even slower. However, only about 30% of DM contained in L, B and F were effectively degraded compared with approximately 50% in low quality grass stem (Chaves, 2003). This is largely due to ADF and NDF being virtually non-degradable in flax, whereas in pasture more than 50% of ADF and NDF is degradable (Burke, 2004). The protein content of flax was low compared with pasture but this protein had similar degradability to grass protein (Burke, 2004).

Fresh mincing of forage samples for *in vitro* and *in sacco* digestions more closely reflect normal digestion (Barrell *et al.,* 2000). However due to the fibrous nature of flax, freeze-drying and sieve-grinding were the only practical preparation methods and this is likely to have over-estimated degradability by approximately 10% (Barrell *et al.,* 2000). Even for this dried-ground material, *in vitro* measurements and BestFeed™ modelling showed that digestion of fibrous flax by-products would not progress to completion without additional energy and protein sources. Overseas studies showed that a diet of 40% linen flax straw, ammoniated to increase nitrogen content, and 60% barley grew dry dairy cows at 1 kg/hd/day (Mann *et al.,* 1988). Concentrate supplements however, are not normally economically viable in New Zealand.

BestFeed™ revealed insufficient savings in pasture to warrant feeding fibrous flax by-products. The only potential role for fibrous flax by-products is as an additive to the diet for stock grazing fibre-deficient forages. This was beyond the scope of the BestFeed™ simulation to model.

In conclusion, the nutrition costs to the ruminant of digesting fibrous flax by-products were too high to warrant feeding to livestock. However, flax stripped green matter has nutritive potential as a supplement to pasture fed animals.

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