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The additive effects of condensed tannins on the disappearances of protein, cell wall and lignin from semi-arid browse foliage *in sacco*

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Introduction

Condensed tannin (proanthocyanidin or PCy) in the foliage of trees and shrubs (browse) can reduce the degradability of browse in the rumen, in particular its neutral-detergent (ND) fibre, fractions of ND fibre, and crude protein.

We have observed that over 90% of butanol-HCl PCy in browse is soluble in ND with sulphite. We hypothesized that: (a) removing ND soluble matter from browse would improve the rumen degradability of ND fibre and its fractions *in sacco* (from nylon bags); (b) the improvement would be proportional to the PCy content of the browse; (c) ND soluble protein not degraded in the rumen would also vary with PCy content; and (d) the effects of PCy on the degradabilities of the various fractions would be correlated. These hypotheses were tested with the freeze-dried foliage of ten browse species collected from Charleville in subtropical semi-arid Queensland, Australia, in spring and autumn.

Material and methods

Seven rumen-cannulated Merino sheep were given 17 g dry matter (DM) per kg live weight per day of bird-resistant sorghum stover with urea and minerals for a 20-day period, the *in sacco* trial occupying the last 6 days. There were 2 × 2 × 2 bags of each browse species, there being two types of preparation (*X*, extracted in ND or *W*, washed in warm water at 60 to 70°C), two samples of browse, and two durations of incubation (48 or 96 h). Six nylon bags were incubated in each sheep at once. The browse species were: MLG, mulga (*Acacia aneura*); IWD, ironwood (*A. excelsa*); MYL, myall (*A. pendula*); BEL, belalie (*A. stenophylla*); KRJ, kurrajong (*Brachychiton populneum*); BTT, bottle tree (*B. rupestre*); BMB, bumbil (*Capparis mitchellii*); LWD, leopardwood (*Flindersia maculosa*); WLG, wilga (*Geijera parviflora*); and RWD, rosewood (*Heterodendron oleifolium*), and were sampled in autumn and spring except for BEL, BMB and RWD (autumn only).

PCy was measured in a spectrophotometer at 550 nm after heating in 95 : 5 : 5 : 5 butan-1-ol : HCl : methanol : water at 95°C for 2 h (Bate-Smith, 1981), using mulga condensed tannin as a standard. For fibre analysis (Goering and Van Soest, 1970), sintered glass crucibles of porosity 2 (0·020 to 0·040 mm) were used. Nylon bags had a pore size of 0·035 mm, were 90 x 170 mm internally and contained 2·2 g DM of *W* or an equivalent mass of *X*. All material was allowed to become equilibrated with air before sampling from nylon bags or crucibles. The residue of *W* was then ND extracted for 2 h, and *X* was ND extracted for 1 h, to make the total ND extraction times 2 h for both *W* and *X*. Acid-detergent fibre and Klason lignin were determined on the ND residue, from which hemicellulose and cellulose were calculated.

Results and discussion

The yields of hot water-washed and ND-extracted browse DM are shown in Table 1. In all species proportionately over 0·9 of the PCy was soluble in ND and was therefore located mainly in cell contents. Of the nitrogen in the browse, most (9 to 19 mg/g browse DM, or 24 mg in bumbil) was removed by ND; the residue of this fraction after rumen incubation is shown in Table 1. The ND-insoluble nitrogen fraction was 5 to 7 mg/g browse DM in *Acacia* spp. or 2 to 4 mg in others; about half of it was associated with lignin.

Between 0·16 and 0·27 of ND fibre disappeared from washed browse (*W*) in 48 or 96 h, with the exception of wilga (0·43). From browses that had previously been extracted in ND (*X*), the disappearance of ND fibre ranged from two to three times higher than from *W* (*Brachychiton* spp.), through 1·5 times higher (bumbil and rosewood), down to no significant difference (*Acacia* spp., leopardwood and wilga). Hemicellulose and cellulose were the main components of cell wall disappearing, but in wilga lignin alone was affected.

Table 1 *Composition of browse before incubation and residue after incubation, and effects of prior ND-extraction on fibre residues (mg/g original browse dry matter (DM))*

	Genus									
	Acacia				*Brachychiton*		Other			
Species†	MLG	IWD	MYL	BEL	KRJ	BTT	BMB	LWD	WLG	RWD	s.e.
Composition of original browse											
Hot water-washed browse DM (*W*)	803	787	716	596	851	817	640	809	670	747	
ND-extracted browse DM (*X*)	479	440	473	394	413	352	315	245	214	307	
Proanthocyanidin	40	54	30	36	103	147	14	74	89	94	
Residue after incubation in the rumen‡											
ND-soluble N × 6·25	32	32	23	26	16	26	0	13	32	35	3·2
Difference in quantity of residue after incubation between *W* and *X*											
Hemicellulose	15	3	7	8	32*	53***	11	8	6	34*	10·8
Cellulose	10	2	2	4	85***	39***	15	6	1	21*	9·4
Lignin	0	1	0	–7	6	16	14	19	24*	4	7·3
ND fibre	24	5	6	–3	101***	112***	38*	25	29	48*	15·6

† See **Material and methods**

‡ 0·27 of ND soluble N × 6·25 was not degraded in bumbil; this fraction was subtracted from the undergraded ND-soluble N × 6·25 of all browses.

Three possible effects of PCy were studied: *E1*, undegraded ND-soluble N × 6·25; *E2*, increase in undegraded cellulose and hemicellulose; *E3*, increase in undegraded lignin. The correlations (*r*) of these *E* values with PCy concentration were –0·09, 0·79 ($P < 0·05$) and 0·65, respectively. Within PCy levels, the correlations between the *E* values were negative: –0·23 (*E1*, *E2*), –0·18 (*E1*, *E3*) and –0·73 ($P < 0·05$) (*E2*, *E3*), indicating that the effects of PCy were mutually exclusive. Klason lignin residues were associated with cell wall disappearance, consistent with the hypothesis that the extra 'lignin' was inactivated tannin (Goodchild and McMeniman, 1992). By multiple regression, the combined effects of *E1*, *E2* and *E3* were highly correlated with PCy:

$$\text{PCy (mg/g)} = 17·1 + 0·85E1\ (P > 0·05) + 0·73E2\ (P < 0·01) + 2·07E3\ (P < 0·01)$$
$$(r = 0·952;\ P < 0·001).$$

There are several possible reasons for between-species differences in the effect of tannin on components such as ND soluble nitrogen, (hemi) cellulose and lignin. These include species variation in: (a) chemistry or microstructural inter-relationships of components; (b) specificity of tannins, either for components or for rumen microbes and their enzymes; (c) localization of tannins in plant tissues. Since favourable effects of condensed tannins on ruminant nutrition are known, future work should optimize the relationship between tannins, nutritional components, plant tissues and rumen microbes.

References

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