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Review article

The implications of condensed tannins on the nutritive value of temperate forages fed to ruminants

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New methodology for measuring forage condensed tannin (CT) content is described and the effects of CT upon forage feeding and nutritive value for ruminant animals are reviewed. CT react with forage proteins in a pH-reversible manner, with reactivity determined by the concentration, structure and molecular mass of the CT. Increasing concentrations of CT in *Lotus corniculatus* and *Lotus pedunculatus* reduce the rates of solubilization and degradation of fraction 1 leaf protein in the rumen and increase duodenal non-NH_3 N flow. Action of medium concentrations of total CT in *Lotus corniculatus* (30–40 g/kg DM) increased the absorption of essential amino acids from the small intestine and increased wool growth, milk secretion and reproductive rate in grazing sheep without affecting voluntary feed intake, thus improving the efficiency of food conversion. High concentrations of CT in *Lotus pedunculatus* (75–100 g/kg DM) depressed voluntary feed intake and rumen carbohydrate digestion and depressed rates of body and wool growth in grazing sheep. The minimum concentration of CT to prevent rumen frothy bloat in cattle is defined as 5 g/kg DM and sheep grazing CT-containing legumes were shown to better tolerate internal parasite infections than sheep grazing non CT-containing forages. It was concluded that defined concentrations of forage CT can be used to increase the efficiencies of protein digestion and animal productivity in forage-fed ruminants and to develop more ecologically sustainable systems of controlling some diseases under grazing.

Condensed tannins: Forage: Protein digestion

Temperate forages grazed in the leafy vegetative state contain high concentrations of metabolizable energy (11·5 MJ/kg DM) and total N (30 g/kg DM). Rumen digestion of readily fermentable and structural carbohydrate is efficient on such diets (Ulyatt & MacRae, 1974), but with N digestion duodenal flow of non-NH_3 N (NAN) is only about 65 % of the N eaten (MacRae & Ulyatt, 1974). This is due to the extent of degradation of forage proteins to NH_3 by rumen micro-organisms (70–80 %) being much faster than the rate that NH_3 can be incorporated into microbial protein, resulting in high absorption of N as NH_3 from the rumen (Ulyatt *et al*. 1975). Subsequent research, using post-ruminal infusions of proteins and amino acids or dietary supplementation with undegraded proteins has identified absorption of essential amino acids (EAA) from the small intestine as limiting productivity in ruminants fed entirely on diets of high quality fresh forages *ad libitum* (Barry, 1981). A search then commenced for plant compounds that would reduce the degradation of proteins in the rumen and increase EAA absorption in ruminants fed on fresh forages. This paper reviews progress with forage condensed tannins (CT) for fulfilling this objective.

Condensed tannin structure and reactivity

Condensed tannins or proanthocyanidins comprise polymerized flavan-3-ol-units, and those occurring in temperate forages have a relative molecular mass of 2000–4000 comprising ten to twelve units condensed together (Foo *et al*. 1982) (Fig. 1). They normally occur in plant vacuoles. The chemistry of CT is complex. First, there are differences in the hydroxylation of the B-ring of the flavan-3-ol monomer units. The stereochemistry of the heterocyclic C-rings can take the form of 2,3-*cis* or 2,3-*trans* and this dictates

Abbreviations: CT, condensed tannins; EAA, essential amino acids; NAN, non-ammonia nitrogen; Rubisco, ribulose-bisphosphate carboxylase/oxygenase; VFI, voluntary feed intake.

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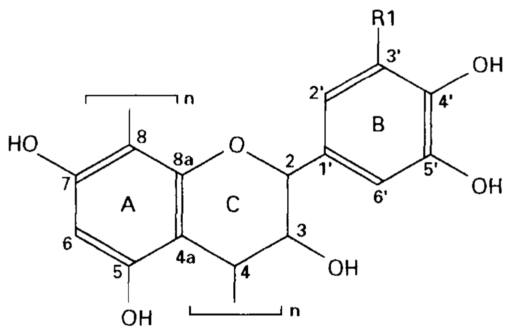

Fig. 1. Constituent flavan-3-ols of the condensed tannins in *Lotus corniculatus* and *Lotus pedunculatus*. When R1 is H, the condensed tannin is (epi)catechin (procyanidin); when R2 is OH, the condensed tannin is (epi)gallocatechin (prodelphinidin).

how the monomeric units are attached relative to one another. The monomeric units can be linked by C4/C8 or C4/C6 interflavanoid linkages which effects the shape of the CT polymer chain. Finally, the number of monomeric units (i.e. the degree of polymerization) can vary (Foo *et al*. 1996, 1997). Together these differences can produce an infinite variety of chemical structures, which in turn affect the biological properties of the CT.

CT from *Lotus corniculatus* (Birdsfoot trefoil) and *Lotus pedunculatus* (Big trefoil) differ considerably in their chemical structures (Foo *et al*. 1996, 1997). The average relative molecular mass of the CT from *Lotus pedunculatus* is 2200 and this is slightly higher than the relative molecular mass of CT from *Lotus corniculatus* (1900). Furthermore, the CT from *Lotus pedunculatus* contains a predominance of prodelphinidin-type subunits with epigallocatechin (0·64) being the prevalent subunit (Foo *et al*. 1997). Conversely, the CT from *Lotus corniculatus* has predominantly procyanidin-type subunits with epicatechin (0·67) dominating this CT (Foo *et al*. 1996).

CT bind strongly to proteins, and it has been proposed that some plants evolved CT production as a chemical defence, first against invasion by pathogenic micro-organisms, then against being eaten by insects and finally against being eaten by grazing herbivores (Swain, 1979). Jones & Mangan (1977) first showed in laboratory studies that reactivity between CT and forage protein was pH-dependent, with stable complexes being formed at pH 3·5–7·5, but the complexes dissociating and releasing protein at pH $<$ 3·5. Much research with animals (to be reviewed here) then followed, examining this reactivity as the basis for increasing undegradable protein and EAA absorption in ruminants fed entirely on diets of fresh forages.

For many years, CT was extracted from plants with acetone–water (70 : 30, v/v) and it was assumed that this extracted all the CT. However, the development of subsequent protein-bound and fibre-bound steps (Terrill *et al*. 1992*a*) (Table 1) showed that this was not the case, with extractable CT representing on average about 70–80 % of total CT. In New Zealand, it was originally thought that the only CT-containing forages were *Lotus* species, sulla (*Hedysarium coronarium*) and sainfoin (*Onobrychis vicifolia*). However, the newer methodology has shown the presence of very low CT concentrations in the common grasses, legumes and herbs used in temperate grazing systems (Table 1) and this has been confirmed using detection of ^{13}C by NMR spectrometry and by anthocynanidin formation (Jackson *et al*. 1996).

Some browsing animals, notably deer, have evolved production of CT-binding proline-rich salivary proteins, as a means of counteracting the plants' chemical defence against defoliation and of reducing the anti-nutritional effects of high CT concentrations (Austin *et al*. 1989). In some cases these salivary proteins can be highly specific, as in the case of moose (*Alces alces*), and only bind the type of CT that is present in the normal diet eaten and will not bind other types of CT (Hagerman & Robbins, 1993). However, in studies conducted to date, domesticated sheep and cattle (i.e. grazers) do not produce CT-binding proteins in their saliva (Austin *et al*. 1989). This is indeed fortunate, as it means dietary CT can be used to manipulate N digestion in sheep and cattle fed on fresh forages. The situation is less clear with domesticated farmed deer, and this will be referred to later.

Table 1. The extractable and bound condensed tannin content of legumes, grasses and herbs fed to ruminants in temperate grazing systems, measured by the butanol–HCl method*

	Condensed tannin (g/kg DM)			
Forage	Extractable	Protein-bound	Fibre-bound	Total
Legumes				
Big trefoil (*Lotus pedunculatus*)	61	14	1	77
Birdsfoot trefoil (*Lotus corniculatus*)	36	9	2	47
Sulla (*Hedysarum coronarium*)	33	9	3	45
Sainfoin (*Onobrychis vicifolia*)	29			
Red clover (*Trifolium pratense*)	0·4	0·6	0·7	1·7
Lucerne (*Medicago sativa*)	0·0	0·5	0·0	0·5
Grasses				
Perennial ryegrass (*Lolium perenne*)	0·8	0·5	0·5	1·8
Herbs				
Chicory (*Chicorium intybus*)	1·4	2·6	0·2	4·2
Sheeps burnet (*Sanguisorba minor*)	1·0	1·4	1·0	3·4

* From Terrill *et al.* (1992*b*); Jackson *et al.* (1996).

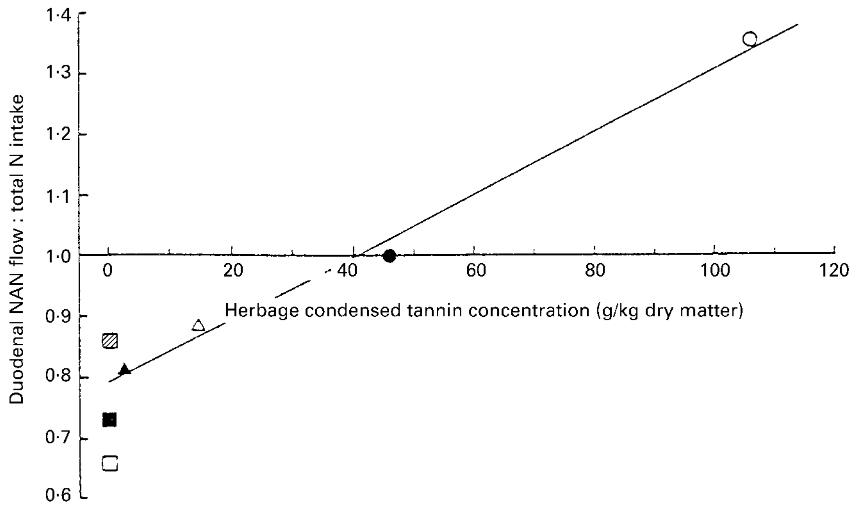

Fig. 2. Duodenal non-ammonia nitrogen (NAN) flow per unit total nitrogen intake as a function of herbage condensed tannin concentration in sheep fed on *Lotus* species. (○), High- and (●) low-tannin *Lotus pedunculatus*; (△), high- and (▲) low-tannin *Lotus corniculatus*. Results are compared with the non-tannin-containing herbages; (▨), short rotation ryegrass; (□), perennial ryegrass; (■), white clover. All results are for a nitrogen intake of 28 g/d and refer to fresh forages. From Barry & Manley (1984).

Feeding value

Ulyatt (1973) defined feeding value as the animal production from grazing a forage under unrestricted conditions, with its components being voluntary feed intake (VFI), the digestion process and the efficiency of utilization of digested nutrients. The effects of CT on each of these processes will be reviewed, using the sheep as the experimental animal unless stated otherwise. Data will mainly be quoted from work with fresh *Lotus* legume species, as most work in New Zealand has been done with these species. Effects of CT have been deduced by comparing unsupplemented sheep (CT acting) with a group of sheep supplemented with PEG (relative molecular mass 3350), as PEG specifically binds and inactivates CT without affecting microbial or digestive enzymes (Jones & Mangan, 1977; Barry & Manley, 1986).

Voluntary feed intake

High CT concentrations in *Lotus pedunculatus* (63 and 106 g/kg DM) substantially depressed VFI in sheep (–27 %), in accordance with plant CT production being a defence against consumption by herbivores (Barry & Duncan, 1984). Smaller depressions in VFI (–12 %) were produced by 55 g CT/kg DM in *Lotus pedunculatus* (Waghorn *et al*. 1994). However, medium CT concentrations in sulla (45 g/kg DM) and in *Lotus corniculatus* (34 and 44 g/kg DM) had no effect upon VFI (Terrill *et al*. 1992*b*; Wang *et al*. 1996*a*,*b*).

Digestion of nitrogen and of carbohydrate

Duodenal NAN flow (Fig. 2) can be used as an index of protein-N leaving the rumen. With perennial ryegrass (*Lolium perenne*), short-rotation ryegrass (*Lolium multiflorum*

Table 2. Effect of condensed tannin (CT) upon the intake and absorption of essential amino acids from the small intestine of sheep fed on fresh *Lotus corniculatus* and *Lotus pedunculatus*, containing respectively 22 and 55 g CT/kg DM*

	Lotus corniculatus†		*Lotus pedunculatus*‡	
	CT acting	PEG supplemented	CT acting	PEG supplemented
Rumen ammonia (mg N/l)	367	504	175	458
CT intake (g/d)	98·9	98·9	103·2	116·8
Abomasal flow:				
g/d	84·7	55·5	121·1	105·6
Proportion intake	0·86	0·56	1·17	0·90
Apparent absorption from small intestine:				
g/d	58·8	36·2	81·4	83·5
Proportion abomasal flow	0·67	0·67	0·66	0·79
Proportion intake	0·59	0·37	0·79	0·72

* From Waghorn *et al.* (1987, 1994).
† Excluding arginine.
‡ Including arginine.

$\times$ *Lolium perenne*) and white clover (*Trifolium repens*), which contain only traces of CT, duodenal NAN flow is only about 0·75 of N intake, illustrating the extensive absorption of NH_3 from the rumen. However with *Lotus* species duodenal NAN flow increased linearly with increasing CT concentration and equalled N intake at a CT concentration of approximately 40 g/kg DM (Fig. 2). *In situ* and *in vitro* experiments have shown that this is due to action of CT in *Lotus pedunculatus* and *Lotus corniculatus* slowing the rates of both solubilization and degradation of forage proteins by rumen micro-organisms, especially that of the principal leaf protein, ribulose-bisphosphate carboxylase/oxygenase (*EC* 4.1.1.39) (Rubisco; fraction 1 leaf protein) (McNabb *et al.* 1996; BR Min, WC McNabb, TN Barry and JS Peters, unpublished results).

Effects of CT upon apparent absorption of EAA from the small intestine differed between *Lotus corniculatus* (22 g extractable CT/kg DM; Waghorn *et al.* 1987) and *Lotus pedunculatus* (55 g extractable CT/kg DM; Waghorn *et al.* 1994) and are shown in Table 2. When expressed as a proportion of N intake, action of CT in *Lotus corniculatus* increased both abomasal flow (+53 %) and the net absorption of EAA from the small intestine (+59 %), with no effect on apparent digestibility (proportion abomasal flow) in the small intestine. However, whilst action of CT increased abomasal flow in sheep fed on *Lotus pedunculatus* (+30 %), this was counteracted by reduced apparent digestibility in the small intestine, with there being only a small increase in apparent absorption of EAA from the small intestine (+10 %). These effects with *Lotus pedunculatus* could be due to effects of the CT in not releasing some amino acids in the small intestine, increasing endogenous protein secretion or inactivating digestive enzymes. These are all areas for future research. In addition to differences in CT concentration between the two species, reactivity of CT with plant proteins differs between *Lotus corniculatus* and *Lotus pedunculatus* for other reasons, such as differences in CT structure.

Although the differences in CT structure between *Lotus corniculatus* and *Lotus pedunculatus* were insufficient to cause any appreciable difference in the *in vitro* precipitation of Rubisco when these CT extracts were reacted with total soluble leaf protein from white clover (McNabb *et al.* 1998), the CT extract from *Lotus pedunculatus* was more effective at reducing Rubisco degradation by rumen micro-organisms than the CT extract from *Lotus corniculatus* (Aerts *et al.* 1999*b*). Taken together, the results of Aerts *et al.* (1999*b*) and McNabb *et al.* (1998) suggest that protein precipitation by CT may be more responsive to the relative molecular mass of the CT, and to a lesser extent, effected by the prodelphinidin content, whilst the effect of CT on the degradation of protein by rumen micro-organisms may be more responsive to differences in the flavan-3-ol composition of the CT. This certainly warrants further investigation.

In UK work, digestion of both fresh and dried sainfoin (which contains CT) produced greater duodenal NAN flow and increased amino acid absorption from the small intestine relative to digestion of fresh white clover and dried lucerne (*Medicago sativa*) (Thompson *et al.* 1971; Beever & Siddons, 1986), with the authors attributing these effects to the CT in sainfoin.

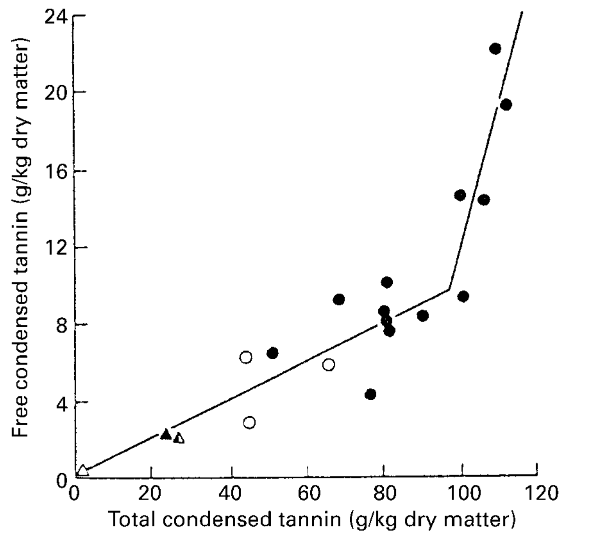

Fig. 3. Free condensed tannin concentration as a function of total condensed tannin concentration in macerates of fresh legumes. *Lotus pedunculatus* (cv. Grasslands Maku) grown under (○), high and (●), low soil fertility conditions; *Lotus corniculatus* cultivars (△), Winnar, (▲), El Boyero and (◭), Granger grown under low soil fertility conditions. From Barry & Manley (1986).

High concentrations of CT in *Lotus pedunculatus* (95 and 106 g/kg DM) depressed rumen digestion of readily fermentable carbohydrate (soluble sugar + pectin) and hemicellulose, but this was counteracted by increased post-ruminal digestion (Barry & Manley, 1984; Barry *et al.* 1986). Carbohydrate digestion in sheep fed on *Lotus corniculatus* (25–35 g CT/kg DM) was not affected by CT (Waghorn *et al.* 1987).

Effects of CT upon rumen fermentation of carbohydrate and protein can be explained by the concept of ‘free tannin’, defined as the CT not precipitated in high-speed centrifugation of plant mascerates (Barry & Manley, 1986; Fig. 3). Up to a total CT concentration of approximately 90 g/kg DM, 90 % of the CT was precipitated with plant constitutuents (i.e. protein) and 10 % was free in solution, whereas increments in total CT concentration above 90 g/kg DM were all released as ‘free tannin’. Thus, for *Lotus* species almost all the CT reacted with proteins in the host plant until the binding capacity of this system had been saturated (at about 90 g CT/kg DM). It is proposed that insoluble CT functions through reducing plant protein degradation in the rumen, whilst free CT can react with and inactivate microbial enzymes, explaining why high levels of free CT reduce rumen carbohydrate digestion (Fig. 4). This concept also

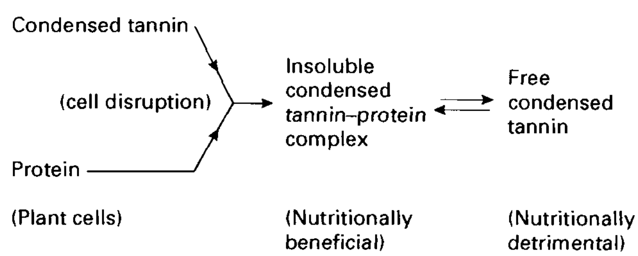

Fig. 4. Proposed mechanism of condensed tannin reaction with plant proteins and free tannin formation during cell disruption, and the suggested roles of insoluble and free condensed tannin in ruminant nutrition. From Barry & Manley (1986).

explains why mixing CT-containing and non CT-containing temperate forages seldom produces a beneficial outcome (Beever & Siddons, 1986), as the CT will preferably react with proteins in the forage of the CT-containing plant. It also explains why there is limited transfer of CT-induced protein-precipitating activity through rumen fluid from CT-containing to non CT-containing plants (BR Min, WC McNabb, TN Barry and JS Peters, unpublished results). Beneficial effects of forage mixing can only be expected if the CT content is extremely high and the protein content relatively low in the CT-containing plant, thus releasing some 'free' CT to bind with proteins in the non CT-containing plant. These conditions occur with some tropical legume forages and legume shrubs, especially if grown under low soil-fertility conditions, and some advantage may occur if they are added as a supplement to a diet of non CT-containing tropical grasses.

Digestion of condensed tannins

There are analytical problems in measuring the CT content of animal digesta, even with methods that measure bound CT, as CT added to digesta cannot be quantitatively recovered (Terrill *et al*. 1994). However, [^{14}C]CT-labelling studies showed that there was no absorption of the label in the small and large intestine of sheep fed on the temperate forage *Lotus pedunculatus* (Terrill *et al*. 1994), but that 45 % of the CT eaten was lost (presumed absorbed) from the rumen and 40 % of that flowing at the abomasum was lost from the small and large intestine (presumed absorbed) in sheep and goats fed on the tropical legume *Desmodium intortum* in Australia (Perez-Maldonado & Norton, 1996). This difference may be explained by differences in relative molecular mass, structure and reactivity between the two types of CT.

Metabolism of absorbed nutrients

^{35}S-labelling studies with sheep fed on both *Lotus pedunculatus* and *Lotus corniculatus* have shown that action of CT had no effect on the irreversible loss rate of methionine from jugular-blood plasma, but substantially increased the irreversible loss rate of cystine and reduced that of inorganic sulfate (McNabb *et al*. 1993; Wang *et al*. 1994). Interconversions of these three metabolites are shown in Fig. 5. In sheep fed on both forages, action of CT approximately doubled the amount of cystine used for body synthetic reactions. The mechanism for this comprised an increase in cystine entry rate to the plasma cystine pool, together with increased trans-sulfuration of methionine to cystine

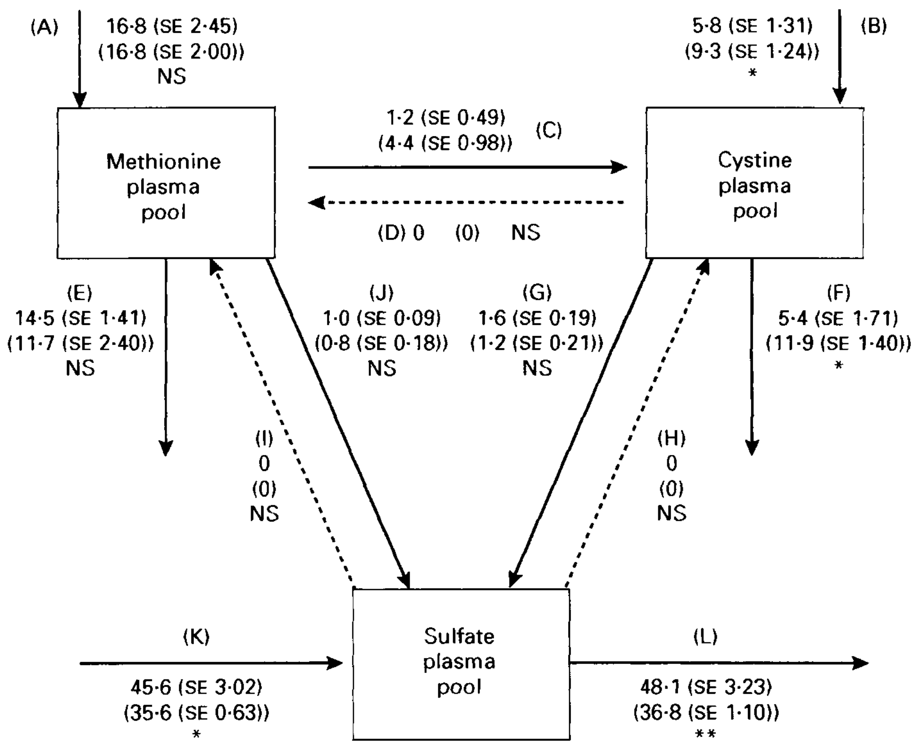

Fig. 5. A general three-pool, compartmentalized model for sulfur amino acid transactions in the post-hepatic plasma of sheep fed on fresh *Lotus corniculatus*. Mean values with their standard errors are shown for six PEG-infused sheep, with the corresponding values for six condensed tannin-acting sheep given in parentheses; $^{*}P < 0{\cdot}05$, $^{**}P < 0{\cdot}01$. → and ⇢, Rates of flow (μmol/min). The pathways are: (A) methionine entering the plasma pool from whole-body protein turnover and absorption from the small intestine; (B) cystine entering the plasma pool from whole-body protein turnover and absorption from the small intestine; (C) transulfuration of methionine to cystine; (D) conversion of cystine to methionine, which does not occur in mammalian tissue; (E) methionine leaving the plasma and being utilized for productivity processes and maintenance; (F) cystine leaving the plasma and being utilized for productivity processes and maintenance; (G) cystine oxidized to sulfate (and carbon dioxide); (H) plasma sulfate reassimilated as cystine. This cannot occur directly in mammalian tissue, but sulfate re-entering the rumen via saliva may be absorbed as cystine from microbial protein; (I) plasma sulfate reassimilated as methionine. This cannot occur directly in mammalian tissue, but sulfate re-entering the rumen via saliva may be absorbed as methionine from microbial protein; (J) methionine oxidized to sulfate (and carbon dioxide); (K) sulfate entering the plasma, chiefly from oxidation of sulfide absorbed from the rumen, but also sulfate and oxidation of sulfide absorbed from the intestine; (L) sulfate leaving the plasma chiefly in urine, but also recycled directly to the intestine and rumen via saliva. From Wang *et al*. (1994).

Table 3. Voluntary feed intake, live-weight gain, carcass gain and wool growth in lambs grazing the forage legumes *Lotus corniculatus* (34 g CT/kg DM) and lucerne (*Medicago sativa*) (0·3 g CT/kg DM)*

	Lotus corniculatus		Lucerne		
	CT acting	PEG supplemented	CT acting	PEG supplemented	SE
Rumen ammonia (mg N/l)	255	370	555	535	
Voluntary feed intake (kg organic matter/d)	1·19	1·20	1·32	1·34	0·06
Live-weight gain (g/d)	203	188	185	178	6
Carcass gain (g/d)	79	75	68	63	3
Wool growth (g/d)	12·1	10·9	10·8	10·2	0·4

CT, condensed tannins.
* From Wang *et al.* (1996*b*).

and reduced oxidation of both methionine and cystine. This indicates a high demand for cystine, which can be explained by these New Zealand sheep being selected for high rates of wool growth and wool protein containing a very high content of cystine (130 g/kg). This shows that action of forage CT can be used to increase the supply of the amino acid which is most limiting for wool growth.

Effects of condensed tannins upon forage feeding value

The effects of CT upon forage feeding value can be regarded as the sum of its effects upon VFI, upon the digestive process and upon the metabolism of absorbed nutrients. In growing lambs (initial live weight 22·4 kg) grazing *Lotus corniculatus* for 4 months during summer and autumn, action of CT (i.e. unsupplemented sheep *v*. PEG-supplemented sheep) increased wool growth by 12 % without affecting rate of body growth or VFI (Wang *et al*. 1996*b*; Table 3), indicating that EAA supply was limiting wool growth but not body growth in these lambs. This infers that body growth was restricted by energy (i.e. metabolizable energy) intake in these forage-fed animals. The diet selected contained respectively 31 and 34 g N and CT/kg DM and the organic matter digestibility was 0·73. There was no response to PEG supplementation in comparable sheep grazing lucerne, containing only traces of CT (0·3 g/kg DM), confirming that PEG supplementation only produces animal responses when the diet fed contains substantial concentrations of CT.

Action of CT in lactating ewes rearing twin lambs that were grazing *Lotus corniculatus* had no effect upon milk secretion in early lactation (Fig. 6), but increased the secretion rates of whole milk (by 21 %), lactose (by 12 %) and protein (by 14 %) during mid- and late-lactation (Wang *et al*. 1996*a*). The diet selected was also of high nutritive value, containing 36 and 44 g N and CT/kg DM, with an organic matter digestibility of 0·73. Milk protein concentration was unaffected by CT, but action of CT substantially reduced milk fat content (approximately 10 g/kg). These results have implications for the dairy industry, as a nutritional treatment which simultaneously increases the efficiency of milk protein production whilst reducing fat content would be very desirable for human nutrition. Experiments of this type with dairy cows are therefore needed.

A review of many years data implicated a role for protein nutrition in the ovulation rate of ewes (Smith, 1991), and this was illustrated by an increase in ewes showing multiple ovulations in response to abomasal infusions of lactalbumin

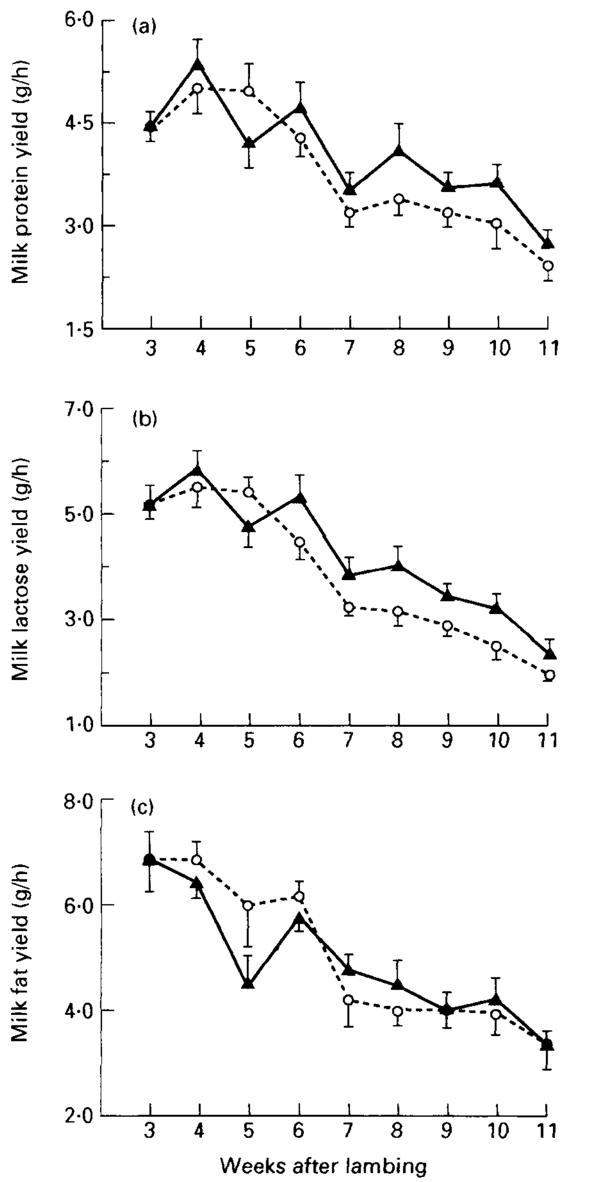

Fig. 6. Yields (g/h) of (a) protein, (b) lactose and (c) fat in the milk of twin-bearing lactating ewes grazing *Lotus corniculatus* (Birdsfoot trefoil; cv. Grasslands Goldie). (-▲-), Condensed tannin acting ewes; (-○-), ewes given twice-daily oral administration of PEG (relative molecular mass 3500). From Wang *et al.* (1996*a*).

and soyabean protein isolate (73 *v*. 55 %; Cruickshank *et al*. 1988). Subsequent work correlated this response to an increase in plasma concentration of branched-chain amino acids (valine + leucine + isoleucine; Waghorn *et al*. 1990) and intravenous infusion of branched-chain amino acids over the last 5 d of the oestrus cycle before luteolysis increased mean ovulation rate from 1·5 to 2·4 (Downing *et al*. 1995). Grazing trials were then carried out for 6 weeks, with ewes grazing perennial ryegrass/white clover pasture or *Lotus corniculatus*, containing 1 and 23 g CT/kg DM respectively, to study if this effect on ovulation rate could be induced by CT. Mean ovulation rates of ewes fed on *Lotus corniculatus*, with and without PEG supplementation (1·56 and 1·78 respectively) were greater than those of ewes grazing pasture (1·36 and 1·33 respectively), showing a clear response to CT (Min *et al*. 1999). Subsequent birth rank at lambing was much greater for the CT-acting *Lotus corniculatus* group (1·70) than for the other three groups (1·36–1·42) and it was shown that action of CT increased lambing percentage through increasing fecundity (number of corpa lutea : ewe ovulating) with no effect on fertility (ewes ovulating : ewes mated). Feeding on *Lotus corniculatus* increased plasma concentration of branched-chain amino acids more than that of any other EAA (57 %), with most of the increase in plasma amino acid concentration being directly attributed to the effects of CT. Recent reviews have also implicated plasma NH_3 as a possible cause of reduced ovulation and embryo survival when dietary rumen-degradable protein concentrations are high (Kaur & Arora, 1995), and CT is well known to reduce fermentation of rumen-degradable protein to NH_3. Further research is needed into the mechanism of how action of CT in *Lotus corniculatus* improves reproduction performance in grazing ewes.

In contrast to the increased productivity obtained from CT in *Lotus corniculatus*, action of CT in *Lotus pedunculatus* containing 76–90 g CT/kg DM markedly depressed rates of both body growth and wool growth (Barry, 1985), further illustrating the ecological role of high CT concentrations as a chemical defence.

The role of forage condensed tannins in the sustainable nutrition of grazing animals

Ruminants grazing forage diets are subject to a number of diseases, some of which have a nutritional component. Two such conditions are rumen frothy bloat in cattle and internal parasite infections in grazing sheep, cattle, deer and goats. Both are currently controlled by regular oral administration of chemicals, detergents in the case of bloat to disperse the foam and anthelmintic drenches in the case of internal parasites to kill the parasites. These remedies control both conditions in the short term but have long-term problems. First, they treat the symptoms and not the cause. Second, they cause consumer concerns about sustained use of chemicals and possible product residues, leading to longer withholding periods. Finally, in the case of anthelmintics, sustained regular use over many years has led to the development of parasites that are resistant to the drugs used. These effects have led to a reconsideration of control measures, in particular the development of new control measures that are more nutritionally based and ecologically sustainable.

Bloat is caused by very high solubility of forage proteins leading to the development of a stable foam in the rumen, and is very prevalent in cattle fed on legumes, especially in spring (Mangan, 1959). Because of their protein-precipitating properties, grazing CT-containing legumes has long been known to eliminate bloat (Jones *et al*. 1973). However, the minimum plant CT concentration needed to make forage bloat-safe was not known; this has recently been proposed to be 5 g CT/kg DM or greater (Li *et al*. 1996). Most common legumes and grasses used in temperate agriculture have CT concentrations well below this value (Table 1), and both conventional plant breeding and genetic engineering techniques are now being examined to try and increase these levels (Aerts *et al*. 1999*a*).

Parasitism of the abomasum and small intestine causes extensive protein losses in sheep (Kimambo *et al*. 1988), and re-directs protein synthesis away from skeletal muscles and into repair of gut tissues, leading to reduced N retention (MacRae, 1993). Increasing dietary protein intake and abomasal infusion of protein results in the animal being much better able to tolerate these infections and improves N retention (Brown *et al*. 1991; Coop & Holmes, 1996), with the main effect of increased protein supply being to increase the rate of acquisition of immunity. Subsequent studies have shown that lambs grazing CT-containing forages (sulla and *Lotus pedunculatus*) are better able to tolerate parasite infections than lambs grazing non CT-containing forage (lucerne), and show both increased growth and lower gut-worm burdens (Niezen *et al*. 1995). Two possible mechanisms could be involved. First, improved EAA supply from the action of the CT may counteract the protein loss caused by gut parasitism and may stimulate the immune system, and second the CT may directly react with and inactivate parasite larvae during passage through the gut. Forages containing CT may offer a nutritionally-based system for controlling the effects of parasites which is ecologically sustainable, thus allowing use of anthelmintic drugs to be reduced.

Forage condensed tannins and different ruminant species

The work reviewed has shown detrimental nutritional effects to ruminants with diets containing high CT concentrations (70–100 g/kg DM), as also found for single-stomached species fed on diets containing CT (Mangan, 1988), and consistent with CT being a plant defence against herbivory. However, in contrast, medium concentrations of CT such as found in *Lotus corniculatus* (30 g/kg DM) have substantially improved the efficiency of protein digestion and have improved sustainable productivity of grazing ruminants. It is important to realize, however, that nutritional manipulations such as this can only be made with forage CT if the ruminant species concerned does not produce salivary CT-binding proteins. As domestic cattle do not produce these proteins, the conclusions reached for domestic sheep should apply equally well to cattle. Deer do produce salivary CT-binding proteins and the implications of this can be seen in Table 4. CT can be 90 % recovered from plant material chewed and swallowed by sheep (i.e. oesophageal extrusa) but only 25 % recovered in deer,

Table 4. Extractability of forage condensed tannins (CT) from oesophageal (OF) extrusa samples of sheep and red deer fed on *Lotus corniculatus* and sulla (*Hedysarum coronarium*), compared with the original plant material. CT content was determined by the butanol–HCl method*

	CT concentration (g/kg DM)			
	Original plant material	OF extrusa	Plant fed	Reference
Sheep				
Extractable CT	31·3	10·0	Sulla	Terrill *et al.* (1992*a*)
Protein-bound CT	13·1	25·5		
Fibre-bound CT	1·7	5·0		
Total CT	46·1	40·3 (87 %)†		
Extractable CT	17·0	2·8	*Lotus corniculatus*	Min *et al.* (1998)
Protein-bound CT	12·3	22·8		
Fibre-bound CT	0·5	1·2		
Total CT	29·8	26·8 (90 %)†		
Red deer				
Extractable CT	36·1	ND	*Lotus corniculatus*	Min *et al.* (1997)
Protein-bound CT	10·9	10·6		
Fibre-bound CT	1·2	2·5		
Total CT	48·2	13·1 (27 %)†		
Extractable CT	10·0	0·2	*Lotus corniculatus*	Adu *et al.* (1998)
Protein-bound CT	10·3	3·8		
Fibre-bound CT	0·3	1·3		
Total CT	21·2	5·2 (25 %)†		

ND, not detected.
* Methodology from Terrill *et al.* (1992*b*); Jackson *et al.* (1996).
† Percentage of CT in corresponding manually-harvested sample.

showing tighter binding of CT in deer oesophageal extrusa. The significance of this on degradation of protein in the rumen and its release in the small intestine needs to be established with farmed deer; it may be that there is much less opportunity to manipulate N digestion with CT in farmed deer than there is with domesticated sheep and cattle.

Conclusions

When used under defined conditions, forage CT can be used to improve the efficiency of N digestion in ruminants fed on fresh forage diets. Improvements found to date include increases in wool growth, milk protein secretion, ovulation rate and the development of more nutritionally-based and ecologically-sustainable systems for disease control in grazing animals.

New methodology has shown the presence of trace amounts of CT in most of the common grasses and legumes grazed in temperate agriculture (1–2 g/kg DM). However, results to date show strongly that this is too low to reduce protein solubility and degradation in the rumen (BR Min, WC McNabb, TN Barry and JS Peters, unpublished results) and a minimum concentration of 5 g/kg DM or greater is suggested. Evaluation of traditional plant selection techniques and genetic engineering techniques for increasing CT concentration in common legumes such as white clover, red clover (*Trifolium pratense*) and lucerne offers exciting future possibilities.

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