FORAGE QUALITY OF SHRUB LEGUMES EVALUATED IN ACID SOILS[1]

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ABSTRACT

The forage quality of shrub legume species planted in acid soils of the llanos and forest margins of Colombia was evaluated by the Tropical Forages Program (TFP) of CIAT. Results showed differences in quality among and within species. Species free of tannins such as *Cratylia argentea* and *Desmodium velutinum* had medium in vitro dry matter digestibility (IVDMD) and high crude protein (CP). In contrast, species with tannins such as *Flemingia macrophylla*, *Tadehagi* spp., *Dendrolobium* spp. and *Codariocalyx gyroides* had adequate CP but low IVDMD. In these legumes, IVDMD of different accessions within species was negatively correlated with level of extractable condensed tannins (ECT). However, only 20 to 30% and 40 to 70% of the variation in IVDMD could be explained by level of ECT in the wet and dry seasons, respectively. The capacity of ECT to complex protein was variable among shrub legumes, and was related to degree of polymerization of CT. Extractable CT and protein-bound CT were negatively correlated (r=-0.5) with IVDMD, but no relationship was found between fiber-bound CT and IVDMD. Of the species evaluated, *C. argentea* and *D. velutinum* have high potential as fodder for livestock. Other species evaluated appear to have low feeding value due to the presence of CT and not well defined phenols and cell wall constituents which could also affect digestibility. Future studies should investigate factors responsible for low digestibility of some nitrogen fixing shrub legumes and determine the effects of level, structure and form of condensed tannins on ruminant nutrition.

FORQUALI
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INTRODUCTION

Nitrogen fixing tree and shrub legumes provide fodder for livestock (Blair, 1990) and also contribute to soil enhancement through nutrient cycling (Catchpoole and Blair, 1990). Common woody legume species used in the tropics, such as *Leucaena leucocephala,* and *Glyricidia sepium* are not well adapted to acid-infertile soils with high aluminum saturation prevalent in large areas of tropical America (Perdomo, 1991; Shelton et al., 1991). Thus, the TFP (Tropical Forages Program) of CIAT (Centro Internacional de Agricultura Tropical) gave high priority to the selection of nitrogen fixing shrub legumes for acid soils.

A collection of shrub legume species maintained in the Genetic Resources Unit of CIAT was evaluated in acid soils for dry matter yield, reaction to pests and diseases, and forage quality. In this paper we summarize the results on forage quality and their implication on future evaluation of shrub legumes with tannins. Finally, we discuss the fodder value of shrub legumes evaluated.

SHRUB LEGUME SPECIES EVALUATED AND THEIR QUALITY

As part of on-going activities in germplasm development for acid soils, shrub legumes species were planted at the Carimagua research station (Oxisol pH 4.0-4.5 and >85% Al saturation), Llanos of Colombia during 1989 and 1990. Species planted were: *Desmodium velutinum* (105 accessions), *Flemingia macrophylla* (63 accessions), *Tadehagi* spp. (40 accessions), *Dendrolobium* spp. (25 accessions), *Cratylia argentea* (10 accessions), *Uraria* spp. (4 accessions). In addition, *Codariocalyx gyroides* (27 accessions) was planted in 1992 in a humid forest environment of the Amazon Piedmont (Ultisol pH 4.5-5; and >80% Al saturation) of Colombia

(Florencia, Caquetá).

One of our objectives was to make an early assessment of quality differences among and within shrub legume species planted in acid soils. Initial results showed that there were some differences in crude protein (CP) and large differences in vitro dry matter digestibility (IVDMD) among species. Some variability within species was also observed, particularly in IVDMD (Table 1). Accessions of *D. velutinum, C. argentea, Uraria* spp. and *C. gyroides* had high levels of CP, and intermediate values of IVDMD. In contrast, accessions of *Dendrolobium* spp., *F. macrophylla* and *Tadehagi* spp. showed extremely low IVDMD values.

(Insert Table 1)

As a follow-up to this initial characterization of shrub legumes, we wanted to determine relationships: (1) between level of extractable condensed tannins (ECT) and IVDMD, and (2) between level of ECT and capacity of plant tannins to complex protein.

RELATIONSHIP BETWEEN TANNINS AND DIGESTIBILITY

It is commonly accepted that condensed tannins are associated with depressed forage digestibility and low animal performance (Barry and Duncan, 1984; Pritchard et al., 1988; Reed et al., 1990). Our results also showed that ECT and IVDMD were negatively correlated (Table 2). However, the degree of association varied among legume species and between seasons of the year. Only 20 to 30% and 40 to 70% of the variation in IVDMD within legume species was related to level of ECT in the wet and dry seasons, respectively. Thus, other phenolics and non-starch polysaccharides (NSP) could also be associated with the low digestibility of some of the shrub legumes evaluated. Longland et al. (1994) analyzed the level of different non-starch polysaccharides (NSP) in tropical shrub legumes provided by ILCA and CIAT. Results showed that total NSP level was higher in shrub legumes with tannins of low digestibility (i.e. *Tadehagi* spp. and *F. macrophylla*) as compared to shrub legumes with tannins, but of higher digestibility (i.e. *Leucaena leucocephala*, *Sesbania sesban*). Among the NSP fractions measured, xylose was high in legumes of low digestibility and was also the least degradable monomer. Digestibility of the shrub legumes evaluated, measured as in vitro loss of NSP by Longland et al. (1994), was inversely proportional to total phenolics ($r=-0.86$ $P<0.01$) and this relationship was greater than that observed with ECT ($r=-0.64$ $P<0.05$). The digestibility of sorghum leaves from bird resistant lines was shown to be negatively correlated with butin, a phenolic compound (Mueller-Harvey et al., 1993).

(Insert Table 2)

We do not know the extent to which xylose, other non-starch polysaccharides and non-tannin phenolic compounds are contributing to the low digestibility of some of the shrub legumes that we evaluated. Thus, future work on quality of tropical shrub legumes should carefully examine their cell wall chemistry and the extent of digestion of different cell wall constituents.

RELATIONSHIPS BETWEEN TANNIN LEVEL AND STRUCTURE AND PROTEIN PRECIPITATION ACTIVITY

A well known property of tannins is their ability to form complexes with proteins in pH dependent reactions (Fahey and Jung, 1989). This biological property of CT results in reduced rumen ammonia levels and increased flow of protein to the duodenum of animals fed tropical legumes with tannins (Carulla, 1994; Powell and Lascano, unpublished results). Thus, in the characterization of shrub legumes it is important to examine the protein precipitation activity of plant tannin extracts.

Results shown in Table 3, indicate large differences among shrub legumes species in level of ECT and in the ability of tannins from plant extracts to complex protein (bovine serum albumine). However, protein complexed by tannins was not related to the level of ECT in the plant tissue of shrub legumes evaluated. For example, *F. macrophylla* and *Tadehagi* spp. had similar levels of ECT on a dry matter basis, but the amount of protein complexed by tannin extracts from *Tadehagi* spp. was 3 times higher. On the other hand, the amount of protein precipitated by tannin extracts of *F. macrophylla* and *Dendrolobium* spp. was similar, even though the level of ECT was higher in *F. macrophylla*.

(Insert Table 3)

Results from a study involving five legume species, showed that ECT in *Tadehagi* spp. were less polymerized (6.5 units of a monomer) than ECT in *Phyllodium* spp. (9.5 units of a monomer) (Cano et al., 1994). Correlation analysis performed in the data of Cano et al. (1994), indicated that protein complexed by ECT was negatively related (r=-0.8 <P.001) with degree of polymerization of ECT. In grapes, tannins with 3 to 8 units of a monomer (i.e. lower molecular weight) were more reactive with protein than tannins with more than 8 units of a monomer (i.e. higher molecular weight) (Oh and Hoff, 1979).

We suggest that in order to make a better assessment of the quality of tropical shrub legumes, there is a need to consider not only the level of condensed tannins, but also the structure of the tannins present in the forage. Future research should attempt to determine the significance of different structures of tannins on digestibility and rumen escape protein in ruminants. One alternative is to measure digestibility and escape protein in animals fed shrub legume species with tannins of contrasting molecular weight, using a tannin binding agent such as PEG (polyethylene glycol) to generate different levels of tannins in the forage offered.

FORMS OF CONDENSED TANNINS IN LEAF TISSUE OF SHRUB LEGUMES

A technique developed by Terrill et al. (1992) allows quantitative analysis of extractable and protein or fiber-bound condensed tannins (CT). In a wide range of temperate legumes, approximately 65% of the CT were extractable and 32% and 3% were bound to protein and fiber, respectively (Terrill et al., 1992). Results with some tropical legumes showed that on average 79% of the CT were extractable, 14% were bound to protein and 7% were bound to fiber (Cano et al., 1994). However, with tropical legumes, there were some extreme cases. In *C. gyroides* only 42% of the CT were extractable with the remaining being bound to protein (25%) and fiber (33%) (CIAT, unpublished results). In contrast, in *Tadehagi* spp., 97% of the CT were extractable and only 3% were bound (Cano et al., 1994).

The significance of extractable and bound CT in quality of tropical shrub legumes has not been clearly established. In the case of *C. gyroides*, our results showed that extractable and protein-bound CT had similar negative correlations ($r = -0.5$, $P<.005$) with IVDMD, and that there was no relationship ($r= 0.2$, NS) between fiber-bound CT and IVDMD. Longland et al. (1994) also found similar correlations between in vitro loss of non-starch polysaccharides and extractable ($r=-0.64$, $P<0.05$) and bound ($r=-0.73$, $P<0.02$) CT in tropical shrub legumes. In sheep fed *Desmodium ovalifolium*, duodenal flow of protein and fiber-bound CT was similar to the intake of these forms of tannins (Carulla, 1994). These results suggest that bound tannins are stable in the rumen and to the change in pH at the duodenum.

Therefore, it would seem the extractable CT is the fraction in the plant that reacts with protein and thus could have important implications in ruminant nutrition. Shrub legume species with a lower proportion of bound CT could result in more rumen escape plant protein than legume species with similar levels of total CT, but with a higher proportion of bound CT. This hypothesis needs to be tested in future studies.

FODDER VALUE OF SOME SHRUB LEGUMES EVALUATED

Eventhough nitrogen fixing legumes can have multiple roles in farming systems of the tropics, farmers will most likely view their value as fuelwood and as fodder for livestock. Based on our initial quality assessment of shrub legumes evaluated in acid soils, *D. velutinum* and *C. argentea* offered the greatest potential as a source of fodder. Because of the oustanding agronomic performance of *C. argentea* in different testing sites with acid soils, we initiated work to determine its feeding value with sheep and milking cows. In addition, we have conducted some feeding trials using mixtures of shrub legumes with and without tannins.

Fodder value of *Cratylia argentea*

Our initial objective was to evaluate *C. argentea* in a protein bank with milking cows. However, it soon became evident that cows with access to the protein bank refused to consume immature *C. argentea*, but consumed the legume when the forage was cut and wilted.

Therefore, we proceeded to assess the effect of wilting and sun-drying on intake rate of *C. argentea* by sheep in metabolism crates. Results indicated that intake rate of *C. argentea* was affected by post-harvest treatment when the legume was immature, but not when it was mature (Raaflaub and Lascano, submitted for publication). With immature *C. argentea* intake rate was two times higher when the forage was wilted and dried than when fed fresh. When sheep were given the choice of selecting fresh or dried *C. argentea* they also consumed more dried forage, regardless of plant maturity, age or previous experience of the animals. In subsequent laboratory analysis, we found the presence of hydroxycoumarins and terpens in immature leaves of *C. argentea*, but we do not know if these compounds are responsible for the low intake of immature *C. argentea*. Thus, future research should determine if an adverse plant chemical constituent is in fact responsible for the low acceptability of young regrowth of *C. argentea*.

Following the work with sheep, we used wilted *C. argentea* as a cut and carry protein supplement (1% of BW on a DM basis) for milking cows grazing a grass only pasture. Results, showed an 8 to 13% increase in milk yield due to *C. argentea* in the wet and dry season, respectively (Table 4). Milk composition was not affected by supplementation of *C. argentea*.

(Insert Table 4)

From these results we concluded that *C. argentea* has great potential as a dry season supplement in cut and carry systems of smallholders.

Fodder value of mixtures of shrub legumes

It is known that low levels of condensed tannins (1 to 2% of DM) are beneficial in ruminant diets, since they reduce protein degradation in the rumen and increase protein flow to the small intestine (Barry et al., 1986). By feeding mixtures of shrub legumes with and without tannins we could dilute tannins in the forage offered and thus obtain benefits in the nutrition of ruminants in the tropics.

We tested this concept by supplementing mixtures of *C. argentea* (i.e. no tannins and highly degradable protein) and *F. macrophylla* (i.e. high tannins and slowly degradable protein) to sheep on a low quality grass basal diet. Results indicated less rumen ammonia and more nitrogen flow to the small intestine when the legume mixture was fed as compared to feeding only *C. argentea* (Powell and Lascano, unpublished results). However, with increased proportion of *F. macrophylla* in the mixture there was a reduction in dry matter and fiber digestibility and an increase in fecal nitrogen (Fässler and Lascano, submitted for publication).

From our initial results, it is clear that mixtures of *C. argentea* and *F. macrophylla* can contribute to escape protein in ruminants fed low quality grass diets. However, this otherwise positive effect is offset by reduced digestibility. Future studies with mixtures of tropical shrub legumes should consider not only tannin level but also their digestibility. For example, it would be of interest to evaluate mixtures of shrub legumes with and without tannins and with contrasting digestibility.

CONCLUSIONS

Of the shrub legumes evaluated in acid-infertile soils, *C. argentea* and *D. velutinum* have the greatest potential as fodder for ruminant animals. Other legumes studied had low forage quality, associated with variable levels of condensed tannins which negatively affected digestibility. However, we suggest that the negative effect of condensed tannins on digestibility of some tropical shrub legumes has been overemphasized in the past. Future studies should examine the effect of different non-tannin phenols and non-starch cell wall polysaccharides on digestibility. In addition, there is a need to determine the significance of different structures and forms of condensed tannins in the plant tissue. This information should prove useful in the selection of shrub legumes for fodder and in formulating legume mixtures to supplement ruminant animals fed low quality grasses or agriculture by-products in the tropics.

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Table 1. Crude protein (CP) and in vitro dry matter digestibility (IVDMD) of leaves from different shrub legumes harvested in the wet season in Carimagua, Llanos and Florencia, Caquetá, Colombia (CIAT, unpublished results).

Legume species*	No. of accessions	CP (%)	IVDMD (%)
Llanos			
Desmodium velutinum	59	16.1 ± 1.9**	51.7 ± 3.6**
Flemingia macrophylla	41	16.5 ± 2.1	20.1 ± 4.7
Tadehagi spp.	40	12.5 ± 1.3	25.0 ± 5.3
Dendrolobium spp.	17	13.5 ± 1.9	16.7 ± 5.3
Cratylia argentea	10	18.6 ± 1.5	52.7 ± 1.8
Uraria spp.	4	16.9 ± 1.5	56.9 ± 3.6
Caquetá			
Codariocalyx gyroides	27	19.0 ± 0.6	43.6 ± 3.1

*Regrowth: 12 weeks.

**Standard deviations of the means.

Table 2. Correlations between level of extractable condensed tannins (ECT) and in vitro dry matter digestibility (IVDMD) of leaves from shrub legumes evaluated in acid soils (CIAT, unpublished results).

Legume species	No. of accessions	Dry season ECT vs IVDMD (r)	Wet season ECT vs IVDMD (r)
Codariocalyx gyroides	27	---	-0.52 (P<0.005)
Dendrolobium spp.	17	-0.81 (P<.001)*	---
Flemingia macrophylla	19	-0.74 (P<.001)	-0.52 (P<0.02)
Tadehagi spp.	34	-0.62 (P≤.001)	-0.43 (P<.02)

*Level of significance

Table 3. Level of extractable condensed tannins (ECT) and protein complexed by tannin extracts from leaves of shrub legumes evaluated in acid soils (CIAT, unpublished results).

Legume species	No. of accessions	ECT* (% of DM)	Protein complexed by tannins** (g/100 g DM)
Dendrolobium spp.	17	9.5 ± 4.8***	7.6 ± 3.8 (0.8)****
Flemingia macrophylla	38	15.4 ± 3.2	7.1 ± 1.7 (0.5)
Tadehagi spp.	34	15.7 ± 2.4	23.0 ± 5.0 (1.5)

*Butanol-HCL (Porter et al., 1986).

**Radial diffusion assay (Haggerman, 1987).

***Standard deviation of the means.

****Figures in parenthesis are protein complexed per unit of ECT.

Table 4. Milk yield and composition of cows grazing *Brachiaria dictyoneura* pastures and supplemented with chopped-wilted *Cratylia argentea* (CIAT, unpublished results).

Treatment	Season	Yield (kg/cow/day)	Fat (%)	Non-fat solids (%)
B. dictyoneura		5.4	4.2	8.4
B. dictyoneura + *C. argentea**	Dry	6.1	4.6	8.5
	Difference	(13%)		
B. dictyoneura		9.0	3.6	8.3
B. dictyoneura + *C. argentea***	Wet	9.7	3.7	8.5
	Difference	(8%)		

*Intake = 2.9 kg DM/cow/day

**Intake = 3 kg DM/cow/day