# Protein binding capacities of tannins from various *Leucaena* genotypes

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*Leucaena leucocephala* is a high quality, high protein forage. Several new cultivars are becoming available from this genus, all of which contain condensed tannin (CT). CT are defined as polyphenolics with the ability to precipitate protein and as such interfere with protein digestion. There is evidence that CT from other forage species differ in their ability to precipitate protein[1]. The aim of the present study was to assess the protein binding capacities of CT within the *Leucaena* genus as compared to those from other nutritionally significant forages.

Binding affinities (astringency) of CTs extracted from three *Leucaena* genotypes (*L. diversifolia* (OFI 53/88), *L. pallida* (CQ3439) and *L. leucocephala* cv. Tarramba) were compared to *Lotus pendunculatus* cv. Maku and *Acacia aneura* (Mulga). Fresh leaf was collected onto dry ice, lyophilised, ground and the CT extracted with 70% acetone and purified with a Sephadex LH-20. Individual CTs from the *L. diversifolia* extract were separated using a Fractogel TSK HW-40 size exclusion column and the four most obvious peaks, detected at 350 nm, were collected. Protein binding capacities of the nine samples was examined by adding sequential amounts of CT (0-0.8 mg) to 0.75 mg of bovine serum albumin. Reactions were performed in 0.2 M sodium acetate buffer with 0.17 M NaCl at pH 5. Amounts of protein bound were measured by hydrolysing the protein in the precipitates and assaying with ninhydrin. Binding parameters were determined by fitting the sigmoid equation $y = a/(1+\exp(-(x-b)/c))$ where "a" is the maximal amount of protein precipitated and "b" is the concentration of CT at 50% of maximal precipitation.

Among species, *L. pallida* was more astringent than *L. diversifolia* and both were substantially more astringent than *L. leucocephala*. Lotus and *L. pallida* were similarly astringent, while Mulga and *L. diversifolia* were less astringent than the former. Of the individual CTs separated from the *L. diversifolia* extract, the earlier eluting and hence the heavier CTs (peaks 1& 2) were more astringent than the next eluting CT (peak 3), which in turn was more than peak 4.

| Tannin source | b | se |
|---|---|---|
| *Leucaena pallida* | $0.138^{a}$ | 0.011 |
| *Lotus pendunculatus* | $0.141^{ab}$ | 0.011 |
| *L. diversifolia* (peak 1) | $0.151^{ab}$ | 0.010 |
| *L. diversifolia* (peak 2) | $0.170^{abc}$ | 0.010 |
| *Leucaena diversifolia* | $0.171^{bc}$ | 0.008 |
| *Acacia aneura* | $0.186^{cd}$ | 0.010 |
| *L. diversifolia* (peak 3) | $0.209^{d}$ | 0.011 |
| *L. diversifolia* (peak 4) | $0.293^{e}$ | 0.014 |
| *Leucaena leucocephala* | $0.295^{e}$ | 0.015 |

Values lacking common superscript letter differ ($P<0.05$)

These data show that the relative astringencies of CT from *Leucaena* can vary by at least two-fold. Such variability may be associated with differences in molecular weights of the CTs and could have implications for the nutritive value of proteins in various types of *Leucaena*.

1. Perez-Maldonado RA; Norton BW; Kerven GL Factors affecting in vitro formation of tannin protein complexes. J Sci Food Agric 1995;69:291-298