The potential for using the alkanes and long-chain alcohols of plant cuticular wax to distinguish the contribution of different plant species to a mixed root mass

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Introduction In mixed pastures, plants compete below ground for soil water and nutrients, just as they compete above ground for light. Quantifying below-ground competition is difficult, partly because of the difficulty of measuring the contribution of different plant species to a mixed root mass. For some years, the hydrocarbons (alkanes) of plant cuticular wax have been used to quantify the species composition of the diet of herbivores (see Mayes & Dove, 2000). More recently, the long-chain aliphatic alcohols (LCOH) of plant wax have also proved useful markers (Bugalho *et al*., 2004). Plant roots also contain cuticular alkanes and these may be used to discriminate between roots coming from different species (Dawson *et al*., 2000). We report an extension of this concept, using a combination of cuticular alkanes and LCOH to discriminate between root tissues from plant species commonly found in or sown as pastures in southeastern Australia.

Materials and methods Nine plant species (Figure 1) were grown in pots in a glasshouse, harvested while still vegetative and separated into root and shoot material. Cuticular alkanes and LCOH were extracted from freeze-dried, ground root material (12 samples/species) and assayed by gas chromatography (Bugalho *et al.,* 2004). Differences in alkane or LCOH profiles between plant species were examined using canonical variates analysis (CVA). Canonical variates scores from alkanes and LCOH were compared by orthogonal Procrustes rotation to determine if the discriminatory information provided by LCOH was additional to that provided by alkanes.

Results Alkanes with carbon-chain lengths C21-C35 were detected and, in confirmation of earlier work (Dawson *et al*., 2000), their concentrations were <20 mg/kg OM, an order of magnitude lower than those typical of shoot tissue (Bugalho *et al*., 2004). Root LCOHs with chain lengths C20-C32 were detected; concentrations (5-400 mg/kg OM) were also lower than those reported in shoots (Bugalho *et al*., 2004), but were in general much higher than root alkane concentrations. The CVA showed that the concentration profiles of both alkanes and LCOH in root tissue from the nine pasture species differed, and that they significantly discriminated between roots from the different species (P<0.001). Orthogonal Procrustes rotation indicated that the discrimination based on LCOH was additional to that based on alkanes. CVA based on the combined use of alkanes and LCOH achieved complete discrimination between roots from the 9 species (Figure 1: P<0.001).

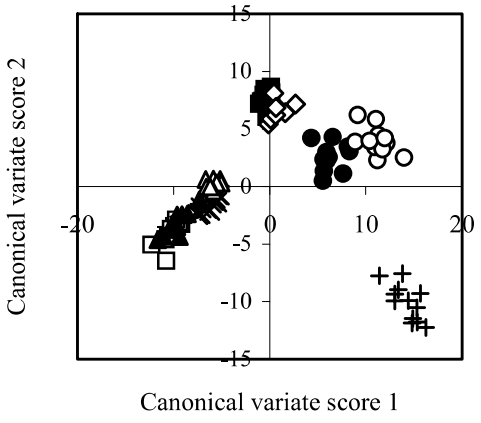

Figure 1 Canonical variates analysis of combined data for alkanes and LCOH in root tissue. The species listed were significantly discriminated by their canonical scores 1, 2 and (in parenthesis) score 3 (all P<0.001). X *Lolium perenne* (1.76); □ *Vulpia bromoides* (6.69); Δ *L. rigidum* (-3.99); ▲ *Dactylis glomerata* (-2.33); ■ *Austrodanthonia richardsonii* (-1.72); + *Trifolium repens* (-2.97); ● *Medicago sativa* (-2.73); ◊ *Phalaris aquatica* (-4.74); ○ *T.subterraneum* (8.46)

Conclusions Between-species differences in alkanes and LCOH concentrations in root tissue from pasture plants, separately and especially in combination, are sufficient to provide a chemical approach for assessing the contribution of different plant species to a mixed root mass. In further work, we are validating this technique using known mixtures of roots from different species.

References

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