SELECTING ACID-SOIL TOLERANT WHITE CLOVER

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ABSTRACT
A two-stage, tandem, recurrent-selection program is proposed to improve acid-soil tolerance of white clover, *Trifolium repens* L. In stage one, we evaluate root growth of very young seedlings, grown in an highly-acid Al toxic soil. Although the relationship between the acid-soil response of very young seedlings and that of mature white clover is unknown, the failure of white clover to become established on a highly-acid, Al-toxic soil suggests that establishment is a critical stage of plant development on such soils. In stage two we reevaluate genotypes selected in stage one, in a small-pot study at two lime levels, to separate the effects of plant vigor and acid-soil tolerance, account for any effects of growth stage on evaluation results and ensure that all genotypes selected are vigorous in soils with a higher pH.

KEYWORDS
Al tolerance, recurrent selection, tandem selection

INTRODUCTION
Efforts to improve acid-soil tolerance of small-seeded legumes through recurrent selection have been minimal. In alfalfa, *Medicago sativa* L., important but limited improvement was achieved (Devine et al., 1976; Brooks et al., 1982; Campbell et al., 1988). However, Campbell et al. (1988) suggested that selection may have been more effective for vigor than for Al tolerance.

Work with white clover has been limited to characterization of germplasm and selection of Al tolerant or sensitive genotypes. Variation among cultivars has been small at best (e.g., Dodd et al., 1992). Aluminum tolerant and susceptible genotypes were selected from ‘Grasslands Huia’ (Caradus et al., 1991). They were similar morphologically, but differed in response to P (Crush and Caradas, 1992). Based on a six clone diallel, Caradus et al. (1991) suggested that Al tolerance in white clover was a recessive character and that selection within adapted populations was a more effective improvement strategy then attempting to identify new sources of Al tolerance.

We propose a two-stage, tandem, recurrent-selection program to develop acid-soil tolerant white clover populations.

Stage 1. We evaluate young clover seedlings grown in acid soil. This is important because: 1) Establishment is a critical stage in plant persistence; 2) Seedling plants can be evaluated quickly and easily allowing evaluation of large numbers of individuals; and 3) The amount of soil required is small.

Caradas et al. (1987) found that white clover germination was relatively insensitive to Al concentration. They reported also that shoot growth of established seedlings was more adversely impacted by Al than was initial establishment. However, they did observe establishment differences among accessions (Mackay et al., 1990).

Our field observations indicated that few clover plants became established on a highly-acid soil (pH 4.2), even with reduced competition, unless they were transplanted after initial growth in a greenhouse. Once transplanted they persisted, but spread only slowly, for periods of a few months to two years. On an adjacent limed site (pH 5.2), seedling clover plants occurred frequently.

Large numbers of plants must be evaluated if the proportion of individuals selected is low and the number selected high. The number selected must be high when a second stage of selection is planned to avoid inbreeding depression. For cross-pollinated clovers, at least 100 plants should be selected. If 15 percent of those evaluated are selected in both stages, 675 plants need to be evaluated in stage two and 4,500 in stage one.

Selection at the young seedling stage has limitations. Each genotype can only be grown at one Al level, thus, selection could be more for vigor than acid-soil tolerance. Also, the relationship between seedling and mature plant tolerance of white clover is unknown. Finally, young seedling tests are easily affected by soil bulk density and water content (Belesky et al., 1991).

We developed a soil-on-agar technique (Morris and Voigt, 1996; Voigt et al., 1996) for the first stage of selection. The procedure is simple and uses only small amounts of soil, 6 kg for 5,000 seedlings. Use of pregerminated seed, selected for uniform radical length, minimizes seedling vigor differences. Also, the experimental unit is small (3.5 by 8.5 cm surface), thus ensuring relative soil uniformity within each unit. Selections are made from each flask (similar to grid selection in field studies) to minimize effects of soil differences between units.

Stage 2. Seedlings from stage 1 are saved, propagated from stolon tips, and reevaluated in stage 2. Two replications of each genotype are grown in acid and in limed soil for about 30 d. Selection is based on adventitious root growth (weight and estimated length) and top growth (weight and number of leaves). Campbell et al. (1988) proposed a similar second-stage of selection in alfalfa to obtain better estimates of Al tolerance and to separate the effects of vigor and Al tolerance.

Large amounts of soil are required to conduct this stage. To minimize soil and labor costs we use a selection percentage of about 6% in stage one and 33% in stage two. This increases the number of genotypes evaluated at stage one to 5940 and reduces the number evaluated at stage two to 330 and the amount of soil required to about 630 kg. Our approach should allow us to make selection progress for acid-soil tolerance while avoiding inadvertent selection for inferior performance in limed soil.

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