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**Soil nitrate promotes growth of an exotic grass more than native forbs.**

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**Introduction.** Recent research has shown that soil nitrate concentrations regulate the abundance of problematic exotic annuals in threatened temperate grassy white box (*Eucalyptus albens* Benth.) woodlands in NSW (Prober et al. 2002, 2005). Soil nitrate concentrations in autumn were high (5-30 mg/kg) in remnants dominated by exotic annuals, and very low (< 3 mg/kg) in high-quality remnants with sparse exotics (Prober et al. 2002). Addition of carbohydrate (sugar) to deplete soil nitrate temporarily reduces exotic annual biomass, which in turn can promote establishment of native plants (Prober et al. 2005). Ultimately, successful restoration of degraded woodland communities may depend on long-term reduction in soil nitrate concentrations, otherwise exotic grasses are likely to re-establish and suppress native forbs (Prober & Thiele 2005). However, little research has been conducted on the responses of native forbs to altered nutrient concentrations.

Forb diversity is typically high in woodland remnants with low soil nutrients (Prober et al. 2002), and Smallbone et al. (in press) showed that reducing soil nitrate concentrations can promote establishment of forbs from sown seeds in degraded remnants. Conversely, high nutrient concentrations promote the growth of many native forbs under horticultural conditions (Wrigley 1988; Barker et al. 2002). This may seem paradoxical, however, the field response is easily interpreted if high nutrient conditions promote exotic plants more than native forbs, leading to strong competitive effects.

In this study we compared the growth responses of four common perennial woodland forbs – Common Everlasting Daisy (*Chrysocephalum apiculatum* (Labill.) Steetz), Nodding Chocolate Lily (*Dichopogon fimbriatus* (R.Br.) J.F. Macbr.), Yam Daisy (*Microseris lanceolata* (Walp.) Schultz-Bip.) and Tufted Bluebell (*Wahlenbergia communis* Carolin), an exotic annual grass - Wild Oats (*Avena fatua* L.), and a native perennial grass - Kangaroo Grass (*Themeda australis* (R.Br.) Stapf.), when grown under a range of soil nitrate concentrations. These species include a range of life-forms, from geophytic lilies to tussock grasses. We hypothesised that high nitrate concentrations would promote growth of all species, but that the exotic annual grass would grow taller and produce more biomass under high nitrate levels than the native species. We discuss the implications of these results for restoration and management of degraded remnants.

**Methods.** In a heated glasshouse, we compared the growth of the six species grown in monoculture across five nitrate concentrations, each with six replicates arranged in a randomized complete block design. Ten centimetre diameter pots were filled with washed
river sand and planted with a single seedling of one species. At the time of planting, seedlings were 2–4 months old at similar stages of growth, although Wild Oat plants with above ground material (hereafter referred to as ‘shoots’) or roots > 8 cm long were pruned before planting to maintain uniformity in initial plant sizes. Five rates of NaNO₃ were added as treatments (0, 4.1, 10.9, 21.8 and 81.9 mg per pot in 100 ml tap water). This was intended to approximate a range of nitrate concentrations (0, 1.5, 4, 8 and 30 mg/kg) found by Prober et al. (2002) in degraded and intact white box remnants in the field. Nitrate levels were maintained by watering with a 100 ml solution every 4-7 days. Analysis of nitrate concentrations (using the exchangeable NO₃ extraction method, Mulvaney 1996) in four replicate, unplanted pots per treatment indicated mean nitrate concentrations for the five treatments as follows: 1.5±0.39, 3.6±0.24, 6.1±0.27, 10.8±0.44 and 42.3±0.89 mg/kg nitrate N. These slightly higher than predicted values may have resulted from the presence of small amounts of nitrate in tap water and/or soil moisture levels of greater than field capacity at the time of measurement. Phosphorus and potassium were also supplied in the 100 ml solution, as 15.8 mg of KH₂PO₄ approximating phosphorus concentrations found in degraded White Box woodlands (8 mg/kg, Prober et al. 2002). Trace elements were added at the beginning of the experiment using Micromax granular at 250 mg per pot.

Plant height was measured in week 1 and at 2 week intervals for 8.5 weeks, at which stage all roots and shoots were harvested, oven dried at 70°C for 24 hours and weighed. Nutrient effects on root and shoot growth of each species were analyzed using generalized least squares regression (Genstat 2003), including a species-nutrient interaction term. A quadratic component was included in the model where significant. Separate analyses were performed for final root and shoot dry weights, and for the proportional change in plant height, calculated as the final height divided by the initial height of each plant. Nitrate levels were log (ln (x + 1)) transformed for analysis. Plant diameter rather than height was used in the Tufted Bluebell analysis, reflecting the different growth form of this species.

Results. Three species showed a significant positive response in either shoot, root or height growth to increasing nitrate levels: Wild Oats, Common Everlasting Daisy and Yam Daisy (Fig. 1). Wild Oats responded most strongly, with the most rapid rate of increase in shoot biomass of all species as soil nitrate increased (Fig. 2). Root biomass and height also increased most rapidly in Wild Oats, although the rate of increase declined at the higher two nitrate levels, possibly because the plants were becoming limited by the size of pots (Figs. 1 & 2).

Of the native plants, Common Everlasting Daisy root biomass responded positively up to the second highest nitrate concentration (Fig. 1), and height growth of Common Everlasting Daisy and Yam Daisy was significantly enhanced by increasing nitrate, although to a much lesser extent than Wild Oats (Figs 1 & 2). A number of Nodding Chocolate Lily plants perished during the experiment, and no significant increase in biomass or height with nitrate concentration was detectable. Kangaroo Grass did not respond to increasing nitrate, whereas Tufted Bluebell roots surprisingly showed a significant negative response to nitrate (Figs 1 & 2).

Discussion. Results demonstrate that all five native species were less responsive to increasing nitrate concentrations than the exotic annual grass Wild Oats, although at least two native forbs (Common Everlasting Daisy and Yam Daisy) were more responsive than the native perennial grass, Kangaroo Grass. Other studies have found that exotic Mediterranean annuals respond more vigorously to elevated resource levels (including
phosphorus and soil moisture) than many native perennial species (Allcock 2002; Lenz and Facelli 2005). Indeed, the ability of many exotic annuals to survive and reproduce under a wide range of nutrient concentrations is a major reason for their success as invasive species (Booth et al. 2003).

In white box woodlands in central NSW, available nitrate concentrations in intact, diverse remnants are usually 0.1–2.5 mg/kg (Prober et al. 2002), comparable to the low concentrations in this glasshouse trial. In the present study, Wild Oat plants did not increase in biomass under the lowest nitrate treatment, and this may explain why Wild Oats is rarely problematic in intact remnants (Prober & Thiele 1995; Prober et al. 2002). In a similar fashion, Lenz and Facelli (2005) found a related species, Slender Oats (Avena barbata), was largely restricted to high resource patches in two native grasslands in South Australia, in particular to patches with high soil moisture levels.

The positive response of forbs such as Common Everlasting Daisy and Yam Daisy to nitrate addition contrasts with their response in the field. In lowland grassy woodlands, Yam Daisy is largely restricted to high quality remnants where available nitrate concentrations are extremely low (Prober & Thiele 1995). Further, in a field restoration experiment (Smallbone et al., in press), initial establishment of Yam Daisy from sown seed and survival of Common Everlasting Daisy transplants were strongly promoted when nitrate concentrations were reduced through carbon additions. In the field, forbs such as Common Everlasting Daisy and Yam Daisy appear to face a trade-off between an innate ability to respond positively to higher nitrate concentrations, and a reduced ability to compete against vigorous exotic weeds in high nutrient soils.

In conclusion, our results support suggestions that: (1) increasing soil nutrients in remnant woodlands are more likely to promote the growth of annual exotics such as Wild Oats than native herbaceous species; and (2) woodland restoration may be assisted by depleting soil available nitrate concentrations, and consequently, the biomass of annual exotics, because the disadvantage to vigorous, nitrophilic exotics is likely to be significantly greater than to native species. Further development of practical techniques for reducing available soil nutrients and re-introducing forbs into degraded woodlands would thus be worthwhile.

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References


**Figure 1.** Effect of log ($\ln (X + 1)$) soil nitrate concentration on shoot and root dry weights and proportional change in height of four native forbs, a native grass (Kangaroo Grass) and an exotic grass (Wild Oats). $P$ is shown for linear (Lin) and where relevant, quadratic (Quad) terms of the model. Log nitrate levels indicate the predicted $\text{NO}_3$ concentrations based on the amount of $\text{NaNO}_3$ added to each pot (these were slightly lower than measured levels, see text). Note different scales on y axis for Nodding Chocolate Lily and Tufted Bluebell. Proportional change in growth represents the final plant height (or rosette width for Tufted Bluebell) divided by initial plant height (or rosette width).

**Figure 2.** Parameter estimates and 95% confidence intervals for the response of shoot biomass, root biomass and proportional change in growth to log ($\ln (X + 1)$) soil nitrate concentration for Wild Oats (W), Common Everlasting Daisy (C), Nodding Chocolate Lily (N), Yam Daisy (Y), Kangaroo Grass (K) and Tufted Bluebell (T). Estimates for the quadratic term are shown where it was included in the model. Lin = Linear response, Quad = Quadratic response.
Figure 1

Dry weight (mg)

Shoots

Wild Oats
Lin < 0.001

Yam Daisy
Lin = ns

Kangaroo Grass
Lin = ns

Tufted Bluebell
Lin = ns

Roots

Wild Oats
Lin < 0.001

Common Everlasting Daisy
Lin = ns

Yam Daisy
Lin = ns

Kangaroo Grass
Lin = ns

Tufted Bluebell
Lin = ns

Proportional change in growth

Wild Oats
Lin < 0.001

Common Everlasting Daisy
Lin = 0.009

Yam Daisy
Lin = 0.020

Kangaroo Grass
Lin = ns

Tufted Bluebell
Lin = ns

Change in rosette width (cm)

Log Nitrate (mg/kg)

Shoots

Wild Oats
Lin < 0.001

Yam Daisy
Lin = ns

Kangaroo Grass
Lin = ns

Tufted Bluebell
Lin = ns

Roots

Wild Oats
Lin < 0.001

Common Everlasting Daisy
Lin = ns

Yam Daisy
Lin = ns

Kangaroo Grass
Lin = ns

Tufted Bluebell
Lin = ns

Proportional change in growth

Wild Oats
Lin < 0.001

Common Everlasting Daisy
Lin = 0.009

Yam Daisy
Lin = 0.020

Kangaroo Grass
Lin = ns

Tufted Bluebell
Lin = ns

Change in rosette width (cm)

Log Nitrate (mg/kg)