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Effectiveness of lupins as a cover crop for reducing damage by browsing mammals
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Public

Effectiveness of lupins as a cover crop for reducing damage by browsing mammals

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Project C4: Strategies to reduce vertebrate browsing damage
Deliverable 4.6: Effectiveness of lupins as cover crops for reducing browsing damage

Project B1: Site productivity
Deliverable 1.12: Screen lupins as a browsing deterrent

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Summary

Managing non-tree vegetation has been suggested as a potential option for minimizing browsing damage in eucalypt plantations. We tested this hypothesis by growing sweet and bitter types of lupins (*Lupinus angustifolius* cv. 'Wonga' and *L. albus* cv. 'Lupini', respectively) with eucalypt seedlings (*Eucalyptus nitens*) in (a) enclosures with Tasmanian pademelons (*Thylogale billardierii*) as the browser, and (b) the field close to a high population of native and introduced browsers (mainly pademelons and rabbits). A control treatment of eucalypt seedlings was included that simulated operational establishment practices for ex-pasture sites. Both types of lupins reduced the incidence and severity of browsing damage in the enclosure experiment, where the browser was introduced after both lupins had become established. In the field, the sweet lupin germinated, but its young plants were totally browsed. Bitter lupins established successfully in the field in 5 of the 6 replicates, and they significantly reduced browsing damage. In the other replicate, the bitter lupin was heavily browsed, there was prolific weed growth in the control treatment, and browsing of eucalypt seedlings was most severe in the bitter lupin treatment. Scat counts indicated that all treatments were visited similarly by the browsers. We concluded that (a) the sweet lupin was unsuitable because of heavy browsing soon after germination, (b) even the bitter lupin was palatable to some unidentified browsers under some conditions, (c) if non-tree vegetation is taller than the tree seedlings, there was a significant reduction in browsing damage, and (d) use of bitter lupins in most replicates led to reduced browsing damage and taller height growth of non-browsed seedlings. Use of lupins might have also improved tree form and reduced the number and size of branches, but this was not assessed. Further research is warranted to refine the methods of managing bitter lupins and other vegetation, and to quantify its longer-term benefits.

1. Introduction

Browsing in newly established plantations is an economically important problem in Australia (Coleman et al., 1997; Montague, 1996), through its impact on growth and tree form (Bulinski and McArthur, 1999; Wilkinson and Neilsen, 1995). Managing browsing frequently involves lethal control of browser populations using "1080" poison (sodium monofluoroacetate) (Tasmania only) or shooting. Non-lethal methods are being developed to reduce reliance on lethal methods. Silvicultural practices to alter the selective use of patches of vegetation by herbivores need to be investigated as a potentially useful component of non-lethal management of browsing.

Recent research at the CRC has shown that some plant species, such as grasses, increase the probability of browsing damage to plantation seedlings, when measured both at the level of patches within plantations (Pietrzykowski, 2000), and at the between-plantation scale (Bulinski, 1999). However, natural vegetation that hides seedlings, including bracken and shrubs, results in reduced probability of detection, and hence reduced damage (Gilbert, 1961; Pietrzykowski, 2000). In some plantations, particularly on ex-pasture sites, there is the potential to grow "cover" crops specifically to manipulate the feeding behaviour of herbivores and hence reduce damage. If chosen carefully, cover crops could also have the added purpose of suppressing weeds, such as grasses, and further, if they are nitrogen-fixing, adding some nutritional benefit to the site (Turvey and Smethurst, 1983). In farm forestry, the use of cover crops such as lupins might also provide the opportunity to harvest and sell the lupin seed, off-setting the costs of browsing control and other components of plantation establishment.

This project specifically tested, on an ex-pasture site and in animal enclosures, whether or not one type of crop, i.e. lupins, was beneficial in this role of reducing browsing damage. In previous experiments, lupins have been relatively easy to establish and, if an annual species was chosen, they did not represent a large competitive threat to plantation seedlings during a dry summer (Smethurst et al., 1986). If established several weeks before planting tree seedlings, annual lupins could reduce damage during the same early post-planting phase that is currently protected through lethal methods. Lupins can be either "sweet" or "bitter", the latter with a relatively high alkaloid content. Potentially, sweet lupins could function by both hiding tree seedlings and providing an alternative food source. Bitter lupins, if unpalatable, could function by hiding tree seedlings and reducing the overall quality of the plantation to browsers, hence reducing use of plantations for foraging. We investigated one sweet and one bitter lupin, i.e. *Lupinus angustifolius* cv. "Wonga" and *L. albus* "Lupini", respectively, and, in the field only, we included a supplementary treatment of a lupin of intermediate bitterness, i.e. *L. angustifolius* cv. "New Zealand bitter blue".

In this project, our aims were to:

(1) Determine whether these lupins could be established successfully on an ex-pasture forestry plantation,

(2) Measure how effectively they reduced browsing damage to *Eucalyptus nitens* seedlings compared with current silvicultural practices, and

(3) If successful as a technique, provide preliminary silvicultural prescriptions for using lupins to reduce browsing damage.

The research had two components:

(1) An enclosure experiment, using captive Tasmanian pademelons (*Thylogale billardierii*), to determine the potential of established lupins to reduce browsing by individual herbivores, and

(2) A field experiment to determine whether lupins would (a) establish on an ex-pasture site, and (b) reduce browsing under realistic field conditions.

2. Materials and methods

2.1 Enclosure experiment

2.1.1 Experimental area, animals and basal diet

An existing fenced area of 28 m × 15 m was used in the Animal Enclosure at the School of Zoology, University of Tasmania, Hobart. The area consisted of 10 individual pens (each 5.6 m x 7 m), five on either side of a soil path that provided access to all pens. Each pen had a sheltered area with bedding straw and a water container. Nine pens were used in the trial, and the tenth was used as a holding pen.

Six pademelons, three female and three male (average body mass 7.0 kg s.e. 1.3 and 8.1 kg s.e. 2.2, respectively) were tested consecutively. Each animal was kept in a holding pen before the experiment. Each was fed a daily basal diet of unlimited pasture replacement pellets (suppliers: Pivot Nutrition, Launceston; composition: 2.3% nitrogen, 24.0% neutral detergent fibre, 13.2% acid detergent fibre, 5.5% lignin, 0.5% ash) and lucerne chaff, 1 carrot, 1 apple, 4 leaves of silver beet and celery.

2.1.2 Treatments and experimental design

Nine of the 10 pens were used for the experiment, with free access between pens. Each pen was randomly allocated one of three treatments (control, bitter lupin or sweet lupin), within three 3-pen blocks. Control pens were left untended, apart from some manual weeding. The main weed that grew in these pens was wild mignonette (*Reseda luteola*). Fertilizer (NPK 3:15:13, with Mo) was applied to the area at 360 g.enclosure^{-1}. For pens containing one or other lupin treatment, the soil surface and large clods were broken up using a mattock. Seed was sown in a 3 m x 4 m area within each pen. Bitter lupin was sown at a rate of 60 seeds m^{-2} and sweet lupin at 97 seeds m^{-2}. At day 45, sweet lupin had to be re-sown because vermin (mice and rats) had eaten the seed. One week before re-sowing, 10 g of rat poison ('Wax blocks' Roberts Ltd) was spread throughout the pens. Seed was re-sown at three times the original rate (~290 seeds m^{-2}).

Experiments with pademelons began 81 days after the original sowing of lupins. *Eucalyptus nitens* seedlings, supplied by Forestry Tasmania's Perth Nursery, were planted six per pen, 1 m x 1 m apart. Each animal was exposed to the 9-pen experiment (total 54 seedlings) for 48 hours. New tree seedlings were planted for each animal.

2.1.3 Lupin growth assessments

Two 1 m^2 quadrats were randomly allocated within each pen to assess lupin growth. Once past the cotyledon stage, quadrats were scored for lupin health, percentage 'lupin', 'grass' and 'other vegetation' cover. Lupin health was scored on a scale from 0-3 (0=healthy, with no sign of nutrient deficiency; 1 = 0-33% of the plant necrotic; 2 = 34-66% of the plant necrotic; 3 = 67-100% of the plant necrotic and dead). Percentage lupin cover was scored on a 0-6 scale (0 = 0% cover; 1 = 1-5% cover; 2 = 6-25% cover; 3 = 26-50% cover; 4 = 51-75 cover; 5 = 76-95% cover; 6 = 96-100% cover). Height of living vegetation in the plot was also measured in 16, evenly spaced positions in each quadrat. Scoring of these plots continued on a weekly basis throughout the entire experiment.

2.1.4 Tree seedling browsing damage and height assessments

Eucalyptus nitens seedlings were assessed at 0, 24 and 48 hours exposure to each animal. Severity of browsing damage to each seedling (percentage foliage removed) was given a score of 0-6, which was a visual estimate of the percentage of foliage removed by herbivores:

Score	0	1	2	3	4	5	6
% range	0	1-5	6-25	26-50	51-75	76-95	96-100

Seedlings that were pulled out and were found lying beside the planting hole were also given a score of 6, and were included in the dead category. Apical bud damage was recorded as either damaged by mammals or not. Seedling heights were also measured at 0, 24 and 48 hours.

2.1.5 Statistical analysis

Lupin germination and growth assessments: We analysed data from the lupin treatments with Stat-Graphics© software. The analyses for treatment (bitter *vs.* sweet lupin) and block effects were performed on averages of two 1 m^2 quadrats per plot for percentage cover, health scores and height (cm) at 21, 29, 38, 45, 52, 64, 79, 93, 107 and 121 days since sowing.

Seedling assessments: We tested whether the dependent variables (browsing severity, number of seedlings browsed and change in seedling height) were a function of treatment (control, bitter or sweet lupin). Scores of browsing severity were converted to the midpoint of the range in percentage of foliage removed for each score before analysis. Results were analysed using PROC GLM (SAS Institute Inc., 1989). For statistical purposes, the unit of replication was the treatment and each animal was considered as a block. Severity, number of seedlings damaged and reduction in height data were averaged for all seedlings in each treatment. Data did not need to be transformed. *Post hoc* pairwise comparisons of treatments were made using the Tukey-Kramer adjustment.

2.2 Field experiment

2.2.1 Site and site preparation

The field experiment was established on an ex-pasture plantation (Walker Tree Farm, Gunns Limited), south of Franklin in southern Tasmania (S43º, E146º). The entire plantation was 92.7 ha, but the experiment comprised a total of 4.5 ha in selected areas. The experiment was divided into six blocks, each containing three plots (each 30-50 m wide x 50 m deep). Two blocks were in the north west of the plantation and four blocks were in the north east.

Gunns plantation personnel prepared the experimental area by first cultivating the area, forming rows and inter-rows. Four of the six blocks were aerially sprayed with herbicides, i.e. simazine (12.5 L ha^{-1}) and glyphosate (2.5 L ha^{-1}). The remaining two blocks were sprayed by hand, due to difficult access for aerial spraying. During this period we applied fertilizer (NPK 3:15:13, with Mo) to the entire experimental area at a rate of 300 kg product ha^{-1}.

2.2.2 Treatments and experiment design

The experiment was set up in a randomized block design, with the three treatments (control, bitter lupin and sweet lupin, as for the enclosure experiments) randomly allocated to plots within each block. The control treatment only received herbicide. For the two lupin treatments, lupin seed was inoculated with "Nitrogerm 500" before sowing. Sweet lupin seed was then sown by a manual seed disperser and by hand. All bitter lupin plots were hand-sown, as seed was too large to be sown by a manual spreader. We applied the sweet lupin at a rate of 97 kg ha^{-1} (57 seeds m^{-2}) and the bitter at 400 kg ha^{-1} (57 seeds m^{-2}, based on advice from its supplier, i.e. Andrew Youl, Simmons Plains, Tasmania). Lupin treatments were sown on 2nd and 3rd October 2001, i.e. nine weeks before (week –9) planting of eucalypt seedlings.

Gunns plantation personnel planted the experimental area with *Eucalyptus nitens* seedlings (~ 25 cm tall at planting, supplied by Gunns nursery) on 5th December 2002, once the average height of most bitter lupin plots had reached ~50 cm. Seedlings were planted at 2.5 m spacing. The experimental area was planted two months later than the rest of the plantation.

2.2.3 Lupin germination and growth assessments

In each plot, two 1 m^2 quadrats were randomly placed on mounds and two on the adjacent inter-mounds, to allow assessment of lupin growth on two types of soil tilth. The lupin crop was scored initially for number of seeds germinated m^{-2} and soil tilth (scored from 1 to 4, ranging from crumbs to clods). Weekly scoring of these quadrats continued throughout the entire experiment for (a) % lupin cover, and (b) five random lupin heights (cm).

2.2.4 Growth of a supplementary bitter lupin: NZ Bitter Blue

A supplementary treatment of an alternative bitter lupin (*Lupinus angustifolius* cv. 'New Zealand Bitter Blue') was included in the design, but these plots were very small (28 m^2) and they were used only to assess lupin germination and growth. The seed was sown at the same rate as the sweet lupin, i.e. 97 kg ha^{-1} (57 seeds m^{-2}).

2.2.5 Vegetation assessments around seedlings

Additional vegetation assessments were made within a 1 m^2 quadrat around each of the 21 seedlings assessed per plot for browsing and growth (see Section 2.2.7). Percentage of vegetation cover was estimated for lupin, grasses, fireweed, bare ground and "other". We also measured sixteen heights (4 x 4 rows) at uniform, standard positions within each quadrat, to calculate average height of vegetation around the seedling. In bitter lupin plots, vegetation was assessed three times: (1) 5th –7th December 2001, when eucalypt seedlings were planted, (2) 3rd January 2002, week 4 after eucalypt seedlings were planted, and (3) 12th February 2002, week 10 after eucalypt seedlings were planted. Vegetation assessments were completed for all three treatments at the third time.

2.2.6 Indices of animal abundance

One 15 m scat transect was established in each plot, running perpendicular from the forest/plantation boundary into each plot in blocks 1-4, and from the paddock/plantation boundary into each plot in blocks 5 and 6. Scats were initially cleared from transects one month after lupins were sown (week –5 in relation to planting of eucalypt seedlings). Scats were then cleared from 1 m^2 quadrats along the transects, counted, identified and removed at three subsequent monthly intervals (week –1, week 3 and week 7 in relation to planting of eucalypt seedlings). Scats were counted as individual pellets for each type; piles were not counted because they were not clearly defined entities.

2.2.7 Seedling browsing damage and growth assessments

We marked and measured 21 seedlings per treatment plot, in three rows of seven extending in from the plantation edge. Seedling locations were determined using a GPS that later allowed us to generate weekly maps showing damage in the experimental areas. Measurements of height were made at 0, 0.2, 0.6, 1, 3, 4, 5, 6, 7, 9, 11 and 12 weeks after planting. Dead seedlings were also recorded at each time period. Only 14 seedlings of the total 378 scored died (3.7% mortality), so survival is not considered further. Extent and severity of browsing and apical bud damage by browsing mammals was measured at planting, then at the same intervals as for heights. Insect damage was also scored, as for mammal damage. However, as only one seedling of the total 378 scored had an insect damage score > 1, we do not consider it further.

2.2.8 Statistical analyses

Treatments in block 1 showed consistent differences from the other blocks, due to the failure of the bitter lupin to establish, and the failure of herbicide in the control plot to prevent weed growth. As a result, this block was analysed separately from the other five (blocks 2-6) for all but the lupin germination results.

Lupin germination and growth assessments: Lupin germination and growth data were analysed by analysis of variance for a replicated split-plot design, using treatments as main plots, mound position as sub-plots, and quadrats as sub-sub-plots.

Vegetation assessments around seedlings: Plot means of vegetation height and percentage cover of lupin were calculated for the bitter lupin plots in blocks 2-6 at week 0, 5 and 6, and for sweet lupin and control plots at week 6 only. Plot means were used as the unit of replication in analyses. Data were square-root arcsine transformed before analysis to satisfy assumptions of normality and homoscedasticity (Zar, 1996). Treatment and block effects were tested using the general linear model procedure PROC GLM (SAS Institute Inc., 1989). Pairwise comparisons of treatments were made after using the Tukey-Kramer adjustment for multiple comparisons. For block 1 at week 6, treatment means were calculated from the 21 assessments within each plot.

Indices of animal abundance: For analysis, scats were pooled for each plot for each species from the three collection periods (-1, 3 and 7 weeks from planting). There were only sufficient data for further analysis of pademelon and rabbit scats. Effects of treatment and block were tested in blocks 2-6 using the general linear model procedure PROC GLM (SAS Institute Inc., 1989), after square root transformation to satisfy assumptions of normality and homoscedasticity (Zar, 1996). Pairwise comparisons of treatments were made as above.

Seedling browsing damage and growth assessments: Scores of severity of browsing were converted to the midpoint of the range in percentage of foliage removed for each score before analysis. Extent of browsing was analysed as the number of seedlings with any mammal damage per plot, and the occurrence of apical bud damage was analysed as the number of seedlings with apical bud damage per plot. For analyses of browsing severity, extent, apical bud damage and height, the unit of replication for blocks 2-6 was the plot. In block 1, the unit of replication was the individual seedling within a plot.

We obtained least-squares means (lsm) of browsing severity, browsing extent, apical bud damage and seedling height over time for each treatment, using the general linear model procedure PROC GLM (SAS Institute Inc., 1989). For browsing severity, extent and apical bud damage, we then tested for effects of treatment and block (between subject effects), and time (week since planting) plus its interactions (within-subjects effects), using PROC GLM for repeated measures (SAS Institute Inc., 1989). We only included data once the mean browsing level exceeded 5% (from 9 weeks since planting), because testing effects when browsing was minimal was biologically meaningless. The test for sphericity of orthogonal components was significant for browsing severity, so the Greenhouse-Geisser (G-G) adjusted P value was used for within-subjects tests of significance for this variable. Severity data were square-root arcsine transformed before analysis to satisfy assumptions of normality and homoscedasticity (Zar, 1996). We tested for treatment effects on change in height from planting to the end of the experiment, using PROC GLM. Pairwise comparisons of treatments were made as above.

The effect of bitter lupin on tree seedling growth in the absence of mammal browsing was examined using the regression procedure (PROC REG) (SAS Institute Inc., 1989). Data included only seedlings in bitter lupin plots and only seedlings with no mammal damage at week 6 and/or week 12. The dependent variable was either seedling height at week 6 or week 12 (for short and longer term effects respectively) and the independent variable was the percentage lupin cover at week 6. No data transformations were needed.

3. Results

3.1 Enclosure experiment

3.1.1 Germination and growth of lupin

Bitter lupin germinated faster than the sweet variety, but there was no significant difference in % germination (c. 90% germination for all types of lupin). There were no significant differences in percentage lupin cover between treatments at any time after sowing (Figure 1). However, there was a decrease in sweet lupin cover at day 45 to zero, after mice and rats had eaten the lupin. Once re-sown, the sweet lupin cover rapidly increased again.

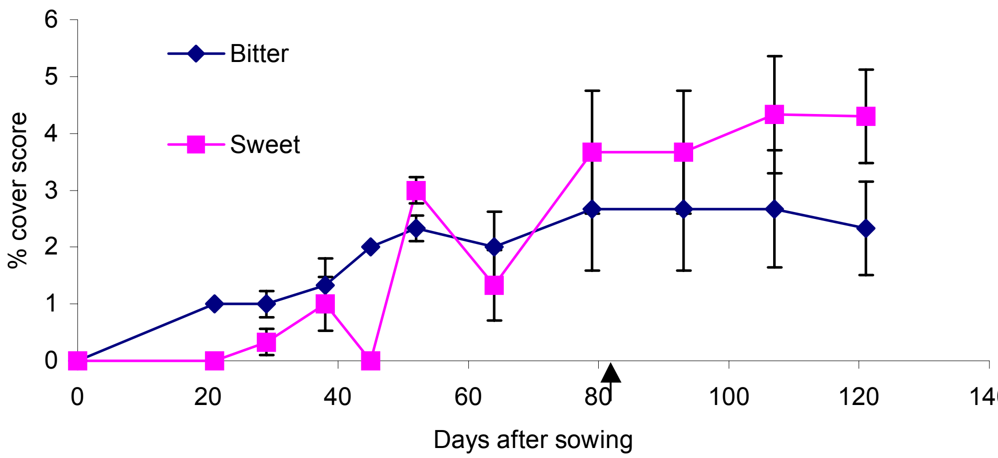

Figure 1. Score of percentage cover of bitter and sweet lupin over time after sowing in the enclosure experiment (least-squares means with s.e.). Arrow signifies the start of the captive animal experiments (81 days after sowing).

Once germinated, both lupin varieties were generally healthy (Score 1 or 2) (Figure 2). Sweet lupin established slightly later than the bitter lupin, and the higher sweet lupin health score at day 38 ($F_{1,5} = 16.0$, $P = 0.0572$) reflected its poorer health resulting from browsing by rats and mice. In one of the three blocks, the bitter lupin was slightly deformed and density was low due to water-logging (data not shown). Both bitter and sweet lupin began to flower at 107 days (10th January 2002) since sowing; thereafter seedpods began to form. By day 121, both varieties were either in full flower, with fully enlarged seedpods or senescing.

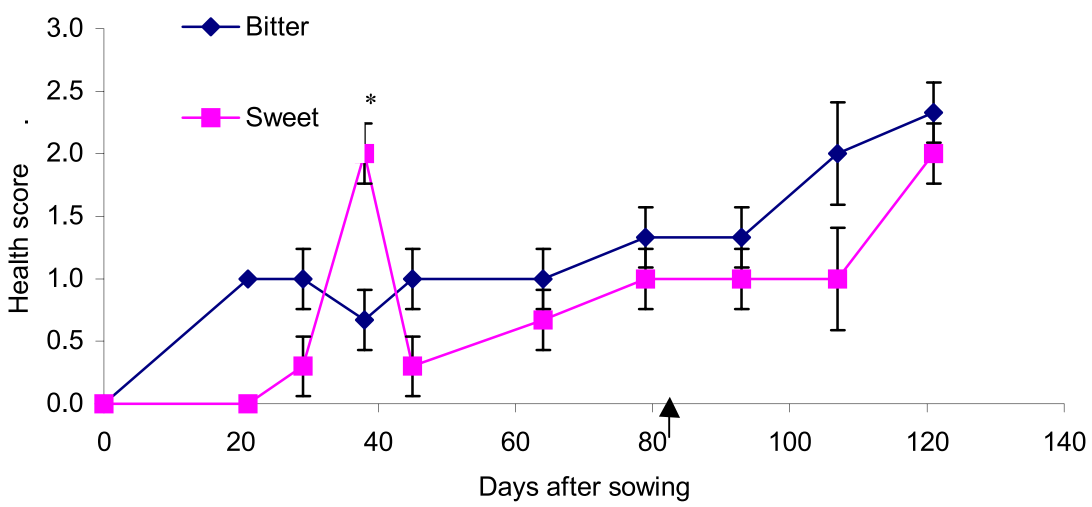

Figure 2. Health of bitter and sweet lupin in relation to time after sowing in the enclosure experiment (least-squares means with s.e.). The arrow indicates the start of captive animal experiments (81 days after sowing). '*' indicates values are significantly different (α =0.05) at that time. Note that the lower the score, the healthier the lupin.

Before the vermin control on day 45, bitter lupin tended to be taller than sweet lupin (significant at day 21, $F_{1,5} = 1$, $P = 0.0144$) (Figure 3). Once sweet lupin re-established, bitter and sweet lupin heights remained similar (Figure 3).

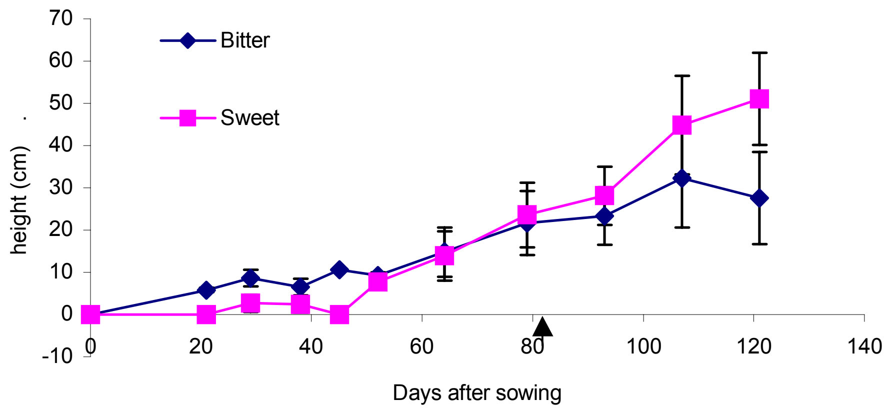

Figure 3. Height of bitter and sweet lupin over time since sowing in the enclosure experiment (least-squares means with s.e.). Arrow indicates the start of captive animal experiments (81 days after sowing).

3.1.2 Mammal browsing and height of tree seedlings

There was a significant treatment effect ($F_{2,10} = 7.84$, $P = 0.0089$) on percentage foliage browsed from *E. nitens* seedlings. Mean severity of browsing to *E. nitens* was higher in control patches (10.2%, s.e 1.8) than bitter lupin (2.1%, s.e 1.8) or sweet lupin (1.2%, s.e 1.8) patches after 48 hours (Figure 4).

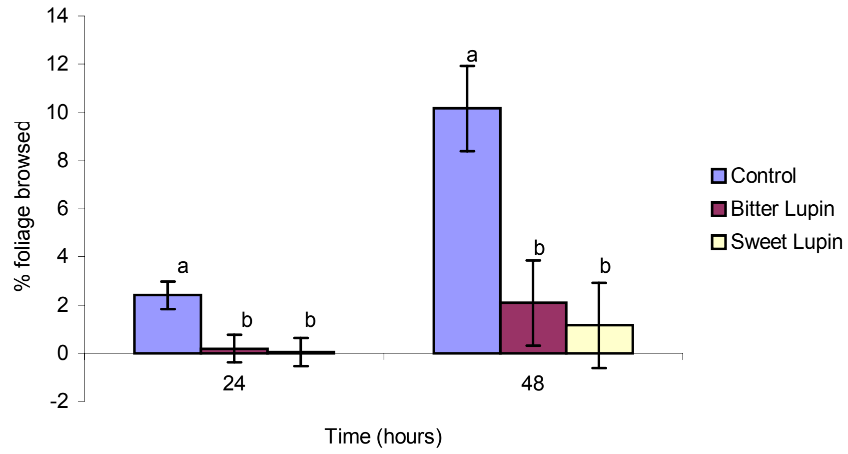

Figure 4. Browsing severity to *E. nitens* seedlings in control, bitter lupin and sweet lupin treatments after 24 and 48 hours in the enclosure experiment. Units are percent foliage browsed (removed) by Tasmanian pademelons (least-squares means with s.e bars). Letters that differ indicate significant differences (α=0.05 after Tukey-Kramer adjustment).

Similarly, there was a significant treatment effect ($F_{2,10} = 22.79$, $P = 0.0002$) on the number of seedlings browsed after 48 hours. In the control treatment, the mean number of seedlings with any damage was 5.7 (s.e 0.5), compared to 1.5 (s.e 0.5) in the bitter lupin treatment and 1.2 (s.e 0.5) in the sweet lupin treatment (Figure 5).

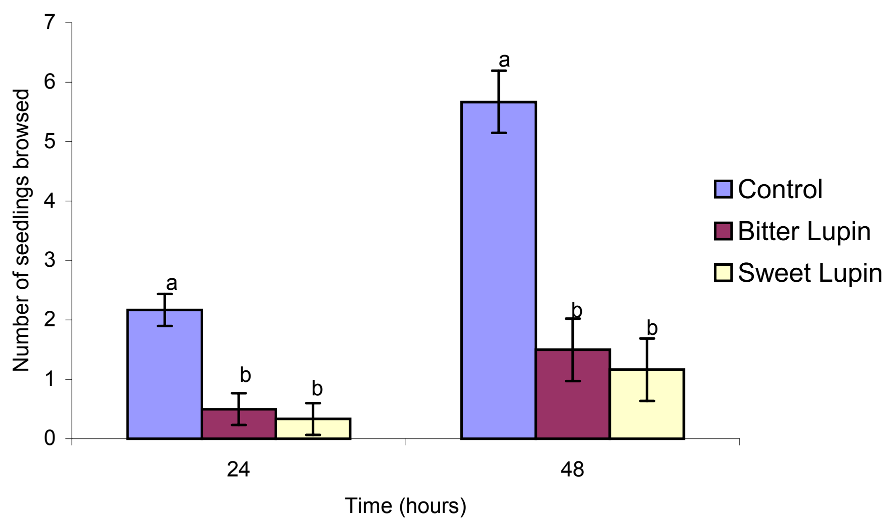

Figure 5. Number of *E. nitens* seedlings damaged by Tasmanian pademelons in control, bitter lupin and sweet lupin treatments after 24 and 48 hours in the enclosure experiment. Letters that differ indicate significant differences (α=0.05 after Tukey-Kramer adjustment).

There was a significant treatment effect ($F_{2,10} = 6.31$, $P = 0.0169$) on the reduction in height of seedlings after 48 hours. Seedlings lost more height in the control treatment than either of the lupin treatments (Figure 6).

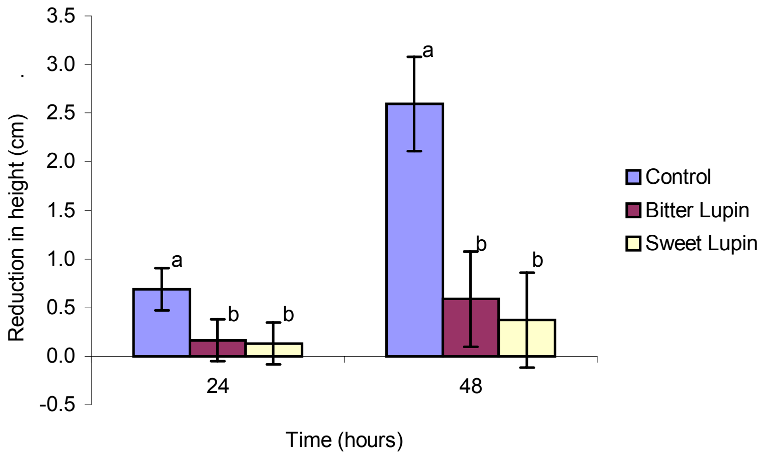

Figure 6. Reduction in height (least-squares means with s.e) of *Eucalyptus nitens* seedlings in control, bitter lupin and sweet lupin treatments after 24 and 48 hours in the enclosure experiment. Letters that differ indicate significant differences (α=0.05 after Tukey-Kramer adjustment).

3.2 Field experiment

3.2.1 Germination and growth of lupin

Germination rate of all lupin types was high (c. 90%). Soon after germination, 'New Zealand Bitter Blue' was totally browsed; no further results of this cultivar are presented. The grand mean for germination rate of the other two lupins (bitter and sweet) was 89.6% (s.e 18.6), and there was no significant effect due to, or interaction between, lupin treatment, block, or mound position. However, lupins appeared to be generally healthier on mounds than on the inter-mounds. There were also no significant differences in lupin growth with various soil conditions on any assessment date. After initial germination, the sweet lupin was eaten by animals and so no more lupin growth data were collected for this lupin. Height and cover of the bitter lupin in blocks 2-6 increased progressively after sowing (Figs. 7 and 8). Conversely, in block 1, height increased initially, but then decreased after day 92, and lupin was non-existent by day 120 (Figs. 7 and 8).

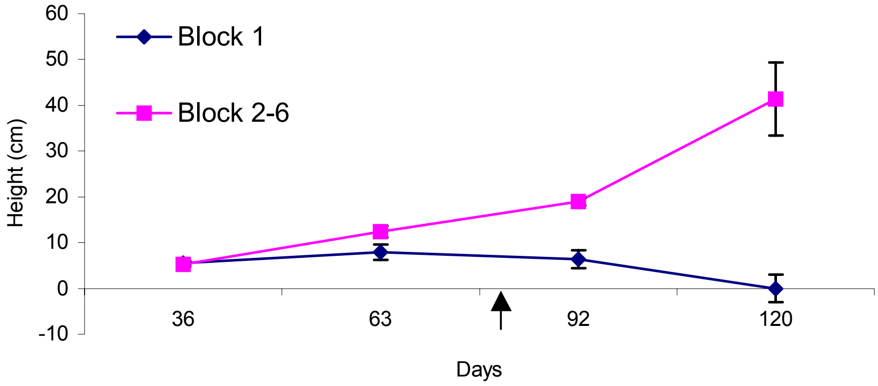

Figure 7. Height of bitter lupin over time since sowing for block 1 and combined data from blocks 2-6 (least-squares means with s.e). Arrow indicates approximate time when *E. nitens* seedlings were planted.

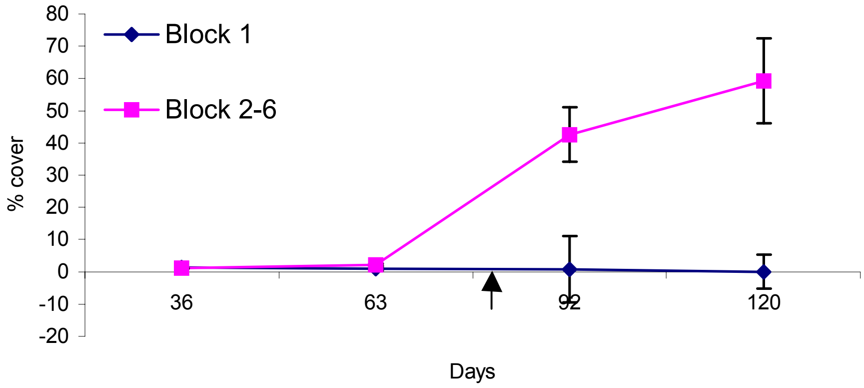

Figure 8. Percentage cover of bitter lupin over time since sowing for block 1 and combined data from blocks 2-6 (least-squares means with s.e). Arrow indicates approximate time when *E. nitens* seedlings were planted.

3.2.2 Vegetation characteristics

Bitter lupin established reasonably well in blocks 2-6, but failed to establish in block 1 (Table 1, Figures 7, 8, 9 and 10). Sweet lupin failed to establish in any block (Figure 9). In blocks 2-6, fireweed and grass were the two most abundant plant groups, except in the bitter lupin plots (Figure 9). The percentage cover of fireweed tended to be greatest in control plots and least in bitter lupin plots ($F_{2,8} = 3.234$, $P = 0.0932$). There was no significant difference between treatments in the % cover of grasses ($F_{2,8} = 0.65$, $P = 0.5473$) or "other" plants ($F_{2,8} = 0.19$, $P = 0.8294$). In block 1, cover of both fireweed and grass was greatest in the control plot, intermediate in the sweet lupin plot and least in the bitter lupin plot (Figure 9).

Table 1. Vegetation height (mean ± 1s.e) and percentage cover of bitter lupins (mean ± 1 s.e.), in blocks 2-6 and block 1, at 0, 5, and 6 weeks after tree seedlings were planted.

	Blocks 2-6		Block 1	
Week	Veg. Height (cm)	% Cover lupin	Veg. Height (cm)	% Cover Lupin
0	6.5 ± 1.4	28.5 ± 3.1	2.1 ± 0.5	4.9 ± 1.2
5	15.3 ± 2.8	36.3 ± 4.8	1.8 ± 0.6	0.1 ± 0.1
6	42.7 ± 5.6	45.2 ± 9.4	3.7 ± 1.3	0.0 ± 0.0

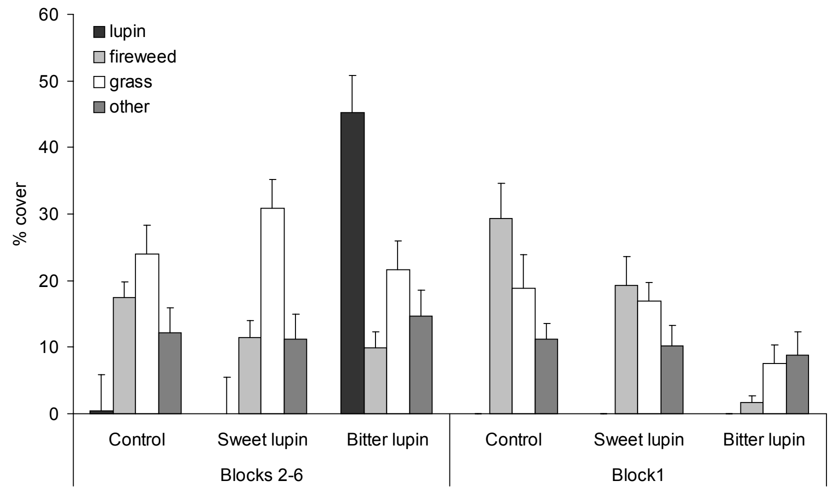

Figure 9. Percentage cover (mean + s.e) of major vegetation types within 1 m^2 area surrounding *E. nitens* seedlings in the three treatments in blocks 2-6 and block 1 at week 6 after *E. nitens* seedlings were planted.

In blocks 2-6, there was a significant treatment effect on vegetation height at week 6 ($F_{2,8}$ = 7.14, P=0.0166). Vegetation was taller in the bitter lupin plots than in either the sweet lupin or control plots (Figure 10). In block 1, vegetation was tallest in the control plot, intermediate in the sweet lupin plot and least in the bitter lupin plot (Figure 10).

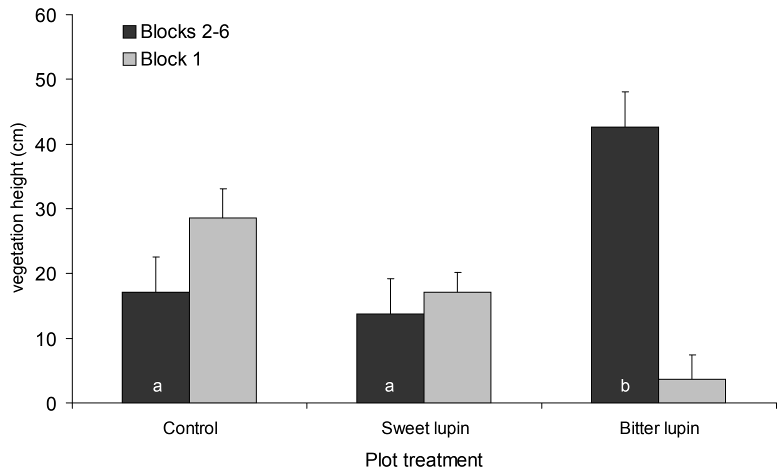

Figure 10. Height of vegetation (mean + s.e) in 1m^2 area surrounding *E. nitens* seedlings in the three treatments in blocks 2-6 and block 1, at week 6 after *E. nitens* seedlings were planted. Within blocks 2-6, letters that differ indicate significant differences (α=0.05 after Tukey-Kramer adjustment).

3.2.3 Indices of animal abundance

A total of 745 pademelon, 322 rabbit, 26 hare, 17 brushtail possum, 4 Bennett's wallaby and 3 carnivore scats as found on blocks 1-6 from the three collection periods at week –1, 3 and 7 from the date when tree seedlings were planted. The numbers of pademelon and rabbit scats for each treatment in blocks 2-6 and block 1 are shown in Table 2. There were no treatment or block effects on the counts for pademelon scats ($F_{2,8}$ = 0.16, P=0.8574; $F_{4,8}$ = 1.70, P=0.2417 respectively) or rabbit scats ($F_{2,8}$ = 0.41, P=0.6739; $F_{4,8}$ = 1.45, P=0.3041 respectively). Scats from other species were not analysed due to insufficient data.

Table 2. Cumulative counts of pademelon and rabbit scats for each treatment in blocks 2-6 (mean ± s.e) and block 1.

	Blocks 2-6		Block 1	
Treatment	Pademelon	Rabbit	Pademelon	Rabbit
Control	51.6 ± 15.7	16.0 ± 15.0	48	0
Sweet lupin	34.4 ± 15.7	15.8 ± 15.0	8	0
Bitter lupin	47.8 ± 15.7	32.6 ± 15.0	20	0

3.2.4 Mammal browsing and growth of tree seedlings

In blocks 2-6, the severity of mammal damage to *E. nitens* seedlings was relatively low during the first four weeks after planting (Figure 11a). It then increased over time, and remained consistently higher in control and sweet lupin plots than in bitter lupin plots. When the average severity was greater than 5% (weeks 9 to 12 after planting), treatment, block and time effects were significant ($F_{2,8}$ = 4.68, P=0.0451; $F_{4,8}$ = 5.55, P=0.0194; $F_{2,16}$ = 13.87, P=0.0048 respectively), but the interactions of treatment and block with time were not ($F_{4,16}$ = 0.42, P=0.6795; $F_{8,16}$ = 1.98, P=0.1842 respectively). In contrast, mammal damage to *E. nitens* seedlings in block 1 was greatest in the bitter lupin plot, intermediate in the sweet lupin plot and least in the control plot (Figure 11b).

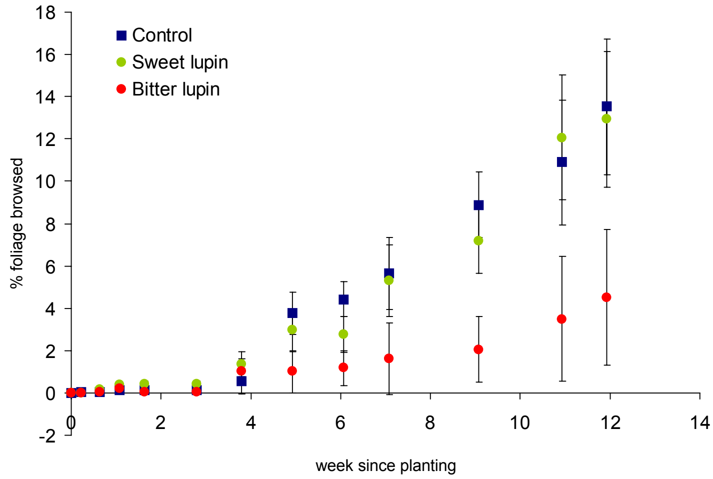

Figure 11a. Mammal browsing severity, as the percentage of foliage browsed from *E. nitens* seedlings (least-squares means ± s.e), over time since planting, in blocks 2-6.

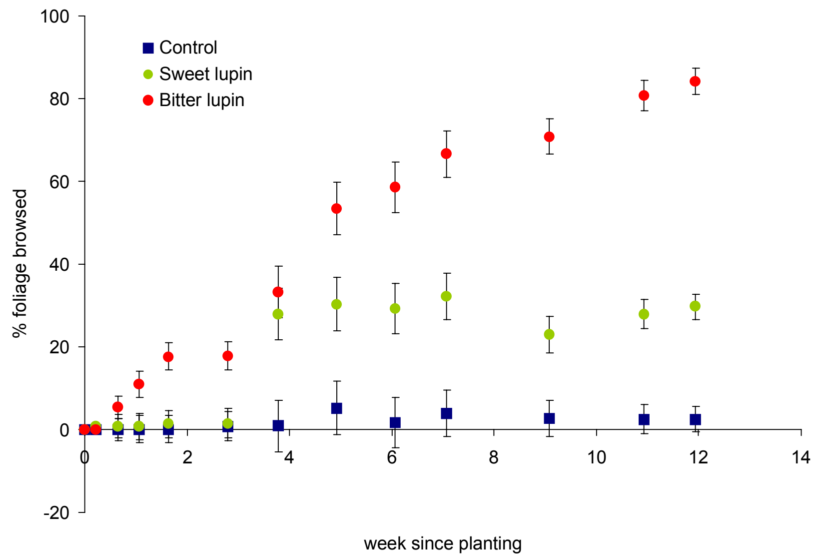

Figure 11b. Mammal browsing severity, as the percentage of foliage browsed from *E. nitens* seedlings (least-squares means ± 1 s.e.), in relation to time after planting in block 1.

When mammal damage in blocks 2-6 was expressed as extent of damage, i.e., the number of *E. nitens* seedlings browsed (Figure 12), irrespective of how severely, patterns reflected severity data. Fewer seedlings were browsed in the bitter lupin treatment than in the other treatments. When the average severity was greater than 5% (weeks 9-12 after planting), treatment, block and time effects on extent of damage were significant ($F_{2,8} = 6.54$, $P = 0.0207$; $F_{4,8} = 12.30$, $P = 0.0017$; $F_{2,16} = 37.37$, $P = 0.0001$ respectively), the interaction of time with block was significant ($F_{8,16} = 4.07$, $P = 0.0081$, but the interaction of time with treatment was not ($F_{4,16} = 0.14$, $P = 0.9650$).

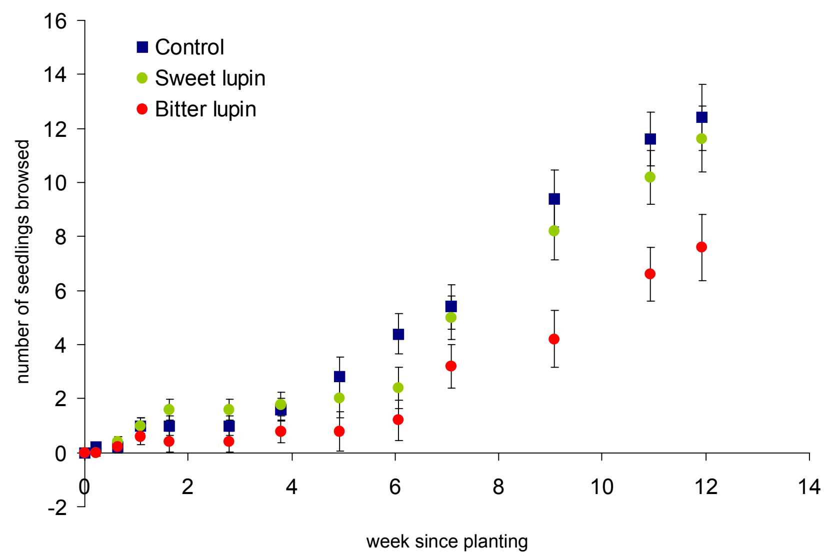

Figure 12. Number of *E. nitens* seedlings browsed by mammals per plot (least-squares means ± s.e) over time since planting in the three plot treatments, in blocks 2-6.

When mammal damage in blocks 2-6 was expressed as the number of *E. nitens* seedlings with apical bud damage (Figure 13), patterns again reflected severity data. Fewer seedlings had apical bud damage in the bitter lupin treatment than in the other treatments. When the average severity was greater than 5% (weeks 9-12 after planting), treatment, block and time effects on apical bud damage were significant ($F_{2,8}$ = 5.03, P=0.0387; $F_{4,8}$ = 9.82, P=0.0035; $F_{2,16}$ = 13.88, P=0.0003 respectively), the interaction of time with block was significant ($F_{8,16}$ = 4.33, P=0.0061, but the interaction of time with treatment was not ($F_{4,16}$ = 1.61, P=0.2211).

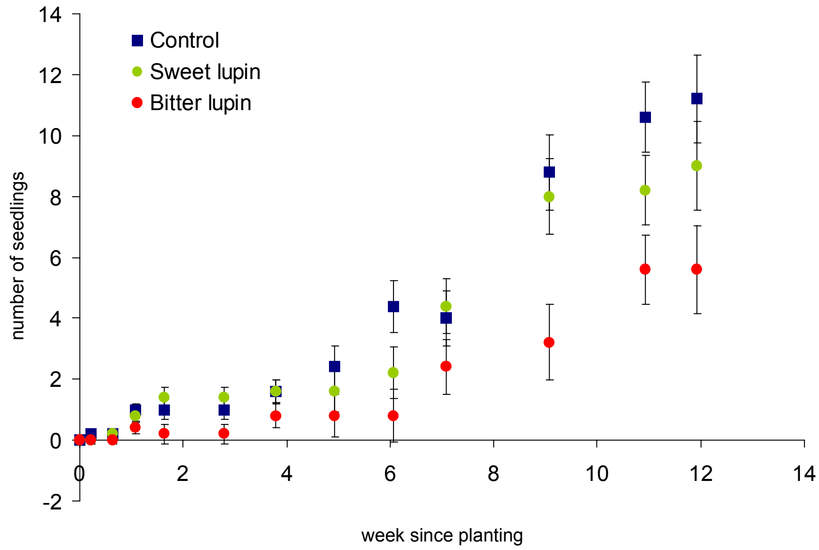

Figure 13. Number of *E. nitens* seedlings with apical bud damage per plot (least-squares means ± s.e) over time since planting, in blocks 2-6.

In blocks 2-6, the mean increase in *E. nitens* seedling height from planting to the end of the study 12 weeks later was 12.3 cm (42%), 15.4 cm (53%) and 18.6 cm (64%) for control, sweet lupin and bitter lupin treatments respectively (Figure 14a). There was a significant treatment effect on this change in height ($F_{2,8}$ = 4.63, P=0.0461) and a significant block effect ($F_{4,8}$ = 19.84, P=0.0003). In contrast, *E. nitens* seedlings in block 1 grew on average 11.5 cm in the control plot, 1.4 cm in the sweet lupin plot, but lost 7.6 cm in the bitter lupin plot over 12 weeks (Figure 14b).

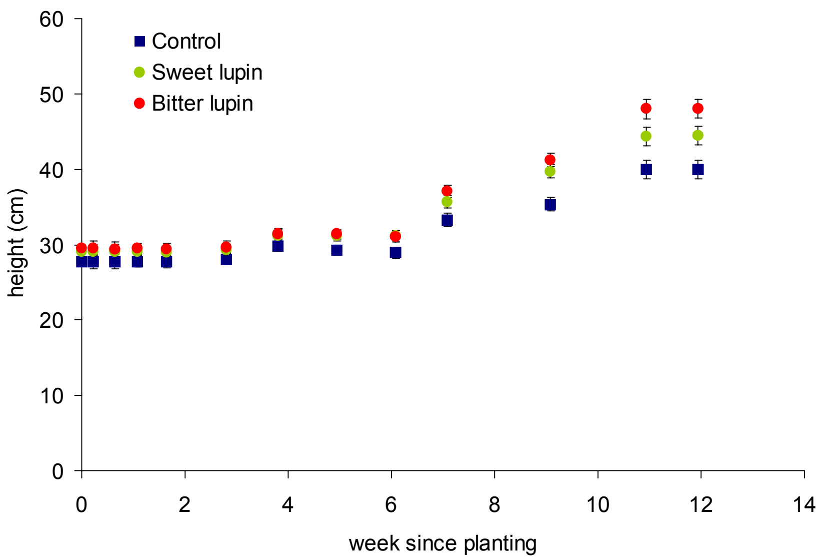

Figure 14a. Height (cm) of live *E. nitens* seedlings (least-squares means ± s.e) over time since planting in blocks 2-6.

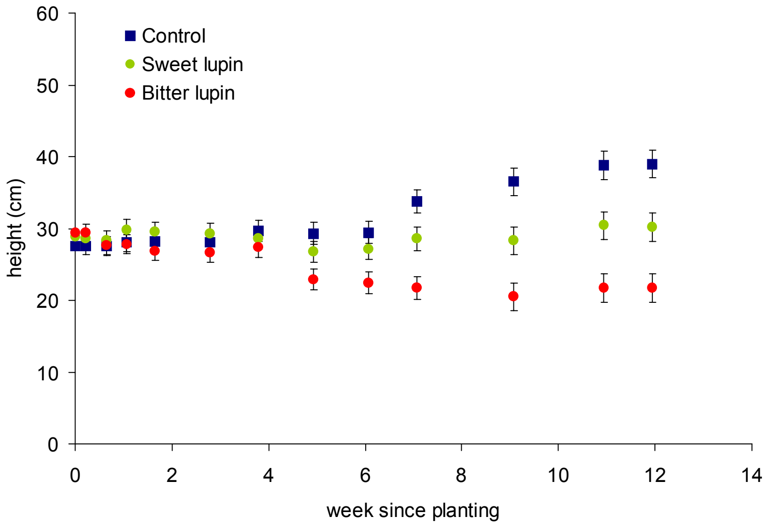

Figure 14b. Height (cm) of live *E. nitens* seedlings (least-squares means ± s.e) over time since planting in block 1.

Irrespective of the major plant type immediately surrounding a tree seedling, there was a clear relationship between vegetation height and severity of mammal browsing (Figure 15). When vegetation height was equal to or greater than the seedling height (i.e. 30 cm), no seedling was severely browsed and only 1.8% (2 of 113 seedlings) had lost 38% of foliage through browsing. In contrast, when surrounding vegetation was shorter than the seedling, 12.1% of seedlings had lost at least 38% of foliage through browsing, and the browsing severity ranged up to 100%.

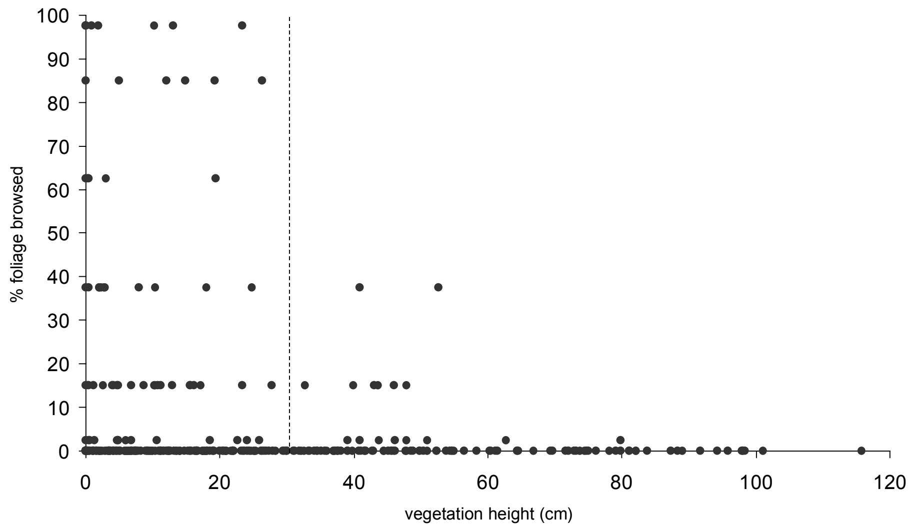

Figure 15. Mammal browsing severity, as the percentage of foliage browsed from *E. nitens* seedlings in all blocks at week 6 after planting, in relation to the mean vegetation height in an area of $1m^2$ around the seedling at week 6, irrespective of vegetation type (e.g. lupin, fireweed etc). Vertical dashed line indicates the mean height of *E. nitens* seedlings at that time.

3.2.5 Effect of bitter lupin on tree seedling growth in the absence of mammal browsing

At week 6, the effect of lupin cover on *E. nitens* seedling height was not significant (adjusted $r^2 = 0.0065$, $F_{1,97} = 1.638$, P=0.2038), using seedlings that had no mammal damage at that time. However, when the lupin cover at week 6 was compared to *E. nitens* seedling height at week 12, the effect was significant (Figure 16). Seedling height at week 12 increased as lupin cover at week 6 increased up until about 90% cover, beyond which there appeared to be a slight reduction. The model that best fitted the data (adjusted $r^2 = 0.1928$, $F_{1,58} = 8.164$, P=0.0008) included a quadratic function to take this into account:

$$height = 41.052 + 0.448 lupin - 0.0026 lupin^2$$

where *height* is seedling height (cm) at week 12, and *lupin* is the % cover of bitter lupin in the $1m^2$ quadrat around a seedling at week 6, using only seedlings with no mammal browsing at both weeks 6 and 12.

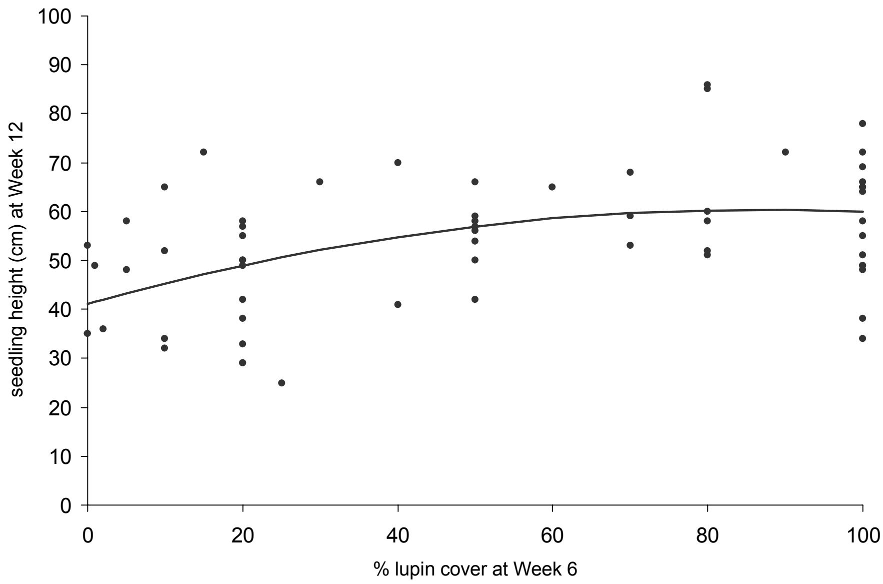

Figure 16. Height (cm) of unbrowsed *E. nitens* seedlings at week 12 after planting in relation to the percentage cover of bitter lupin at week 6 in a 1 m^2 area around the seedling.

This model indicated that, seedlings with 90% bitter lupin cover at week 6 were 47% (19 cm) taller at week 12 than those with no lupin cover.

3.2.6 Effect of NZ bitter blue lupin on tree growth

Tree data collected from the NZ bitter blue plots were not analyzed as the seed was eaten and lupin did not establish an adequate cover.

4. Discussion

4.1 Germination and establishment of lupin cover crops

In both enclosure and field experiments, the germination rate of bitter and sweet lupin was generally high, though somewhat more variable for the bitter lupins (data not presented).

Rodents and possibly birds contributed to poor establishment of sweet lupins in enclosure experiments, but once these problems were overcome, it established at least as successfully as the bitter lupin. In the field, however, sweet lupin failed to establish at all because it was browsed immediately after germination. We speculate that the difference in establishment success of the two varieties was related to alkaloid content; expected alkaloid concentrations in seeds of the lupins tested were sweet lupins 0.002%, bitter 'lupini' 1.5-2.5%, and New Zealand bitter blue 1.3-1.7% (G. Dean, A. Youl and NSW Agriculture). Alkaloid content of foliage has not been quantified.

It is encouraging that no special cultivation appears to be needed for lupin establishment. Note that lupins also can be successfully established on 2nd rotation plantation sites with thick litter layers and with no more cultivation than that required for the routine establishment of tree seedlings (Smethurst et al., 1986). Hence, lupin establishment may cost as little as that required to purchase and spread the seed. However, although not specifically assessed, bitter lupin appeared to be sensitive to waterlogging and germinated poorly in wet areas of both enclosure and field experiments.

Bitter lupin establishment appeared to fail in block 1 of the field experiment due to heavy browsing. However, scat counts did not indicate marked differences in animal numbers between block 1 and blocks 2-6 (Table 2), so it is unclear why the browsing level was so different between the blocks. This failure suggests a risk in growing lupins that is yet to be fully understood or quantified.

4.2 Effectiveness of lupins in reducing mammal browsing damage to eucalypt seedlings

In enclosure experiments, *E. nitens* seedlings in both bitter and sweet lupin treatments were less damaged than those in the control treatment. In the field experiment, results analysed from combined blocks 2-6 data showed that seedlings in the bitter lupin treatment were less damaged than seedlings in the sweet lupin and control treatments. The difference in effectiveness of the sweet lupin treatment in the enclosure versus field experiment is clearly a result of its failure to establish in the latter.

The only apparently conflicting result in the field experiment was in block 1, in which browsing damage to seedlings was highest in the "bitter" plot, intermediate in the "sweet" plot and lowest in the "control" plot (Figure 11b). We suggest this occurred as a result of failure of either lupin variety to establish in this block, along with the failure of the herbicide in the control plot: vegetation cover and height in block 1 plots were essentially the opposite of the other blocks (Table 1, Figures 10 and 11). When we related browsing damage to seedlings with average vegetation height around the seedling, there was strong evidence from all blocks that browsing damage was substantially less in taller vegetation, irrespective of vegetation type (Figure 15). These results are consistent with the enclosure experiment results, where both sweet and bitter lupins had adequate height to reduced browsing damage. These results are also consistent with our earlier studies, showing that pine (*Pinus radiata*) seedlings were less damaged by mammalian herbivores when planted in bracken than in low grass patches, and less in unclipped bracken or shrub patches than in clipped bracken or shrub patches (Pietrzykowski, 2000; Pietrzykowski et al., submitted). Similarly, results from Figure 12 indicate that part of the explanation for reduced severity of browsing when bitter lupin plots establish successfully, is that herbivores take longer before each new seedling is first damaged. This too is consistent with our earlier results (Pietrzykowski et al., submitted). That the sweet lupins were not browsed significantly in the enclosure experiment suggests either that sweet lupins at that stage of growth are not particularly attractive to pademelons, or that the time frame was too short to detect any intake.

4.3 Effect of bitter lupin on short-term height growth of eucalypt seedlings

The presence of bitter lupin seemed to result in competition for light between it and the tree seedlings, but this was potentially a benefit because it forced the tree seedlings to increase in height faster (Figure 16), helping to reduce the risk of browsing and potentially improving tree form with presumably straighter stems with fewer and smaller branches. It is important to note, however, that the period of the experiment, i.e. spring-summer 2001-2002, was particularly wet and the site was a relatively fertile ex-pasture site. Hence, competition between lupins and eucalypt seedlings for water and nutrients was unlikely to be important. Under drier or less fertile conditions, competition between lupins or other competing vegetation and tree seedlings can be intense (Smethurst and Nambiar, 1989; Smethurst et al., 1986), necessitating the use of spot or strip weed control around the tree seedlings.

5. Recommendations

5.1 Preliminary prescriptions for managing lupins

Based on this study, we do not recommend using sweet lupin as a cover crop. We suggest three factors related to lupin management are important for the successful establishment of bitter lupin and the subsequent growth of eucalypt seedlings:

1. Site preparation: Although lupins require minimal cultivation, some cultivation of mounds and intermounds is likely to be beneficial . Routine mound ploughing accompanied by light discing of the inter-row is likely to be adequate.
2. Sowing rate of the lupin: We sowed the lupin seeds at a rate based on commercial practices and previous research (Andrew Youl pers. comm., Smethurst et al., 1986). This rate, i.e. bitter lupin 400 kg ha^{-1} (57 seeds m^{-2}), can probably be considered the maximum required.
3. Lupin control around each tree seedling: Under conditions where competition for water or nutrients is likely to be important, preventing the establishment of lupins by manual or chemical weeding in a zone of 60-100 cm diameter around each seedling should be considered. Timely applications of simazine or glyphosate may be appropriate, but these options need to be tested. We caution against the use of strip applications of herbicide down the tree row, because there is the potential that such strips will become preferred paths for browsers that will lead them directly to the tree seedlings and result in increased browsing damage. The potentially positive and negative effects of competition for light need to be resolved before we can recommend related precautions, but we suggest as a precaution that complete over-topping of the tree seedlings should be avoided if possible.

5.2 Refining the method

This study should be seen as the initial step in determining the success of cover crops, such as bitter lupin, for reducing browsing damage to tree seedlings in plantations. A number of topics need to be addressed to refine the method:

1. Determine the risk of bitter lupin becoming a weed, and recommend conditions for which this risk will be minimal.
2. Determine the minimum effective density or seeding rate required for bitter lupin that results in sufficient vegetation cover to minimize browsing.
3. Define the minimum area around each seedling that should remain vegetation-free to minimize both browsing and detrimental competition.
4. Estimate the benefits of combining cover crops with other browsing control methods, such as the use of seedlings with low palatability.
5. Determine suitable geometric layouts of deterrent vegetation, e.g. lupin patch size.
6. Explore the use of other vegetation management options, e.g. retention of selected native species.
7. Quantify the long-term effects of lupin or other suitable deterrents on the growth and form of tree seedlings in the presence and absence of mammal browsing.

Acknowledgments

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