RELATIONSHIPS BETWEEN SEED MASS, SEED NUTRIENTS, AND SEEDLING GROWTH IN *BANKSIA CUNNINGHAMII* (PROTEACEAE)

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We examined relationships between seed mass, seed nutrients, and seedling growth in two populations of *Banksia cunninghamii* to determine whether large seed mass provided an advantage for seedling establishment on nutrient-poor soils. N and P content of seeds increased more than proportionally with increasing seed mass, indicating that large seeds had greater concentrations of these nutrients than small seeds. K content of seeds increased either proportionally or more than proportionally with seed mass, depending on population. In a common garden, larger seeds produced larger seedlings, although seed mass had little effect on seed germination, seedling survival, root : shoot ratios, or relative growth rate. In a nutrient-deprivation experiment, seedlings were smaller under low nutrients than under high nutrients. Under low nutrients in one population, the increase in seedling size with seed mass was greater than proportional, indicating that large seed mass minimized the effect of nutrient deprivation. Under high nutrients in this population and under both nutrient regimes in the other population, the increase in seedling size with seed mass was only proportional. Overall, our findings highlight the advantage of large seed mass on nutrient-poor soils, probably by rendering seedlings less reliant on soil nutrients.

Keywords: *Banksia*, nutrient poor soils, Proteaceae, seed mass, seed nutrients, seedling allocation patterns.

Introduction

Variation in seed mass occurring within or between species is commonly interpreted as a trade-off between seed number and mass. Although large seeds have an increased chance of establishing seedlings, they are necessarily produced in fewer numbers than small seeds when resources are finite (Westoby et al. 1996). Large and well-resourced seeds have been shown to provide an advantage when seedlings are competing with neighbors (Black 1958; Stanton 1984; Parrish and Bazzaz 1985; Eriksson 1999; Turnbull et al. 1999). In addition, seedlings from large seeds often have a specific advantage under a variety of adverse environmental conditions, including shading, burial under litter, drought, and defoliation (reviewed by Westoby et al. 1996).

The advantage of large seed mass in nutrient-poor environments has received little attention (Milberg et al. 1998). Enhanced nutrient reserves in large seeds and their translocation from cotyledons to seedlings during early growth can reduce the reliance of seedlings on external supplies of nutrients, which would be advantageous on infertile soils (Stock et al. 1990; Milberg and Lamont 1997). In some instances, seeds selectively store particular nutrients so that seed nutrient reserves complement nutrients available in the soil (Fenner and Lee 1989; Stock et al. 1990). In addition, larger seeds often produce larger seedlings with lower relative growth rates than

small seeds (Westoby et al. 1992). An increase in either the absolute size of root systems or relative allocation to roots promotes access to soil nutrients, while lower growth rates reduce the requirement for nutrients by seedlings (Stock et al. 1990; Milberg et al. 1998; Lloret et al. 1999).

If large nutrient-rich seeds are advantageous for seedling establishment on nutrient-poor soils, then we would expect such seeds to have higher success and be more common in low nutrient environments than high nutrient environments. Several studies have reported increased survival and/or growth of seedlings of large-seeded compared to small-seeded species in nutrient-poor environments (Stock et al. 1990; Jurado and Westoby 1992; Milberg et al. 1998; Lloret et al. 1999). Studies of closely related species have also found relationships between increased seed size and infertile soils. Lee and Fenner (1989) found that species in the grass genus *Chionochloa* from infertile soils had larger seeds with more seed nutrients and produced seedlings with smaller shoots than species from fertile sites. Similar relationships between seed mass, seed nutrient content, and soil fertility were reported between closely related species pairs in the family Proteaceae (Esler et al. 1989; Mustart and Cowling 1992). However, contrary to expectations, surveys that correlate soil fertility with seed mass over broad geographic scales have generally found a lack of evidence for larger seeds on poor soils (Westoby et al. 1990; Hammond and Brown 1995; Leishman et al. 1995).

Here, we examine whether large seed mass provides an advantage for seedling establishment on nutrient-poor soil in two populations of *Banksia cunninghamii* (Proteaceae). First, we examine the relationship between seed mass and the N, P, and K content of seeds. Second, we grow seedlings in a common

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garden and examine relationships between seed mass and seedling growth. Finally, we grow seedlings under high and low nutrient regimes and determine whether large mass provides an advantage for seedling growth and survival.

Material and Methods

Study Species and Sites

The shrub *Banksia cunninghamii* (formally *Banksia spinulosa* var. *neoanglica*) occurs in fire-prone heath or woodland on nutrient-poor soils near the Great Dividing Range in northern New South Wales, Australia (Harden 1991). Plants have a lignotuber and are able to resprout after fire. Fruit set is low (<10%), and seeds are predominantly outcrossed (Vaughton and Carthew 1993). Seed mass varies fivefold within populations, and up to 55% of the variation occurs within individual plants (Vaughton and Ramsey 1997). Fruits are serotinous, and high temperatures during fires stimulate seed release. Most seedling recruitment occurs in high light conditions in the first few months after fire.

Seeds were collected from *B. cunninghamii* plants at Gibraltar Range National Park (GR; lat. 29°36'S, long. 152°16'E, 1000 m a.s.l.) and New England National Park (NE; lat. 30°30'S, long. 152°27'E, 1300 m a.s.l.). The sites are ca. 100 km apart and experience similar climatic and rainfall regimes: summers are warm, winters are cold, and rainfall occurs throughout the year. Soils at GR are derived from granite, while those at NE are from trachyte; both are shallow, acidic, and nutrient poor. Mean seed mass does not differ significantly between the populations (mean ± SE; GR: 12.74 ± 0.08 mg; NE: 12.19 ± 0.09 mg; Vaughton and Ramsey 1997).

Differences in soil fertility between the study populations were examined by collecting four soil samples (5 cm diameter and 10 cm depth) during the summer from patches between shrubs in each population. Each sample consisted of two subsamples that were collected from within 2 m of each other and mixed. Samples were dried at 25°C for 3 d and sieved through 2 mm mesh. Soil analyses were conducted by Lanfax Laboratories, Armidale, New South Wales, and methods followed Rayment and Higginson (1992). Samples were analyzed for total nitrogen (N, method 7A1), available N (NH_4^+ and NO_3^-, method 7C1), available phosphorus (P, method 9E), and potassium (K, method 15D3). We used multivariate analysis of variance (MANOVA) to test for differences in these nutrients between populations. We restricted our analysis to these three macronutrients because they often limit plant growth.

Seed Nutrient Analyses

For seed nutrient analyses, we individually weighed 30 seeds with intact seed coats from bulk seed collections from each population containing at least 20 maternal plants (total $n = 60$). The same seeds were used for the P and K analyses, and different seeds were used for the N analyses. Seed N content was measured by steam distillation using a Tecator Kjeltec 1035 nitrogen analyzer. Seed P and K contents were determined by inductively coupled plasma-optical emission spectrometry. Methods followed Vaughton and Ramsey (1998). We assessed the effects of seed mass and population on seed nutrient content with analyses of covariance (ANCOVA), with seed mass

as a covariate. All interactions between seed mass and population were significant, and we calculated separate regressions for each population. We assessed whether slopes exceeded 1 ($b > 1$) using one-tailed t-tests (Sokal and Rohlf 1995).

Relationships between Seed Mass and Seedling Traits

To assess the effect of seed mass on seedling traits, we used a glasshouse common garden experiment with 160 seeds from each population (total $n = 320$). Seeds were selected from bulk seed collections containing at least 20 maternal plants and were placed on the soil surface of plastic tubes (200 cm³) containing soil comprised of equal parts loam, sand, and peat. Tubes were placed in trays on a laboratory bench (ca. 22°C) and kept moist. Seeds were monitored daily for 30 d to assess germination. A seed was considered germinated when the radicle emerged from the testa. To control for possible effects of seedling age on seedling traits, only those seedlings that germinated within the same 5-d period were used (final $n = 134$ and 147 seedlings population⁻¹). After 3 wk, seedlings were moved to a glasshouse, were watered every second day, and were fertilized every 4 wk with a half-strength solution of liquid fertilizer with trace elements (23% N, 4% P, 18% K; Aquasol, Hortico, Laverton North, Australia). Trays were moved each week to avoid bench position effects. Seedlings were harvested when they were ca. 12 wk old; under natural field conditions most mortality occurs before this age. All cotyledons were green and were measured with an area meter (Delta-T Devices, Burwell, Cambridgeshire, U.K.). The number of fully expanded leaves was counted. Shoots (cotyledons + leaves + stem) and roots of each seedling were harvested separately, dried at 65°C for 7 d, and weighed. Root : shoot ratios using dry masses were calculated. Relative growth rates (RGR) were calculated as (ln seedling mass − ln seed mass)/weeks of growth.

We assessed whether the probability of seed germination and seedling survival was dependent on seed mass and population using logit models and analyses of deviance (Collett 1991). Three seed mass categories were used: small (<11.0 mg), medium (11.0–13.5 mg), and large (>13.5 mg). We also assessed the effects of seed mass and population on cotyledon area, number of leaves, shoot mass, root mass, total seedling mass, root : shoot ratios, and RGR using ANCOVAs with seed mass as a covariate. In addition, we calculated regressions between seed mass and seedling traits for both populations.

Effects of Low and High Nutrient Supply

We determined the effect of seed mass on seedling growth under high and low nutrient conditions using 128 seeds selected from bulk seed collections from each population (total $n = 256$). Seeds were placed on the soil surface of plastic tubes (200 cm³) containing washed sand. Tubes were placed in trays on a laboratory bench (22°C) and kept moist. When the majority of seeds had germinated, trays were moved to a glasshouse, watered every 2 d with tap water, and moved weekly to avoid position effects. To control for the effect of age on growth parameters, we used only those seedlings that germinated within the same 5-d period (final $n = 58$–62 seedlings treatment⁻¹ population⁻¹).

When seedlings were ca. 3 wk old, tubes were randomly assigned to either high or low nutrient treatments. High nu-

Table 1

Concentration of Macronutrients in Soils (mg kg⁻¹) from the Populations (Gibraltar Range [GR] vs. New England [NE])

Macronutrients	GR	NE
Nitrogen (total)	850.0 ± 86.6	2800.0 ± 445.0
Nitrogen (available)	17.3 ± 4.5	30.7 ± 4.5
Phosphorus (available)	1.3 ± 0.1	2.1 ± 0.3
Potassium	72.9 ± 12.5	206.2 ± 41.0

Note. Data are means (± SE) from four replicates at each site.

trient conditions were created by placing eight granules of controlled-release fertilizer containing trace elements (17% N, 1.6% P, 8.7% K; Osmocote Plus, Scotts (Europe) Heerlen, The Netherlands) on the soil surface and by watering seedlings every 14 d with a half-strength solution of Aquasol. In the low nutrient treatment, seedlings were not fertilized. In both treatments, we applied a full-strength solution of chelated iron (Hortico, Australia) every 4 wk to prevent chlorosis due to iron deficiency. Seedlings were harvested when they were ca. 16 wk old. Shoots (cotyledons, leaves, and stems) and roots were harvested separately, dried at 65°C for 7 d, and weighed.

We assessed the effects of seed mass, nutrients, and population on root and shoot masses using ANCOVAs, with seed mass as a covariate. Regressions between seed mass and seedling traits were calculated for nutrient treatments for each population. We assessed whether slopes exceeded 1 ($b > 1$) or were less than 1 ($b < 1$), using one-tailed t-tests (Sokal and Rohlf 1995).

Analyses

Seed mass and other variables were log-transformed to normalize data distributions and stabilize error term variances. For regressions, such transformations linearize allometric relationships. In our study, seed mass (x) and seed or seedling traits (y) are quantified as power functions of the form $\log y = \log a + b \log x$, where a is the y-intercept and b is the regression coefficient (slope), which estimates the exponent of the untransformed allometric relationship, $y = ax^b$. In such models, $b = 1$ indicates a linear increase in y with increasing x, and if $b > 1$, y increases more rapidly than x; if $b < 1$, y increases less rapidly. In our study, both x- and y-variables were measured with error, and Model II regressions are appropriate. However, to be consistent with our use of ANCOVAs and to allow us to examine allometric relationships, we use Model I (least squares) regressions (Sokal and Rohlf 1995).

All analyses were initially run with full models, and when interactions were not significant, they were pooled with the error. Adjusted means (± SE) in relation to seed mass are given (Sokal and Rohlf 1995).

Results

Soil Nutrients

Soils at GR contained significantly lower concentrations of N, P, and K than those at NE (table 1; MANOVA, $F_{4,3} =$

10.71, $P = 0.04$). Concentrations of individual nutrients were 38%–70% less at GR than NE.

Seed Nutrient Analyses

The N, P, and K content of seeds increased significantly with increasing seed mass (fig. 1; table 2). Seed mass explained ≥97% of the variation in the N and P content of seeds but explained less of the K content (ca. 86%). For all nutrients, there were significant differences between populations, and the
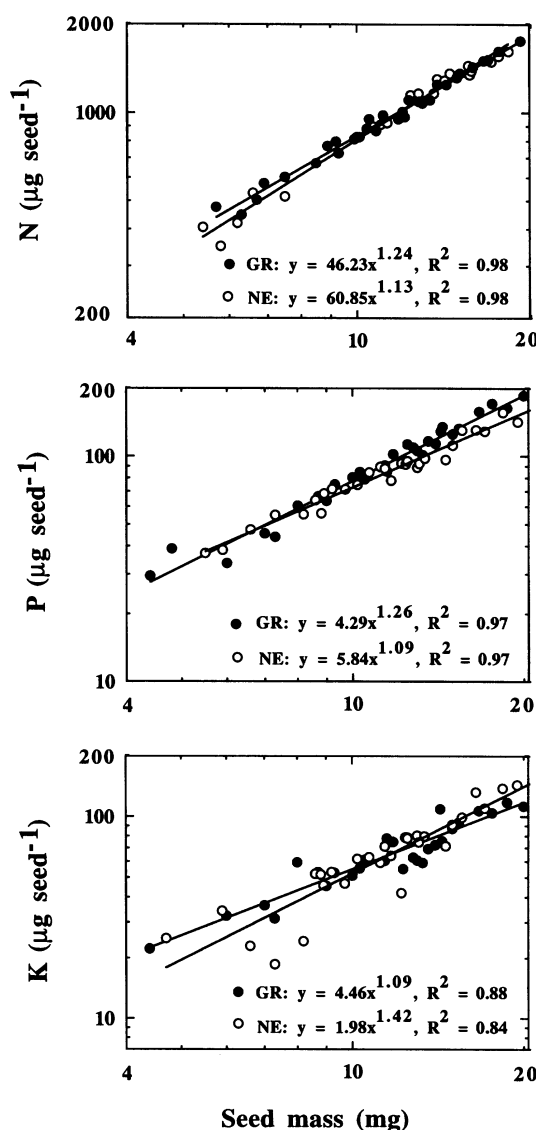
Fig. 1 Relationships between seed mass (mg) and the nitrogen (N), phosphorus (P), and potassium (K) content of seeds (μg seed⁻¹) for the Gibraltar Range (*GR*) and the New England (*NE*) populations of *Banksia cunninghamii*. Data are plotted on a log-log scale. For all nutrients, slopes differed significantly between populations. See table 1 for analyses.

Table 2

ANCOVAs of the Effects of Seed Mass (Covariate) and Population (Gibraltar Range [GR] vs.
New England [NE]) on the Nitrogen, Phosphorus, and Potassium Contents of Seeds

Nutrient	Seed mass	Population	Population × seed mass	GR	NE
Nitrogen	3250.74***	7.19*	6.59*	1080.8 ± 26.5	1054.3 ± 41.3
Phosphorus	1721.69***	4.90*	8.11**	106.3 ± 2.9	91.0 ± 4.3
Potassium	308.91***	5.37*	5.36**	71.8 ± 3.1	68.9 ± 1.3

Note. Data for GR and NE (μg seed^{-1}) are adjusted means ($\pm$ SE). F ratios for seed mass, population, and population × seed mass are given (all df = 1, 56). Regressions for seed mass are given in figure 1.

* $P < 0.05$.
** $P < 0.01$.
*** $P < 0.001$.

N, P, and K contents of seeds were 3%, 17%, and 4% greater for GR than NE seeds (table 2). Relationships between seed mass and seed nutrients differed between populations, as indicated by significant population × seed mass interactions (table 2).

In both populations, the slopes of the relationships between seed mass and the N and P contents of seeds were significantly greater than 1, indicating that nutrient content increased at a greater rate in larger seeds (fig. 1; all b = 1.09, all $t_{28} > 2.60$, $P < 0.01$). The increase in nutrients with seed mass, however, was greater at GR than at NE, as indicated by the greater slopes (b ± SE; N: 1.24 ± 0.03 vs. 1.13 ± 0.03; P: 1.26 ± 0.04 vs. 1.09 ± 0.04; fig. 1; table 2). For K, the slope of the relationship between seed mass and nutrient content was significantly greater than 1 for NE but not GR seeds (b ± SE; NE: 1.41 ± 0.12, t_{28} = 3.42, P = 0.001; GR: 1.09 ± 0.08, t_{28} = 1.13, P = 0.135; fig.1; table 2).

Relationships between Seed Mass and Seedling Traits

Of 320 seeds used in the common garden experiment, 93.4% germinated. Seed germination was independent of population and seed mass (population, χ^2 = 1.95, df = 1, P = 0.163; seed mass, χ^2 = 0.627, df = 2, P = 0.731; population × seed mass interaction, χ^2 = 4.66, df = 2, P = 0.097). Survival of the 299 germinated seeds was dependent on population (χ^2 = 4.20, df = 1, P = 0.040) but not seed mass (χ^2 = 0.70, df = 2, P = 0.704). For GR, 97% of seedlings survived compared to 91% of NE seedlings. The population × seed mass interaction was not significant (χ^2 = 0.55, df = 2, P = 0.760).

Seedling size was positively related to seed mass in both populations (all $b > 0$; tables 3, 4). Seedlings derived from larger seeds had more leaves and larger shoots, roots, and cotyledon areas than seedlings derived from smaller seeds. In both populations, RGR was unrelated to seed mass (table 3). Populations differed significantly for the mean seedling traits (table 3). GR seedlings had larger roots, root : shoot ratios, and cotyledons than NE seedlings. NE seedlings had more leaves, larger shoots, and greater RGR and total mass.

Only the slopes of the relationships between seed mass, root mass, and root : shoot ratio differed between populations (tables 3, 4). For root mass, the slope for GR was significantly less than for NE (b ± SE; 0.65 ± 0.09 vs. 1.00 ± 0.12). For

root : shoot ratio, the slope for GR was weakly negative (-0.26 ± 0.07), but the slope for NE did not differ from 0 (-0.02 ± 0.08).

Effects of Low and High Nutrient Supply

All seedlings survived for the duration of the experiment. Shoot and root masses were positively related to seed mass (all $b > 0$; table 5; fig. 2). Nutrient deprivation significantly reduced shoot mass (table 5; fig. 2). For GR, adjusted mean masses under high versus low nutrients were 139.4 ± 8.4 mg versus 117.1 ± 1.5 mg. Corresponding values for NE were 148.5 ± 7.1 mg versus 105.2 ± 4.1 mg. For shoot mass, the interaction between nutrients and population was significant, and nutrient deprivation had a greater effect on NE than GR seedlings. Compared to high nutrients, shoot mass of NE seedlings was reduced by 29% under low nutrients compared to only 16% for GR seedlings. The significant nutrient × seed mass interaction indicated that the slope of the regression between seed mass and shoot mass was greater under low nutrients than under high nutrients (fig. 2; table 5). For GR seedlings, the slope was significantly greater than 1 under low nutrients, indicating a greater than proportional increase in shoot mass with increasing seed mass (b ± SE; low: 1.29 ± 0.08, t_{59} = 3.55, P = 0.000; fig. 2). Under high nutrients, the slope was marginally less than 1 (0.84 ± 0.10, t_{60} = 1.67, P = 0.051; fig. 2). For NE seedlings, the slopes did not differ from 1 under both low and high nutrients (low: 0.99 ± 0.08, t_{56} = 0.089, P = 0.465; high: 0.91 ± 0.07, t_{58} = 1.21, P = 0.116; fig. 2). The population × seed mass interaction was not significant, indicating that the effect of seed mass on shoot mass pooled over nutrient treatments was similar in both populations (table 5).

Nutrient deprivation also significantly reduced root mass (fig. 2; table 5). For GR, adjusted mean masses under high versus low nutrients were 59.4 ± 4.7 mg versus 26.7 ± 0.5 mg. Corresponding values for NE were 55.5 ± 2.6 mg versus 24.6 ± 1.5 mg. The nutrient × population interaction was not significant (table 5). Root mass of GR and NE seedlings was reduced by 55% and 56%, respectively, under high compared to low nutrients. The nutrient × seed mass interaction was marginally significant. Similar to shoot mass, the slope of the regression between root mass and seed mass tended to be greater under low nutrients than high nutrients (fig. 2; table 5). For GR seedlings, there was a greater than proportional

Table 3

ANCOVAs of the Effects of Seed Mass (Covariate) and Population (Gibraltar Range [GR] vs. New England [NE]) on Seedling Growth under Common Garden Conditions

Parameter	Seed mass	Population	Population × seed mass	GR		NE	
Cotyledon area (cm²)	676.05***	18.20***	ns	0.67	± 0.014	0.61	± 0.013
Number of leaves	108.16***	182.51***	ns	5.88	± 0.07	7.35	± 0.10
Shoot mass (mg)	383.24***	9.84**	ns	53.49	± 1.28	57.24	± 1.50
Root mass (mg)	126.68***	6.35*	5.81*	14.58	± 0.54	14.26	± 0.37
Total mass (mg)	346.73***	4.57*	ns	68.06	± 1.57	71.50	± 1.93
Root : shoot ratio	6.27*	7.94**	5.84*	0.28	± 0.006	0.25	± 0.006
RGR (mg mg⁻¹ wk⁻¹)	1.42 ns	4.51*	ns	0.168	± 0.002	0.175	± 0.001

Note. Data for GR and NE are adjusted means ($\pm$ SE). F ratios for seed mass, population, and population × seed mass are given. Nonsignificant interactions (ns = $P > 0.05$) were pooled with the error. For these analyses, df = 1,278, and for other analyses, df = 1,277. Seed mass regressions are given in table 4.

* $P < 0.05$.
** $P < 0.01$.
*** $P < 0.001$.

increase in root mass with increasing seed mass under low nutrients but a marginally less than proportional increase under high nutrients ($b \pm$ SE; low: 1.18 ± 0.08, $t_{59} = 2.13$, $P = 0.018$; high: 0.76 ± 0.16, $t_{60} = 1.51$, $P = 0.068$). For NE seedlings, there was a marginally less than proportional increase under low nutrients and a proportional increase under high nutrients (low: 0.86 ± 0.08, $t_{56} = 1.71$, $P = 0.047$; high: 0.93 ± 0.12, $t_{58} = 0.56$, $P = 0.291$). The population × seed mass interaction was not significant (table 5).

Discussion

Our results for *Banksia cunninghamii* highlight the advantage of large seed mass for seedling establishment on nutrient-poor soils. There were strong positive relationships between seed mass and seed nutrient content. For N and P, nutrient content increased disproportionately with increasing seed mass ($b > 1.0$) in both populations, indicating that large seeds had a greater concentration of N and P (μg mg⁻¹ of seed tissue) than small seeds. For K, nutrient content increased disproportionately with increasing seed mass for NE seeds, whereas for GR seeds increases were proportional ($b \approx 1.0$). N and P are generally considered to be the main nutrients limiting plant growth in Australian sclerophyllous vegetation (Specht 1963; Beadle 1966; Lamont 1995; Specht and Specht 1999). For seedlings, P availability is considered especially important because of the insoluble nature of this nutrient, and the inability of seedlings to obtain sufficient quantities with their small root systems. Species of Proteaceae typically produce large seeds with high N and P concentrations (Kuo et al. 1982; Pate et al. 1986). Most seedling establishment occurs after fire when K and other cations are abundant in the soil. High concentrations of N and P in seeds complement the soil supplies of K, providing conducive conditions for seedling growth (Stock et al. 1990).

Relationships between seed mass and seed nutrients differed between the *B. cunninghamii* populations. For N and P, the increase in the nutrient content of seeds with increasing seed

mass was greater for GR than NE, indicating that large GR seeds had higher concentrations of N and P than large NE seeds. Conversely for K, large NE seeds had a higher nutrient concentration than large GR seeds. There were also differences between populations in the mean nutrient content of seeds. Despite a difference of only 4.5% in seed mass between the populations (Vaughton and Ramsey 1997), GR seeds contained on average 17% more P than NE seeds, indicating that seedlings from GR should be more tolerant of low P conditions than NE seedlings.

Table 4

Regressions between Seed Mass and Traits of Gibraltar Range (GR) and New England (NE) Seedlings Grown under Common Garden Conditions

Trait and population	Regression	R^2
Cotyledon area (cm²):		
GR	$y = -1.10x^{0.86}$	0.76
NE	$y = -1.13x^{0.86}$	0.72
Number of leaves:		
GR	$y = 0.40x^{0.35}$	0.33
NE	$y = 0.52x^{0.32}$	0.23
Shoot mass (mg):		
GR	$y = 0.74x^{0.91}$	0.58
NE	$y = 0.65x^{1.02}$	0.58
Root mass (mg):		
GR	$y = 0.45x^{0.65}$	0.26
NE	$y = 0.05x^{1.00}$	0.36
Total mass (mg):		
GR	$y = 0.91x^{0.85}$	0.55
NE	$y = 0.75x^{1.02}$	0.56
Root : shoot ratio:		
GR	$y = 0.29x^{-0.26}$	0.07
NE	ns	...
RGR (mg mg⁻¹ wk⁻¹):		
GR	ns	...
NE	ns	...

Note. All regressions were significantly greater than 0 ($P < 0.05$). Analyses comparing slopes and intercepts are given in table 3. $n = 147$ and 133 at GR and NE, respectively; ns = $P > 0.05$.

Table 5

ANCOVAs of the Effects of Seed Mass (Covariate), Nutrients (High vs. Low), and Population (GR vs. NE) on Shoot and Root Masses of Seedlings

Source of variation	Shoot mass $F_{1, 235}$	Root mass $F_{1, 236}$
Seed mass	577.71***	242.41***
Nutrients	20.67***	19.63***
Population	0.75 ns	0.61 ns
Nutrients × population	5.85**	ns
Nutrients × seed mass	10.44**	2.63[†]
Population × seed mass	ns	ns

Note. Seed mass regressions are given in figure 2. Nonsignificant interactions were pooled with the error; ns = $P > 0.05$.
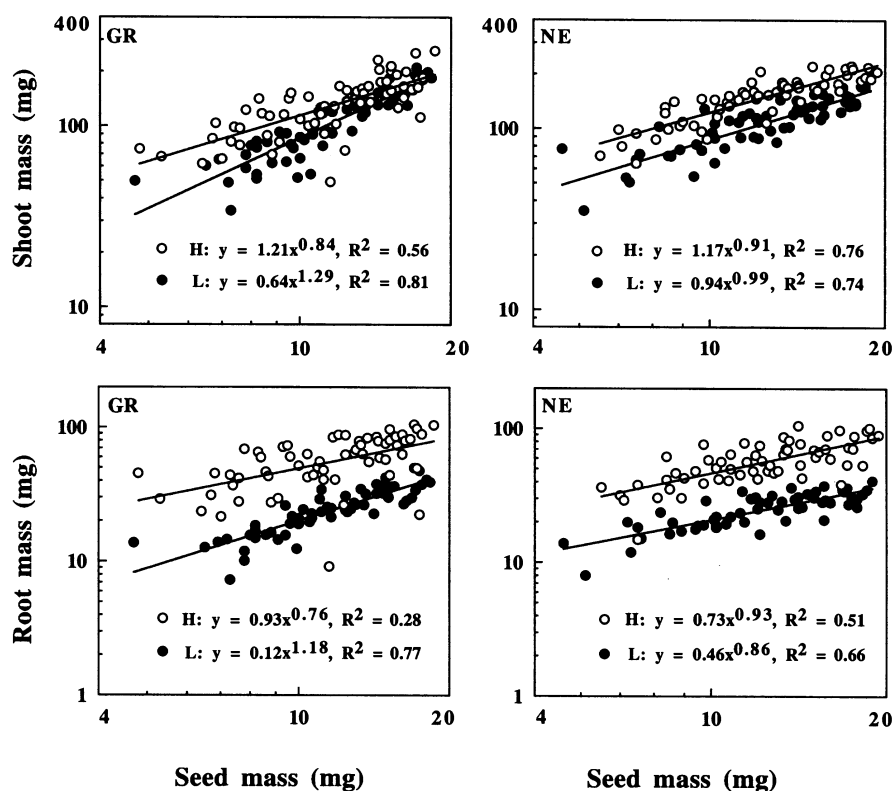
[†] $P = 0.10$.

** $P < 0.01$.

*** $P \leq 0.001$.

The size of B. cunninghamii seedlings was positively related to seed size, which concurs with findings for other species (Wulff 1986; Moegenburg 1996; Bonfil 1998; Vaughton and Ramsey 1998; Castro 1999; Eriksson 1999). The general effect of seed size on seedling size may reflect either increased carbon or increased nutrient reserves in large seeds. In our study, relationships between seed mass and seedling mass were generally stronger under low than high nutrients, indicating that the higher nutrient content of large seeds probably enabled the production of large seedlings. In other Proteaceae, nutrients are translocated from the cotyledons into the seedlings primarily during the first few months of growth, thereby rendering seedlings less dependent on soil nutrients during this period (Milberg and Lamont 1997; Milberg et al. 1998). The decreased growth of B. cunninghamii seedlings under low nutrients compared to high nutrients in both populations contrasts with other studies of Banksia species that have found no effects of increased nutrients on growth or adverse effects when nutrients were added in excess (Specht 1963; Groves and Keraitis 1976; Siddiqi et al. 1976; Milberg et al. 1998). This difference may reflect differences in seed mass among species. Seeds of B. cunninghamii are small (mean mass, 12.1–12.7 mg) compared to other Banksia species that have been examined (mean seed mass of six species, 25.0–113.7 mg; Milberg et al. 1998) and probably have smaller nutrient reserves.

In contrast to seedling size, there was little or no effect of seed mass on seed germination, seedling survival, root : shoot ratio, or RGR. Similar findings were reported for Banksia marginata (Vaughton and Ramsey 1998). High survival of B. cunninghamii seedlings under low nutrient conditions supports conclusions of Stock et al. (1990) that Proteaceae species are surprisingly tolerant of infertile soils. It is possible, however, that differences in seedling survival may have occurred under low nutrient conditions if our experiments had been conducted for longer. The duration of our study also potentially influenced root : shoot ratios, which were low compared to reports for other Proteaceae. In other species, proteoid root development occurs in 80–200-d-old seedlings (Stock et al. 1990). We harvested seedlings after 84–112 d, and root : shoot ratios would be expected to increase as proteoid roots were produced.

The common garden experiment revealed probable genetic differences in seedling allocation patterns between populations that would potentially affect establishment on nutrient-poor soils. Cotyledon area was greater for GR seedlings than NE seedlings, which may result in more photosynthates for nutrient translocation and initial plant growth before true leaves are produced (Milberg and Lamont 1997). GR seedlings also had larger roots and greater relative allocation to roots, whereas NE seedlings had larger shoots and increased RGR. Larger root systems would provide greater exposure to soil nutrients and moisture, while increased relative allocation to roots would allow a root system to develop more rapidly. Similarly, reduced shoot mass and the consequent reduction in RGR would conserve seed nutrients and, therefore, delay the need for seedlings to access soil nutrients. Such traits increase fitness under nutrient-poor conditions and are common in plants from infertile habitats (Stock et al. 1990; Milberg et al. 1998; Lloret et al. 1999; Wright and Westoby 1999; but see Marañón and Grubb 1993).

Consistent with the observed differences in seed nutrient content and seedling allocation patterns between the populations, GR seedlings were more tolerant of nutrient deprivation than NE seedlings. Under low nutrients, shoot mass of NE seedlings was reduced by 29% compared to only 16% for GR seedlings. This difference between populations under low nutrients may reflect the greater P content of GR seeds, although differences in seedling allocation patterns may have also played a role. Seeds with larger nutrient stores would be expected to exhaust their reserves more slowly after germination under low nutrients than seeds with smaller reserves, making them more tolerant of nutrient scarcity (Milberg et al. 1998). For GR seedlings, shoot and root masses increased disproportionately with increasing seed mass under low nutrients ($b > 1.0$) but increased proportionally under high nutrients ($b \approx 1.0$). Thus, large seed mass minimized the effect of nutrient deprivation on the size of GR seedlings. In contrast, increases for NE seedlings were proportional ($b \approx 1.0$) under both nutrient regimes, indicating that the effect of nutrient deprivation was similar for all seed sizes.

The differences in seed nutrients and seedling allocation patterns between the study populations may be related to differences in habitat-specific selection pressures. We found that GR soils had lower concentrations of N, P, and K than NE soils. Differences in seed nutrients and seedling growth between the populations are consistent with selection for resource allocation patterns to ensure seedling establishment on lower-nutrient soils at GR compared to NE. In support of this suggestion, studies of other Proteaceae have reported larger seed mass and higher nutrient content in species occurring on nutrient-poor soil compared to closely related taxa at more fertile sites (Esler et al. 1989; Mustart and Cowling 1992). However, environmental factors besides soil fertility (Westoby et al. 1996) or random genetic effects could also contribute to the observed differences between populations in our study, and further work in other B. cunninghamii populations that differ in soil fertility is now required. Our experimental design also does not exclude the possibility of maternal effects, which may influence seed quality and seedling growth (Roach and Wulff 1987). Banksia cunninghamii takes 5–6 yr to flower, and it was not possible to generate seeds under common garden con-

The GR shoot mass panel shows: O H: $y = 1.21x^{0.84}$, $R^2 = 0.56$; ● L: $y = 0.64x^{1.29}$, $R^2 = 0.81$.

The NE shoot mass panel shows: O H: $y = 1.17x^{0.91}$, $R^2 = 0.76$; ● L: $y = 0.94x^{0.99}$, $R^2 = 0.74$.

The GR root mass panel shows: O H: $y = 0.93x^{0.76}$, $R^2 = 0.28$; ● L: $y = 0.12x^{1.18}$, $R^2 = 0.77$.

The NE root mass panel shows: O H: $y = 0.73x^{0.93}$, $R^2 = 0.51$; ● L: $y = 0.46x^{0.86}$, $R^2 = 0.66$.

Fig. 2 Relationships between seed mass (mg) and shoot and root masses (mg) of seedlings grown under low (*L*) and high (*H*) nutrients for the Gibraltar Range (*GR*) and the New England (*NE*) populations of *Banksia cunninghamii*. Data are plotted on a log-log scale. See table 5 for analyses.

ditions, which would be necessary to discount such maternal effects.

Although large seed mass and high seed nutrient content should increase the likelihood of seedling establishment on infertile soils at GR, a potential cost of this strategy in a nutrient-poor environment is the production of fewer seeds per plant. We did not compare seed production in the GR and NE populations, but we predict that fewer seeds would be produced at GR, consistent with the greater cost of seed provisioning in this population. Compared to other woody plants, fruit : flower ratios are low in all *B. cunninghamii* populations, which is, in part, due to the generally infertile soils on which they occur (Vaughton 1991). Selection for the ability to es-

tablish on infertile soils could be an important determinant of trade-offs between seed size and number in *B. cunninghamii*, as has been suggested for other Proteaceae (Esler et al. 1989; Stock et al. 1990; Mustart and Cowling 1992).

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