DEMOGRAPHY AND INDIVIDUAL GROWTH OF TWO INTRODUCED *BROMUS* SPECIES

By

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DEMOGRAPHY AND INDIVIDUAL GROWTH OF TWO

INTRODUCED *BROMUS* SPECIES

Abstract

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Comparative studies of congeners have been identified as valuable tools to understand why some introduced species become invasive and others do not. I compared individual and population growth characteristics of two annual grasses, invasive *Bromus tectorum* and naturalized *B. briziformis*, in greenhouse and field experiments. Smaller pot size, lower water availability and lower rates of nutrient addition all reduced maximum plant size, decreased growth rates, and delayed the period of most rapid plant growth of both species in greenhouse studies. Ontogenetically controlled analyses of biomass allocation revealed that plants had higher root to shoot ratios in less favorable growing conditions (limited soil volume or low nutrient addition), compared to more favorable growing conditions. *B. tectorum* biomass allocation was more root-heavy compared to *B. briziformis* biomass allocation in all treatments. Neither species adjusted its allocation to roots in response to water availability but greater root allocation by *B. tectorum* compared to *B. briziformis* throughout development may increase the drought tolerance of that species.

I used logistic and linear regression analysis of matrix population-model parameters to examine the response of *B. tectorum* and *B. briziformis* population growth rates to individual plant nitrogen and water status at two sites. *B. tectorum* population growth rates were 2 to 3.5 times greater than *B. briziformis* growth rates across water and nutrient treatments, and across study sites. *B. tectorum* plants were larger than *B. briziformis* plants, had lower leaf nitrogen content and produced nearly three times as many seeds as *B. briziformis* plants. Fertilized plants were larger and produced more seeds than unfertilized plants, but only at one site. Watered plants were more depleted in ^{13}C than unwatered plants only at one site. However, fertilizer and water treatment effects on plant size, seed production, or $\delta^{13}C$ did not translate into differences in population growth rates. Population growth rates differed only by species and sites.

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INTRODUCTION

Invasive species are an important component of global environmental change (Vitousek et al. 1996), costing an estimated $120 billion in the United States each year, (Pimental et al. 2005) disrupting ecosystem services and ranking second among threats to endangered species. There is considerable interest in developing the ability to predict which species might become invasive so that their impacts can be mitigated or avoided (Mack 1996).

Investigations of invasive species may be conducted at the level of species physiology (e.g. Hierro and Callaway 2003, Maricle and Lee 2006), anatomy (e.g. Maricle and Lee 2002), whole plant performance (e.g. D'Antonio 1993, Gentle and Duggin 1997), genotype performance (e.g. Novak et al. 1993, Saltonstall 2002, Siemann and Rogers 2003), or population performance (e.g. McEvoy and Coombs 1999, Koop and Horvitz 2005, Lambrecht-McDowell and Radosevich 2005). Although studies at all levels of biological organization are useful, ecological effects can be scale dependent (Stohlgren et al. 1999 , DeLucia et al. 2001, Knight and Reich 2005, Alvarez-Cobelas and Cirujano 2007), so effects that are observed at one level of biological organization might not be important at other levels of biological organization. Understanding the causes of harmful species invasions at any level of biological organization may suggest management techniques to reduce their prevalence.

Phenotypic plasticity, the ability of an organism to alter the expression of its genotype depending on environmental conditions, allows plants to survive in environments that vary in time and space (Rice and Mack 1991, Meyer et al. 1997). Plastic responses have also been implicated as traits that confer invasive potential on introduced species (Weber and D'Antonio 1999, Gerlach and Rice 2003). If plants are plastic with respect to biomass allocation, optimal

partitioning theory predicts that carbon should be allocated to above and below-ground structures so that all resources are equally limiting to plant growth (Chapin et al. 1986).

In this dissertation I compare two introduced annual grasses, *Bromus tectorum* and *B. briziformis* at three levels of biological organization. Chapter one investigates differences in the plasticity of individual plant allocation of biomass to roots and shoots. In chapter two I use nonlinear curve fitting to compare the responses of whole-plant growth to differences in soil water

and soil volume. And in chapter three I use matrix population models to examine population-level responses to fertilizer and water additions to planted individuals.

In chapter one I compare root to shoot allocation of *B. briziformis* and *B. tectorum* in response to experimental water, nutrient or soil volume treatments. Root to shoot ratios have been shown to increase (Aronson et al. 1992, Heschel et al. 2004, chapter 1) or remain unchanged (McConnaughay and Coleman 1999) in response to low water or low nutrient treatments, depending on the species tested and the experimental protocols (McConnaughay and Coleman 1999). Plant responses to restricted rooting volume differ between studies, and even within studies (McConnaughay and Bazzaz 1991 and references therein, chapter 1). I observed greater[*] root to shoot ratios in smaller pots compared to larger pots, but the reasons were unclear. Both species adjusted their allocation patterns in response to our experimental treatments, but *B. tectorum* had larger root to shoot ratios compared to *B. briziformis* across treatments, and the two species were not different in the degree to which biomass allocation changed in response to water, nutrients or soil volume.

[*] Throughout this dissertation, any report of a species or treatment difference implies a statistical test with $p < 0.05$. Conversely, 'no difference' implies $p \geq 0.05$.

Measures of whole-plant growth such as leaf area or biomass are often used to gauge plant performance in studies comparing two or more species. Generally, this follows the rationale that larger plants are better at capturing resources and should produce more offspring than smaller plants (but see Neytcheva and Aarssen 2008), suggesting that they are more competitive or fit than smaller plants. The use of plant size as a measure of plant performance becomes problematic in greenhouse studies where pot size may have a greater affect on plant size than treatment effects (McConnaughay and Bazzaz 1991, McConnaughay et al. 1993, Ronchi et al. 2006). As an alternative to plant size alone, some authors suggest multiple harvests and an analysis of the whole plant growth curve. The major advantage of a curve-fitting approach is that the whole pattern of plant growth is captured by a few model parameters that can be interpreted in terms of the underlying species biology (Zhao et al. 2005).

In chapter two I used the Richards growth model (Causton and Venus 1981), which describes a sigmoid growth curve, to evaluate the effects of water availability on growth of *B. tectorum* and *B. briziformis*. I found no differences in growth curves between the two species at any water availability or pot size. Larger pots produced larger plants, but watering effects were inconsistent across pot sizes. The analyses in this chapter may have benefited from a harvest schedule that concentrated data during the period of most rapid plant growth (Causton and Venus 1981).

Population-level comparisons between invasive and non-invasive congeners are appealing because invasion is ultimately a population growth rate at a problematic level (Parker 2000), and the literature is replete with examples of such studies (e.g. Schierenbeck et al. 1995, Radford and Cousens 2000, Smith and Knapp 2001, Kercher and Zedler 2004)

Population demographic studies have been used to investigate life-history traits that are associated with invasiveness (e.g. Radford and Cousens 2000, Gerlach and Rice 2003, Koop and Horvitz 2005, Lambrecht-McDowell and Radosevich 2005). Life table response experiments (Caswell 1989) have been used to examine the effects of experimental treatments on population dynamics (e.g. Walls et al. 1991, Davis et al. 2004, Williams and Crone 2006), but more commonly, demographic parameters are reported along with physiological data (e.g. Donovan and Ehleringer 1994, Allen 1998, Erneberg 1999, Goodwin et al. 1999, Andersson et al. 2002). The absence of a direct statistical linkage between physiological data and population demographic parameters makes it difficult to develop a predictive framework that links these two levels of biological organization.

In chapter three I examine the population growth responses of *B. briziformis* and *B. tectorum* to supplemental fertilizer and water at two study sites. I use logistic and linear regression to describe the relationships between population model parameters (plant growth, survival and fecundity) and proxy measures of plant gas exchange ($\delta^{13}C$) and nitrogen status (tissue %N). I subsequently used regression relationships to generate simulated populations with nitrogen and water status adjusted from field observations. This approach allows me to examine the effects of individual plant water and nitrogen status on population growth rates.

The empirical population growth rates showed large differences in population growth rates between the two species, with *B. tectorum* populations increasing faster, compared to *B. briziformis* populations, as expected. There was also a substantial difference in population growth rates between the study sites, possibly due to a difference of two weeks in planting dates. Supplemental water and nitrogen produced measurable effects in our measures of plant water and nitrogen status, and also in plant size, but did not translate into differences in population

growth rates among the treatments. Similarly, none of the simulated populations were responsive to changes in $\delta^{13}C$. Simulated populations responded to tissue %N only at one site, and the response was species-specific.

Across the three experiments I observed differences between the species in biomass allocation to roots versus shoots, and differences between the species in population growth rates. Whole-plant growth rates for both species were responsive to experimental treatments with no difference between the species responses to water, nutrients and soil volume. Plant-level effects that I observed in response to supplemental water and fertilizer did not translate to differences in population growth rates in field trials, and did not translate into consistent differences in population growth rates for simulated populations. *Bromus tectorum* allocated more biomass belowground compared to *B. briziformis*, which may enhance its ability to capture water during the onset of summer drought. *B. tectorum* population growth rates were higher compared to *B. briziformis* population growth rates.

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CHAPTER ONE

PLASTIC ROOT TO SHOOT ALLOCATION AND ONTOGENETIC DRIFT IN TWO ANNUAL *BROMUS* SPECIES IN RESPONSE TO SOIL RESOURCES.

1.1 ABSTRACT

We assessed biomass allocation in two introduced annual grasses, *Bromus tectorum* and *B. briziformis*, to test for differences in phenotypic plasticity and ontogenetic drift in response to soil volume, nutrient availability, and water availability. Greater pot size, higher water availability and higher rates of nutrient addition all increased the growth rates of *B. tectorum* and *B. briziformis*. Ontogenetic drift was evident in both species, but failing to account for drift resulted only in subtle effects on the analysis of biomass allocation. Plants allocated more biomass to roots in less favorable growing conditions (limited soil volume or low nutrient addition), compared to more favorable growing conditions. *B. tectorum* biomass allocation was more root-heavy than *B. briziformis* biomass allocation. Neither species adjusted its allocation to roots in response to water availability but greater root allocation by *B. tectorum* throughout development may increase drought tolerance.

1.2 INTRODUCTION

Phenotypic plasticity in growth and morphology allows plants to survive in environments that vary in time and space. An understanding of plasticity within populations therefore is beneficial to an understanding of the ecological role and responses of those populations. Plastic responses to the environment are known to be important in species interactions (Agrawal 2001, Callaway et al. 2003), survival in variable environments (Rice and Mack 1991, Meyer et al. 1997, Agrawal 2000), and invasive potential of introduced species (Weber and D'Antonio 1999, Gerlach and Rice 2003).

According to optimal partitioning theory, plants should allocate biomass to above-ground and below-ground tissues so that above ground and below-ground resources are equally limiting to plant growth (Chapin et al. 1986). Previous work has investigated allocational shifts in response to nutrient availability (Gedroc et al. 1996, Arredondo and Johnson 1999, McConnaughay and Coleman 1999, Bonifas et al. 2005), light intensity (McConnaughay and Coleman 1999), CO_2 concentration (Bernacchi et al. 2000), water availability (Aronson et al. 1992, McConnaughay and Coleman 1999, Heschel et al. 2004), and available soil volume (McConnaughay and Bazzaz 1991, McConnaughay et al. 1993). However the results have not been consistent among resource gradients, nor even within species. This may be due to different experimental protocols between studies investigating the same species (McConnaughay and Coleman 1999), or to differences between species or populations if phenotypic plasticity represents an evolutionary cost in stable environments (Via and Lande 1985).

Flawed experimental design has contributed to our lack of understanding of plant species' plastic responses to resource conditions: most species adjust allocation to above-ground and

below-ground tissues as a normal part of development, a phenomenon termed ontogenetic drift (McConnaughay and Coleman 1999). Annual forbs typically decrease root to shoot ratios as they grow and develop, particularly as they enter the reproductive phase of their life cycles (Hunt 1978). Therefore, if a series of experimental treatments produces plants with differing growth or developmental rates, the analysis of root to shoot ratios at fixed points in time may appear to indicate phenotypic plasticity simply because the individual plants were sampled at different stages of development (Bernacchi et al. 2000).

McConnaughay and Coleman (1999) present an improved approach to such experiments. They suggest analysis of biomass allocation in an allometric framework such that root to shoot ratios are regressed on total plant mass (e.g. figure 1.1), rather than on plant age. Ontogenetic drift occurs when the relative allocation to roots and shoots changes as the plant grows and is indicated by a slope that differs from zero. We will refer to this as the trajectory of root/shoot allocation. Phenotypic plasticity occurs when plants respond to different treatments by adjusting their allocation trajectories (different slopes), by adjusting the root to shoot ratios of young plants (different intercepts; we will refer to this as initial root to shoot ratio), or both.

Here we test for differences in phenotypic plasticity and ontogenetic drift between two introduced Eurasian annual grass congeners: *Bromus tectorum*, which is an abundant, highly invasive species and *B. briziformis*, which is introduced and naturalized, but less abundant and not invasive (Hulbert 1955). A high degree of phenotypic plasticity is frequently cited as a common trait among invasive species (Gedroc et al. 1996, Weber and D'Antonio 1999, Maurer and Zedler 2002) and congener studies have been identified as valuable tools to determine whether the attributes of a particular invader are important determinants of invasion success (Mack 1996). We conducted two greenhouse experiments to test whether *B. tectorum* and *B.*

briziformis exhibit phenotypic plasticity in response to soil volume, nutrient availability, or water availability, and if so whether they differ in their responses to these gradients in ways that might explain why *B. tectorum* is invasive and *B. briziformis* is merely naturalized. Our design also allowed us to assess the importance of ontogenetic drift as a factor that may obscure true phenotypic plasticity when the data are improperly analyzed.

1.3 METHODS

Seeds of *B. tectorum* and *B. briziformis* were collected from Elk Creek Falls Recreation Area in north-central Idaho (46˚ 44' W, 116˚ 10' N) within the *Pseudotsuga menziesii* / *Holodiscus discolor* habitat type (Daubenmire and Daubenmire 1968) in August 2005. Seeds were collected from several hundred individuals of each species, thoroughly mixed, and stored at room temperature until planting.

In the first experiment we investigated the effects of soil volume and water availability on the biomass allocation of our two study species. Individual *B. tectorum* or *B. briziformis* seeds were planted into 164 mL, 410 mL, or 2830 mL pots (hereafter called small, medium, and large; SC10 Cone-tainers, D25LW Deepots, or Tall One Treepots, respectively; Stuewe & Sons, Corvallis OR). Pots were filled with potting medium (Sun-Gro Sunshine Professional potting mix, Bellevue, WA) that had been mixed with 25%, 45% or 70% sand by volume. The soil texture treatments were imposed to effect a water-availability treatment, assuming coarsely textured soils are more drought-prone than finely textured soils (Smith and Smith 2001). We recognize that the coarser soils may also have had lower nutrient availability than the finer soils

(Smith et al. 2001). Pot racks were placed haphazardly on the benches and individual pots were assigned randomly to positions within these racks; the pots were not moved during the study.

Seeds were planted in pots on 6 March 2006 and maintained under natural lighting in a glasshouse. The glasshouse was maintained with temperatures of 4 - 8˚C at the beginning of the study then gradually increased to 12 - 16˚C over the course of the study to encourage normal phenologic development. Supplemental fertilizer was never added to the potting medium.

All plants within each pot size were watered to excess with tap water when approximately 50% of the plants in that treatment appeared to be wilting. One pot per treatment was randomly selected for the purpose of monitoring soil water. Pots for monitoring soil water were weighed immediately prior to watering and one to two hours after watering (after excess water had drained) to determine water loss between watering events. Total water added to each pot size-soil mixture was calculated based on weight of water added; treatments with more sand generally received less water than those with less sand (table 1.1).

In studies designed to examine patterns of biomass partitioning, it is more effective to harvest fewer plants more frequently than to conduct mass harvests infrequently (McConnaughay and Coleman 1999). Therefore we harvested one randomly-selected individual from each treatment on days 25, 30, 34, 38, 43, 54, 57, 65, 72, 77, 85, 94, 100 and 113 after planting. The final eight harvests required two or three days to complete.

In a second experiment we tested the effects of water availability and nutrient amendments on biomass partitioning to roots and shoots. We planted *B. tectorum* and *B. briziformis* into 410 mL pots with potting medium that had been mixed with 25%, 45% or 70% sand by volume, again to impose a water-availability treatment. Supplemental nutrients (Peter's Professional 20-20-20 mix; Marysville, OH) were added weekly as 100 mL of tap water with

fertilizer mix diluted to provide 0, 25 or 250 ppm nitrogen. Plants were watered as in the soil volume study above (table 1.1).

The nutrient experiment was planted on 7 March, 2006 and maintained in a glass house with day time temperatures of 22 – 25˚C and night time temperatures of 15 – 19˚C. Supplemental lighting provided a 12-hour photoperiod. Pots were randomly assigned to bench positions but were not moved during the course of the study.

Harvests were initiated on days 13, 15, 18, 20, 25, 32, 36, 41, 50, and 59 after planting. The final three harvests required three days to complete.

Procedures were similar for all harvests in both experiments. Shoot material was clipped at the soil surface and separated into live shoot and dead shoot material; only live shoot material was used for analyses. Soil was washed from the roots with running water over a 1.4 mm screen and captured on a 150 micron screen. The material in both screens was then hand-sorted to retrieve root material. All plant components were further washed in a sonicator with detergent for 30 minutes, rinsed with tap water and dried at 80˚C for at least 72 hours before weighing.

Data were analyzed with a multiple-slopes analysis of covariance (MS-ANCOVA, Proc GLM, SAS 9.1; SAS Institute, Cary, NC). All analyses included species, sand content of the soil, and pot size or nutrient level with interactions as class variables. Time was considered a continuous variable and plant total mass as the dependent variable to test for differences in growth rates among treatments and species. We used root to shoot ratio as the dependent variable and plant total mass as the independent variable to test for ontogenetic drift and phenotypic plasticity in response to our experimental conditions. We repeated the analysis using time as the independent variable to examine the impact of ontogenetic drift on our findings. Continuous variables were natural log-transformed to meet assumptions of normality, resulting

in covariates with negative values (e.g. figure 1.1a). We adjusted values of the covariates by a constant so that all values were zero or positive (e.g. figure 1.1b); this allowed us to interpret ANCOVA intercepts as the root to shoot ratio of small plants; we refer to these as initial root to shoot ratios. We report the highest-order significant interaction(s) for each analysis. Multiple comparisons between slopes and intercepts were made using a Bonferroni-Holm adjustment (Wright 1992) to preserve an experiment-wise error rate of 0.05. We report only those differences for comparisons with $p < 0.05$ and take $p > 0.05$ to indicate no difference between treatments. Plots of MS-ANCOVA results were difficult to read due to the large number of treatments (e.g. figure 1.1); therefore we present the results in histogram plots showing fitted intercepts and slopes from the ANCOVA models (figures 1.3, 1.4, 1.5, 1.6).

1.4 RESULTS

We observed phenotypic plasticity in both species: greater pot size, greater water availability and higher rates of nutrient addition all increased the growth rates of *B. tectorum* and *B. briziformis*. In both experiments, root to shoot ratio trajectories generally increased in less favorable growing conditions (limited soil volume or low nutrient addition), compared to more favorable growing conditions. In the soil volume study *B. tectorum* root to shoot allocation trajectories were more root-heavy than *B. briziformis* trajectories. Ontogenetic drift was detected in half of our treatments but failing to account for drift would not substantially have altered the conclusions drawn here.

Plant growth patterns were similar across treatments (figure 1.2). Plant growth rates were highest in large pots and lowest in the small pots (table 1.2, figure 1.3) and greater with 25% or

45% sand treatments compared to 70% sand treatments (table 1.2, figure 1.3). *B. briziformis* initial root to shoot ratio was greater in 70% sand compared to *B. briziformis* in 25% sand and greater than *B. tectorum* in any sand treatment (table 1.3, figure 1.4c). The allocation trajectory of *B. tectorum* favored roots more than that of *B. briziformis* across all treatments (table 1.3, figure 1.4d). Both species' allocation trajectories became more root-heavy as pot size decreased (table 1.3, figure 1.4d).

In the nutrient experiment, plant growth rates were highest in the high-nutrient treatment and the medium- and low-nutrient treatments did not differ from one another (table 1.4, figure 1.5). *B. tectorum* grew more slowly in the 70% sand potting medium than either species in the 25% sand potting medium (table 1.4, figure 1.5).

Initial root to shoot ratios for both species were greater in the high nutrient 70% sand treatment compared to the medium nutrient 70% sand treatment (table 1.5, figure 1.6c). Plants receiving medium or low nutrients in 70% sand had more root-heavy allocation trajectories than plants receiving high nutrients in 70% sand (table 1.5, figure 1.6d).

Ontogenetic drift was apparent (i.e. regression slopes differed significantly from zero) in six treatments in the soil volume study (figure 1.4d) and in nine treatments in the nutrient study (figure 1.6d). Failing to account for ontogenetic drift in the soil volume experiment produced results that indicated larger initial root to shoot ratios in *B. briziformis* compared to *B. tectorum* (table 1.6, figure 1.4a) and smaller initial root to shoot ratios in the low water treatment compared to the high water treatment (table 1.6, figure 1.4a). In contrast, the ontogenetically controlled analysis indicated a species by water interaction (see above; figure 1.4c). Failing to account for drift in the nutrient experiment detected no significant effects on initial root to shoot ratios (table 1.7, figure 1.6a), as opposed to the water by nutrient interactions indicated by the

ontogenetically controlled analysis (see above; figure 1.6c). Trajectories of root versus shoot allocation were similar between ontogenetically controlled and uncontrolled analysis in both experiments (figures 1.4b, d, 1.6b, d).

1.5 DISCUSSION

Our results demonstrate that 1) the root to shoot ratios of both *Bromus* species are plastic in response to all of the resource gradients we imposed, 2) nutrient effects on root to shoot ratio depended on water availability, and 3) failing to account for ontogenetic drift produced analyses that subtly obscured or misrepresented the plastic responses to the treatments.

Bromus briziformis exhibited a plastic response to soil moisture availability by increasing initial root to shoot ratios with low available water compared to medium or high available water (figure 1.4c). Both species had allocation trajectories that were more root-heavy in low-water, low nutrient treatments (figure 1.6d). Aronson et al. (1992) and Heschel et al. (2004) also report larger root to shoot ratios in response to low-water treatments in the winter-annual mediterranean mustard, *Erucaria hispanica*, and from three populations of the annual *Polygonum persicaria*, respectively. In contrast, (McConnaughay and Coleman 1999) found no plastic response of root to shoot ratios in response to available water for three annual old-field species. They suggested that plants may respond to reduced water availability by adjusting water use efficiency, rather than by shifting biomass allocation belowground, an observation unsupported by our data or that of Rice et al. (1992) from eastern Washington. Plasticity in biomass allocation to ensure survival and reproduction would clearly be adaptive in arid environments (Via and Lande 1985, Rice et al. 1992, Heschel et al. 2004).

Soil volume may affect plant growth independently of nutrient or water availability, but no clear pattern has been shown. Root to shoot ratios may increase, decrease or remain unchanged in response to restricted rooting volume (this study, McConnaughay and Bazzaz 1991 and references therein). The greater allocation to roots we observed in smaller pots (figure 1.4d) may result from inhibited shoot growth as roots became space-limited. Alternatively, shoot biomass may have senesced as soil resources became limiting (Hunt 1978). In either case, the net result was that fewer plants in small pots flowered compared to plants in medium or large pots (data not shown).

Low nutrient availability in our study produced higher root to shoot ratios (figure 1.6d), as predicted by optimal partitioning theory (Chapin et al. 1986). However, this effect was observed only at the lowest water availability, possibly because nutrient limitation tends to be exaggerated by low soil water content (McConnaughay and Coleman 1999). When soil water content is low, nutrient movement in the soil is decreased and it may be necessary for plants to explore a greater volume of soil to meet their nutrient requirements (Chapin et al. 1986).

Ontogenetic drift was apparent in half of our treatments for both experiments (figures 1.4d, 1.6d). Ontogenetically uncontrolled analysis of the soil volume experiment indicated that both species increased their initial root to shoot ratios in response to low water and that *B. briziformis* had higher initial root to shoot ratios than *B. tectorum*. Analysis controlling for ontogenetic drift found that only *B. briziformis* increased its initial root to shoot ratio with limited soil water (figure 1.4). Failing to account for ontogenetic drift in the nutrient experiment resulted in no significant effects of the treatments on initial root to shoot ratios. Biomass allocation trajectories were similar between the ontogenetically controlled and uncontrolled analyses (figures 1.4d, 1.6d). Differences between the ontogenetically controlled and

uncontrolled analyses of our data are subtle, but consistent with McConnaughey and Coleman's (1999) conclusion that failing to account for drift may produce results that misrepresent the magnitude or direction of effects on plant biomass allocation.

Annual plants generally decrease allocation to roots relative to shoots as they enter the reproductive phase of the life cycle (Hunt 1978, McConnaughay and Coleman 1999). In our soil volume study *B. briziformis* had a more shoot-heavy allocation trajectory than *B. tectorum* (figure 1.4d), suggesting either a stronger or earlier ontogenetic shift towards decreased root production in *B. briziformis* compared to *B. tectorum*. Of several hundred plants that remained after harvests were complete, only a single *B. briziformis* individual flowered, compared to 85 flowering *B. tectorum* individuals. If *B. tectorum* maintains a higher root to shoot ratio than *B. briziformis* through the reproductive phase of the life cycle, it may have better access to late-spring soil moisture resulting in higher reproductive success than *B. briziformis*. Greater drought tolerance of *B. tectorum* compared to *B. briziformis* may explain its greater invasive potential.

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1.7 TABLES

Table 1.1. Total water added to soil mixture and pot size treatments during the experiment.

Pot size	Sand	Water added ± s.d. (mL)
Soil volume study		
Small	25%	322 ± 62.2
Small	45%	335.5 ± 13.4
Small	70%	289 ± 24.0
Medium	25%	1102 ± 98.3
Medium	45%	1008 ± 112
Medium	70%	831 ± 101
Large	25%	3359 ± 659
Large	45%	2735 ± 319
Large	70%	2398 ± 364
Nutrient study		
Medium	25%	1272 ± 141
Medium	45%	1192 ± 162
Medium	70%	1059 ± 222

Table 1.2. Analysis of covariance with log total plant mass as the response variable, species, pot size and potting medium as treatment variables, and log plant age as the covariate. Highest-order significant terms are indicated in the final column.

SOURCE	DF	SS	F	P	Significant
Intercept components					
Sand	2	3.0161	4.56	0.0115	*
Species	1	0.3884	1.17	0.2797	
Pot size	2	41.5799	62.87	< 0.0001	*
Sand * Species	2	0.3369	0.51	0.6016	
Sand * Pot size	4	0.0965	0.07	0.9903	
Species * Pot size	2	0.1215	0.18	0.8323	
Sand * Species * Pot size	4	0.2081	0.16	0.9595	
Slope components					
Age	1	1051.9666	3181.22	< 0.0001	
Age * Sand	2	4.7453	7.17	0.0010	*
Age * Species	1	0.4538	1.37	0.2428	
Age * Pot size	2	54.3581	82.19	< 0.0001	*
Age * Sand * Species	2	0.3481	0.53	0.5915	
Age * Species * Pot size	2	0.0969	0.15	0.8638	
Age * Sand * Pot size	4	0.0534	0.04	0.9969	
Age * Sand * Species * Pot size	4	0.1530	0.12	0.9769	

Table 1.3. Analysis of covariance with log root to shoot ratio as the response variable, species, pot size and potting medium as treatment variables, and log total plant mass as the covariate. Highest-order significant terms are indicated in the final column.

SOURCE	DF	SS	F	P	Significant
Intercept components					
Sand	2	1.7674	6.28	0.0022	
Species	1	2.8438	20.21	< 0.0001	
Pot size	2	0.0264	0.09	0.9105	
Sand * Species	2	0.8774	3.12	0.0463	*
Sand * Pot size	4	0.4771	0.85	0.4965	
Species * Pot size	2	0.2125	0.76	0.4712	
Sand * Species * Pot size	4	0.6019	1.07	0.3727	
Slope components					
Plant mass	1	0.7137	5.07	0.0254	
Plant mass * Sand	2	0.5007	1.78	0.1714	
Plant mass * Species	1	0.8151	5.79	0.0170	*
Plant mass * Pot size	2	4.6559	16.54	< 0.0001	*
Plant mass * Sand * Species	2	0.5435	1.93	0.1476	
Plant mass * Species * Pot size	2	0.1273	0.45	0.6368	
Plant mass * Sand * Pot size	4	0.3356	0.60	0.6658	
Plant mass * Sand * Species * Pot size	4	0.5111	0.91	0.4602	

Table 1.4. Analysis of covariance with log total plant mass as the response variable, species, nutrient treatment and potting medium as treatment variables, and log plant age as the covariate. Highest-order significant terms are indicated in the final column.

SOURCE	DF	SS	F	P	Significant
Intercept components					
Sand	2	1.6377	4.61	0.0114	
Species	1	0.0220	0.12	0.7251	
Nutrient level	2	2.3772	6.69	0.0017	*
Sand * Species	2	1.1376	3.2	0.0435	*
Sand * Nutrient level	4	0.3470	0.49	0.7440	
Species * Nutrient level	2	0.1558	0.44	0.6457	
Sand * Species * Nutrient level	4	0.2996	0.42	0.7927	
Slope components					
Age	1	722.7704	4070.81	< 0.0001	
Age * Sand	2	2.6542	7.47	0.0008	
Age * Species	1	0.0043	0.02	0.8763	
Age * Nutrient level	2	3.3677	9.48	0.0001	*
Age * Sand * Species	2	1.1409	3.21	0.0431	*
Age * Sand * Nutrient level	4	0.5087	0.72	0.5821	
Age * Species * Nutrient level	2	0.1977	0.56	0.5742	
Age * Sand * Species * Nutrient level	4	0.3063	0.43	0.7858	

Table 1.5. Analysis of covariance with log root to shoot ratio as the response variable, species, nutrient treatment and potting medium as treatment variables, and log total plant mass as the covariate. Highest-order significant terms are indicated in the final column.

SOURCE	DF	SS	F	P	Significant
Intercept components					
Sand	2	0.0266	0.08	0.9196	
Species	1	0.1749	1.11	0.2949	
Nutrient level	2	0.7357	2.32	0.1015	
Sand * Species	2	0.3572	1.13	0.3264	
Sand * Nutrient level	4	1.7341	2.74	0.0310	*
Species * Nutrient level	2	0.4127	1.30	0.2747	
Sand * Species * Nutrient level	4	1.1764	1.86	0.1210	
Slope components					
Plant mass	1	14.7156	92.96	< 0.0001	
Plant mass * Sand	2	0.6050	1.91	0.1516	
Plant mass * Species	1	0.0923	0.58	0.4465	
Plant mass * Nutrient level	2	2.8166	8.90	0.0002	
Plant mass * Sand * Species	2	0.3048	0.96	0.3843	
Plant mass * Sand * Nutrient level	4	2.8642	4.52	0.0018	*
Plant mass * Species * Nutrient level	2	0.2446	0.77	0.4637	
Plant mass * Sand * Species * Nutrient level	4	0.8862	1.40	0.2370	

Table 1.6. Analysis of covariance with log root to shoot ratio as the response variable, species, pot size and potting medium as treatment variables, and log plant age as the covariate. Highest-order significant terms are indicated in the final column.

SOURCE	DF	SS	F	P	Significant
Intercept components					
Sand	2	1.3103	4.53	0.0119	*
Species	1	3.3846	23.38	< 0.0001	*
Pot size	2	0.0923	0.32	0.7273	
Sand * Species	2	0.6154	2.13	0.1219	
Sand * Pot size	4	0.3628	0.63	0.6441	
Species * Pot size	2	0.3683	1.27	0.2824	
Sand * Species * Pot size	4	0.4112	0.71	0.5857	
Slope components					
Age	1	0.9255	6.39	0.0122	
Age * Sand	2	0.3586	1.24	0.2919	
Age * Species	1	1.1980	8.28	0.0044	*
Age * Pot size	2	7.0876	24.48	< 0.0001	*
Age * Sand * Species	2	0.3813	1.32	0.2701	
Age * Species * Pot size	2	0.1851	0.64	0.5286	
Age * Sand * Pot size	4	0.3038	0.52	0.7177	
Age * Sand * Species * Pot size	4	0.4197	0.72	0.5759	

Table 1.7. Analysis of covariance with log root to shoot ratio as the response variable, species, nutrient treatment and potting medium as treatment variables, and log plant age as the covariate. Highest-order significant terms are indicated in the final column.

SOURCE	DF	SS	F	P	Significant
Intercept components					
Sand	2	0.0770	0.23	0.7910	
Species	1	0.1205	0.74	0.3925	
Nutrient level	2	0.7086	2.16	0.1188	
Sand * Species	2	0.4075	1.24	0.2915	
Sand * Nutrient level	4	1.4872	2.27	0.0646	
Species * Nutrient level	2	0.4774	1.46	0.2364	
Sand * Species * Nutrient level	4	1.1017	1.68	0.1575	
Slope components					
Age	1	12.8847	78.62	< 0.0001	
Age * Sand	2	0.2313	0.71	0.4955	
Age * Species	1	0.1180	0.72	0.3975	
Age * Nutrient level	2	2.2765	6.95	0.0013	
Age * Sand * Species	2	0.3973	1.21	0.3005	
Age * Sand * Nutrient level	4	2.2599	3.45	0.0100	*
Age * Species * Nutrient level	2	0.3306	1.01	0.3672	
Age * Sand * Species * Nutrient level	4	0.7214	1.10	0.3587	

1.8 FIGURE CAPTIONS

Figure 1.1. Analysis of covariance with log root to shoot ratio as the response variable, nutrient level, soil texture and species as treatment variables and log total plant mass as the covariate. Log plant mass values are raw (a), or increased by a constant value (b; see text). Intercepts and slopes in figures 1.3 – 1.6 were derived from figures such as this.

Figure 1.2. Log plant mass plotted against time for the soil volume study (a, b) and for the nutrient study (c, d). Plant growth trajectories were of a similar form across treatments; representative curves were constructed by averaging across species and pot size (a), species and sand content (b, d) or species and nutrient level (c).

Figure 1.3. Regression slopes from the analysis of covariance with log plant mass as the response variable, pot size, sand content of soil and species as treatment variables and log plant age as the covariate. No species differences were detected. Slopes differed between all pot sizes (uppercase letters). Sand treatments with the same lowercase letters above them were not significantly different.

Figure 1.4. Regression intercepts (a, c) and slopes (b, d) from the analysis of covariance with log root to shoot ratio as the response variable and log plant age (a, b) or log total plant mass + 6.1 (c, d) as the covariate; both analyses included pot size, soil texture and species as treatment variables. Groups with the same capital letter above them do not differ across pot size treatments, groups with the same lowercase letter above them do not differ between sand treatments (a) or sand by species treatments (c). Species effects were detected in panels a, b, and d. Slopes with asterisks beneath them (d) were significantly different from zero, indicating ontogenetic drift.

Figure 1.5. Regression slopes from the analysis of covariance with log plant mass as the response variable, nutrient level, sand content of soil and species as treatment variables and log

plant age as the covariate. Nutrient levels with the same uppercase letters above them are not significantly different from one another. Sand by species treatments with the same lowercase letters above them do not differ significantly within nutrient levels.

Figure 1.6. Regression intercepts (a, c) and slopes (b, d) from the analysis of covariance with log root to shoot ratio as the response variable and log plant age (a, b) or log total plant mass + 5.6 (c, d) as the covariate; both analyses included nutrient level, soil texture and species as treatment variables. Sand by nutrient treatment combinations with the same letters above them do not differ significantly from one another. No species effects were detected. Slopes with asterisks beneath them (d) were significantly different from zero, indicating ontogenetic drift.

1.9 FIGURES

Figure 1.1

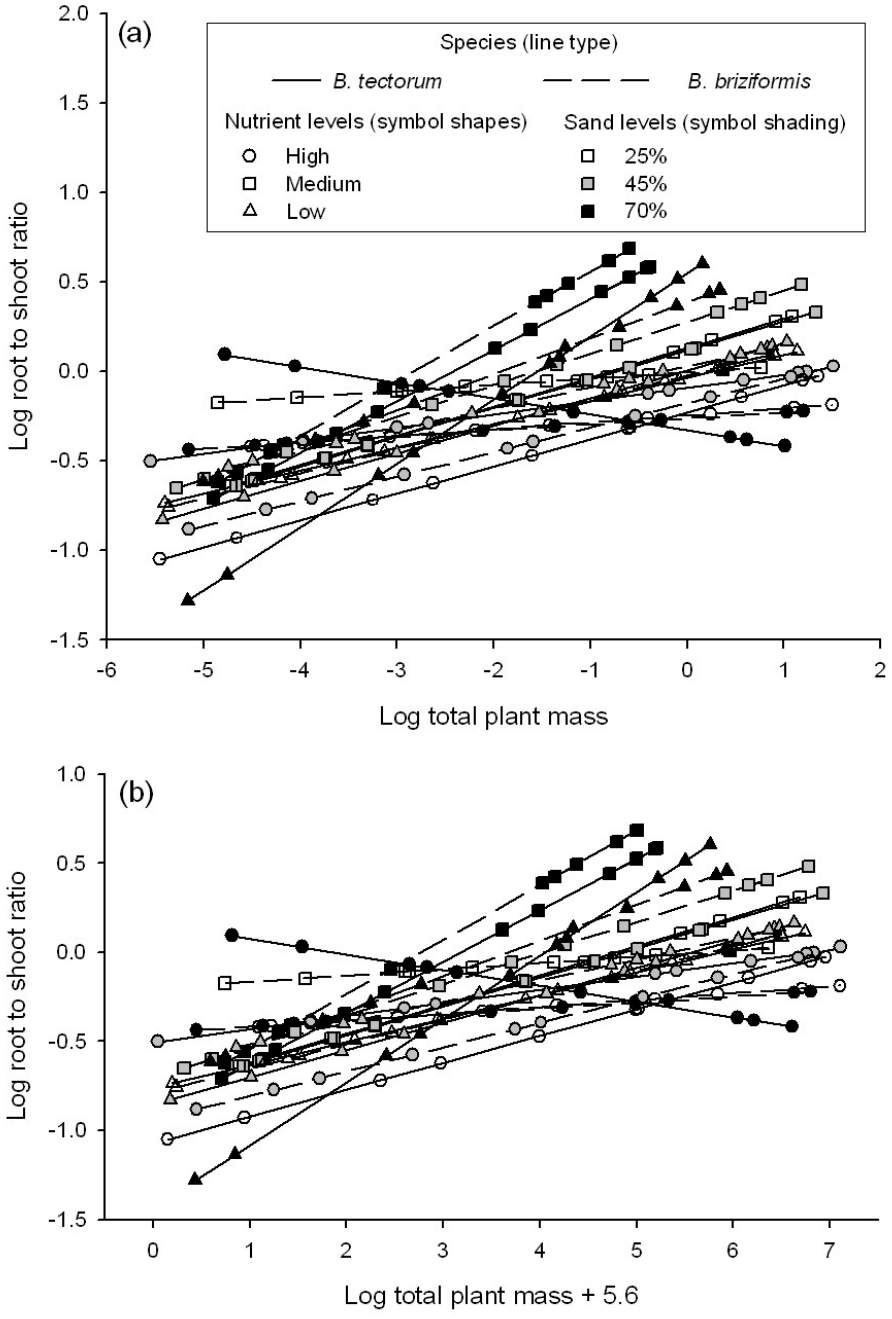


Figure 1.2

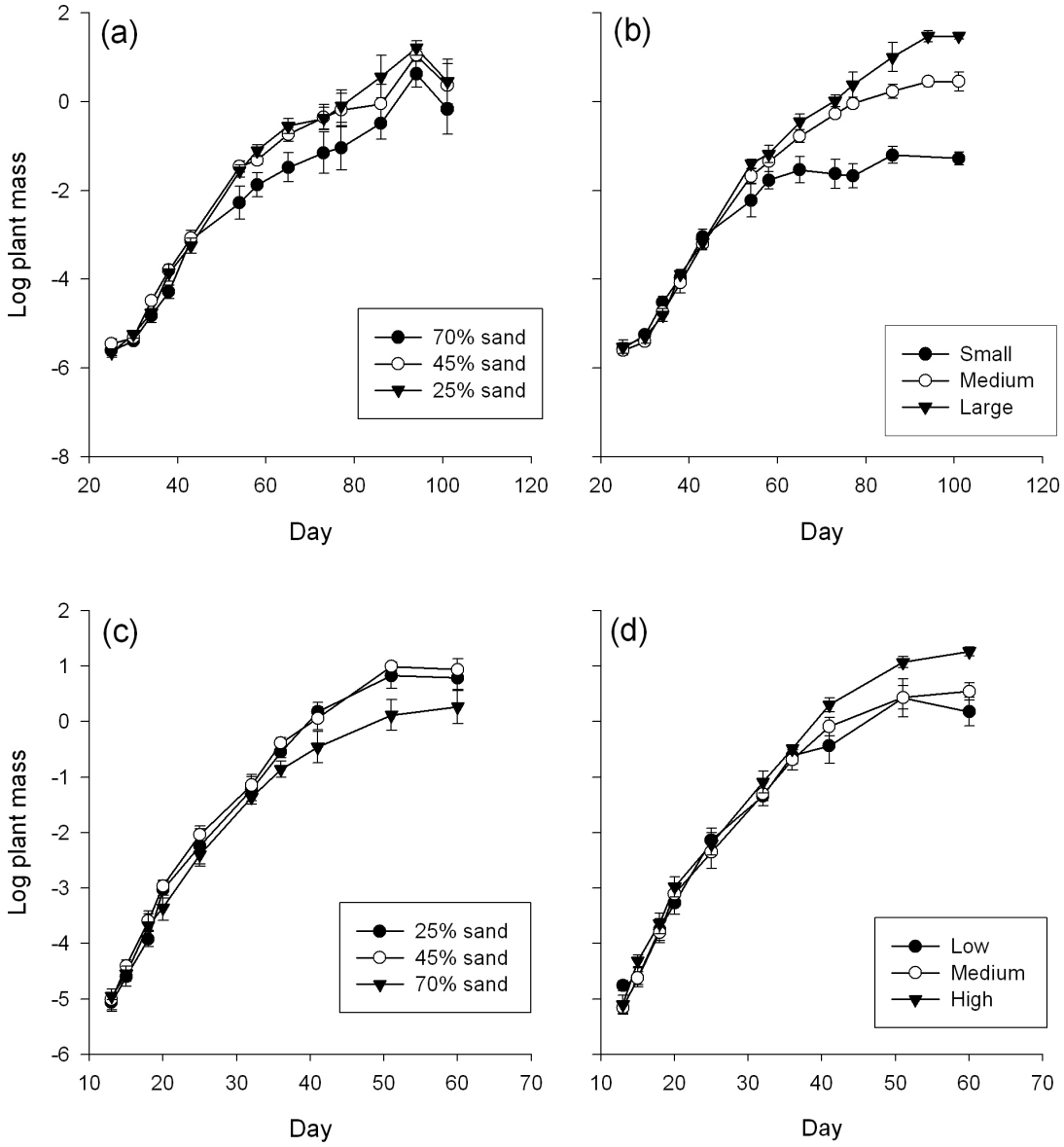

(a)
Log plant mass
Day
70% sand
45% sand
25% sand
(b)
Day
Small
Medium
Large
(c)
Log plant mass
Day
25% sand
45% sand
70% sand
(d)
Day
Low
Medium
High

Figure 1.3

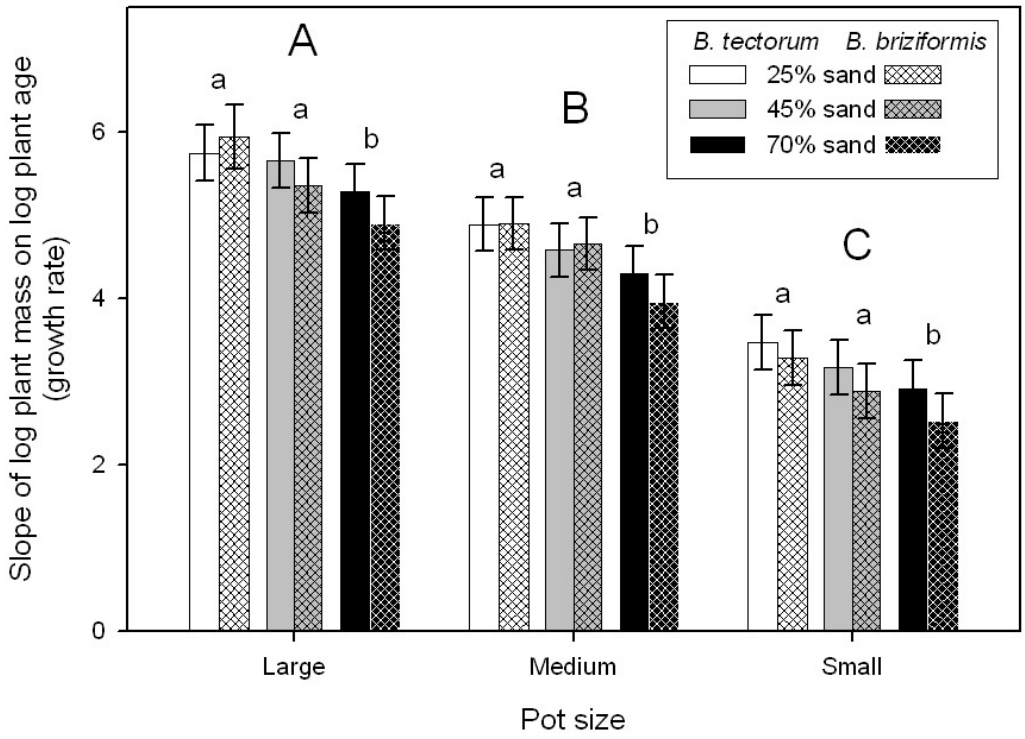

A
B
C
a
a
b
a
a
b
a
a
b
B. tectorum
B. briziformis
25% sand
45% sand
70% sand
Slope of log plant mass on log plant age (growth rate)
0
2
4
6
Large
Medium
Small
Pot size

Figure 1.4

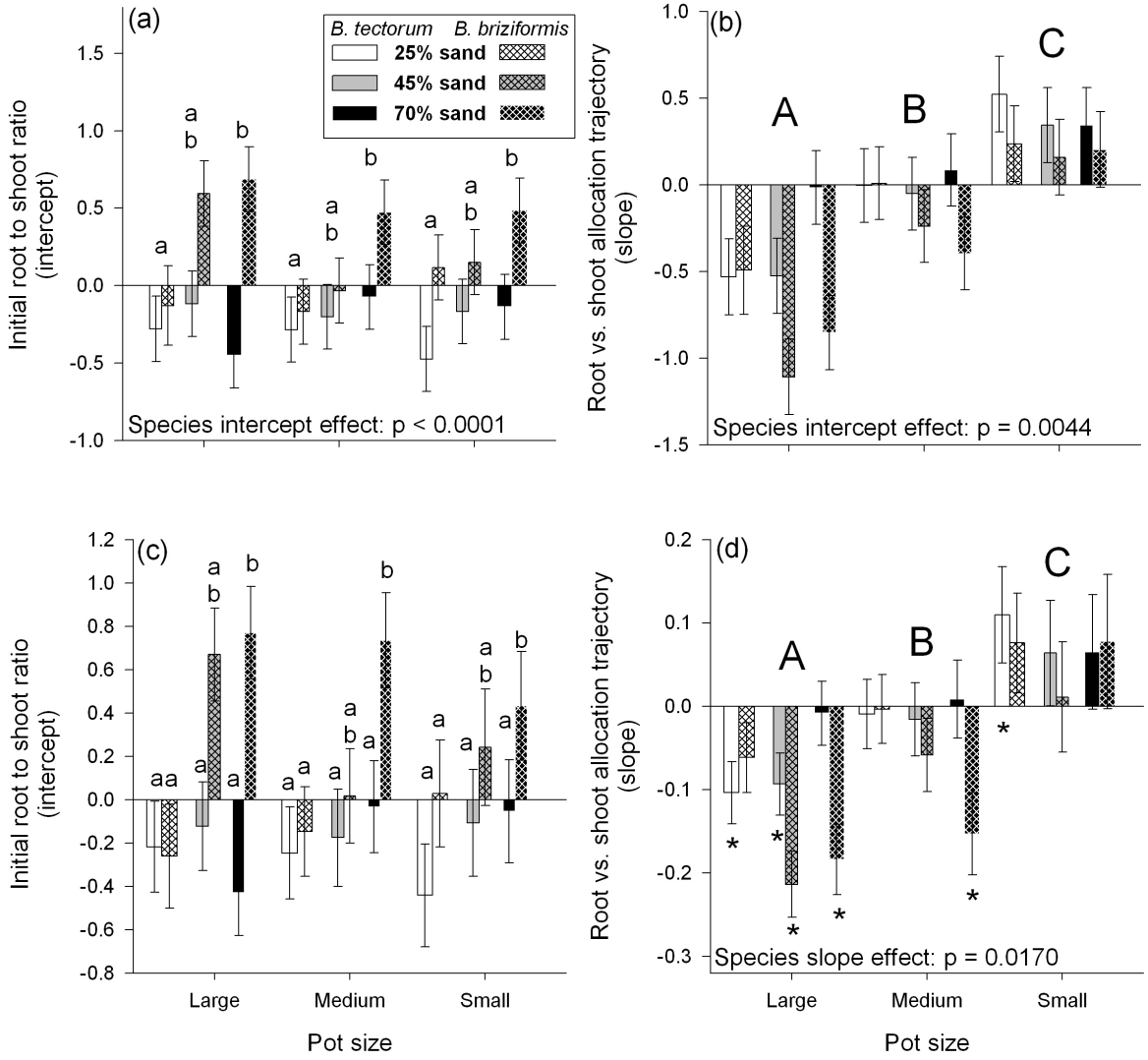

(a)
B. tectorum B. briziformis
25% sand
45% sand
70% sand
Initial root to shoot ratio (intercept)
Species intercept effect: p < 0.0001
(b)
A
B
C
Root vs. shoot allocation trajectory (slope)
Species intercept effect: p = 0.0044
(c)
Initial root to shoot ratio (intercept)
Large
Medium
Small
Pot size
(d)
A
B
C
Root vs. shoot allocation trajectory (slope)
Species slope effect: p = 0.0170
Large
Medium
Small
Pot size

Figure 1.5

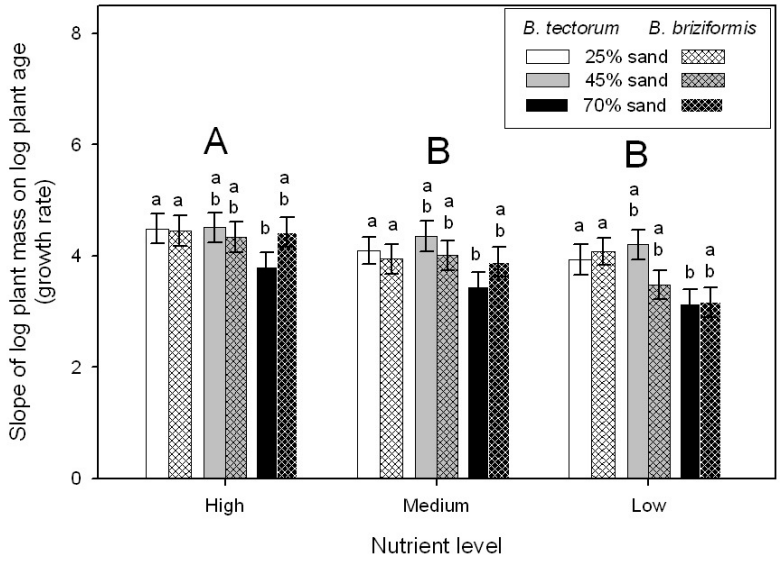
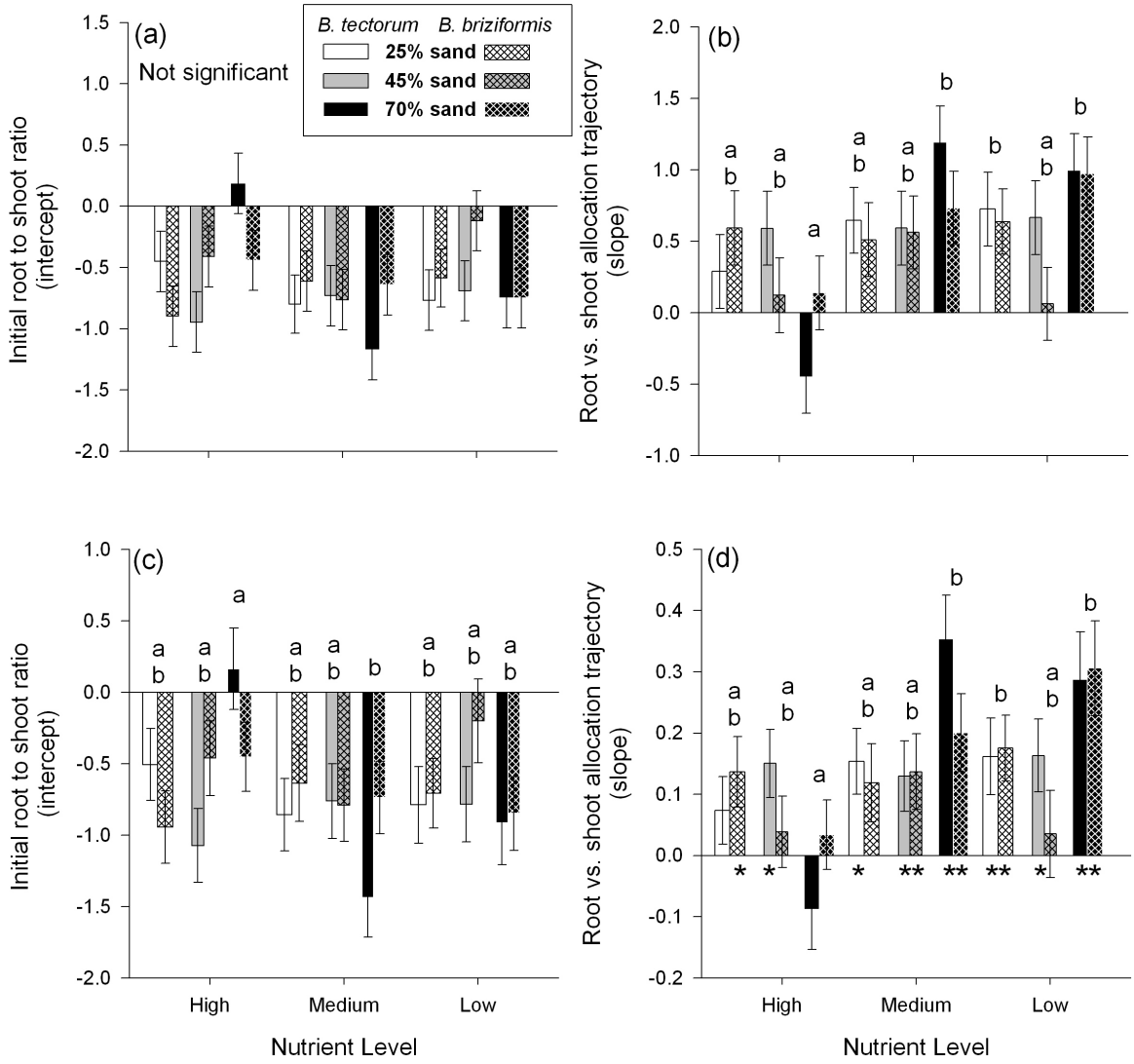

B. tectorum
B. briziformis
25% sand
45% sand
70% sand
Slope of log plant mass on log plant age (growth rate)
Nutrient level
High
Medium
Low
A
B
B

Figure 1.6


(a)
Not significant
B. tectorum B. briziformis
25% sand
45% sand
70% sand
Initial root to shoot ratio (intercept)
(b)
Root vs. shoot allocation trajectory (slope)
(c)
(d)
High
Medium
Low
Nutrient Level

.

CHAPTER TWO

NEGLIGIBLE EFFECTS OF POT SIZE ON GROWTH CURVE ANALYSIS FOR TWO ANNUAL *BROMUS* SPECIES IN A WATER-AVAILABILITY EXPERIMENT.

2.1 ABSTRACT

Greenhouse conditions can introduce environmental artifacts into plant growth data. We compared growth curves of two introduced annual grass congeners, *Bromus tectorum* and *B. briziformis* in response to water availability and pot size. The water availability treatments were designed to test whether invasive *B. tectorum* performs better under droughty conditions than naturalized but non-invasive *B. briziformis.* We used three levels of water availability in each of three pot sizes to determine whether small pot sizes would obscure the effects of water availability on logistic growth curves. We detected no differences in growth curves between the two species at any level of water or pot size. Small pots produced plant growth curves with maximum plant sizes reduced more than 95% compared to growth curves from plants in large pots and more than 80% compared to growth curves from plants in medium pots. Low water availability generally reduced maximum plant size, reduced plant growth rate and delayed the period of most rapid plant growth compared to medium or high water availability, but results were not always consistent across pot sizes.

2.2 INTRODUCTION

Greenhouse experiments are often used to assess plant responses to treatments because conditions in a greenhouse are relatively easy to manipulate so that the effects of a specific treatment on plant performance can be isolated. A necessary consequence of a greenhouse setting is that growth conditions are unlike anything experienced by wild plants and artifacts may be introduced into the experimental data. Among these are well-known pot effects (McConnaughay et al. 1991, McConnaughay et al. 1993, Ronchi et al. 2006) which may alter biomass allocation, root or shoot morphology or plant growth. The pot effect becomes a serious problem for experiments that use plant mass over time as a response variable. Plant mass increases as plants capture more resources and so is thought to indicate competitive ability or capacity to produce seed (but see Neytcheva et al. 2008).

An alternative approach is to fit nonlinear growth curves to plant mass data obtained from sequential harvests. The advantage of this approach is that it captures the entire growth pattern of the plant (Zhao et al. 2005). The popular Richards growth model describes sigmoid growth (Causton et al. 1981):

$$[1] \qquad w(t) = a\,(1 + (d - 1)\,e^{\,k\,(g - t)})^{1\,/\,(1 - d)},\ d \neq 1$$

where $w(t)$ is the mass of the plant at time t, a is the maximum asymptotic plant size (hereafter, 'size parameter'), d is a shape parameter that determines the size at which the plant will experience the highest absolute growth rate (i.e. where the growth curve is steepest; hereafter, 'shape parameter'), k is a growth rate (hereafter, 'growth rate') and g determines the age at which the plant will have the highest absolute growth rate (hereafter, 'date of maximum plant growth'). The size parameter serves to scale the rest of the function (which is bounded by zero and one) to

a size appropriate to the mature plant. It is often impossible to fit the Richards growth model when there are few data points near the value of the size parameter (Causton and Venus 1981), but this problem could be avoided by estimating the size parameter independently, potentially allowing the use of smaller pots for greenhouse experiments.

The application of a mathematical model to growth data has the advantage of reducing complex growth patterns to a few model parameters (Zhao et al. 2005). Additionally, if the model is appropriately specified, the parameters can be interpreted in terms of the biological phenomena underlying the response curve (Meredith et al. 1991); for example, the growth rate parameter in the Richards model above is proportional to the average relative growth rate over the growth interval from plant germination to maximum asymptotic plant size (Causton and Venus 1981). However, Potvin et al. (1990) found that two different models for photosynthetic light response curves produced substantially different parameter estimates and could have led to biologically different interpretations. Consequently, they advocate qualitative comparisons of whole curves rather than interpretation of model parameters.

The objectives in this study were to examine the effects of a pot-size limitation on growth model performance, and to test whether these effects could be avoided by using early-harvest data from small plants in conjunction with a fixed, arbitrarily large size parameter. We also discuss qualitative interpretations of model output based on the shape of the curves compared to more systematic interpretation of the model parameters directly.

We used growth data from *Bromus tectorum* (cheatgrass) and *Bromus briziformis* (rattlesnake brome). Both species are native to Eurasia but *B. tectorum* is highly invasive in the Pacific Northwest and *B. briziformis* is naturalized in western North America, but is not considered invasive (Hulbert 1955). Both species decline in abundance along a moisture

gradient running from eastern Washington (mesic) to central Washington (semiarid), and *B. tectorum* extends its range further into the semiarid part of the state than *B. briziformis* (Hulbert 1955). Within the framework described above, we ask whether the growth patterns of these introduced congeners respond differently to variation in water availability and pot size.

2.3 METHODS

Seeds of *B. tectorum* and *B. briziformis* were collected from Elk Creek Falls Recreation area in north-central Idaho (46˚ 44' W, 116˚ 10' N) within the *Pseudotsuga menziesii* / *Holodiscus discolor* habitat type (Daubenmire et al. 1968) in August 2005. Seeds were collected from several hundred individuals of each species, thoroughly mixed, and stored at room temperature until planting.

B. tectorum or *B. briziformis* seeds were planted into 164 mL, 410 mL, or 2830 mL pots (hereafter called small, medium, and large; SC10 Cone-tainers, D25LW Deepots, or Tall One Treepots, respectively; Stuewe & Sons, Corvallis OR). Pots were filled with potting medium (Sun-Gro Sunshine Professional potting mix, Bellevue, WA) that had been mixed with 25%, 45% or 70% sand by volume. The soil texture treatments were imposed to effect a water-availability treatment, assuming coarsely textured soils are more drought-prone than finely textured soils (Smith et al. 2001). We recognize that the coarser soils may also have had lower nutrient availability than the finer soils (Smith et al. 2001). Pot racks were placed haphazardly on the benches and individual pots were assigned randomly to positions within these racks; the pots were not moved during the study.

Seeds were planted in pots on 6 March 2006 and maintained under natural lighting in a greenhouse. The greenhouse was maintained with temperatures of 4 - 8˚C at the beginning of

the study then gradually increased to 12 - 16˚C over the course of the study to encourage normal phenotypic development. Supplemental fertilizer was not added to the potting medium.

All plants within each pot size were watered to excess with tap water when approximately 50% of the plants in that treatment appeared to be wilting. One pot per treatment was randomly selected for the purpose of monitoring soil water. Pots for monitoring soil water were weighed immediately prior to watering and one to two hours after watering (after excess water had drained) to determine water loss between watering events. Total water added to each pot size-soil mixture was calculated based on weight change; treatments with more sand generally received less water than those with less sand (table 2.1).

We harvested one randomly-selected individual from each treatment on days 25, 30, 34, 38, 43, 54, 57, 65, 72, 77, 85, 94, 100 and 113 after planting. The final eight harvests required two or three days to complete. Shoot material was clipped at the soil surface and separated into live shoot and dead shoot material; only live shoot material was used for analyses. Soil was washed from the roots with running water over a 1.4 mm screen and captured on a 150 micron screen. The material in both screens was then hand-sorted to retrieve root material. All plant components were further washed in a sonicator with detergent for 30 minutes, rinsed with tap water and dried at 80˚C for at least 72 hours before weighing.

We used R statistical software (R Development Core Team 2006) to fit Richards growth curves to whole-plant mass data for each of the 18 treatments separately and for the data set pooled across all treatments. Of these 19 models only seven converged on a solution; those that did converge produced an error-weighted mean value of 2.37 for the shape parameter (*d*; data not shown). The logistic growth curve is a special case of the Richards growth curve with the shape parameter set to 2: with this value of the shape parameter, the logistic curve has the highest

absolute growth rate at 50% of maximum asymptotic plant size. A shape parameter value of 2.37 causes the highest absolute growth rate to occur at 53% of the asymptotic maximum plant size. Therefore we fit the logistic model to our data; this model is generally easier to parameterize and more robust than the Richards' growth model (Causton and Venus 1981).

To test treatment effects on the growth curves, models were compared in a framework that is analogous to analysis of variance for linear models (Potvin et al. 1990). The statistical model is

[2] $y_{ij} = \mu_i + \varepsilon_{ij}$,

for $i = 1, 2, \ldots, m$ treatments and $j = 1, 2, \ldots, n$ observations within each treatment, and μ is a nonlinear function, in this case the logistic growth function:

[3] $w(t) = a\,(1 + e^{k\,(g - t)})^{-1}$,

where the parameters are the same as for the Richards' growth model. This approach differs from a one-way means model analysis of variance in that the mean is replaced by a nonlinear curve fit and the degrees of freedom must be adjusted to account for the parameters in the nonlinear model:

[4] $MST = (SS\mu_0 - \Sigma\, SS\mu i) / (mp - p + m - 1)$,

[5] $MSE = SS\mu_0 / (N - m - mp)$,

where *MST* is the mean square for treatments, $SS\mu_0$ is the sum of squared ε_j for the reduced model (i.e. a single set of model parameters fitted to all data), $\Sigma\, SS\mu i$ is the sum of squared ε_{ij} summed over models fitted to each of the m treatment groups, p is the number of parameters in the nonlinear model, *MSE* is the mean square for error, and N is the total number of observations in all m treatments. This approach erroneously assumes that the parameters in the model are independent of one another, but the resulting error is conservative (i.e. will underestimate the F

statistic). If the overall F-test was significant, fitted curves were compared in a pair-wise fashion using the same approach and a Bonferroni-Holm correction for multiple comparisons was applied (Wright 1992). For pairs of curves that were significantly different from one another, the two sets of model parameters were compared using a test that is analogous to Hotelling's T^2 test for the location of a multivariate mean vector (Johnson et al. 2002):

[6] $T^2 = p\,(\mathbf{X_1} - \mathbf{X_2})'\ \mathbf{S}^{-1}\,(\mathbf{X_1} - \mathbf{X_2})$,

where p is the length of the vectors, $\mathbf{X_1}$ and $\mathbf{X_2}$ are the parameter vectors and **S** is the covariance matrix of the two vectors.

To examine the feasibility of small-pot data for growth curve analysis we repeated the analysis using arbitrarily large values of the maximum plant-size parameter. We calculated an error-weighted mean value for the size parameter (*a*) from all six of the large-pot treatments and from all six of the medium-pot treatments; the values were 5.72 and 1.96, respectively (models for which the size parameter was fixed are hereafter called 2-parameter models). Because we were interested in avoiding pot-limitation of plant growth, we excluded data from the later harvests when fitting these models. We refitted all the models several times, deleting the final data point between each fit until only five data points from each treatment remained. We then chose the level of data truncation that produced the smallest confidence intervals across all parameters, assuming that this represented a trade-off between sparse data and plants that were becoming too large, relative to maximum plant size (data not shown). This resulted in a truncated data set including the first nine data points (72 days) from each treatment. These models and their parameter values were analyzed using the same analysis of variance framework described above. We report only differences for which we obtained statistical comparisons with $p < 0.05$; we take $p > 0.05$ to indicate no difference between treatments.

2.4 RESULTS

Small pots reduced plant size compared to medium or large pots and the mean growth rate was generally lower for plants with less water compared to plants with more water, and greater for plants in smaller pots compared to plants in larger pots. Plants in less favorable growing conditions reached their period of maximum growth sooner than those in more favorable growing conditions.

We observed a treatment effect on the plant growth curves (table 2.2), but it was impossible to make eleven of the 153 pair-wise curve comparisons because the null models could not be fitted to the data (see figure 2.1 caption). Growth curves of *Bromus briziformis* and *B. tectorum* within the same treatment never differed significantly from one another (figure 2.1) so the data were pooled across species for all subsequent analyses.

Plant growth curves were significantly different between the treatments when the data were pooled across species (figure 2.2, table 2.2). The null model needed to compare the growth curve of plants in large pots with low water availability to the growth curve of plants from small pots with medium water availability did not converge (figure 2.2), but all other comparisons converged. Growth curves always differed between plants grown in different pot sizes (figure 2.2). Growth curves of plants grown with low water availability always differed from growth curves of plants grown with medium or high water availability, but growth curves of plants grown with medium water availability differed from those of plants grown with high water availability only in large pots (figure 2.2).

The size parameter (a) decreased with decreasing pot size and always differed between pot sizes (figure 2.3a). Plants grown with high and medium water availability were significantly

larger than those grown with low water in the small and medium pot sizes but in the large pots, the lowest water availability treatment produced the largest plants (figure 2.3a). The rate parameter (k) for plants grown in the largest pots with the highest water availability was more than twice as large as any other rate parameter and the rate parameter for plants grown in the smallest pots with the lowest water availability was smaller than any other rate parameter (figure 2.3b). Growth rates of plants in large and small pots decreased significantly with reductions in water availability but plants in the medium pots with high or low water availability had higher growth rates than those grown with medium water availability (figure 2.3b). Decreasing water availability delayed the date of maximum plant growth in large and small pots but plants with the lowest water availability had the earliest date of maximum growth in the medium pots (figure 2.3c).

We detected fewer differences between overall growth curves when the maximum plant size parameter was fixed at 5.7 or 2.0 (figures 2.4, 2.5), and the pattern of differences between the fitted curves was the same for both analyses. Growth curves of all plants in small pots differed from those of all plants in large pots, and from those of plants with medium or high water availability in medium pots (figures 2.4, 2.5). Growth curves of plants in medium pots with medium or low water availability differed from those of plants in large pots with high or medium water availability, and plants in medium pots with high water availability differed from those in large pots with medium water availability (figures 2.4, 2.5). Growth curves of plants with low water availability were significantly different from those of plants grown with high water availability in medium and small pot sizes (figures 2.4, 2.5) and growth curves of plants grown with low water availability differed from those of plants grown with medium water availability in large and small pots (figures 2.4, 2.5).

Magnitudes of the parameters differed between the analysis that fixed the maximum plant size parameter at 5.72 compared to the analysis that fixed the maximum plant size parameter at 1.96 but the pattern of significant differences between model parameters did not change between the two analyses (figures 2.6, 2.7). Growth rates were lower for plants with low water availability compared to plants with high water availability in large and small pots, but the difference was reversed for plants in medium pots (figures 2.6, 2.7). Growth rates of plants in medium and large pots were higher than those of plants in small pots (figures 2.6, 2.7). The date of maximum plant growth was generally delayed for plants in smaller pots, compared to plants in larger pots. Date of maximum plant growth for plants with low water availability was later than for plants with medium water availability in large pots, later than for plants with high water availability in medium pots, and later than for plants with medium or high water availability in small pots (figures 2.6, 2.7).

2.5 DISCUSSION

We found that 1) a curve fitting approach to plant growth data was sufficient to detect the large effect of pot size on plant size and the smaller effect of water on plant growth rate, 2) interpretation of the underlying parameters was less biologically intuitive but also less subjective than interpretation of whole curves, and 3) the use of small pots does not hinder the interpretation of plant growth curves.

We did not detect differences between *B. tectorum* and *B. briziformis* in growth response to pot size or water availability. Others have found competitive differences (Corbin et al. 2004), different growth rates (Burns 2006) or differences in fecundity (Gerlach et al. 2003) that

appeared to explain differences in the invasive potential between their study species. However some of these differences were context specific (Burns 2006), limited to particular stages in the life histories (Gerlach and Rice 2003) of the study organisms, or did not involve comparisons between congeners (Corbin and D'Antonio 2004). And others have also found no differences in plant growth parameters to explain the invasive nature of the study organisms (Arredondo et al. 1998). *B. tectorum* and *B. briziformis* might not differ with respect to their vegetative growth characteristics; alternatively, our experimental design may have been insufficient to detect different growth responses between the two species.

Smaller pots produced smaller plants with earlier periods of maximum growth (figures 2.2, 2.3a, 2.3c). The volume of soil provided for plant growth can affect plant growth independently of the quantity of nutrient resources contained in that soil (McConnaughay and Bazzaz 1991), and in this study both soil volume and nutrient resources were simultaneously manipulated. Here, plants in smaller pots achieved their period of most rapid growth sooner compared to plants in larger pots (figure 2.3c) and completed most of their total growth over a shorter period of time (figure 2.2) than plants in larger pots. Similar phenological shifts in annual grass species have been observed in response to reduced soil volume (McConnaughay and Bazzaz 1991) and imposed drought stress (Aronson et al. 1992). *B. tectorum* has been demonstrated to be plastic in flowering phenology with resource restriction in the form of higher sowing density (Rice et al. 1991b) or experimentally manipulated soil moisture (Rice et al. 1992), but not in response to year-to-year moisture variation (Rice et al. 1991a).

Plant growth responses to water availability were not consistent across pot sizes. Maximum plant size was smallest for low water availability in the medium and small pots, as expected, but largest for low water availability in the large pots (figure 2.3a). Plant growth rate

declined with declining water availability in the large and small pots, as expected, but not in the medium pots (figure 2.3b, 2.6a, 2.7a). Date of maximum growth was delayed in low-water treatments compared to high water treatments in large and small pots, as expected (figure 2.3c, 2.6b, 2.7b), but date of maximum growth was not consistent between the two-parameter and three-parameter models for medium pots (figure 2.3c, 2.6b, 2.7b). In all cases above, most of the data produce the expected results (low water availability delays and reduces plant growth), but the large number of inconsistencies preclude strong conclusions. The harvest schedule we used concentrated most of the data points during early growth of the plants (see methods), resulting in fewer data points during the period of rapid plant growth and near the maximum plant size. We note that in no cases do the unexpected data come from small pots, and we suggest that a harvesting plants to concentrate data during the period of maximum plant growth data may have improved model fits (Causton and Venus 1981).

Potvin et al. (1990) also fitted curves to plant growth data and advocated a qualitative approach to the interpretation curves, rather than an objective comparison of parameters. In our study, it was not reasonable to make all 36 pair-wise comparisons by visual inspection. For smaller experiments (e.g. Potvin et al. 1990), a direct comparison of the response curves may be feasible but when there are a large number of treatments (nine in the present study), the number of comparisons between treatments necessitate an objective approach to the differences between treatments.

The potential for pot size to obscure (McConnaughay et al. 1993) or influence (McConnaughay and Bazzaz 1991) experimental results for greenhouse experiments is a concern. Although we did observe substantial pot-effects in our experiment, in most cases they affected the magnitude but not the pattern among the three model parameters. If the goal of a

curve fitting procedure is to produce model parameters that can be interpreted in terms of the underlying biology, then a successful model will produce parameters that vary consistently across a gradient of treatments, such as water availability. By that measure, we achieved the highest degree of success with plants in the small pots because all three parameters varied consistently across the watering treatments (figure 2.3). We achieved intermediate success with plants in the large pots because two of three parameters varied consistently across the watering treatments, but there was no consistent pattern in the size parameter (figure 2.3a). Plants in medium pots produced the poorest growth curves because only the maximum plant size parameter (figure 2.3a) responded consistently to the watering treatments. Previous work on the effect of pot size on plant growth has indicated pot effects on biomass yield (McConnaughay and Bazzaz 1991, McConnaughay et al. 1993) and biomass allocation (McConnaughay and Bazzaz 1991), and that such pot effects may be species specific.

Our attempt to mitigate pot effects on the analysis by fitting models only to the early-growth data with arbitrarily large values of the maximum plant size parameter produced two undesirable effects and no desirable effects. First, it eliminated maximum plant size as a response variable, and there were small but significant differences between maximum plant sizes across the water gradient (figure 2.3a). Second, although the 2-parameter model results for the small and large pots were consistent with the three-parameter models (figures 2.3b, 2.3c, 2.6, 2.7), this was not the case for the medium pots. Growth rates of plants in medium pots increased with decreasing water availability in the two-parameter model but showed no consistent trend in the three-parameter model. And date of maximum growth for plants grown with low water availability was latest in the two-parameter model but earliest in the three-parameter model. If the growth model is appropriately specified, the early-growth data should contain all of the

information necessary to fit the whole curve (Hao Zhang, personal communication) but the logistic growth model is particularly difficult to fit when the data do not include values near the maximum plant size (Causton and Venus 1981). Apparently the high degree of correlation between the model parameters (Causton and Venus 1981) causes the growth rate and date of maximum growth parameters to be sensitive to the value of the maximum size parameter.

Although pot size in our study had a strong effect on plant size, growth rate and date of maximum growth, the effects of water on those parameters were still generally significant in all three pot sizes. Curve fitting may be a good analytic method to use when pot effects are a concern because unlike techniques that compare biomass yield or other measures of growth at one to several points in time, growth curves reduce the pattern of biomass accumulation to a few parameters (Zhao et al. 2005) that capture the fundamental elements of plant growth.

2.6 LITERATURE CITED

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2.7 TABLES

Table 2.1. Total water added to soil mixture and pot size treatments during the experiment.

Pot size	Sand	Water added ± s.d. (mL)
Small	25%	322 ± 62.2
Small	45%	335.5 ± 13.4
Small	70%	289 ± 24.0
Medium	25%	1102 ± 98.3
Medium	45%	1008 ± 112
Medium	70%	831 ± 101
Large	25%	3359 ± 659
Large	45%	2735 ± 319
Large	70%	2398 ± 364

Table 2.2. One-way analyses of variance with fitted logistic growth curves as the responses.

Source	SS	df	MS	F	P
Full model, all parameters free					
Treatments	173.04	68	2.55	28.882	< 0.0001
Error	13.656	155	0.88		
Data pooled across species, all parameters free					
Treatments	168.325	32	5.26	54.70	< 0.0001
Error	18.267	191	0.10		
Data pooled across species, a = 5.72					
Treatments	2.859	23	0.124	20.179	< 0.0001
Error	0.837	135	.0062		
Data pooled across species, a = 1.96					
Treatments	2.852	23	0.1240	22.179	< 0.0001
Error	0.756	135	.0056		

2.8 FIGURE CAPTIONS

Figure 2.1. Logistic growth curves fitted to plant mass data. Curves with the same letters above them are not statistically different from one another. Curves indicated with an asterisk could not be statistically compared to *Bromus briziformis* in large pots with low water. Curves indicated with two asterisks could not be statistically compared to curves of either species large pots with low water; see text.

Figure 2.2. Logistic growth curves fitted to plant mass data pooled across species. Open circles: *Bromus briziformis*, closed circles: *Bromus tectorum*. Curves with the same letters above them are not statistically different from one another. Parameter values are given in figure 2.3. Growth curves of plants in large pots with low water could not be statistically compared to growth curves of plants in small pots with medium water (indicated with asterisks); see text.

Figure 2.3. Parameter values with confidence intervals for the logistic growth curves in figure 2.2. Parameters with the same letters above them do not differ significantly from one another. Parameters were only tested for equality between treatments if the whole curves differed (see figure 2.2). Growth curves of plants in large pots with low water could not be statistically compared to growth curves of plants in small pots with medium water; see text.

Figure 2.4. Logistic growth curves fitted to plant masses from the first nine harvest dates; the size parameter was fixed at 5.72. Open circles: *Bromus briziformis*, closed circles: *Bromus tectorum*. Curves with the same letters above them are not statistically different from one another. Parameter values are given in figure 2.6.

Figure 2.5. Logistic growth curves fitted to plant masses from the first nine harvest dates; the size parameter was fixed at 1.96. Symbols and lettering as in figure 2.4. Parameter values are given in figure 2.7.

Figure 2.6. Parameter values with confidence intervals for the logistic growth curves in figure 2.4; the maximum plant size parameter was set to 5.72. Parameters with the same letters above them do not differ significantly from one another. Parameters were only tested for differences between treatments when whole curves were statistically different from one another.

Figure 2.7. Parameter values with confidence intervals for the logistic growth curves in figure 2.4; the maximum plant size parameter was set to 1.96. Symbols and lettering as in figure 2.5.

2.9 FIGURES

Figure 2.1

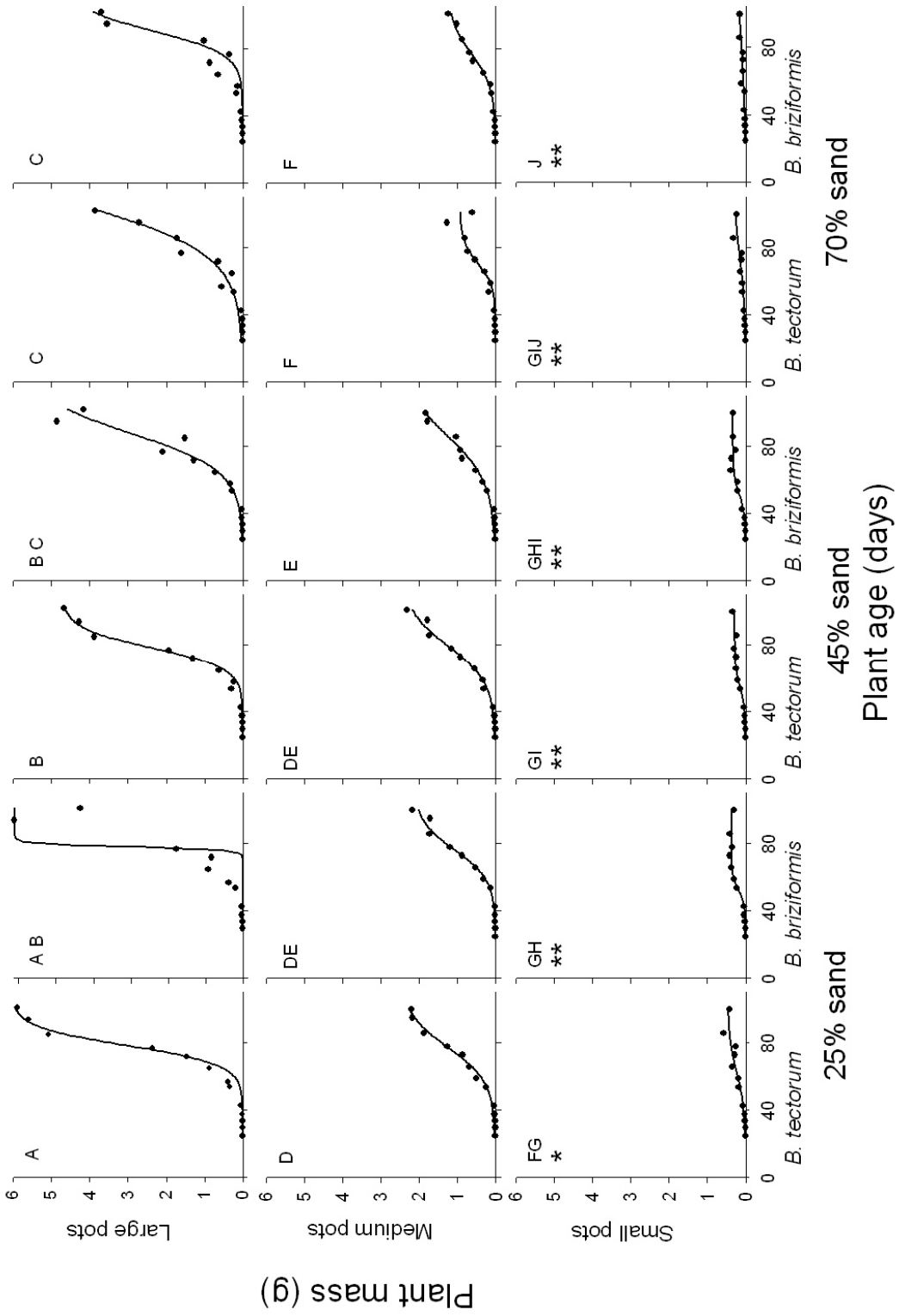


Figure 2.2

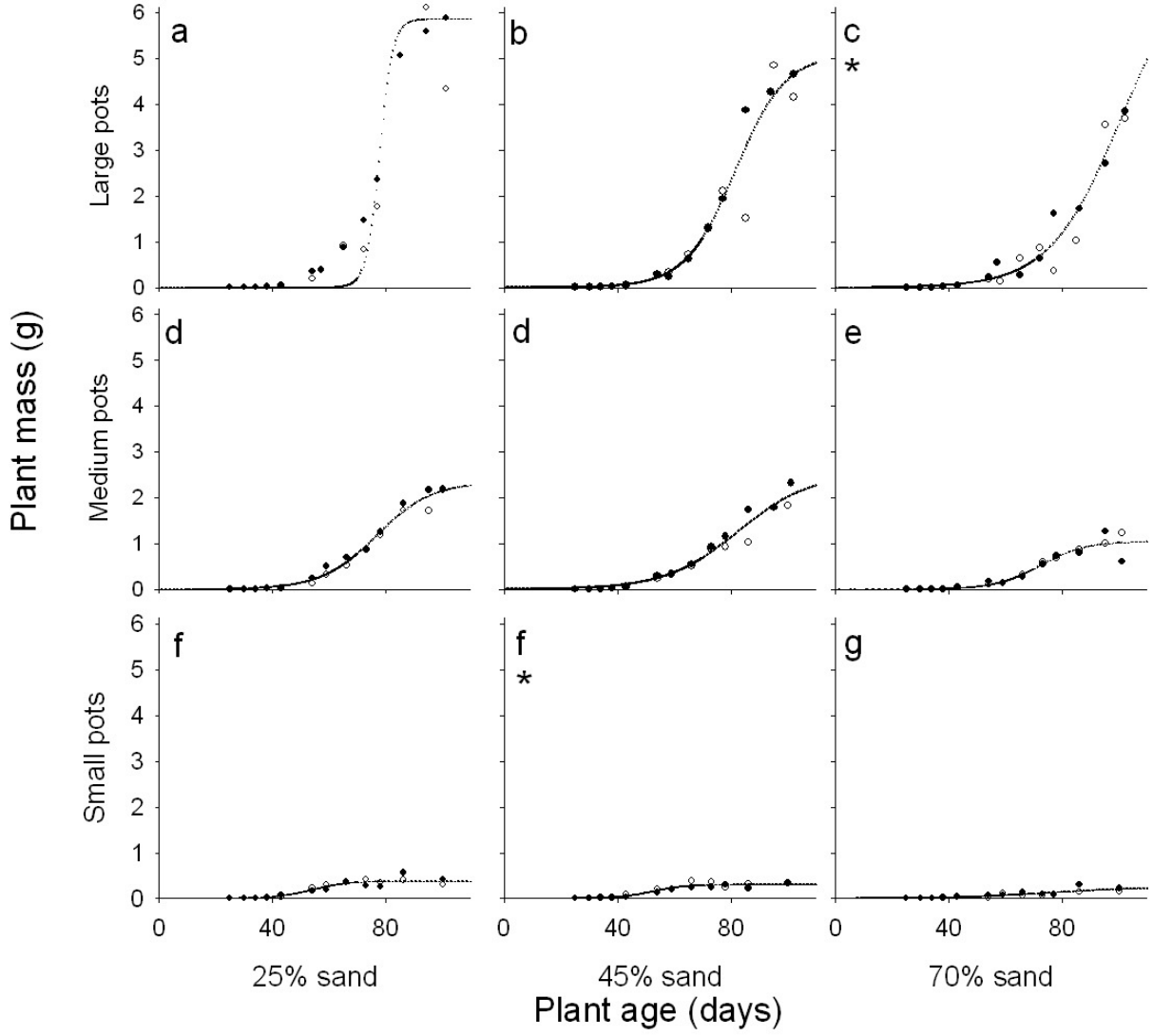

Plant mass (g)
Large pots
Medium pots
Small pots
a
b
c
*
d
d
e
f
f
*
g
0
1
2
3
4
5
6
0
40
80
25% sand
45% sand
70% sand
Plant age (days)

Figure 2.3

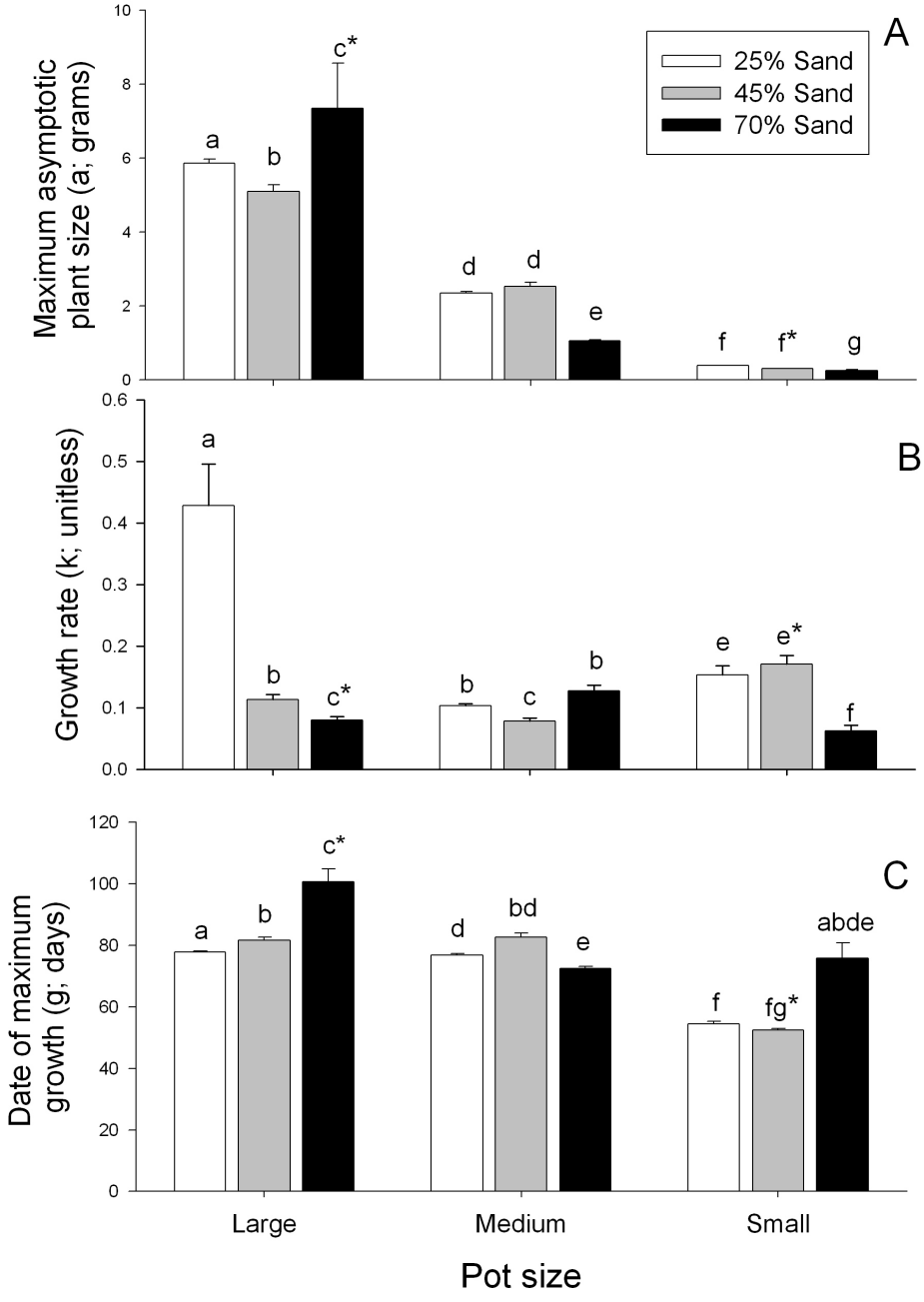

A
25% Sand
45% Sand
70% Sand
Maximum asymptotic plant size (a; grams)
a
b
c*
d
d
e
f
f*
g
B
Growth rate (k; unitless)
a
b
c*
b
c
b
e
e*
f
C
Date of maximum growth (g; days)
a
b
c*
d
bd
e
f
fg*
abde
Large
Medium
Small
Pot size

Figure 2.4

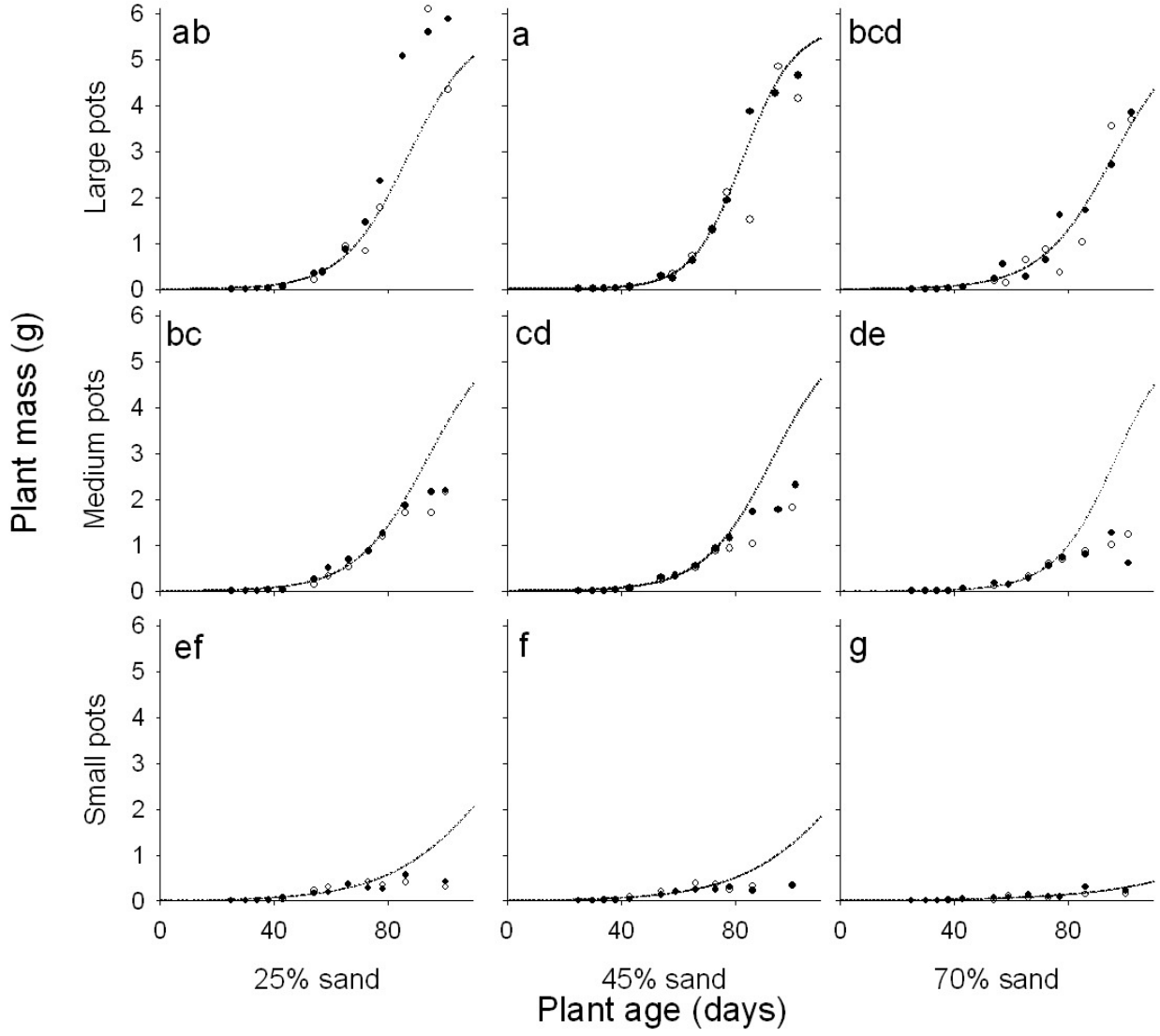

ab
a
bcd
bc
cd
de
ef
f
g
Large pots
Medium pots
Small pots
Plant mass (g)
0
1
2
3
4
5
6
0
40
80
25% sand
45% sand
70% sand
Plant age (days)

Figure 2.5

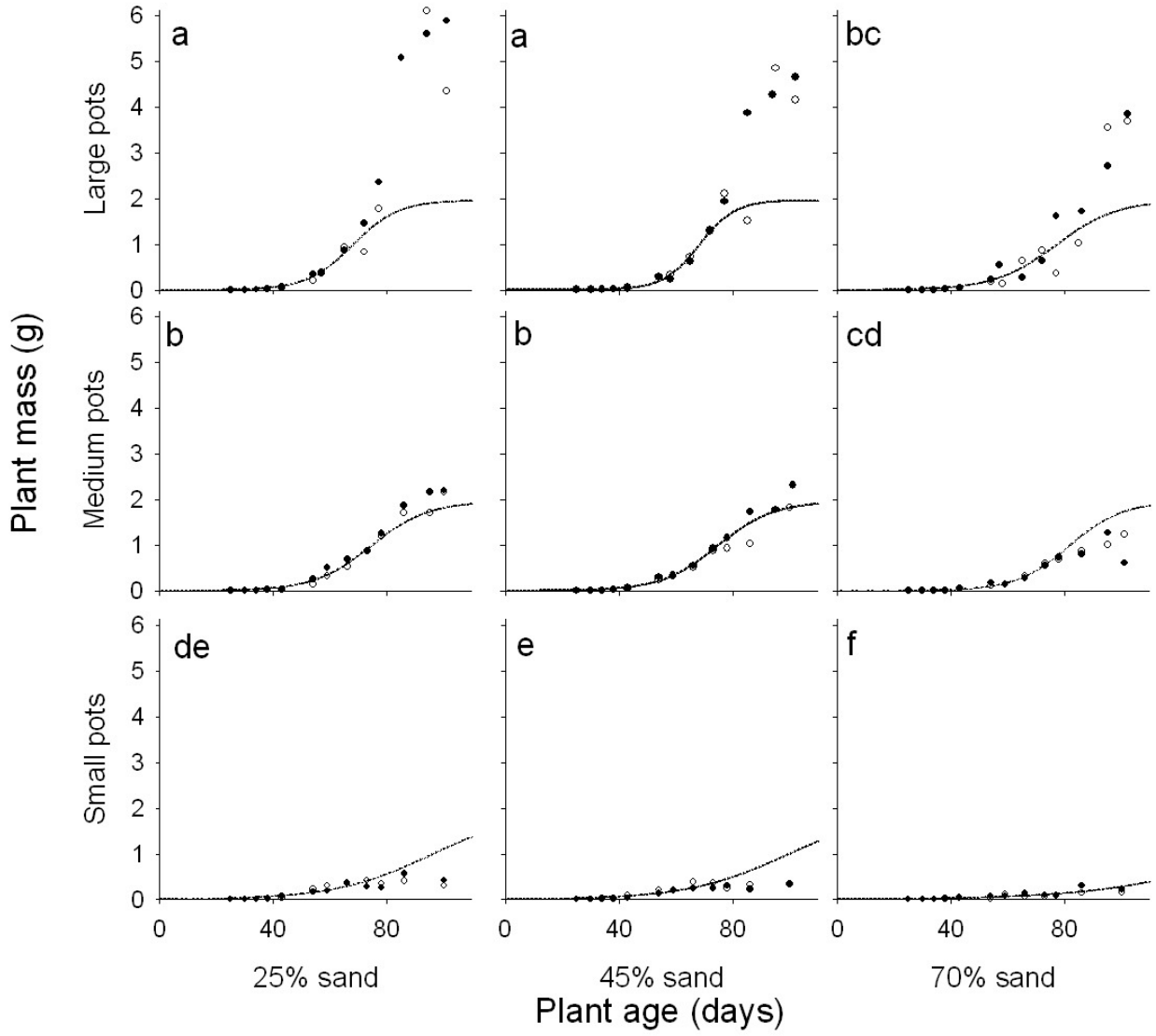

a
a
bc
b
b
cd
de
e
f
Large pots
Medium pots
Small pots
Plant mass (g)
0
1
2
3
4
5
6
0
40
80
25% sand
45% sand
70% sand
Plant age (days)

Figure 2.6

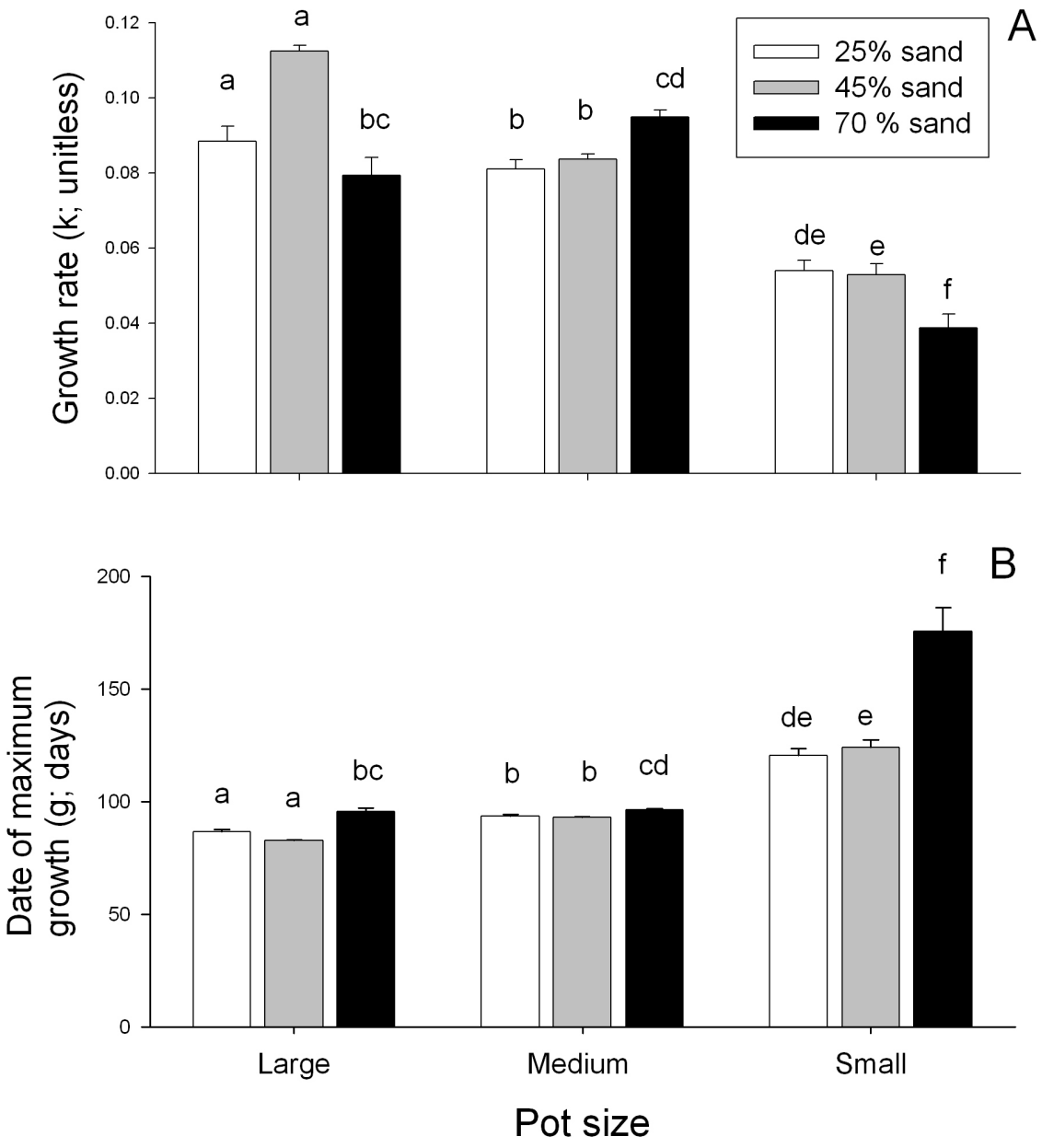
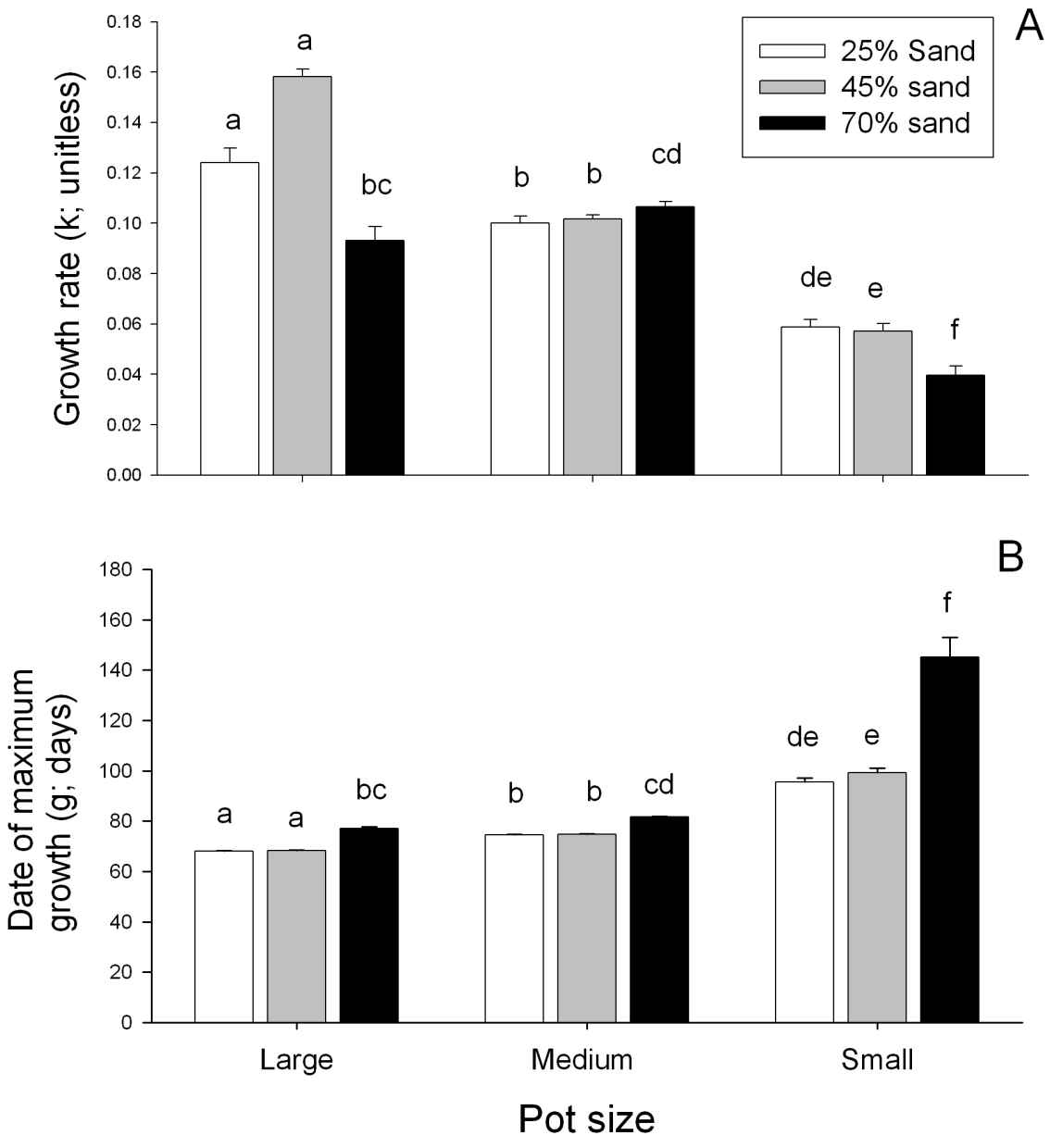

A
25% sand
45% sand
70 % sand
Growth rate (k; unitless)
0.12
0.10
0.08
0.06
0.04
0.02
0.00
a
a
bc
b
b
cd
de
e
f
B
Date of maximum growth (g; days)
200
150
100
50
0
a
a
bc
b
b
cd
de
e
f
Large
Medium
Small
Pot size

Figure 2.7


A
25% Sand
45% sand
70% sand
Growth rate (k; unitless)
0.18
0.16
0.14
0.12
0.10
0.08
0.06
0.04
0.02
0.00
a
a
bc
b
b
cd
de
e
f
B
Date of maximum growth (g; days)
180
160
140
120
100
80
60
40
20
0
a
a
bc
b
b
cd
de
e
f
Large
Medium
Small
Pot size

CHAPTER THREE

FERTILIZER AND WATERING EFFECTS ON SIZE AND SEED PRODUCTION OF TWO INTRODUCED ANNUAL *BROMUS* SPECIES DO NOT TRANSLATE INTO MEASURABLE EFFECTS ON POPULATION GROWTH RATES.

3.1 ABSTRACT

Biological invasion is a population-level phenomenon, but invasive species are often studied at the level of individuals. We investigated population growth responses to plant nitrogen content and $\delta^{13}C$ (an index of metabolic efficiency) in experimental populations of *Bromus tectorum* and *B. briziformis* subjected to watering and fertilization treatments at two sites. We used linear and logistic regression models to predict plant growth and reproduction of simulated populations with nitrogen and $\delta^{13}C$ adjusted relative to observed responses. Observed and simulated population data were analyzed using matrix population models. *B. tectorum* plants were larger than *B. briziformis* plants at all field sampling periods. *B. tectorum* had lower leaf nitrogen content, produced three times as many seeds as and in most cases had higher population growth rates compared to *B. briziformis*. Fertilized plants had higher leaf nitrogen content and were more depleted in $\delta^{13}C$ than unfertilized plants. However, differences in population growth rates were limited to species and site differences and probably resulted from a difference in planting dates for the two sites. The individual-plant responses of size, nitrogen

content, $\delta^{13}C$ and seed-production resulting from our watering and fertilizer treatments did not affect population growth rates.

3.2 INTRODUCTION

Biological invasion is a population-level problem (Parker 2000) that is dependent on interactions between the physiology of the species and the invaded community. Many studies have investigated the effects of environmental conditions on population demographic parameters (i.e. survivorship, growth and fecundity; Kelly 1989, Pierson and Mack 1990, Rice and Mack 1991, Levin et al. 1996, McEvoy and Coombs 1999, Parker 2000, Gotelli and Ellison 2002, Gustafsson and Ehrlén 2003). Some infer physiological tolerances based on observed demographic parameters (D'Antonio et al. 1993, Radford and Cousens 2000), or simultaneously report demographic parameters along with physiological data (Donovan and Ehleringer 1994, Allen 1998, Erneberg 1999, Goodwin et al. 1999, Andersson et al. 2002).

Population growth rates depend on underlying demographic transitions (i.e. growth, survival, and fecundity) that are the inputs to population matrix models (Caswell 2001). Studies that include both physiological and demographic data provide correlative evidence for the linkage between the physiology of the individual and population growth, but often estimate only some of the transitions (e.g. survival or germination, but not fecundity). The importance of selected demographic transitions to population growth without complete demographic data is not clear because differences in transitions might not affect population growth rates (Caswell 1989). Where complete demographic data are available, the absence of statistical relationships between

demographic transitions and individual physiology precludes predictions linking these two levels of biological organization.

Our ability to predict which species may be invasive would be enhanced by studies that link individual-level physiology with population growth. Here, we used matrix population models to compare the population growth rates of two introduced species in response to water and fertilizer treatments. We then used linear and logistic regression to examine the dependence of underlying growth, survival and reproduction on leaf nitrogen concentration and leaf $\delta^{13}C$. We varied leaf nitrogen and $\delta^{13}C$ values and used regression to predict vital rate inputs to population models and generated population growth rates.

STUDY SYSTEM

Bromus tectorum is a Eurasian native naturalized in the continental United States and invasive west of the Rocky Mountains (USDA NRCS 2009, Zouhar 2003). *Bromus briziformis* is also a Eurasian native but is restricted in its US distribution to western and northeastern states and is not listed as a noxious weed (USDA NRCS 2009). *Bromus briziformis* is common in eastern Washington and northern Idaho but rarely forms dense, monospecific stands (pers. obs.).

Little is known about the ecophysiology of *B. briziformis*, but a 1955 study (Hulbert 1955) examined the distribution of several *Bromus* species across a moisture gradient extending from eastern Washington to western Montana. *B. tectorum* occurred across the entire gradient but *B. briziformis* was restricted to mesic sites at low densities. Hulbert (1955) suggested that soil moisture may influence the abundance of the two *Bromus* species in eastern Washington, with the greater abundance of *B. tectorum* being explained by a higher resistance to drought. Nevertheless, other studies within the proposed study area (Mack and Pyke 1983, Mack and

Pyke 1984, Rice and Mack 1991) have shown that population demography of *B. tectorum* is dependent on soil moisture due to drought-induced seedling mortality and reduced fecundity.

We chose $\delta^{13}C$ and leaf nitrogen to indicate plant physiological status (Huxman et al. 2008). In C3 plants if photosynthetic demand for carbon increases relative to stomatal conductance, discrimination against ^{13}C decreases and $\delta^{13}C$ of fixed carbon increases (McCarroll and Loader 2004). Thus $\delta^{13}C$ can be affected both by photosynthetic capacity and by plant water status and may best be interpreted as a time-integrated index of plant function and efficiency (Dawson et al. 2002).

Nitrogen frequently is a limiting resource in grassland ecosystems (Wilson and Tilman 1991, Hook and Burke 1995, Burke et al. 1997). Booth et al. (2003) conducted a competition experiment with *B. tectorum*, *Artemisia tridentata* and *Elymus elymoides*, in the Great Basin of the western United States and concluded that competition for nitrogen was secondary to competition for water in determining competitive interactions. Hulbert (1955) added nitrogen to naturally occurring stands of *B. tectorum* in northern Idaho and observed that individuals in fertilized plots survived longer than individuals in unfertilized plots following the onset of summer drought. He suggested that plants in fertilized plots expanded their root systems to a greater extent than plants in unfertilized plots, providing better access to soil moisture under drought conditions. Leaf nitrogen concentration in *B. tectorum* increases with increasing soil nitrogen availability (Link et al. 1995), possibly due to reduced nitrogen use efficiency (Chapin 1980).

Our objectives were to compare the population growth rates of two introduced species and their responses to supplemental fertilizer and water. We analyzed leaf $\delta^{13}C$ and nitrogen concentrations, and tracked plant size and lifetime seed production to explain the population

growth patterns we observed. Finally we assessed regression-based population simulations to determine the importance of individual leaf nitrogen and $\delta^{13}C$ status for population growth.

3.3 METHODS

Seeds of *B. tectorum* and *B. briziformis* were collected from Elk Creek Falls Recreation area in north-central ID (46˚ 44.2' W, 116˚ 10.5' N) within the *Pseudotsuga menziesii* / *Holodiscus discolor* habitat type (Daubenmire and Daubenmire 1968) in August 2005. Seeds were collected from several hundred individuals of each species, thoroughly mixed, and stored at room temperature until planting. Laboratory germination of seeds in December 2006 was 98 ± 2.6% for *B. briziformis* and 100% for *B. tectorum* (data not shown).

Plots were established within the *Festuca idahoensis/ Symphoricarpos albus* vegetation zone at Smoot Hill (Hudson Biological Reserve at Smoot Hill, Washington State University; 46˚ 49.5' N, 117˚ 14.2' W) and within the *Agropyron spicatum* / *Festuca idahoensis* vegetation zone (Daubenmire 1970) at the Escure Ranch (Rock Creek Management Unit, Bureau of Land Management; 47˚ 0.8' N, 117˚ 56.6' W). The study areas at both sites were on soils that previously had been disturbed. Study areas were plowed with a tractor-mounted plow and then tilled.

Plots at both sites were established on the nodes of an 18 x 18 m grid and each plot was marked. Of the 324 plots at each site, 240 were used for experimental plants (30 replicates x 2 species x 2 fertilizer treatments x 2 water treatments), 16 were used to monitor soil water (4 replicates x 2 fertilizer treatments x 2 water treatments) and the remaining 68 plots (17 replicates x 2 fertilizer treatments x 2 water treatments) at each site were monitored to estimate the

probability of wild-volunteer plants appearing on the experimental plots. Plots were randomly assigned to treatments and planted with a single seed. No volunteer plants were detected at Escure Ranch. At Smoot Hill, seven *B. tectorum* plants germinated in unplanted plots and five *B. tectorum* plants germinated in *B. briziformis* plots. Volunteer *B. tectorum* were included in the experimental populations according to the water and fertilizer treatments their plots received.

The Smoot Hill populations were planted on 17 and 18 November, 2005, and the Escure Ranch populations were planted on 4 December 2005. Grass caryopses of the appropriate species were planted mid-way between the two nails used to mark plots. Fertilizer was applied to treatment plots in a slow-release formulation (Osmocote Outdoor & Indoor Smart-Release Plant Food; The Scotts Company, Marysville, OH) at an approximate rate of 250 kg N/ha. Fertilizer pellets were raked into the top centimeter of soil within a 25-cm diameter circle centered on the plot. Watering treatments received one liter of water weekly between 14 May and 2 July 2006 (eight weeks), except for the week of 4 June 2006, when > 3 cm of rain fell at both sites. During the growing season, plots were weeded to maintain bare soil for a minimum distance of 25 cm from the center of each plot. For brevity, we refer to our treatments as C (control), W (water), F (Fertilizer), and FW (fertilizer plus water).

Plants were censused on 15 January, 2 April, 1 May, 19 May, 6 June, and 23 June, 2006. Plants were recorded if they were within 2 cm of the plot. On 15 January and 2 April, plants were scored as germinated or not germinated. The remaining censuses were photographically recorded for plant size estimates. For these censuses a 25 x 25 cm PVC frame was placed on the ground around the plant and the plot was photographed with a digital camera. The shooting angle between the lens and the ground surface was 60 degrees to maximize visible leaf area in the image (Campbell 1977). The images were imported into MATLAB Image Processing

Toolbox (The MathWorks, Natick, MA) and analyzed. Images were converted to binary images and plant size in pixels was estimated and converted to leaf area. A test set of *B. tectorum* plants were photographed, the aboveground parts harvested, dried and weighed, and the images processed as above. Regression analysis showed that

dry biomass (g) = 0.0064 * leaf area (cm^2) – 0.392, $R^2 = 0.89$, n = 19.

After the 19 May, 6 June, and 23 June censuses, two new (< 2cm long) leaves per plant were marked with a loop of colored wire. After the plants had senesced in late July, marked leaves were collected and analyzed for percent nitrogen and $\delta^{13}C$ using a Eurovector elemental analyzer and Micromass isotope ratio mass spectrometer (Manchester, UK). Carbon isotope (Helliker and Ehleringer 2002, Harlow et al. 2007) and nitrogen content data were assumed to reflect plant status during the census immediately after which the leaves were marked. This assumption is imperfect due to retranslocation of nitrogen from senescing leaves (Chapin 1980) but was considered preferable to destructive harvest of growing leaf tissues.

Between 3 June and 27 July 2006, seeds were collected from individual plants when the inflorescences had lost all green color or when the inflorescence tissues had lost turgor. In most cases it was necessary to collect seeds several times from each plant. Seeds were cleaned by hand and weighed. Seed quantities were estimated based on regressions of seed count on mass from a subsample of our seeds:

B. tectorum seed count = 274.6 * grams of seed + 19.8, $R^2 = 0.72$, n = 19;

B. briziformis seed count = 199.6 * grams of seed – 11.1, $R^2 = 0.97$, n = 15.

Leaf percent nitrogen, leaf $\delta^{13}C$ (19 May, 6 June, and 23 June censuses), and plant leaf area were analyzed in SAS (SAS Institute Inc. 2000) with repeated measures analysis of variance (RM-ANOVA) using plant as the repeated-measures subjects and time, site, species, fertilizer,

and water as fixed effects; we used a Bonferroni-Holm adjustment for multiple comparisons of least squares means (Wright 1992). Leaf percent nitrogen and plant size were log-transformed to meet assumptions of normality. Total seed production ("lifetime fecundity") of plants that survived to produce seed was log-transformed and subjected to analysis of variance with site, species, fertilizer and water as fixed effects and Bonferroni-Holm adjustments for multiple comparisons.

We fitted periodic matrix population models (Caswell 2001) to the seed-production and census data from each of the 16 experimental populations we had established. Size classes were assigned to individuals using an algorithm (Moloney 1986, Caswell 2001) that simultaneously minimizes distribution errors (treating different individuals within a size class as identical) and sampling errors (capturing too few individuals in each size class). Size classes were assigned using the pooled data from all populations to facilitate comparison between models; several of the smaller size classes were later combined to maintain adequate cell counts in the 16 individual population models (table 3.1).

Standard population matrix models use a single transition matrix describing fecundities and size-class transition probabilities to model population growth through one reproductive cycle, often one year:

[1] $\mathbf{N_{t+1}} = \mathbf{A} \times \mathbf{N_t}$

where **N** is a vector of census counts of individuals in each size class in the population, **A** is a square matrix containing fecundities and transition probabilities of all size classes in the population, and **t** is an arbitrary point in time. Periodic matrix models use several seasonal transition matrices to project population growth through a reproductive cycle and are useful in situations where intra-annual variation is important to population dynamics, as is the case for *B.*

tectorum and many other annual species (Mack and Pyke 1983, Caswell 2001). A periodic matrix model for a system with *s* seasons in a reproductive cycle replaces **A** with a series of *s* seasonal matrices that need not be square:

[2] $\mathbf{N_{t+1}} = \mathbf{A_s} \times \mathbf{A_{s-1}} \times \ldots \times \mathbf{A_1} \times \mathbf{N_t}$.

The matrix product of the *s* seasonal matrices,

[3] $\mathbf{A_s} \times \mathbf{A_{s-1}} \times \ldots \times \mathbf{A_1}$

is a square matrix with properties identical to a standard matrix model, including a population growth rate, λ, which is the dominant eigenvalue of the matrix.

Our periodic matrix models included seven transition matrices (Appendix A): one for each of the six censuses, and one to represent seeds produced between the final census and plant senescence near the end of July. Ninety-five percent confidence intervals for λ from these population models were calculated using bias-corrected bootstrap percentile intervals (Caswell 2000) from 4000 bootstrapped samples of the data.

Comparisons of λ between treatments were made using permutation tests (Caswell 2000) with a Bonferroni-Holm correction for multiple comparisons. Four thousand permutation samples of the data were constructed by randomly pairing observed demographic data for each plant with one of the sixteen treatments in the study while preserving sample sizes for each treatment.

For the simulated population models, demographic data were separated into four treatments (two sites by two species) and periodic matrix models were constructed as above (Appendix B). We used linear regression to estimate the contributions of site, species, size class, $\delta^{13}C$ and log percent N to seed production during the 23 June and 27 July censuses (Appendix C1). We used logistic regression to estimate the contributions of site, species, $\delta^{13}C$, and log

percent nitrogen to the transition probabilities during the 19 May, 6 June and 23 June censuses (Appendix C2). We attempted to fit one regression model for each observed transition during those three periods, but for 18 of the 56 observed transitions it was not possible to fit regression models due to insufficient data or perfect separation of the response data.

We generated four simulation data sets by varying the $\delta^{13}C$ or log percent N values by one standard deviation. We then used model parameters from the regressions (Appendix C) to generate seed production and demographic transition data for each plant. Plants were assigned to transitions for which the predicted probability was highest among the available logistic regression models. Where no regression models were available to predict the transition of a simulated plant the simulated plant was assigned as observed for its empirical counterpart in the field. We also used field data to generate transition matrices for the 15 January, 2 April, and 1 May censuses and seed production data for the 19 May census because limited leaf tissue data were available for those censuses. We used the simulation data to construct transition matrices for the remaining censuses and compared these growth models to the observed population models (Appendix B). We constructed bootstrap percentile intervals for population growth rates and compared growth rates among treatments using permutation tests as above.

We report only differences for which we obtained statistical comparisons with $p < 0.05$; we take $p > 0.05$ to indicate no difference between treatments.

3.4 RESULTS

FIELD DATA

Population growth rates of *Bromus tectorum* generally were higher compared to population growth rates for *B. briziformis*. *B. tectorum* individuals were larger, produced more

seeds, and had lower leaf nitrogen content compared to *B. briziformis* individuals. Fertilized plants of both species had higher leaf nitrogen concentrations and at Escure Ranch were larger and produced more seed compared to unfertilized plants. Fertilized *B. tectorum* plants at Escure Ranch had higher population growth rates than any of the *B. briziformis* populations and any Smoot Hill populations. Growth rates of simulated populations were insensitive to leaf $\delta^{13}C$ values but responded negatively to increasing leaf nitrogen content for *B. briziformis* plants at Escure Ranch.

Leaf nitrogen concentrations on all three sampling dates were higher for *B. briziformis* compared to *B. tectorum* in the F and FW treatments (figure 3.1, table 3.2). Leaf nitrogen concentrations were higher for *B. tectorum* in the F and FW treatments compared to *B. briziformis* in the W and C treatments (figure 3.1, table 3.2). In the W and C treatments, leaf nitrogen concentrations were higher for *B. briziformis* compared to *B. tectorum* plants (figure 3.1, table 3.2). On 19 May, plants in the F and FW treatments had higher leaf nitrogen concentrations compared to control plants, and plants in the F treatment had higher leaf nitrogen concentrations compared to plants in the W treatment (figure 3.1, table 3.2). On 6 June and 23 June, plants in the F and FW treatments had higher leaf nitrogen concentrations compared to controls and plants in the W treatment (figure 3.1, table 3.2).

Leaf ^{13}C was more depleted for plants at Escure Ranch compared to plants at Smoot Hill (figure 3.2, table 3.3). Smoot Hill plants in the W and WF treatments were more depleted in leaf ^{13}C compared to plants in the C and F treatments (figure 3.2, table 3.3). Plants in the F and FW treatments did not differ from plants in the W or C treatments on 19 May but had more positive $\delta^{13}C$ compared to plants in the W or C treatments on 6 June and 23 June (figure 3.2, table 3.3). All plants in the W treatment and *B. tectorum* plants in the C treatment were more depleted in

leaf ^{13}C compared to all plants in the F treatment and *B. briziformis* plants in the C and FW treatments (figure 3.2, table 3.3).

B. tectorum plants had more leaf area compared to *B. briziformis* plants on every sampling date, and the size differences increased throughout the summer (figure 3.3, table 3.4). *B. tectorum* plants at Escure Ranch had more leaf area compared to *B. briziformis* plants at Escure Ranch, and more leaf area compared to all plants at Smoot Hill (figure 3.3, table 3.4). Plants at Escure Ranch had more leaf area compared to plants at Smoot Hill on the 1 May sampling date but not on later sampling dates (figure 3.3, table 3.4). Plants in the F and FW treatments at Escure Ranch had more leaf area compared to plants in the W and C treatments at Escure Ranch and all plants at Smoot Hill (figure 3.3, table 3.4).

Among plants that survived to produce seed, *B. tectorum* seed production was nearly three times greater than *B. briziformis* seed production (figure 3.4, table 3.5). Plants in the FW and F treatments at Escure Ranch produced more seeds compared to plants in the W or C treatments at Escure Ranch or plants in the F or FW treatments at Smoot Hill (figure 3.4, table 3.5).

The population growth rates of *B. tectorum* plants in the F and FW treatments at Escure Ranch were greater than growth rates of all *B. briziformis* populations at both sites (figure 3.5; permutation test $p < 0.05$). Population growth rates of *B. tectorum* plants in the F and FW treatments at Escure Ranch also were greater than growth rates of *B. tectorum* at Smoot Hill in the C, F and FW treatments (figure 3.5; permutation test $p < 0.05$). The population growth rate of *B. tectorum* in the W treatment at Escure Ranch was greater than the population growth rate of any *B. briziformis* population at Smoot Hill, and greater than the growth rate of control *B. briziformis* plants at Escure Ranch (figure 3.5, $p < 0.05$). The population growth rate of *B.*

tectorum in the W treatment at Smoot Hill was greater than the population growth rate of *B. briziformis* in the C and W treatments at Smoot Hill (figure 3.5, $p < 0.05$).

MODELING DATA

For the population simulations, demographic data were combined across fertilizer and water treatments and analyzed as four populations (two species at each of two sites). Among these four populations, *B. tectorum* at Escure Ranch had a greater population growth rate than *B. briziformis* at Smoot Hill (figure 3.6, zero offsets; permutation test $p < 0.05$) and no other populations were significantly different from one another. Increasing or decreasing leaf $\delta^{13}C$ by one standard deviation did not change simulated growth rates positively or negatively (figure 3.6a). Simulated growth rates for both species at Smoot Hill were insensitive to leaf nitrogen content (figure 3.6b). At Escure Ranch, increasing leaf nitrogen content caused the simulated growth rate of *B. tectorum* to increase and the simulated growth rate of *B. briziformis* to decrease (figure 3.6b). When log leaf nitrogen contents were increased by one standard deviation population growth rates of *B. briziformis* were significantly lower than *B. tectorum* at Escure Ranch (figure 3.6b, permutation test $p < 0.05$).

3.5 DISCUSSION

We found that *B. tectorum* grew larger, produced more seed and had lower leaf nitrogen contents than *B. briziformis*. Second, watering effects on leaf $\delta^{13}C$ and fertilizer effects on plant growth were site-specific. Watering did not affect plant size or seed production, and watering or fertilizer treatments did not affect population growth rates. Third, differences in population

growth rates between sites probably resulted from differences in winter and spring mortality of seedlings because growth patterns of survivors were not substantially different between sites.

Estimates of population growth rates were between 129 and 1482 (figure 3.5) suggesting rapidly growing populations. At least two factors contributed to the large population growth rates: first, plants were grown in the center of 0.2 m^2 weeded plots and experienced little or no competition. *B. tectorum* seed production generally declines with increasing plant density (Hulbert 1955), so plants grown with competition would likely produce less seed than we report. Second, we estimated demographic parameters between fall and late summer, and did not consider seed survival between seed production and germination. No estimates of first-season seed survival are available, but others (Hulbert 1955, Mack and Pyke 1983, Humphrey and Schupp 2001) have reported seed production or summer seed bank densities comparable to our seed production rates. Our population growth rates, which are generally high, are indicators of relative population performance.

Bromus tectorum plants were larger (figure 3.3, table 3.4), produced more seed (figure 3.4, table 3.5), and had higher population growth rates (figure 3.5) than *B. briziformis*. Few comparative studies of *B. briziformis* with other species exist, but the two that compare *B. briziformis* to *B. tectorum* both found that *B. tectorum* growth was more robust than *B. briziformis* growth (Hulbert 1955, Robocker 1973). Robocker (1973) found that *B. tectorum* in our region produced 40 – 70 % more aboveground biomass than *B. briziformis* during three of four growing seasons. In addition, Hulbert (1955) noted that *B. tectorum* appeared to be more winter-hardy and flowered earlier than *B. briziformis*. In our study, differences in lifetime fecundity (figure 3.4) explained the larger population growth rates of *B. tectorum* relative to *B. briziformis.*

Leaf $\delta^{13}C$ in fertilized plants was less depleted than in unfertilized plants by 0.9‰ and 1.2‰ on 6 June and 23 June, respectively (figure 3.2). Fertilizer additions may have stimulated greater photosynthetic rates relative to unfertilized plants and thus have increased the demand for carbon within plant leaves, assuming stomatal conductance did not change. Carbon isotope discrimination has been positively correlated with yield in wheat (Condon et al. 1987), and negatively correlated with dry matter production in peanuts (Hubick et al. 1988) leading Farquhar et al. (1989) to conclude that the relationship between carbon discrimination and production is difficult to predict. In our study, less discrimination by fertilized plants probably reflects greater photosynthetic demand for carbon by the fertilized plants compared to the unfertilized plants (Dawson et al. 2002). The site-specific difference in $\delta^{13}C$ of 0.6‰ in response to water (figure 3.2), while significant, is too small to be interpreted with confidence (O'Leary et al. 1992).

Fertilized plants had higher leaf nitrogen concentrations than unfertilized plants at both sites (figure 3.1B, 3.1C) but fertilizer effects on plant size (figure 3.3) and lifetime fecundity (figure 3.4) were site-specific. Link (1995) reported increased *B. tectorum* biomass in response to nitrogen amendments, but only when plants were supplemented with both N and water. Hulbert (1955) reported increased seed production by *B. tectorum* in response to N amendments. Hulbert (1955) suggested that N amendments to *B. tectorum* may promote below-ground production. We did not measure below-ground biomass in these plants but found that both *B. tectorum* and *B. briziformis* reduced root-to-shoot ratios in greenhouse pots with more added nitrogen, compared to pots with limited nitrogen (chapter 1).

In spite of responses of leaf $\delta^{13}C$, plant size and lifetime fecundity to water and fertilizer additions, we observed no systematic differences in population growth rates (figure 3.5).

Physiological and ecological effects do not always scale up or down across levels of biological organization (Stohlgren et al. 1999, DeLucia et al. 2001, Knight and Reich 2005). Alvarez-Cobelas et al. (2007) report that individual-plant growth of *Cladium mariscus* (cut-sedge) was positively correlated with water level and phosphorus availability, but showed saturating or negative responses to water and phosphorus at the level of standing crop or plant cover. Our results add to the body of evidence suggesting that the level of biological organization at which responses are measured can limit the conclusions drawn for other levels of organization.

We observed differences in population growth rates between sites (figure 3.5), possibly due to a two week difference in planting dates between the sites, to soil nutrient status, or other unmeasured environmental differences. The populations that were planted earlier (Smoot Hill) were planted before first frost and experienced substantial winter germination and spring mortality of seedlings, whereas the populations that were planted later (Escure Ranch) were planted into frozen soil and experienced less winter germination and lower spring mortality of seedlings (Appendix A). Mack (1984) reported that winter grazing by voles caused considerable mortality among *B. tectorum* populations. Plants in our 2 April and 1 May censuses that were recorded as dead usually were missing entirely (data not shown) suggesting that grazing was a cause of mortality among our *Bromus* populations, too.

Leaf N effects on simulated population growth rates were inconsistent across sites and tended to be species-specific (figure 3.6). In both species, leaf nitrogen concentrations were higher for fertilized plants compared to unfertilized plants (figure 3.1), but increasing nitrogen concentrations in simulated *B. tectorum* populations resulted in greater growth rates, and lesser growth rates in simulated *B. briziformis* populations, compared to controls. It is not clear if our

unexpected result is a modeling artifact or if it is related to the efficiency with which *B. briziformis* retranslocates nitrogen from vegetative to reproductive tissues (Chapin 1980).

Leaf $\delta^{13}C$, plant size, leaf N concentration and fecundity all responded to our water and fertilizer treatments, but these did not translate into population-level responses. Our findings confirm the greater invasive potential of *B. tectorum* relative to *B. briziformis*; a rank ordering of the population growth rates we observed shows that all eight *B. tectorum* populations grew faster than all eight *B. briziformis populations*.

3.6 LITERATURE CITED

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3.7 TABLES

Table 3.1. Size classes for the population matrix models. Size classes for live germinated plants were assigned using Moloney's (1986) algorithm but several of the smaller size classes were combined to form the second size class for our models to maintain cell counts. Seeds and dead plants are biologically distinct demographic categories and were not included in the assignment algorithm

Size class	**Upper limit of leaf area (cm^2)**	**Number of size classes from algorithm**
Seed	Not applicable	Not applicable
1	181.8	1
2	428.9	5
3	885.1	1
4	1246.1	1
5	2958.1	1
7	7000	1
Dead	Not applicable	Not applicable

Table 3.2. Repeated measures analysis of variance with plants as subjects, time as a factor, log leaf nitrogen content (%) as the response variable and site, species, fertilizer and water as treatment variables. Highest order significant terms are indicated in the final column. The site by fertilizer by water interaction that here is indicated as significant collapsed to a simple fertilizer effect when the Bonferroni-Holm correction for multiple comparisons was applied.

Source	DF	F	P	Significant
Site	1,252	1.27	0.261	
Species	1,252	83.49	< 0.0001	
Fertilizer	1,252	155.36	< 0.0001	
Water	1,252	0.31	0.5784	
Time	2,468	2.57	0.0774	
Site*Species	1,252	2.1	0.1486	
Site*Fertilizer	1,252	6.11	0.0141	
Site*Water	1,252	0.89	0.3471	
Site*Time	2,468	3.0	0.0507	
Species*Fertilizer	1,252	6.2	0.0134	*
Species*Water	1,252	0.48	0.4873	
Species*Time	2,468	0.86	0.4248	
Fertilizer*Water	1,252	0.68	0.4099	
Fertilizer*Time	2,468	0.13	0.8775	
Water*Time	2,468	1.55	0.2126	
Site*Species*Fertilizer	1,252	0.15	0.6995	
Site*Species*Water	1,252	1.1	0.2954	
Site*Species*Time	2,468	0.12	0.8854	
Site*Fertilizer*Water	1,252	5.09	0.0249	*
Site*Fertilizer*Time	2,468	0.83	0.436	
Site*Water*Time	2,468	0.37	0.6933	

Table 3.2, continued.

Source	DF	F	P	Significant
Species*Fertilizer*Water	1,252	0.09	0.7604	
Species*Fertilizer*Time	2,468	0.09	0.9128	
Species*Water*Time	2,468	1.11	0.3306	
Fertilizer*Water*Time	2,468	3.47	0.0319	*
Site*Species*Fertilizer*Water	1,252	0.29	0.5929	
Site*Species*Fertilizer*Time	2,468	0.39	0.6788	
Site*Species*Water*Time	2,468	1.02	0.3621	
Site*Fertilizer*Water*Time	2,468	0.44	0.6472	
Species*Fertilizer*Water*Time	2,468	2.8	0.0616	
Site*Species*Fertilizer*Water*Time	2,468	0.42	0.6557	

Table 3.3. Repeated measures analysis of variance with plants as subjects, time as a factor, leaf $\delta^{13}C$ as the response variable and site, species, fertilizer and water as treatment variables. Highest order significant terms are indicated in the final column.

Source	DF	F	P	Significant
Site	1,252	145.89	<.0001	
Species	1,252	27.44	<.0001	
Fertilizer	1,252	48.39	<.0001	
Water	1,252	8.08	0.0048	
Time	2,466	5.12	0.0063	
Site*Species	1,252	0.54	0.4642	
Site*Fertilizer	1,252	3.37	0.0674	
Site*Water	1,252	6.17	0.0136	*
Site*Time	2,466	0.5	0.6061	
Species*Fertilizer	1,252	6.71	0.0101	
Species*Water	1,252	0.11	0.7408	
Species*Time	2,466	0.95	0.3884	
Fertilizer*Water	1,252	6.93	0.009	
Fertilizer*Time	2,466	5.47	0.0045	*
Water*Time	2,466	0.88	0.415	
Site*Species*Fertilizer	1,252	0.26	0.6112	
Site*Species*Water	1,252	0.06	0.8024	
Site*Species*Time	2,466	0.1	0.901	
Site*Fertilizer*Water	1,252	1.53	0.2166	
Site*Fertilizer*Time	2,466	0.11	0.8925	
Site*Water*Time	2,466	0.27	0.7671	
Species*Fertilizer*Water	1,252	7.33	0.0072	*
Species*Fertilizer*Time	2,466	0.6	0.5505	

Table 3.3, continued.

Source	DF	F	P	Significant
Species*Water*Time	2,466	0.67	0.5116	
Fertilizer*Water*Time	2,466	0.4	0.6734	
Site*Species*Fertilizer*Water	1,252	0.25	0.6144	
Site*Species*Fertilizer*Time	2,466	0.88	0.4155	
Site*Species*Water*Time	2,466	0.2	0.8196	
Site*Fertilizer*Water*Time	2,466	1.0	0.3687	
Species*Fertilizer*Water*Time	2,466	0.88	0.4141	
Site*Species*Fertilizer*Water*Time	2,466	0.68	0.5057	

Table 3.4. Repeated measures analysis of variance with plants as subjects, time as a factor, log leaf area (cm^2) as the response variable and site, species, fertilizer and water as treatment variables. Highest order significant terms are indicated in the final column.

Source	DF	F	P	Significant
Site	1,258	21.73	< 0.0001	
Species	1,258	257.43	< 0.0001	
Fertilizer	1,258	1.37	0.2434	
Water	1,258	0.0	0.956	
Time	3,730	966.96	< 0.0001	
Site*Species	1,258	18.33	< 0.0001	*
Site*Fertilizer	1,258	14.12	0.0002	*
Site*Water	1,258	0.04	0.8513	
Site*Time	3,730	24.55	< 0.0001	*
Species*Fertilizer	1,258	0.0	0.9773	
Species*Water	1,258	0.94	0.3344	
Species*Time	3,730	7.23	< 0.0001	*
Fertilizer*Water	1,258	0.33	0.5652	
Fertilizer*Time	3,730	1.11	0.3445	
Water*Time	3,730	1.29	0.2764	
Site*Species*Fertilizer	1,258	0.09	0.7601	
Site*Species*Water	1,258	0.29	0.5938	
Site*Species*Time	3,730	0.82	0.484	
Site*Fertilizer*Water	1,258	0.28	0.5976	
Site*Fertilizer*Time	3,730	0.08	0.9733	
Site*Water*Time	3,730	0.45	0.7141	
Species*Fertilizer*Water	1,258	1.43	0.2324	
Species*Fertilizer*Time	3,730	0.59	0.6232	

Table 3.5, continued.

Source	DF	F	P	Significant
Species*Water*Time	3,730	0.14	0.9372	
Fertilizer*Water*Time	3,730	0.48	0.6938	
Site*Species*Fertilizer*Water	1,258	1.22	0.2702	
Site*Species*Fertilizer*Time	3,730	0.62	0.6038	
Site*Species*Water*Time	3,730	0.24	0.8676	
Site*Fertilizer*Water*Time	3,730	0.11	0.9546	
Species*Fertilizer*Water*Time	3,730	0.03	0.994	
Site*Species*Fertilizer*Water*Time	3,730	0.04	0.9897	

Table 3.5. Analysis of variance with log lifetime seed output of plants that survived to produce seed as the response variable and site, species, fertilizer and water as treatment variables. Highest order significant terms are indicated in the final column.

Source	DF	F	P	Significant
Site	1	1.524	0.219	
Species	1	51.030	< 0.0001	*
Fertilizer	1	1.695	0.195	
Water	1	2.002	0.159	
Site*Species	1	1.963	0.163	
Site*Fertilizer	1	9.678	0.002	*
Site*Water	1	0.638	0.426	
Species*Fertilizer	1	1.006	0.317	
Species*Water	1	0.648	0.422	
Fertilizer*Water	1	3.597	0.060	
Site*Species*Fertilizer	1	0.276	0.600	
Site*Species*Water	1	0.120	0.729	
Site*Fertilizer*Water	1	0.693	0.406	
Species*Fertilizer*Water	1	1.151	0.285	
Site*Species*Fertilizer*Water	1	2.147	0.145	
Error	160			

3.8 FIGURE CAPTIONS

Figure 3.1. Repeated measures analysis of variance with plants as subjects, time as a factor, log leaf nitrogen content (%) as the response variable and site, species, fertilizer and water as treatment variables. Values are back-transformed least-squares means ± the standard deviations. Treatments are Fertilizer and Water (FW), Fertilizer (F), Water (W) and Control (C). Species by fertilizer treatment combinations (effect $p = 0.0134$) with the same lowercase letters above them are not significantly different from one another. Fertilizer by water by time treatment combinations (effect $p = 0.0319$) with the same uppercase letters above them are not significantly different from one another.

Figure 3.2. Repeated measures analysis of variance with plants as subjects, time as a factor, leaf $\delta^{13}C$ as the response variable and site, species, fertilizer and water as treatment variables. Values are least-squares means ± the standard deviations. Treatments as in figure 3.1. Site by water treatment combinations (effect $p = 0.0136$) with the same uppercase letter (A, B, C) below them do not differ significantly from one another. Fertilizer by time treatment combinations (effect $p = 0.0045$) with the same uppercase letter (X, Y) below them are not significantly different from one another. Species by fertilizer by water treatment combinations (effect $p = 0.0072$) with the same lowercase letter below them do not differ significantly from one another.

Figure 3.3. Repeated measures analysis of variance with plants as subjects, time as a factor, log leaf area (cm^2) as the response variable and site, species, fertilizer and water as treatment variables. Values are back-transformed least-squares means ± the standard deviations. Site by species treatment combinations (effect $p < 0.0001$) with the same lowercase letters above them are not significantly different from one another. Treatments as in figure 3.1. Site by

fertilizer treatment combinations (effect $p = 0.0002$) with the same uppercase letters (A, B, C) above them are not significantly different from one another. Site by time combinations (effect $p < 0.0001$) with the same uppercase letters above them (V, W, X, Y, Z) are not significantly different from one another. *Bromus tectorum* plants were larger than *Bromus briziformis* plants at every census ($p < 0.0001$; differences not shown).

Figure 3.4. Analysis of variance with log lifetime seed output of plants that survived to produce seed as the response variable and site, species, fertilizer and water as treatment variables. Values are back-transformed least-squares means ± the standard deviations. Treatments as in figure 3.1. *B. tectorum* produced more seeds than *B. briziformis* ($p < 0.0001$; differences not indicated). Site by fertilizer combinations (effect $p = 0.0022$) with the same letters above them are not significantly different from one another.

Figure 3.5. Population growth rates of experimental populations. Error bars are bias-corrected bootstrap percentile intervals. Treatments as in figure 3.1. Treatments with the same letters above them are not significantly different from one another in permutation tests.

Figure 3.6. Population growth rates (zero offsets) and simulated population growth rates (nonzero offsets) as described in the text. A) Population growth rates predicted by adding or subtracting one standard deviation to leaf $\delta^{13}C$ values. B) Population growth rates predicted by adding or subtracting one standard deviation to leaf log nitrogen content. Error bars are bias-corrected bootstrap percentile intervals. Treatments indicated by the same letters are not significantly different from one another in permutation tests.

3.9 FIGURES

Figure 3.1

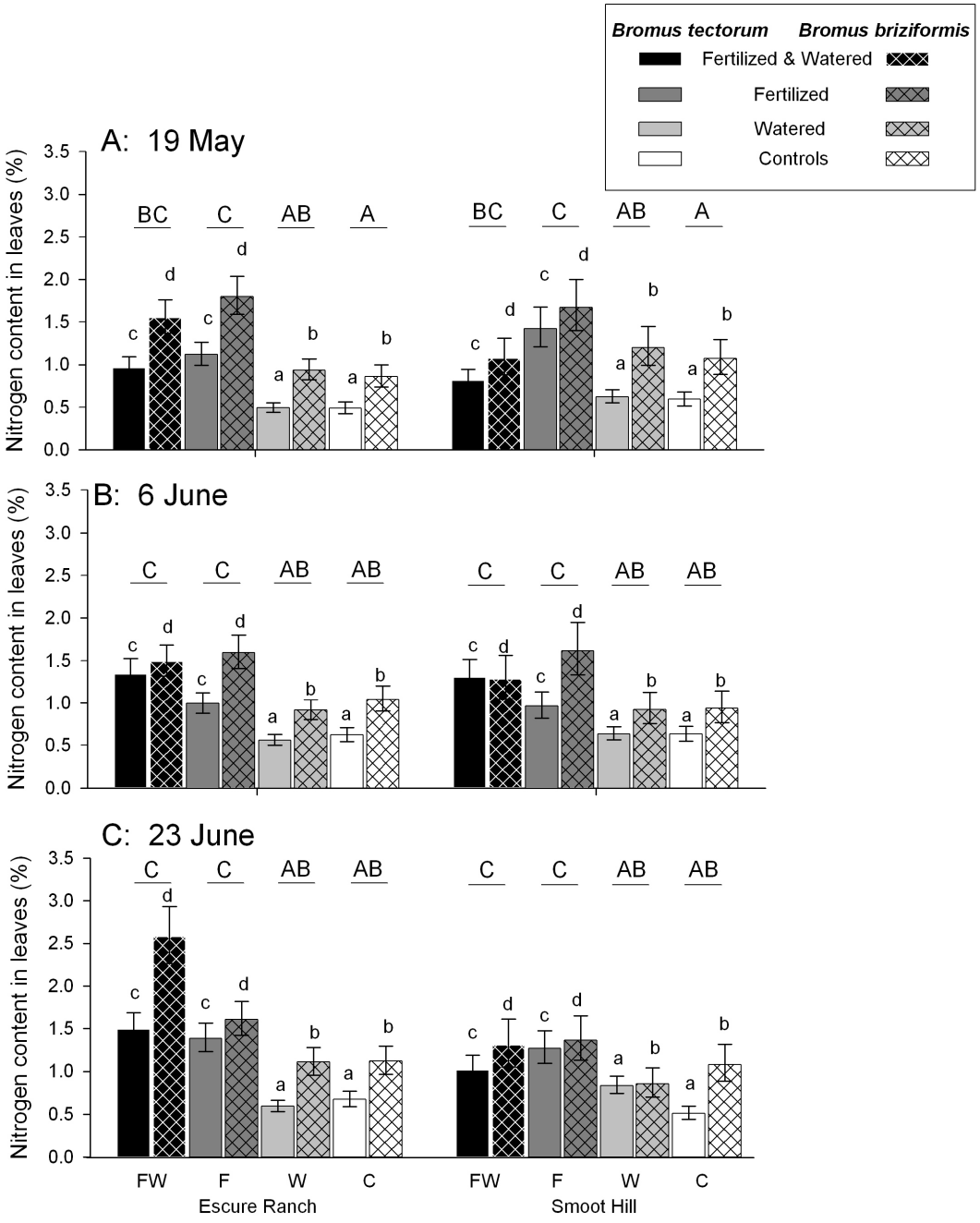


Figure 3.2

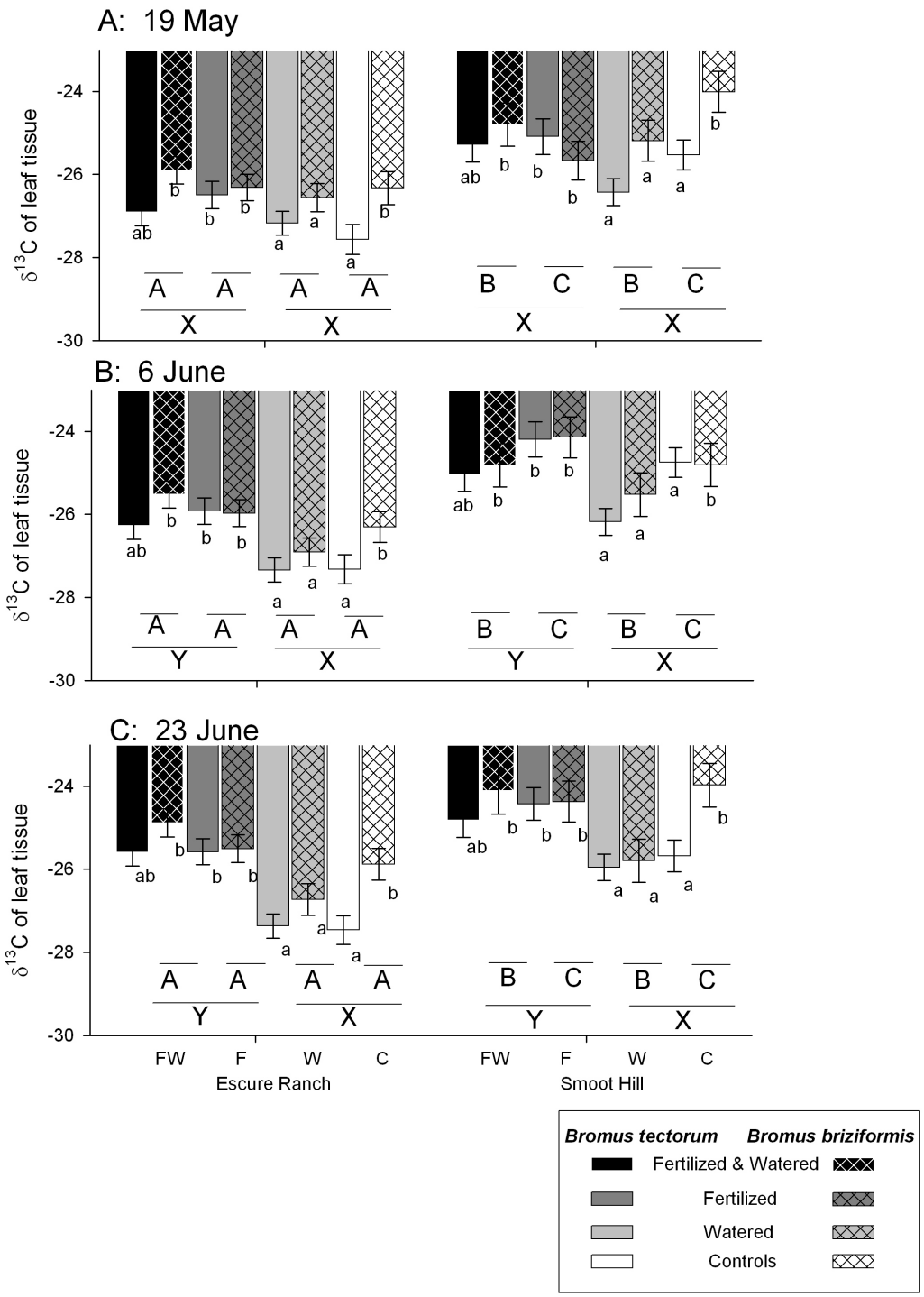

A: 19 May
B: 6 June
C: 23 June
δ13C of leaf tissue
-24
-26
-28
-30
FW
F
W
C
Escure Ranch
Smoot Hill
Bromus tectorum
Bromus briziformis
Fertilized & Watered
Fertilized
Watered
Controls

Figure 3.3 A&B

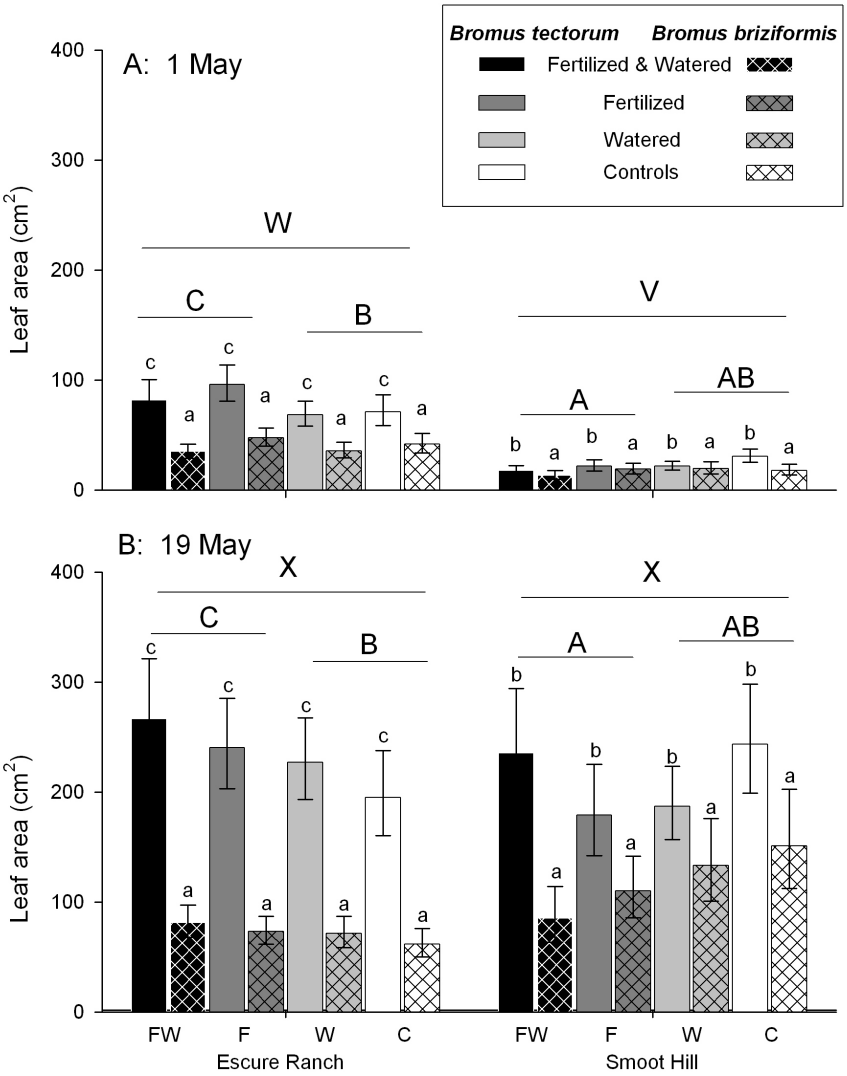

Bromus tectorum
Bromus briziformis
Fertilized & Watered
Fertilized
Watered
Controls
A: 1 May
B: 19 May
Leaf area (cm2)
400
300
200
100
0
W
C
B
V
A
AB
X
FW
F
W
C
Escure Ranch
Smoot Hill

Figure 3.3 C&D

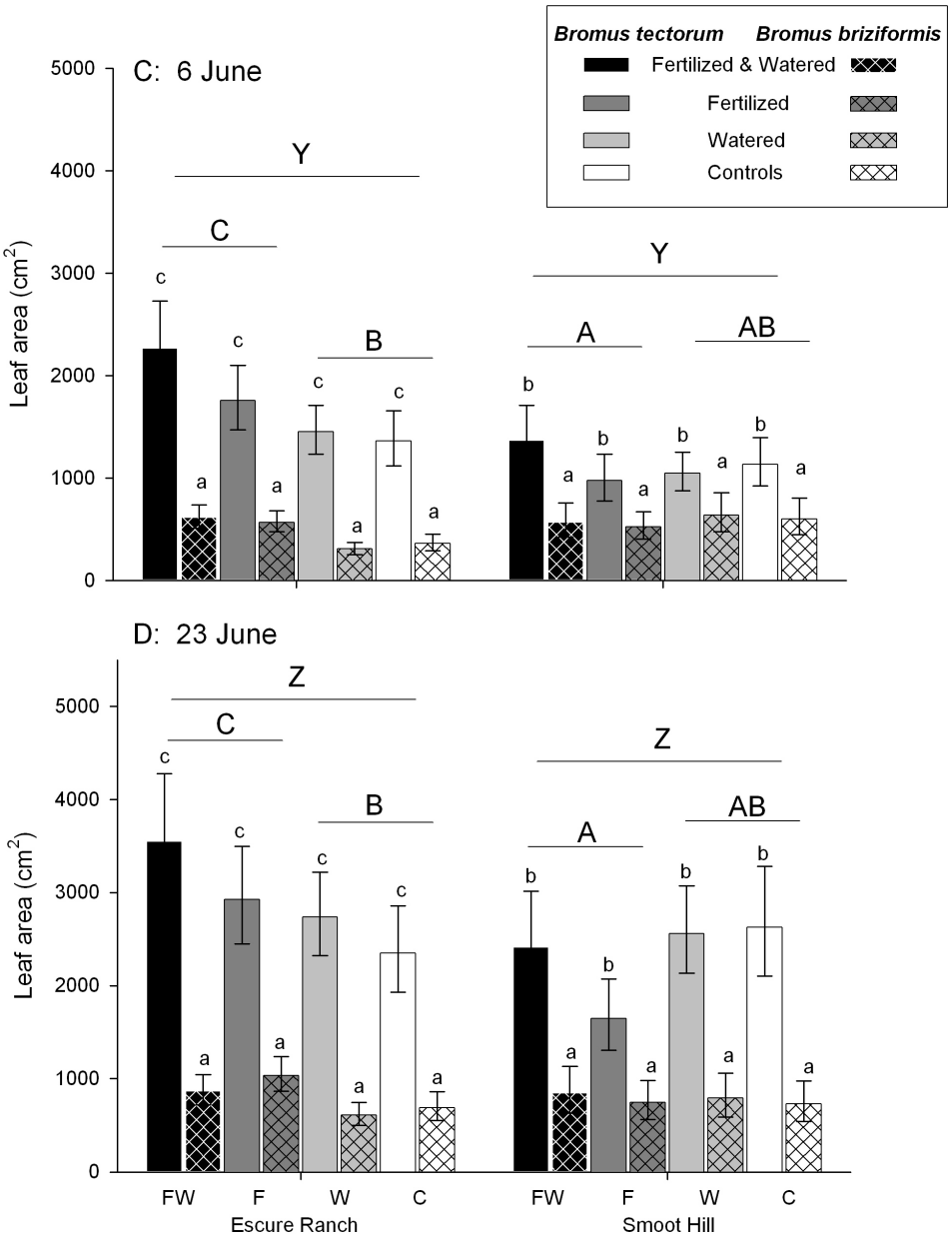

Bromus tectorum
Bromus briziformis
Fertilized & Watered
Fertilized
Watered
Controls
C: 6 June
D: 23 June
Leaf area (cm2)
5000
4000
3000
2000
1000
0
Y
C
B
A
AB
Z
FW
F
W
C
Escure Ranch
Smoot Hill

Figure 3.4

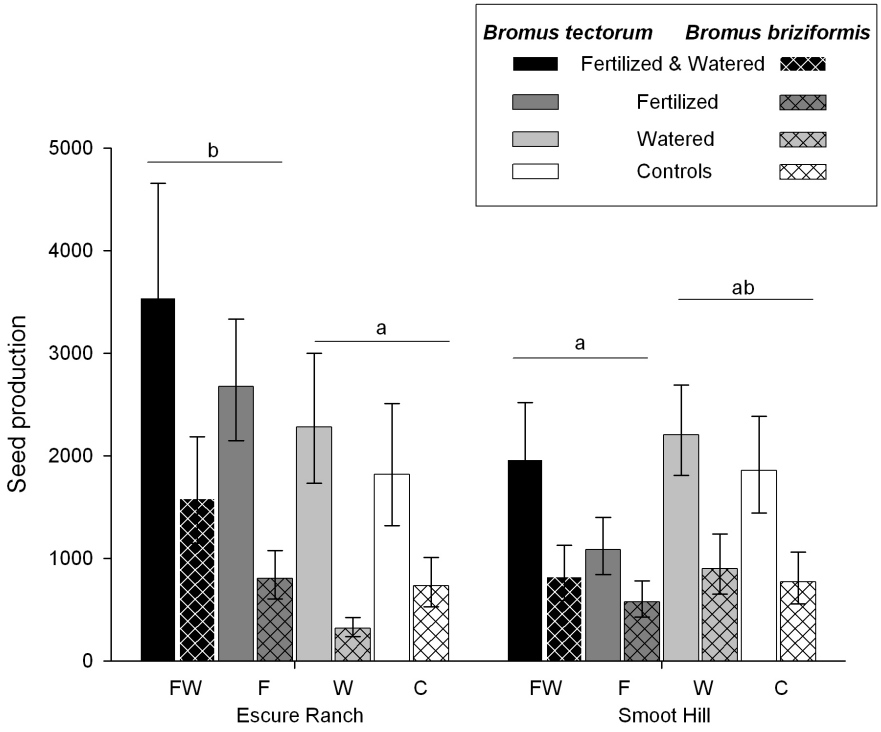

Bromus tectorum
Bromus briziformis
Fertilized & Watered
Fertilized
Watered
Controls
Seed production
5000
4000
3000
2000
1000
0
b
a
a
ab
FW
F
W
C
FW
F
W
C
Escure Ranch
Smoot Hill

Figure 3.5

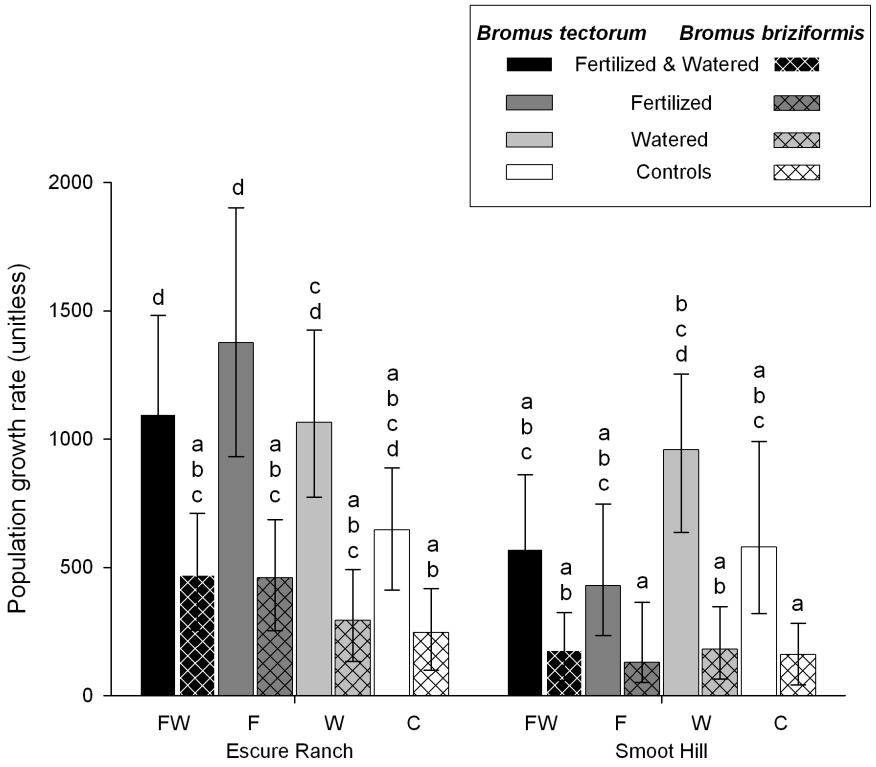
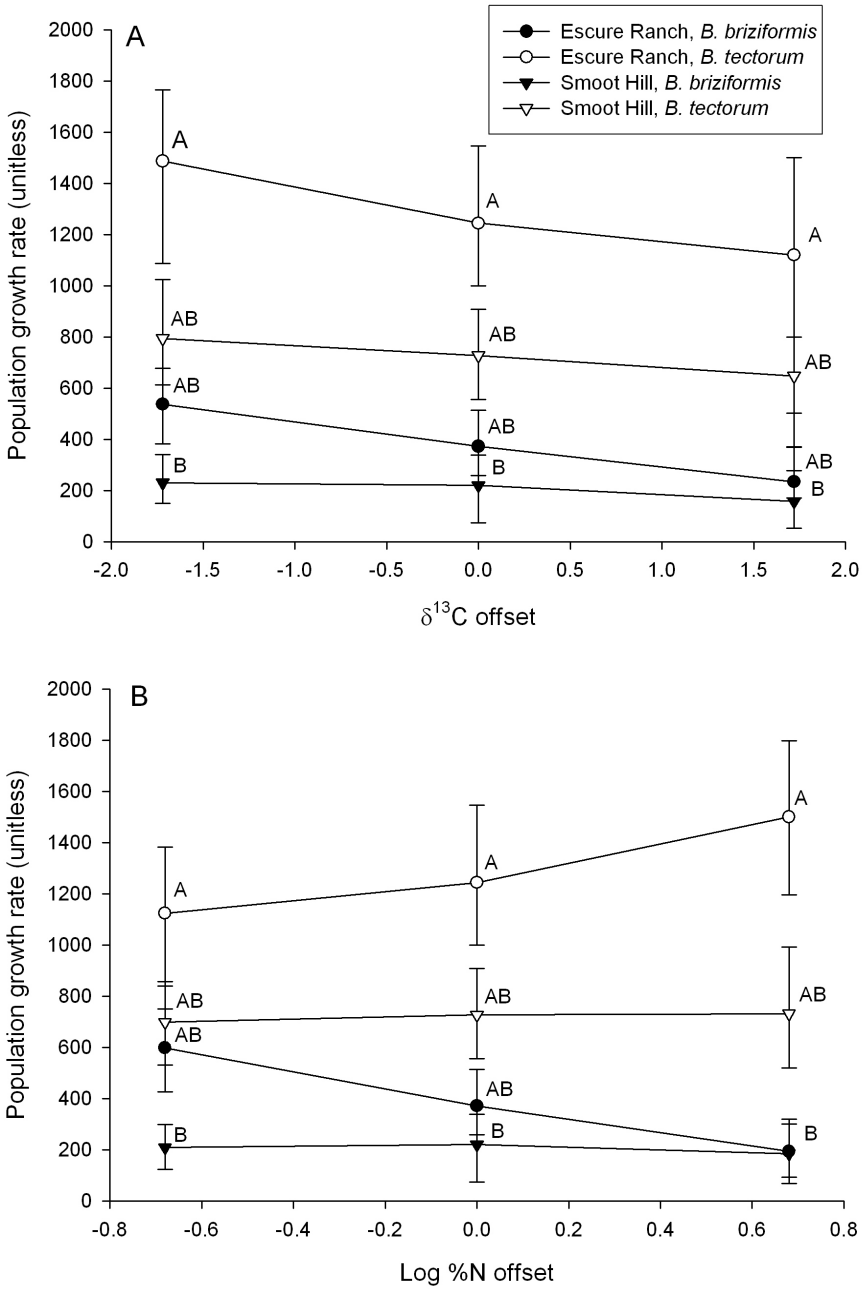

Bromus tectorum
Bromus briziformis
Fertilized & Watered
Fertilized
Watered
Controls
Population growth rate (unitless)
2000
1500
1000
500
0
d
d
a b c
a b c
c d
a b c
a b c d
a b
a b c
a b
a b c
a
b c d
a b
a b c
a
FW
F
W
C
Escure Ranch
FW
F
W
C
Smoot Hill

Figure 3.6


A
Escure Ranch, B. briziformis
Escure Ranch, B. tectorum
Smoot Hill, B. briziformis
Smoot Hill, B. tectorum
Population growth rate (unitless)
δ13C offset
B
Log %N offset

APPENDIX A.

Table A1. Population model transition matrices: Escure Ranch, *Bromus briziformis* control.

15 January		**Seed**
	Seed	0.87
	C1	0.13

2 April		**Seed**	**C1**
	Seed	0.42	0.0
	C1	0.58	0.75

1 May		**Seed**	**C1**
	Seed	1.00	0.0
	C1	0.0	0.94

19 May		**Seed**	**C1**
	Seed	1.00	0.0
	C1	0.0	0.88
	C2	0.0	0.12

6 June		**Seed**	**C1**	**C2**
	Seed	1.00	0.0	0.0
	C1	0.0	0.27	0.0
	C2	0.0	0.13	0.0
	C3	0.0	0.47	0.0
	C4	0.0	0.0	1.00

23 June		**Seed**	**C1**	**C2**	**C3**	**C4**
	Seed	1.00	12.86	0.0	216.9	35.57
	C1	0.0	0.50	0.0	0.0	0.0
	C2	0.0	0.25	0.50	0.14	0.0
	C3	0.0	0.25	0.50	0.43	0.0
	C4	0.0	0.0	0.0	0.14	0.50
	C5	0.0	0.0	0.0	0.29	0.50

27 July		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**
	Seed	1.00	49.80	156.2	632.8	504.8	893.0

Table A2. Population model transition matrices: Escure Ranch, *Bromus briziformis,* no fertilizer, watered.

15 January		**Seed**
	Seed	0.83
	C1	0.17

2 April		**Seed**	**C1**
	Seed	0.36	0.0
	C1	0.64	0.60

1 May		**Seed**	**C1**
	Seed	0.78	0.0
	C1	0.22	0.95

19 May		**Seed**	**C1**
	Seed	1.00	0.0
	C1	0.0	0.85
	C2	0.0	0.15

6 June		**Seed**	**C1**	**C2**
	Seed	1.00	0.12	0.0
	C1	0.0	0.35	0.0
	C2	0.0	0.24	0.0
	C3	0.0	0.35	0.33
	C4	0.0	0.0	0.67

23 June		**Seed**	**C1**	**C2**	**C3**	**C4**
	Seed	1.00	27.00	39.29	99.43	20.57
	C1	0.0	0.50	0.0	0.0	0.0
	C2	0.0	0.17	0.0	0.0	0.0
	C3	0.0	0.17	1.00	0.14	0.0
	C4	0.0	0.0	0.0	0.43	0.0
	C5	0.0	0.0	0.0	0.43	1.00

27 July		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**
	Seed	1.00	22.50	24.33	515.83	737.5	1288.2

Table A3. Population model transition matrices: Escure Ranch, *Bromus briziformis,* fertilized, no water.

15 January		**Seed**
	Seed	0.70
	C1	0.30

2 April		**Seed**	**C1**
	Seed	0.10	0.0
	C1	0.90	0.56

1 May		**Seed**	**C1**
	Seed	1.00	0.0
	C1	0.0	1.00

19 May		**Seed**	**C1**
	Seed	1.00	0.0
	C1	0.0	0.83
	C2	0.0	0.17

6 June		**Seed**	**C1**	**C2**
	Seed	1.00	0.0	0.0
	C1	0.0	0.20	0.0
	C2	0.0	0.10	0.0
	C3	0.0	0.35	0.0
	C4	0.0	0.15	0.50
	C5	0.0	0.10	0.50

23 June		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**
	Seed	1.00	0.14	0.14	91.43	225.00	0.0
	C1	0.0	0.25	0.0	0.0	0.0	0.0
	C2	0.0	0.50	0.0	0.14	0.0	0.0
	C3	0.0	0.25	0.50	0.29	0.0	0.0
	C4	0.0	0.0	0.0	0.29	0.40	0.0
	C5	0.0	0.0	0.50	0.29	0.60	0.25
	C6	0.0	0.0	0.0	0.0	0.0	0.75

27 July		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**	**C6**
	Seed	1.00	6.86	100.57	320.14	534.71	1562.9	1243.0

Table A4. Population model transition matrices: Escure Ranch, *Bromus briziformis,* fertilized, watered.

15 January		**Seed**
	Seed	0.83
	C1	0.17

2 April		**Seed**	**C1**
	Seed	0.28	0.0
	C1	0.72	1.00

1 May		**Seed**	**C1**
	Seed	1.00	0.0
	C1	0.0	0.96

19 May		**Seed**	**C1**
	Seed	1.00	0.0
	C1	0.0	0.68
	C2	0.0	0.32

6 June		**Seed**	**C1**	**C2**
	Seed	1.00	0.0	0.0
	C1	0.0	0.20	0.0
	C2	0.0	0.13	0.0
	C3	0.0	0.33	0.0
	C4	0.0	0.13	0.29
	C5	0.0	0.07	0.71

23 June		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**
	Seed	1.00	0.0	34.83	128.83	0.0	130.33
	C1	0.0	0.67	0.0	0.0	0.0	0.0
	C2	0.0	0.33	1.00	0.0	0.0	0.0
	C3	0.0	0.0	0.0	0.80	0.50	0.0
	C4	0.0	0.0	0.0	0.0	0.25	0.0
	C5	0.0	0.0	0.0	0.20	0.25	0.50
	C6	0.0	0.0	0.0	0.0	0.0	0.50

27 July		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**	**C6**
	Seed	1.00	23.83	115.33	869.00	350.67	1511.3	1405.7

Table A5. Population model transition matrices: Escure Ranch, *Bromus tectorum* control.

15 January		**Seed**
	Seed	0.87
	C1	0.13

2 April		**Seed**	**C1**
	Seed	0.50	0.0
	C1	0.50	0.75

1 May		**Seed**	**C1**
	Seed	0.85	0.0
	C1	0.15	1.00

19 May		**Seed**	**C1**
	Seed	1.00	0.0
	C1	0.0	0.44
	C2	0.0	0.56

6 June		**Seed**	**C1**	**C2**
	Seed	1.00	0.0	0.0
	C1	0.0	0.0	0.0
	C2	0.0	0.0	0.0
	C3	0.0	0.13	0.0
	C4	0.0	0.50	0.20
	C5	0.0	0.38	0.80

23 June		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**
	Seed	1.00	0.0	0.0	23.09	1.36	145.82
	C1	0.0	0.0	0.0	0.0	0.0	0.0
	C2	0.0	0.0	0.0	0.0	0.0	0.0
	C3	0.0	0.0	0.0	0.0	0.0	0.0
	C4	0.0	0.0	0.0	1.00	0.0	0.0
	C5	0.0	0.0	0.0	0.0	1.00	0.73
	C6	0.0	0.0	0.0	0.0	0.0	0.18
	C7	0.0	0.0	0.0	0.0	0.0	0.09

27 July		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**	**C6**	**C7**
	Seed	1.00	0.0	0.0	0.0	55.57	1813.9	258.29	208.21

Table A6. Population model transition matrices: Escure Ranch, *Bromus tectorum,* no fertilizer, watered.

15 January		**Seed**
	Seed	0.77
	C1	0.23

2 April		**Seed**	**C1**
	Seed	0.22	0.0
	C1	0.78	1.00

1 May		**Seed**	**C1**
	Seed	0.80	0.0
	C1	0.20	1.00

19 May		**Seed**	**C1**
	Seed	1.00	0.0
	C1	0.0	0.35
	C2	0.0	0.62
	C3	0.0	0.04

6 June		**Seed**	**C1**	**C2**	**C3**
	Seed	1.00	0.0	0.0	0.0
	C1	0.0	0.0	0.0	0.0
	C2	0.0	0.11	0.0	0.0
	C3	0.0	0.56	0.0	0.0
	C4	0.0	0.22	0.13	0.0
	C5	0.0	0.11	0.75	0.0
	C6	0.0	0.0	0.13	1.00

23 June		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**	**C6**
	Seed	1.00	0.0	2.15	9.15	35.69	91.69	96.54
	C1	0.0	0.0	0.0	0.0	0.0	0.0	0.0
	C2	0.0	0.0	0.0	0.0	0.0	0.0	0.0
	C3	0.0	0.0	0.0	0.40	0.0	0.0	0.0
	C4	0.0	0.0	1.00	0.20	0.0	0.0	0.0
	C5	0.0	0.0	0.0	0.40	1.00	0.23	0.0
	C6	0.0	0.0	0.0	0.0	0.0	0.38	0.0
	C7	0.0	0.0	0.0	0.0	0.0	0.38	1.00

27 July		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**	**C6**	**C7**
	Seed	1.00	0.0	0.0	69.33	148.00	1870.1	1514.3	2527.1

Table A7. Population model transition matrices: Escure Ranch, *Bromus tectorum,* fertilized, no water.

15 January		**Seed**							
	Seed	0.67							
	C1	0.33							
1 May		**Seed**	**C1**						
	Seed	1.00	0.0						
	C1	0.0	0.96						
2 April		**Seed**	**C1**						
	Seed	0.25	0.0						
	C1	0.75	1.00						
	C2	0.0	0.63						
	C3	0.0	0.21						
19 May		**Seed**	**C1**						
	Seed	1.00	0.0						
	C1	0.0	0.17						
	C2	0.0	0.63						
	C3	0.0	0.21						
6 June		**Seed**	**C1**	**C2**	**C3**				
	Seed	1.00	0.0	0.0	0.0				
	C1	0.0	0.0	0.0	0.0				
	C2	0.0	0.0	0.0	0.0				
	C3	0.0	0.50	0.07	0.0				
	C4	0.0	0.25	0.07	0.0				
	C5	0.0	0.0	0.67	0.80				
	C6	0.0	0.0	0.13	0.20				
23 June		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**	**C6**	
	Seed	1.00	0.0	0.0	11.71	18.36	359.57	68.93	
	C1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	
	C2	0.0	0.0	0.0	0.33	0.0	0.0	0.0	
	C3	0.0	0.0	0.0	0.0	0.0	0.0	0.0	
	C4	0.0	0.0	0.0	0.33	0.0	0.0	0.0	
	C5	0.0	0.0	0.0	0.33	1.00	0.43	0.0	
	C6	0.0	0.0	0.0	0.0	0.0	0.21	0.0	
	C7	0.0	0.0	0.0	0.0	0.0	0.36	1.00	
27 July		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**	**C6**	**C7**
	Seed	1.00	0.0	27.00	0.0	151.11	2039.6	1161.3	3426.9

Table A8. Population model transition matrices: Escure Ranch, *Bromus tectorum,* fertilized, watered.

15 January		**Seed**							
	Seed	0.93							
	C1	0.07							
1 May		**Seed**	**C1**						
	Seed	1.00	0.0						
	C1	0.0	0.95						
6 June		**Seed**	**C1**	**C2**	**C3**				
	Seed	1.00	0.0	0.0	0.0				
	C1	0.0	0.0	0.0	0.0				
	C2	0.0	0.0	0.0	0.0				
	C3	0.0	0.0	0.0	0.0				
	C4	0.0	0.0	0.0	0.0				
	C5	0.0	1.00	0.75	0.25				
	C6	0.0	0.0	0.25	0.50				
	C7	0.0	0.0	0.0	0.25				
23 June		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**	**C6**	**C7**
	Seed	1.00	0.0	0.0	0.0	0.0	84.29	88.00	50.07
	C1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
	C2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
	C3	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
	C4	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
	C5	0.0	0.0	0.0	0.0	0.0	0.29	0.0	0.0
	C6	0.0	0.0	0.0	0.0	0.0	0.64	0.20	0.0
	C7	0.0	0.0	0.0	0.0	0.0	0.07	0.80	1.00
27 July		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**	**C6**	**C7**
	Seed	1.00	0.0	0.0	0.0	0.0	716.10	2948.2	2415.0

2 April		**Seed**	**C1**
	Seed	0.32	0.0
	C1	0.68	1.00

19 May		**Seed**	**C1**
	Seed	1.00	0.0
	C1	0.0	0.20
	C2	0.0	0.60
	C3	0.0	0.20

Table A9. Population model transition matrices: Smoot Hill, *Bromus briziformis* control.

15 January		**Seed**
	Seed	0.61
	C1	0.39

2 April		**Seed**	**C1**
	Seed	0.58	0.0
	C1	0.42	0.42

1 May		**Seed**	**C1**
	Seed	1.00	0.0
	C1	0.0	0.77

19 May		**Seed**	**C1**
	Seed	1.00	0.0
	C1	0.0	0.40
	C2	0.0	0.50

6 June		**Seed**	**C1**	**C2**
	Seed	1.00	0.0	0.0
	C1	0.0	0.50	0.0
	C2	0.0	0.0	0.0
	C3	0.0	0.25	0.20
	C4	0.0	0.0	0.60
	C5	0.0	0.0	0.20

23 June		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**
	Seed	1.00	15.67	0.0	142.00	389.00	143.67
	C1	0.0	0.50	0.0	0.0	0.0	0.0
	C2	0.0	0.50	0.0	0.0	0.0	0.0
	C3	0.0	0.0	0.0	0.50	0.33	0.0
	C4	0.0	0.0	0.0	0.0	0.0	0.0
	C5	0.0	0.0	0.0	0.50	0.67	1.00

27 July		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**
	Seed	1.00	0.75	151.50	332.00	0.0	1183.0

Table A10. Population model transition matrices: Smoot Hill, *Bromus briziformis,* no fertilizer, watered.

15 January		**Seed**
	Seed	0.64
	C1	0.36

2 April		**Seed**	**C1**
	Seed	0.67	0.0
	C1	0.33	0.42

1 May		**Seed**	**C1**
	Seed	1.00	0.0
	C1	0.0	0.75

19 May		**Seed**	**C1**
	Seed	1.00	0.0
	C1	0.0	0.78
	C2	0.0	0.22

6 June		**Seed**	**C1**	**C2**
	Seed	1.00	0.0	0.0
	C1	0.0	0.0	0.0
	C2	0.0	0.14	0.0
	C3	0.0	0.71	0.50
	C4	0.0	0.14	0.0

23 June		**Seed**	**C1**	**C2**	**C3**	**C4**
	Seed	1.00	0.0	24.67	187.50	44.17
	C1	0.0	0.0	0.0	0.0	0.0
	C2	0.0	0.0	1.00	0.0	0.0
	C3	0.0	0.0	0.0	0.50	0.0
	C4	0.0	0.0	0.0	0.33	0.0
	C5	0.0	0.0	0.0	0.17	1.00

27 July		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**
	Seed	1.00	0.0	14.00	726.67	672.33	1030.7

Table A11. Population model transition matrices: Smoot Hill, *Bromus briziformis,* fertilized, no water.

15 January		**Seed**	
	Seed	0.71	
	C1	0.29	

2 April		**Seed**	**C1**
	Seed	0.64	0.0
	C1	0.36	0.56

1 May		**Seed**	**C1**
	Seed	0.93	0.0
	C1	0.07	0.77

19 May		**Seed**	**C1**
	Seed	1.00	0.0
	C1	0.0	0.73
	C2	0.0	0.18
	C3	0.0	0.0
	C4	0.0	0.09

6 June		**Seed**	**C1**	**C2**	**C3**	**C4**
	Seed	1.00	0.0	0.0	0.0	0.0
	C1	0.0	0.13	0.0	0.0	0.0
	C2	0.0	0.38	0.0	0.0	1.00
	C3	0.0	0.50	0.0	0.0	0.0
	C4	0.0	0.0	0.0	0.0	0.0
	C5	0.0	0.0	1.00	0.0	0.0

23 June		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**
	Seed	1.00	12.25	67.25	167.00	0.0	236.00
	C1	0.0	0.0	0.0	0.0	0.0	0.0
	C2	0.0	1.00	0.25	0.25	0.0	0.0
	C3	0.0	0.0	0.25	0.0	0.0	0.0
	C4	0.0	0.0	0.25	0.25	0.0	0.50
	C5	0.0	0.0	0.0	0.25	0.0	0.50

27 July		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**
	Seed	1.00	0.0	159.67	106.33	735.33	843.67

Table A12. Population model transition matrices: Smoot Hill, *Bromus briziformis,* fertilized, watered.

15 January		**Seed**
	Seed	0.60
	C1	0.40

2 April		**Seed**	**C1**
	Seed	0.83	0.0
	C1	0.17	0.50

1 May		**Seed**	**C1**
	Seed	0.93	0.0
	C1	0.07	0.78

19 May		**Seed**	**C1**
	Seed	1.00	0.0
	C1	0.0	0.75
	C2	0.0	0.25

6 June		**Seed**	**C1**	**C2**
	Seed	1.00	0.0	0.0
	C1	0.0	0.33	0.0
	C2	0.0	0.0	0.0
	C3	0.0	0.33	0.0
	C4	0.0	0.33	1.00

23 June		**Seed**	**C1**	**C2**	**C3**	**C4**
	Seed	1.00	23.25	0.0	83.00	368.75
	C1	0.0	0.50	0.0	0.0	0.0
	C2	0.0	0.50	0.0	0.0	0.0
	C3	0.0	0.0	0.0	0.50	0.0
	C4	0.0	0.0	0.0	0.0	0.0
	C5	0.0	0.0	0.0	0.50	1.00

27 July		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**
	Seed	1.00	4.00	55.60	121.60	0.0	1322.6

Table A13. Population model transition matrices: Smoot Hill, *Bromus tectorum* control.

15 January		**Seed**
	Seed	0.59
	C1	0.41

2 April		**Seed**	**C1**
	Seed	0.60	0.0
	C1	0.40	0.86

1 May		**Seed**	**C1**
	Seed	0.75	0.0
	C1	0.25	0.90

19 May		**Seed**	**C1**
	Seed	1.00	0.0
	C1	0.0	0.38
	C2	0.0	0.29
	C3	0.0	0.19
	C4	0.0	0.05

6 June		**Seed**	**C1**	**C2**	**C3**	**C4**
	Seed	1.00	1.38	0.0	0.0	3.38
	C1	0.0	0.38	0.0	0.0	0.0
	C2	0.0	0.0	0.0	0.0	0.0
	C3	0.0	0.25	0.0	0.0	0.0
	C4	0.0	0.13	0.17	0.25	0.0
	C5	0.0	0.13	0.83	0.75	1.00

23 June		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**
	Seed	1.00	1.50	0.0	8.90	99.50	397.40
	C1	0.0	0.33	0.0	0.0	0.0	0.0
	C2	0.0	0.0	0.0	0.0	0.0	0.0
	C3	0.0	0.0	0.0	0.0	0.0	0.0
	C4	0.0	0.0	0.0	0.50	0.0	0.0
	C5	0.0	0.0	0.0	0.0	0.67	0.50
	C6	0.0	0.0	0.0	0.50	0.0	0.50

27 July		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**	**C6**
	Seed	1.00	1.29	0.0	0.0	188.29	1546.0	1452.3

Table A14. Population model transition matrices: Smoot Hill, *Bromus tectorum,* no fertilizer, watered.

15 January		**Seed**							
	Seed	0.63							
	C1	0.37							
1 May		**Seed**	**C1**						
	Seed	0.80	0.0						
	C1	0.20	0.91						
6 June		**Seed**	**C1**	**C2**	**C3**				
	Seed	1.00	0.0	0.0	1.40				
	C1	0.0	0.11	0.0	0.0				
	C2	0.0	0.22	0.0	0.0				
	C3	0.0	0.44	0.10	0.0				
	C4	0.0	0.0	0.10	0.0				
	C5	0.0	0.22	0.80	1.00				
23 June		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**		
	Seed	1.00	4.85	15.15	49.69	13.00	294.31		
	C1	0.0	0.0	0.0	0.0	0.0	0.0		
	C2	0.0	0.0	0.0	0.0	0.0	0.0		
	C3	0.0	0.0	0.50	0.0	0.0	0.0		
	C4	0.0	0.0	0.0	0.20	0.0	0.0		
	C5	0.0	0.0	0.50	0.80	1.00	0.15		
	C6	0.0	0.0	0.0	0.0	0.0	0.77		
	C7	0.0	0.0	0.0	0.0	0.0	0.08		
27 July		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**	**C6**	**C7**
	Seed	1.00	0.0	0.0	44.00	137.10	1306.5	2743.1	333.50

Table A15. Population model transition matrices: Smoot Hill, *Bromus tectorum,* fertilized, no water.

15 January		**Seed**							
	Seed	0.32							
	C1	0.68							
1 May		**Seed**	**C1**						
	Seed	1.00	0.0						
	C1	0.0	0.70						

2 April		**Seed**	**C1**
	Seed	0.50	0.0
	C1	0.50	0.71
19 May		**Seed**	**C1**
	Seed	1.00	0.0
	C1	0.0	0.43
	C2	0.0	0.29
	C3	0.0	0.29

6 June		**Seed**	**C1**	**C2**	**C3**
	Seed	1.00	0.0	0.0	0.33
	C1	0.0	0.33	0.0	0.0
	C2	0.0	0.0	0.0	0.0
	C3	0.0	0.67	0.0	0.0
	C4	0.0	0.0	0.25	0.0
	C5	0.0	0.0	0.75	1.00

23 June		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**
	Seed	1.00	5.71	0.0	52.71	41.29	374.29
	C1	0.0	0.50	0.0	0.25	0.0	0.0
	C2	0.0	0.0	0.0	0.0	0.0	0.0
	C3	0.0	0.50	0.0	0.0	0.0	0.0
	C4	0.0	0.0	0.0	0.25	0.0	0.14
	C5	0.0	0.0	0.0	0.25	1.00	0.29
	C6	0.0	0.0	0.0	0.25	0.0	0.43
	C7	0.0	0.0	0.0	0.0	0.0	0.14

27 July		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**	**C6**	**C7**
	Seed	1.00	0.25	0.0	168.25	290.25	1225.0	2032.3	859.00

Table A16. Population model transition matrices: Smoot Hill, *Bromus tectorum,* fertilized, watered.

15 January		**Seed**							
	Seed	0.53							
	C1	0.47							
1 May		**Seed**	**C1**						
	Seed	1.00	0.0						
	C1	0.0	0.83						
2 April		**Seed**	**C1**						
	Seed	0.47	0.0						
	C1	0.53	0.60						
19 May		**Seed**	**C1**						
	Seed	1.00	0.0						
	C1	0.0	0.20						
	C2	0.0	0.67						
	C3	0.0	0.07						
6 June		**Seed**	**C1**	**C2**	**C3**				
	Seed	1.00	0.0	2.00	0.0				
	C1	0.0	0.0	0.0	0.0				
	C2	0.0	0.33	0.0	0.0				
	C3	0.0	0.67	0.10	0.0				
	C4	0.0	0.0	0.0	0.0				
	C5	0.0	0.0	0.90	1.00				
23 June		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**		
	Seed	1.00	0.0	0.0	23.40	0.0	471.00		
	C1	0.0	0.0	0.0	0.0	0.0	0.0		
	C2	0.0	0.0	0.0	0.0	0.0	0.0		
	C3	0.0	0.0	1.00	0.0	0.0	0.0		
	C4	0.0	0.0	0.0	0.33	0.0	0.0		
	C5	0.0	0.0	0.0	0.67	0.0	0.20		
	C6	0.0	0.0	0.0	0.0	0.0	0.70		
	C7	0.0	0.0	0.0	0.0	0.0	0.10		
27 July		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**	**C6**	**C7**
	Seed	1.00	0.0	0.0	36.00	57.00	647.00	2530.0	334.14

APPENDIX B

Table B1. Baseline population model transition matrices for simulations: Escure Ranch, *Bromus briziformis.*

15 January		**Seed**
	Seed	0.81
	C1	0.19

2 April		**Seed**	**C1**
	Seed	0.30	0
	C1	0.70	0.70

1 May		**Seed**	**C1**
	Seed	0.93	0
	C1	0.07	0.96

19 May		**Seed**	**C1**
	Seed	1	0
	C1	0	0.81
	C2	0	0.19

6 June		**Seed**	**C1**	**C2**
	Seed	1	0.03	0
	C1	0	0.25	0
	C2	0	0.15	0
	C3	0	0.37	0.06
	C4	0	0.07	0.50
	C5	0	0.04	0.44

23 June		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**
	Seed	1	10.8	18.7	139.5	75.7	30.1
	C1	0	0.47	0	0	0	0
	C2	0	0.29	0.30	0.08	0	0
	C3	0	0.18	0.60	0.38	0.15	0
	C4	0	0	0	0.23	0.31	0
	C5	0	0	0.10	0.31	0.54	0.40
	C6	0	0	0	0	0	0.60

27 July		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**	**C6**
	Seed	1	27.4	110.6	653.0	609.3	1533.4	816.0

Table B2. Baseline population model transition matrices for simulations: Escure Ranch, *Bromus tectorum.*

15 January		**Seed**	
	Seed	0.81	
	C1	0.19	

2 April		**Seed**	**C1**
	Seed	0.33	0
	C1	0.67	0.96

1 May		**Seed**	**C1**
	Seed	0.91	0
	C1	0.09	0.98

19 May		**Seed**	**C1**
	Seed	1	0
	C1	0	0.28
	C2	0	0.60
	C3	0	0.11

6 June		**Seed**	**C1**	**C2**	**C3**
	Seed	1	0	0	0
	C1	0	0	0	0
	C2	0	0.04	0	0
	C3	0	0.32	0.02	0
	C4	0	0.28	0.09	0
	C5	0	0.32	0.74	0.50
	C6	0	0	0.13	0.40
	C7	0	0	0	0.10

23 June		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**	**C6**	**C7**
	Seed	1	0	0.54	10.3	14.2	173.3	66.4	13.5
	C1	0	0	0	0	0	0	0	0
	C2	0	0	0	0.11	0	0	0	0
	C3	0	0	0	0.22	0	0	0	0
	C4	0	0	1	0.33	0	0	0	0
	C5	0	0	0	0.33	1	0.40	0	0
	C6	0	0	0	0	0	0.37	0.10	0
	C7	0	0	0	0	0	0.23	0.91	1

27 July		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**	**C6**	**C7**
	Seed	1	0	6.8	17.3	96.4	1881.7	1588.3	2240.3

Table B3. Baseline population model transition matrices for simulations: Smoot Hill, *Bromus briziformis.*

15 January		**Seed**
	Seed	0.64
	C1	0.36

2 April		**Seed**	**C1**
	Seed	0.68	0
	C1	0.33	0.47

1 May		**Seed**	**C1**
	Seed	0.96	0
	C1	0.04	0.77

19 May		**Seed**	**C1**
	Seed	1	0
	C1	0	0.66
	C2	0	0.29
	C3	0	0
	C4	0	0.03

6 June		**Seed**	**C1**	**C2**	**C3**	**C4**
	Seed	1	0	0	0	0
	C1	0	0.2	0	0	0
	C2	0	0.16	0	0	1
	C3	0	0.48	0.18	0	0
	C4	0	0.12	0.45	0	0
	C5	0	0	0.27	0	0

23 June		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**
	Seed	1	13.5	29.8	182.2	207.6	98.2
	C1	0	0.40	0	0	0	0
	C2	0	0.60	0.40	0.07	0	0
	C3	0	0	0.20	0.36	0.13	0
	C4	0	0	0.20	0.21	0	0.33
	C5	0	0	0	0.29	0.88	0.67

27 July		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**
	Seed	1	1.8	108.1	341.2	324.8	1305.2

Table B4. Baseline population model transition matrices for simulations: Smoot Hill, *Bromus tectorum.*

15 January		**Seed**	
	Seed	0.52	
	C1	0.48	

2 April		**Seed**	**C1**
	Seed	0.50	0
	C1	0.55	0.74

1 May		**Seed**	**C1**
	Seed	0.87	0
	C1	0.13	0.84

19 May		**Seed**	**C1**
	Seed	1	0
	C1	0	0.36
	C2	0	0.42
	C3	0	0.17
	C4	0	0.01

6 June		**Seed**	**C1**	**C2**	**C3**	**C4**
	Seed	1	0.37	0.67	0.53	0.90
	C1	0	0.23	0	0	0
	C2	0	0.12	0	0	0
	C3	0	0.46	0.07	0	0
	C4	0	0.04	0.10	0.08	0
	C5	0	0.12	0.83	0.92	1

23 June		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**
	Seed	1	3.0	4.9	33.5	36.3	378.3
	C1	0	0.33	0	0.07	0	0
	C2	0	0	0	0	0	0
	C3	0	0.17	0.67	0	0	0
	C4	0	0	0	0.29	0	0.03
	C5	0	0	0.33	0.50	0.80	0.28
	C6	0	0	0	0.14	0	0.63
	C7	0	0	0	0	0	0.08

27 July		**Seed**	**C1**	**C2**	**C3**	**C4**	**C5**	**C6**	**C7**
	Seed	1	0.37	0	50.6	157.4	1233.9	2349.5	337.4

Table C1. Logistic regression parameters for the dependence of demographic transitions on log leaf nitrogen content and δ13C. The transition column indicates the size class at the previous census and the size class at the current census, separated by a dash. Each line is a separate regression. The intercept models the log odds of a transition by a *B. tectorum* individual at Smoot Hill; other parameters adjust the log odds as appropriate. Missing parameters are missing because to include them in the model caused complete separation of the data set, resulting in no maximum likelihood estimate of the parameter. Parameter values significantly different from zero ($p < 0.05$) are indicated with an asterisk. Models with global significance at the $\alpha < 0.05$ level are indicated with an asterisk next to the model log likelihood. Models with global significance at the $\alpha < 0.1$ level are indicated with a dagger symbol next to the model log likelihood.

Census	Transition	Intercept	Escure	*B. briziformis*	δ13c	*B. briz* * δ13c	log (%N)	*B. briz* * log (%N)	Model log likelihood
19 May	C1-C1	0.79	-0.05	-6.25	0.06	-0.31	0.17	0.35	-148.18 *
19 May	C1-C2	0.62	0.34	5.51	0.03	0.26	-0.62 *	0.10	-157.20 *
19 May	C1-C3	-10.78	-1.12 *		-0.34 *		0.22		-71.65
19 May	C1-C4	6.44			0.49		-1.57		-5.84
6 June	C1-C1	11.73	0.39	-12.00	0.57	-0.52	1.53	-0.50	-51.02 *
6 June	C1-C2	-46.59	-0.39	46.24 *	-1.63 *	1.67 *	3.64 *	-3.64 *	-43.21 *
6 June	C1-C3	7.01	-0.90	-12.10	0.28	-0.50	-1.91 *	1.19	-76.03 *
6 June	C1-C4	-2.51	0.71	2.78	-0.02	0.13	-0.23	-0.50	-45.62

Table C1, continued.

Census	Transition	Intercept	Escure	*B. briziformis*	δ13c	*B. briz* * δ13c	log (%N)	*B. briz* * log (%N)	Model log likelihood
6 June	C1-C5	-2.18	1.64	-3.90	-0.01	-0.01	1.21	0.74	-32.39 *
6 June	C2-C3	-13.78	-2.14 *	-6.49	-0.44	-0.30	1.70	-1.43	-18.47
6 June	C2-C4	5.23	0.60	3.02	0.30	0.01	0.03	-2.11	-40.51 *
6 June	C2-C5	4.63	-0.29	-9.30	0.13	-0.27	-1.02 *	3.15	-54.68 *
6 June	C2-C6	-19.77			-0.64 *		1.39 *		-22.45 *
6 June	C3-C4	12.50			0.63				-3.70
6 June	C3-C5	10.42	-1.99		0.32		-0.26		-10.09
6 June	C3-C6	-22.94			-0.82		0.95		-8.28
6 June	C1-Dead	15.16			0.77		0.56		-3.15
6 June	C1-C1	16.73	0.52		0.71 *		1.34		-9.25
6 June	C1-C2	-2.55	-0.28		-0.10		-1.24		-10.99
6 June	C1-C3	-72.51	-1.17		-2.70 *		1.57		-4.57 *
6 June	C2-C2	30.59	1.62		1.32		1.21		-7.58
6 June	C2-C3	-7.25	0.04		-0.26		1.00		-12.00
23 June	C2-C4	-8.51	-0.67		-0.26		-0.42		-5.90
23 June	C2-C5	-15.85	-0.98		-0.54				-5.43
23 June	C3-C1	33.06			1.58 *				-2.73 *

Table C1, continued.

Census	Transition	Intercept	Escure	*B. briziformis*	δ13c	*B. briz* * δ13c	log (%N)	*B. briz* * log (%N)	Model log likelihood
23 June	C3-C2	-12.74	2.50	33.70	-0.30	1.30	2.32	-2.79	-11.42
23 June	C3-C3	-21.67	0.32	22.21	-0.73	0.78	0.81	-0.30	-29.52
23 June	C3-C4	15.60	0.82	-29.94 *	0.66	-1.14 *	-1.18	-0.35	-28.95 †
23 June	C3-C5	-9.13	-0.63	4.69	-0.34	0.20	0.03	0.58	-35.83
23 June	C3-C6	-6.34			-0.07		-2.30		-7.06
23 June	C4-Dead	-8.60			-0.18		1.11		-4.38
23 June	C4-C3	16.04	0.49		0.79		1.26		-7.58
23 June	C4-C4	-0.65			0.06		0.49		-12.39
23 June	C4-C5	-10.92	-2.28		-0.56 *		-0.35		-14.07 †
23 June	C5-C4	13.51			0.71		0.44		-8.10
23 June	C5-C5	6.88	0.84	10.26	0.31	0.39	-0.44	-0.38	-64.31
23 June	C5-C6	0.00	-0.98 *	-28.04 *	-0.02	-1.04 *	0.10	2.73 *	-66.37 †
23 June	C6-C6	37.91			1.58				-2.51

Table C2. Multiple regression model parameters for the dependence of plant seed production on log leaf nitrogen content and δ13C. Each census is modeled with a separate regression. The size class column indicates the size class of the plant at the census prior to seed collection. For the 23 June census the intercept models the log seed output of a *B. tectorum* individual at Smoot Hill of size class 5 and for the 27 July census the intercept models the log seed output of an average *B. tectorum* individual of size class 7; other parameters adjust the log seed output as appropriate. Missing parameters are missing because no plants of the appropriate category produced seed. Parameter values significantly different from zero at the $\alpha < 0.05$ level are indicated with an asterisk. Parameter values significantly different from zero at the $\alpha < 0.1$ level are indicated with a dagger symbol.

Census	Size class	Intercept	Escure	Smoot Hill	B. briziformis	$\delta^{13}C$	*B. briz* * δ13c	log (%N)	*B. briz* * log (%N)
23 June	1	8.62	-4.99	-3.47	-2.95	-0.07	0.00	-7.40	6.07
23 June	2	8.62	-24.34	-21.72	-21.73	-0.65	-0.97	4.29	-4.44
23 June	3	8.62	-5.38	-5.20	3.15	-0.03	0.09	-0.71	0.37
23 June	4	8.62	-1.72	-1.25	2.00	0.07	0.07	0.45	-0.71
23 June	5	8.62	-0.31		-3.10	0.11	-0.12	0.08	-0.56
23 June	6	8.62	1.30		0.00	0.19		1.22	
23 June	7	8.62	-2.07						
27 July	1	9.22	-6.60	-7.27 †	-1.72	-0.14	0.00	-2.76	0.00
27 July	2	9.22	-4.71 †	-4.97 *	-0.13	-0.05	0.00	-0.63 *	

Census	Size class	Intercept	Escure	Smoot Hill	B. briziformis	$\delta^{13}C$	*B. briz* * δ13c	log (%N)	*B. briz* * log (%N)
27 July	3	9.22	-17.25	-17.30	16.15 *	-0.51 *	0.58 *	1.85 *	-1.88 *

Table C2, continued.

Census	Size class	Intercept	Escure	Smoot Hill	B. briziformis	$\delta^{13}C$	*B. briz* * δ13c	log (%N)	*B. briz* * log (%N)
27 July	4	9.22	-1.50	-1.80	-0.88	0.04	-0.04	-0.27	0.26
27 July	5	9.22	-2.80	-3.02	0.54	-0.04	0.03	0.07	-0.06
27 July	6	9.22	-0.11	-0.31	-4.55	0.05	-0.17	0.02	0.08
27 July	7	9.22	0.03			0.04		0.21	