# Plant species traits and capacity for resource reduction predict yield and abundance under competition in nitrogen-limited grassland

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**Summary**

**1.** The objective of this study is to test whether plant traits that are predicted by resource-competition theory to lead to competitive dominance are correlated with competitive response and abundance in a nitrogen-limited grassland. We collected species trait and soil nutrient data on non-leguminous perennial prairie plant species in replicated monoculture plots established for this purpose.

**2.** The soil nitrate concentration of 13 species grown in long-term (5-year) monocultures (a measure of $R^*$) was correlated with their relative yield (a measure of competitive response) and with their abundance in competition. The trait best correlated with a species' relative yield was root length density (RLD), and the trait best correlated with abundance in competition was biomass : N ratio.

**3.** The traits that best predicted nitrate $R^*$ were the biomass : N ratio and allocation to fine roots, where species with higher biomass : N and allocation to fine roots had lower $R^*$. Easily measured species traits may therefore be useful proxy measures for $R^*$.

**4.** The dominance of species with lower nitrate $R^*$ levels and higher RLD and biomass : N in monoculture is qualitatively consistent with the prediction of resource-competition theory that the species most efficient at acquiring, retaining and using the major limiting resource will be the best competitors. Additional mechanisms are needed to explain how these species coexist.

*Key-words*: $R^*$, resource competition, root length density, soil nitrate, tissue nitrogen

## Introduction

Competition is a major factor structuring plant communities (Schoener 1983, Aarssen & Epp 1990; Goldberg & Barton 1992; Gurevitch *et al*. 1992). Plants compete to obtain limiting nutrients, and the productivity of many plant communities is strongly limited by nitrogen (Vitousek & Howarth 1991). Because successful species generally possess a suite of correlated traits (Weiher *et al*. 1999; Garnier *et al*. 2001), it is difficult to determine empirically which traits and mechanisms are responsible for competitive success (Grime 1979; Tilman 1990). The traits associated with dominance in strongly N-limited environments include low tissue N; high root : shoot ratio; long-lived tissue; high nutrient-use efficiency (Grime 1979; Chapin 1980; Vitousek 1982; Tilman 1990; Aerts & Chapin 2000); and the ability to reduce the concentra-

tion of mineral N to lower levels in monoculture (Tilman 1982, 1990; Tilman & Wedin 1991a; Wedin & Tilman 1993). The objective of the current study is to test whether traits that are predicted by resource-competition theory to lead to competitive dominance are correlated with abundance and competitive ability in an N-limited grassland (Tilman 1990).

Based on resource-competition theory, the equilibrial level to which a species can reduce the limiting resource in monoculture (the $R^*$ of the species) should determine the outcome of competition when species are limited solely by that resource (Tilman 1982). Specifically, the species with the lowest $R^*$ for a resource should be the best competitor for that resource. $R^*$ is measured empirically in monocultures as the concentration of available resource in the environment, and is predicted to integrate all the effects a species may have on resource levels, including uptake rates and rates of biomass loss (e.g. senescence, herbivory; Louda *et al*. 1990; Tilman 1990).

Although $R^*$ theory has been used extensively and successfully in aquatic systems (reviewed by Sommer

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2002), there are few tests in terrestrial systems (Miller *et al.* 2005). Notable exceptions include work by Tilman & Wedin (1991a, 1991b); Wedin & Tilman (1990, 1993), where monoculture plots were established to measure *R*\* for five grass species, and pairwise competition plots were established to determine competitive superiority. Soil nitrate concentrations in monoculture were used as a direct measure of *R*\* in these N-limited field experiments. There were significant differences in *R*\* among species, and the species with the lower *R*\* displaced or greatly reduced the other species in competition over 5 years. Thus these experiments support resource-competition theory and the predictive ability of *R*\* (cf. Harpole & Tilman 2006).

In this study, we measured *R*\* for a diverse group of N-limited plant species to test empirically whether the *R*\* values of competing species are correlated with their abundance, not in pairwise interactions, but in interactions among numerous species. Following the theory of Tilman (1982) and the experiments of Tilman & Wedin (1991a, 1991b), we measured *R*\* as soil nitrate concentrations in replicate monocultures of prairie grassland species. Soil nitrate concentrations are a better index of exploitative competition for N than soil ammonium concentrations, because nitrate diffuses more readily and adsorbs to soil exchange sites less readily than ammonium. Soil nitrate concentrations are therefore relatively more sensitive to plant uptake (Tilman 1989; Tinker & Nye 2000) and relatively less sensitive to heterogeneity in soil-exchange capacity (Tinker & Nye 2000). Thus soil nitrate concentrations are less likely than ammonium concentrations to be confounded by covariation with factors unrelated to the strength of exploitative plant competition.

Many different plant traits can influence *R*\* (Tilman 1990), and traits may be measured with differing degrees of ease and accuracy, raising the empirical question of which species traits explain the variation among species in *R*\*, and whether such species traits may be used successfully to predict variation among species in abundance and competitive ability. Specifically, it would be useful to develop proxy measures for *R*\* that do not necessarily require the establishment of equilibrial field monocultures and that are more easily sampled than soil nitrate, which is notoriously variable in space and time (Robertson *et al.* 1988; Robertson & Gross 1994, Cain *et al.* 1999; Hook & Burke 2000).

We focused on traits associated with the production of root biomass and root length, and the efficient use of N. These traits were selected because existing theory and field work show they can influence *R*\*. Tilman & Wedin (1991b) found that the competitively dominant species that most depleted soil nitrate also had high root biomass, as would be expected if high root biomass leads to high N uptake. Compared with root biomass, fine root length density (RLD: root length per volume of soil) might be an even better predictor of soil nitrate because fine roots are the organs responsible for most nutrient uptake. Theory (Tinker & Nye 2000) and

laboratory experiments (Hodge *et al.* 1999; Robinson *et al.* 1999) suggest that species take up N in proportion to their RLD. Thus traits associated with the production of high root biomass and high RLD may be well correlated with *R*\* and competitive ability.

Another factor that significantly influences *R*\* is tissue N concentration, where species that can maximize the production of biomass per unit of limiting resource and minimize nutrient loss via senescence will have lower *R*\* (Tilman 1990; Fargione & Tilman 2002). Biomass : N gives the amount of biomass maintained per unit N in the plant, and is one measure of the efficiency of N use. Biomass : N is also correlated with leaf longevity (Reich *et al.* 1997) such that species with high biomass : N will have long-lived tissues, which also will act to reduce *R*\* (Tilman 1990; Fargione & Tilman 2002). Nutrient-use efficiency (NUE) is a related but distinct measure of the efficiency of nutrient use; here we present analyses using the more easily measured biomass : N. The NUE is defined as the amount of organic matter produced per unit N, and is influenced not only by biomass : N ratios, but also by tissue longevity and retranslocation efficiency (Vitousek 1982). However, because tissue longevity is highly positively correlated with biomass : N (Reich *et al.* 1997), biomass : N ratios may capture much of the variation among species in NUE.

In contrast to previous studies, we tested the hypotheses that *R*\*-nitrate and species traits can predict competitive ability and abundance in mixed-species field plots. We used relative yield as a measure of competitive ability. Relative yield is defined as the ratio of a species' biomass in competition, divided by its biomass in monoculture; better competitors have higher relative yield. We used above-ground biomass in long-term plots planted with 16 species as our measure of abundance in competition. Because the measurement of *R*\* assumes that populations are at equilibrium, we used data from two sources to calculate *R*\*: a data set from short-term monocultures of 21 species (in their third growing season); and a data set from long-term monocultures of a more restricted set of 13 species (averaged over their fifth to ninth growing seasons).

## Methods

### STUDY SITE AND EXPERIMENTAL DESIGN OF SHORT-TERM MONOCULTURES

This study was conducted at Cedar Creek Natural History Area, located on a glacial outwash sandplain 50 km north of Minneapolis, MN, USA. Fertilization experiments have demonstrated that N is the primary limiting resource in these grasslands (Tilman 1987). In the growing season of 2000, 105 1 × 1-m short-term monoculture plots, five replicates of each of 21 non-leguminous prairie species, were established. We collected data in 2002, the experiment's third growing season. Prior to seeding, the ground was disked and treated with methyl bromide to eliminate the seed bank.

Because methyl bromide is also toxic to fungi, we inoculated plots with water that had been mixed with unsterilized soil and filtered through a 250-μm sieve of Nitex fabric to exclude weed seeds. This mesh was large enough allow the passage of most species of fungi (Schenck & Perez 1990), although spore density in our inoculum was not measured. Each plot received ≈1 l of water that had been mixed with ≈0·5 l soil from unsterilized surface soil adjacent to the experiment. Plots were seeded at a rate of 1600 viable seeds m$^{-2}$. This high level of seeding was intended to eliminate recruitment limitation. Bare patches apparent in the summer of 2001 were reseeded at similar densities. Seeds were obtained from Prairie Moon Nursery, Winona, MN, USA. We watered plots daily for 2 weeks after planting to prevent the topsoil from drying out and the seeds from blowing away. Plots were weeded several times each summer to maintain their status as monocultures. Because weeds were removed when small, and there was low weed recruitment (indicating that the methyl bromide treatment successfully eliminated the seed bank), weeding resulted in minimal disturbance.

### DATA COLLECTION FOR TRAITS IN SHORT-TERM MONOCULTURES

Data were collected in July 2002. We harvested above-ground biomass for tissue N measurements in a $10 \times 70$-cm strip in each plot, and sorted it into living and dead plant matter (hereafter above-ground biomass refers to only the living plant matter). We harvested below-ground biomass in two cores of 5 cm diameter and 20 cm deep in each strip. After washing off the soil, below-ground samples were sorted into fine roots (<1 mm), coarse roots (>1 mm) and crowns (the node between roots and shoots, used as a storage organ in some species). All biomass samples were dried and weighed. Below-ground biomass was calculated as the sum of the biomass of fine roots, coarse roots and crowns.

Tissue C and N were measured on a Costech Elemental Combustion System 4010 (Valencia, CA, USA) separately for fine roots, coarse roots and above-ground biomass + crowns. Because plant C concentrations are consistently 45% (Salisbury & Ross 1985; unpublished data), C concentrations <45% were assumed to indicate contamination of samples with soil particles. For such samples, N concentrations were corrected by dividing N concentrations by $[1 - (0.45 - C)]$, where $C$ is carbon concentration. Total tissue N concentrations were calculated as the biomass-weighted average tissue N concentrations in fine roots, coarse roots and above-ground biomass + crowns. Biomass : N ratios are simply the inverse of tissue N concentration.

To determine RLD (length of fine roots per volume of soil, cm cm$^{-3}$), tissue density, and average root diameter, a subsample of the fine root sample was scanned using a root digital image analysis system (WinRhizo, Régent Instruments, Inc., Québec City, Québec, Canada). Soil nitrate was extracted with 0·01 M KCl from two 2·5-cm diameter by 20-cm deep cores per plot and analysed on an Alpkem autoanalyser (Astoria-Pacific, Clackamas, OR, USA).

We also calculated root allocation and fine root allocation. Root allocation is defined as (root biomass)/(above and below-ground biomass); fine root allocation as (fine root biomass)/(total root biomass). These proportions are used rather than ratios for our analyses (e.g. root : shoot ratio) because the proportions are normally distributed and the ratios are not.

### DATA COLLECTION FOR ABUNDANCE IN COMPETITION, COMPETITIVE ABILITY AND LONG-TERM MONOCULTURE AVERAGE SOIL NITRATE

We measured above-ground biomass in competition and relative yield for 13 non-leguminous species using data from a long-term biodiversity experiment adjacent to our short-term monoculture plots (Tilman *et al.* 2001). Above-ground biomass was harvested, sorted to species, dried, and weighed from two $0.1 \times 3.0$-m strips per plot in 2001 and 2002. There were 35 16-species plots; each of the 13 species was planted in 28–34 of these plots. We used each species' 2-year average above-ground biomass from the 16-species plots as a measure of its 'biomass in competition'. To calculate relative yield, we divided the biomass in competition for each species by its 2-year average long-term monoculture biomass. Each species was planted in one to four long-term monoculture plots.

Because soil nitrate values may take longer than 3 years to equilibrate in new monocultures, we calculated average soil nitrate values for the 13 species planted in the long-term monoculture plots. Soil nitrate values were averaged over the years 1998, 1999, 2001 and 2002, the fifth to ninth years of the experiment (Tilman *et al.* 2001; no data available from 2000). This long-term multi-year average of soil nitrate is our best measure of $R*$, but is available only for 13 species. Thus for predicting abundance and competitive ability, which were measured for these same 13 species in the long-term biodiversity experiment, we use soil nitrate concentrations measured in the long-term monocultures of the same biodiversity experiment. For analyses with species trait data for 21 species from the short-term monocultures, we use the soil nitrate measurements from the same short-term monocultures.

### ANALYSES

The relationship between $R*$-nitrate and both measures of dominance (relative yield and biomass in competition) was highly non-linear, so we tested for correlations using a non-parametric rank correlation test. Other relationships were approximately linear and were analysed using linear regressions. Statistical analyses were conducted with JMP ver. 4·0·4 (SAS Institute Inc., Cary, NC, USA).
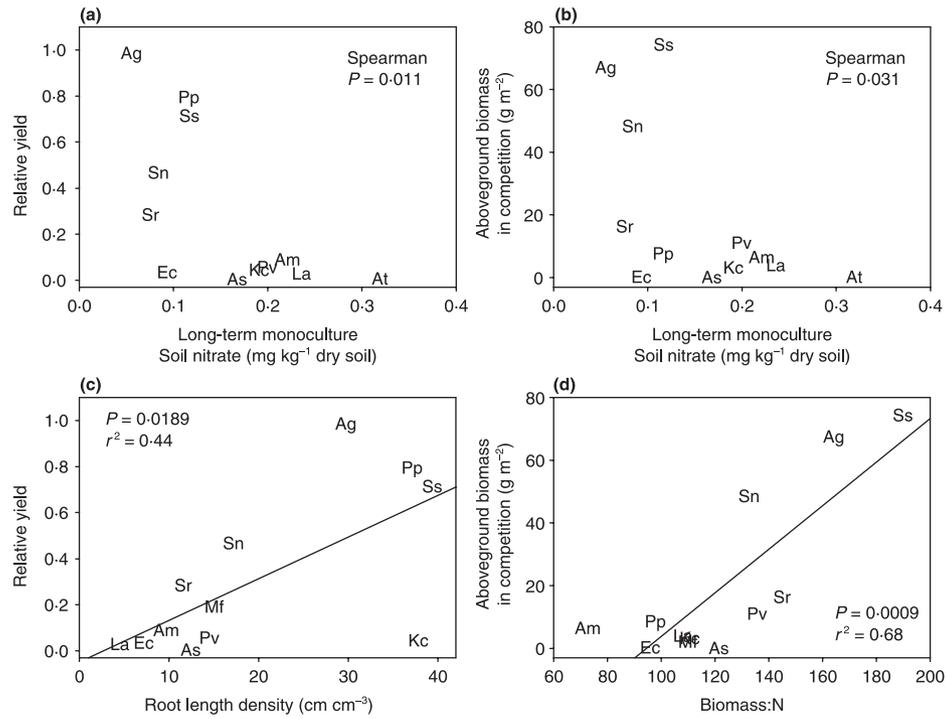
**Fig. 1.** Relative yield and biomass in competition *vs* species traits for 12 species. Relative yield was calculated using averaged data from years 2001 and 2002 of the one- and 16-species plots of an adjacent long-term biodiversity experiment. Soil nitrate is an average from 1998, 1999, 2001 and 2002 from the same long-term biodiversity experiment. See text for further details. Ag, *Andropogon gerardii*; Am, *Achillea millefolium*; As, *Agropyron smithii*; At, *Asclepias tuberosa*; Ec, *Elymus canadensis*; Kc, *Koeleria cristata*; La, *Liatris aspera*; Mf, *Monarda fistulosa*; Pp, *Poa pratensis*; Pv, *Panicum virgatum*; Sn, *Sorghastrum nutans*; Sr, *Solidago rigida*; Ss, *Schizachyrium scoparium*.

## Results

$R^*$-nitrate was negatively correlated with relative yield and biomass in competition (Fig. 1a,b). The relationship between $R^*$-nitrate and both measures of dominance was highly non-linear, so we tested for correlations using a non-parametric rank correlation test (Fig. 1a,b). All dominant species had low $R^*$-nitrate concentrations, but species with low relative yield and biomass in competition had a wide range of $R^*$-nitrate concentrations. The significant correlations between $R^*$-nitrate and

dominance in Fig. 1 used average $R^*$-nitrate data from the long-term monocultures. In contrast, the $R^*$-nitrate values obtained from a single year in our short-term monocultures were not significantly correlated with relative yield or biomass in competition (Spearman's rank correlation $P = 0.375$ and $0.142$ for relative yield and biomass in competition, respectively).

Of all the traits we measured, the best predictors of relative yield and biomass in competition were RLD and biomass : N, respectively (Fig. 1c,d). Most other traits were poorly correlated with abundance and competitive ability, although below-ground biomass in short-term monoculture was a significant predictor of abundance in competition (Table 1).

We conducted separate linear regressions of $R^*$-nitrate on each of eight species traits (Table 2). The traits that best correlated with $R^*$-nitrate were fine root allocation, root tissue density, below-ground biomass, and biomass : N, but these correlations were dependent on whether short- or long-term $R^*$-nitrate measurement was used as a response variable (Table 2).

We expected that $R^*$-nitrate concentrations and RLD would be highly correlated. However, this was not the case: RLD was not a significant predictor of $R^*$-nitrate concentrations in short-term monocultures (Fig. 2; $P = 0.14$, $r^2 = 0.11$). In our data, the presence of species possessing a combination of low RLD and low $R^*$-nitrate concentrations contradicted the predicted negative relationship between RLD and $R^*$-nitrate

**Table 1.** Simple linear regressions of two measures of species dominance *vs* species traits as measured in short-term monocultures for the 12 species for which all predictor and response variables were available

| Parameter | Relative yield | | | Biomass in competition | | |
|---|---|---|---|---|---|---|
| | $t$ | $P$ | $r^2$ | $t$ | $P$ | $r^2$ |
| Biomass : N | 2·12 | 0·060 | 0·31 | 4·63 | **<0·001** | 0·68 |
| Below-ground biomass | 2·13 | 0·059 | 0·31 | 3·78 | **0·004** | 0·59 |
| Root length density | 2·80 | **0·019** | 0·44 | 1·81 | 0·100 | 0·25 |
| Root tissue density | 0·89 | 0·392 | 0·07 | 1·60 | 0·141 | 0·20 |
| Root allocation | 1·45 | 0·177 | 0·17 | 0·20 | 0·842 | 0·00 |
| Average root diameter | −1·2 | 0·240 | 0·13 | −0·62 | 0·551 | 0·04 |
| Fine root allocation | 0·89 | 0·400 | 0·07 | 0·44 | 0·671 | 0·02 |
| Above-ground biomass | −0·56 | 0·587 | 0·03 | −0·03 | 0·979 | 0·00 |

The relationship between soil nitrate and dominance was non-linear and is presented in Figs 1 and 2. Significant $P$ values in bold.

**Table 2.** Linear regressions of $R^*$ measured in short- or long-term monocultures *vs* species traits measured in short-term monocultures

| Parameter | Short-term monoculture $R^*$ | | | Long-term monoculture $R^*$ | | |
|---|---|---|---|---|---|---|
| | $t$ | $P$ | $r^2$ | $t$ | $P$ | $r^2$ |
| Fine root allocation | −0·10 | 0·919 | 0·00 | −3·41 | **0·008** | 0·56 |
| Root tissue density | −0·25 | 0·807 | 0·00 | −2·20 | 0·056 | 0·35 |
| Below-ground biomass | −2·59 | **0·018** | 0·26 | −0·86 | 0·414 | 0·08 |
| Biomass : N | −2·55 | **0·020** | 0·25 | −1·18 | 0·268 | 0·13 |
| Root length density | −1·55 | 0·137 | 0·11 | −0·83 | 0·426 | 0·07 |
| Above-ground biomass | −0·64 | 0·533 | 0·02 | 1·06 | 0·315 | 0·11 |
| Root allocation | 1·12 | 0·278 | 0·06 | −0·94 | 0·373 | 0·09 |
| Average root diameter | 0·02 | 0·987 | 0·00 | 0·07 | 0·944 | 0·00 |

Regressions with $R^*$ in short-term monocultures included all 21 species; those with $R^*$ in long-term monocultures included the 11 species for which all predictor and response variables were available. Significant $P$ values in bold.
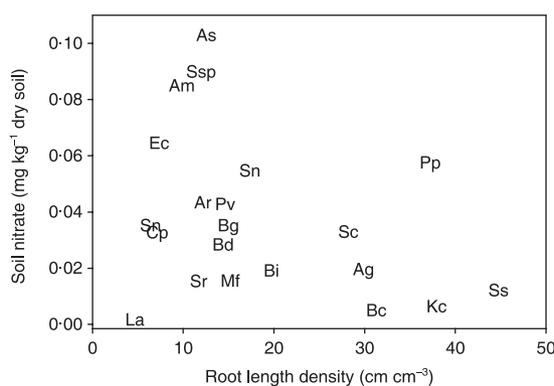


**Fig. 2.** Extractable soil nitrate concentrations *vs* RLD for 21 species; both variables were measured in the short-term monoculture plots. There was no significant linear relationship between these variables ($P = 0·14$, $r^2 = 0·11$). Log transformations did not improve the $r^2$ or $P$ of the fit. Spearman's non-parametric rank test was non-significant ($P = 0·1068$). Species abbreviations as in Fig. 1 and Ar, *Agropyron repens*; Bc, *Bouteloua curtipendula*; Bd, *Buchloe dactyloides*; Bg, *Bouteloua gracilis*; Bi, *Bromus inermis*; Cp, *Coreopsis palmata*; Sc, *Sporobolus cryptandus*; Ssp, *Stipa spartina*.

concentrations (however, no species had both high RLD and high $R^*$-nitrate). Specifically, removal of *Liatris aspera* from the data set resulted in a significant negative correlation between $R^*$-nitrate and RLD ($P = 0·044$, $r^2 = 0·21$). In a separate analysis of the long-term data from the biodiversity experiment, a negative relationship between root biomass and $R^*$-nitrate was observed for $R^*$-nitrate *vs* root biomass in the biodiversity experiment, when multiple years of data were analysed (Fig. 3). Analyses of single years showed similar trends, but were not significant ($P > 0·05$).

To explore the repeatability of $R^*$-nitrate measurements, we compiled data on soil nitrate concentrations for 22 non-leguminous species that were planted in monocultures in at least two experiments at Cedar Creek (Fig. 4). Although variation among experiments was high, there are still significant differences among species in

their $R^*$-nitrate concentrations. Significant differences among species were found in studies reporting data from a single experiment (Tilman & Wedin 1991b; Craine *et al*. 2002; Fargione *et al*. 2003), and we found significant differences among species using only the averages reported in Fig. 4 (42 of 253 pairwise comparisons were significant at $P < 0·05$; this many significant differences would happen by chance with a probability of only $1 \times 10^{-11}$).

We compared the coefficient of variation of each species' $R^*$-nitrate and RLD values in their five short-term monoculture replicates. Comparing all 21 species in a matched-pairs comparison, RLD was less variable than $R^*$-nitrate ($P < 0·0001$). The coefficient of variation for $R^*$-nitrate was, on average, 3·8 times larger than for RLD. For each of the 21 species, RLD was less variable than $R^*$-nitrate. In our study, the higher variation in $R^*$-nitrate could be associated both with higher heterogeneity for soil nitrate than for RLD, and with the smaller area sampled for $R^*$-nitrate (the 3140 cm$^3$ sampled for RLD was 4·1 times greater than the soil volume sampled for $R^*$-nitrate).

## Discussion

Species with high relative yield and biomass in competition had low $R^*$, as measured by soil nitrate concentrations in long-term monocultures. This suggests that the dominant species in this community are those that are most efficient at acquiring and using the primary limiting resource. Although there was substantial residual variation in both response variables, all the dominant species had low $R^*$-nitrate. In addition, biomass : N and RLD were the best predictors of relative yield and biomass in competition, respectively. In total, our results suggest that these species characteristics may provide a useful predictor of dominance on low-N soils.

Surprisingly, biomass in competition was uncorrelated with above-ground biomass in short-term monoculture (Table 1; similar results were found using long-term monoculture biomass, data not shown). This suggests that abundance is more strongly influenced by interactions with other species, such as competition, than by the factors controlling abundance in monoculture, and is consistent with our result that traits associated with competitive ability can predict species abundance. More generally, our results suggest a positive correlation between competitive ability and species abundance.

Low $R^*$, high RLD, and high biomass : N may be part of a suite or 'syndrome' of plant traits that lead to dominance in low-N systems. Our results are consistent with research on the traits of species successful in nutrient-poor habitats (Grime 1979; Chapin 1980; Aerts & Chapin 2000). Species that have high biomass : N and high allocation to roots can produce more roots per unit N, and will therefore achieve a higher equilibrial root biomass in monoculture (Tilman 1990). Species with thinner roots have greater RLD than species with thicker
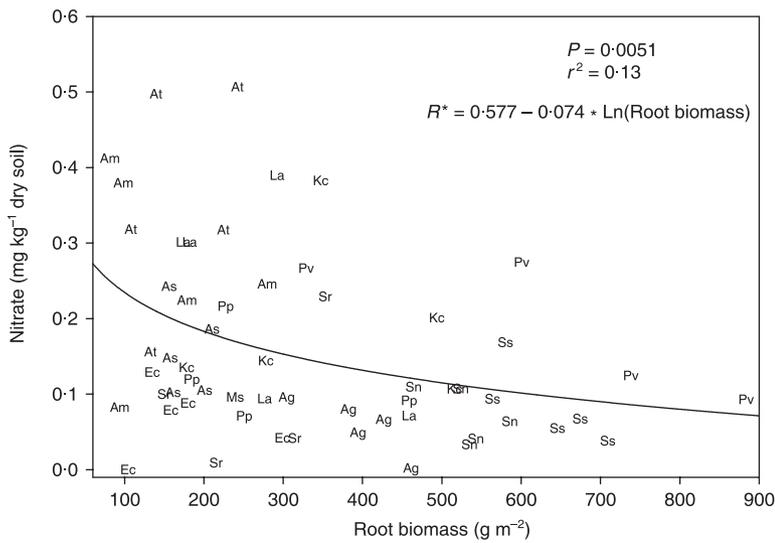
**Fig. 3.** Extractable soil nitrate concentrations *vs* root biomass for 12 species; both variables were measured in the long-term monoculture plots of the biodiversity experiment. Each point represents the value (average of one to four monocultures) for one species from a single year 1997, 1998, 1999, 2001 and 2002. Species abbreviations as in Fig. 1.
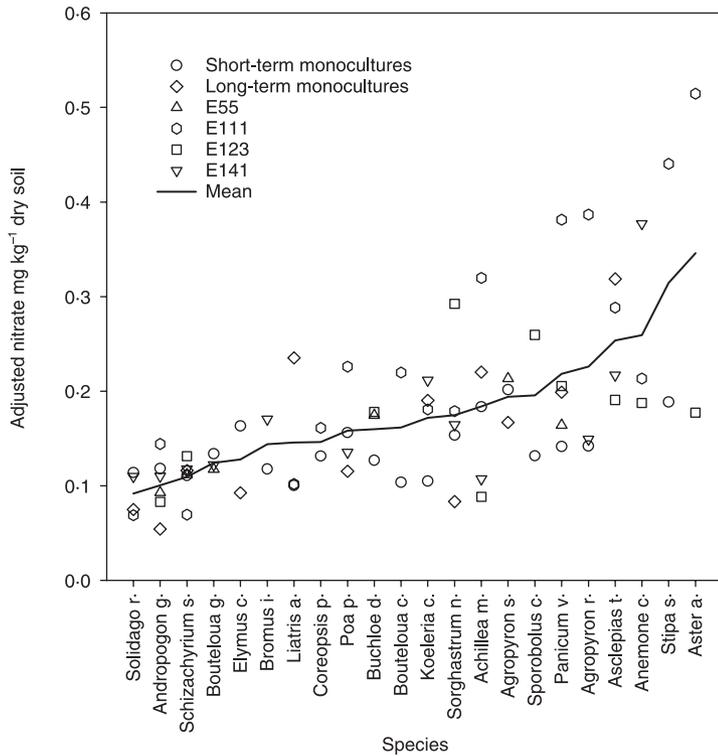


**Fig. 4.** Adjusted extractable soil nitrate concentrations from monocultures of 22 species that occurred in two or more of six experiments at Cedar Creek, sorted in order of increasing average concentration. Because sites differ in fertility, we used mean pairwise differences to adjust all values to be comparable with the values for the long-term monocultures (values used in Fig. 1). Species from experiments that differed from the long-term monocultures were adjusted as follows; +0·0996 for short-term monocultures; –0·0490 for E55; –0·0919 for E123; +0·0969 for E141. Soils in all experiments were extracted with 0·01 м KCl, except for E141 (1 м KCl). References for experiments: E55, Tilman & Wedin (1991b); E111, Craine *et al*. (2002); long-term monocultures: E120, Tilman *et al*. (2001); E123, Tilman *et al*. (1996); E141, (Reich *et al*. 2001); short-term monocultures: E147, this paper. Species are as listed in Figs 1 and 2, plus *Asclepias tuberosa* and *Aster azureus*.

roots (Eissenstat 2000; Wahl & Ryser 2000). Species with high biomass : N ratios also tend to have long-lived tissues (Reich *et al*. 1997). Rates of herbivory on most of these species have been measured under laboratory conditions, and have been shown to be lower for the more abundant, lower tissue-N species (Burt-Smith *et al*. 2003). This suite of traits – including high biomass : N, high fine root allocation, long-lived tissues, and low herbivory – should all lead to high standing biomass, high RLD, low $R^*$, and therefore high competitive ability.

Many studies linking traits with abundance do so across environmental gradients rather than within a habitat, or link traits with abundance or growth rate in short-term bioassays rather than with abundance of species in established field communities. Still, a number of studies have tried to link species traits with abundance in the field. Reader (1998) found significant correlations between the relative abundance of seven species in an infertile abandoned pasture and five of nine traits studied. High root : shoot ratio was correlated with abundance, while root biomass and tissue N concentrations were not. In addition, slow growth rate, low shoot biomass, low herbivory and high mycorrhizal infection were associated with high abundance. Mamolos *et al*. (1995) found that abundance was correlated with tissue N concentrations in an infertile Mediterranean grassland, although above-ground biomass of species significantly differed among years, and these correlations were not found in all years. Tsialtas *et al*. (2001) found that abundance in a Mediterranean grassland was correlated with $\delta^{13}C$ values, indicating that species with higher water-use efficiency were more abundant. Epp & Aarssen (1989) found no relationship between relative abundance of 10 species and five traits in an abandoned hayfield, although they measured traits of seeds and growth rather than roots or nutrients. Theodose *et al*. (1996) found that, for 13 species of tundra plants, high relative abundance was correlated with high root : shoot ratio and total biomass of individual plants, but not with $^{15}N$-uptake rates or tissue N concentrations. Harpole & Tilman (2006) found that, for 27 prairie plants, the relationship between $R^*$ and abundance changed predictably over successional time and with N addition. Thus, although the specific traits differed among studies, a growing number of studies, including this one, have found that traits associated with efficient acquisition and use of limiting resources are correlated with species abundance. Additional studies in this area will allow analysis of the conditions under which different traits predict species abundance.

The best correlation between $R^*$-nitrate and species traits was between $R^*$-nitrate in long-term monocultures and allocation to fine roots (Table 2). This pattern was largely driven by *Panicum virgatum*, which had the highest $R^*$-nitrate and the lowest allocation to fine roots. We expected RLD to be well correlated with $R^*$-nitrate levels because RLD has been shown to predict efficiency of N uptake (Hodge *et al*. 1999; Robinson

*et al.* 1999). *R\**-nitrate and RLD were uncorrelated in the short-term monocultures (Fig. 2), and *R\**-nitrate and root biomass were significantly correlated when multiple years of data were available, with an $r^2$ of 0·13 (Fig. 3). There are two factors that could contribute to the poor correlation between RLD and *R\**-nitrate. First, soil nitrate concentrations are notoriously heterogeneous in space (varying both on the scale of centimetres and between plots) and time (varying seasonally, interannually and with precipitation) (Robertson *et al.* 1988; Davidson *et al.* 1990; Robertson & Gross 1994, Cain *et al.* 1999; Hook & Burke 2000). Obtaining a consistent average value for a species can be difficult because of this heterogeneity, especially with few replicate plots, or with few soil samples per plot.

In addition to sampling problems, there are several possible mechanisms for how species could cause low *R\**-nitrate despite having low RLD, such as occurred especially for *L. aspera* and also possibly for *Solidago rigida* in our data (Figs 2 and 4). It is interesting to note that, in an adjacent experiment, *S. rigida* and *Achillea millefolium* had a high fraction of cell-soluble C, low net mineralization rates, and low levels of soil nitrate, suggesting that root exudates or decomposing roots could promote immobilization (Dijkstra *et al.*, 2006). However, we note that these species have low abundance in the presence of other species, suggesting that reducing soil nitrate via immobilization does not allow a species to become abundant in mixture. Thus these species may shift N from the soil nitrate pool to the microbial pool, but do not prevent competitors from obtaining N when it is remineralized. In contrast, successful competitors in our system had, as predicted by resource-competition theory, efficient nutrient acquisition, use and retention, all of which increase the size of that plant species' N pool rather than the microbial N pool.

Although *R\** and species traits explained some of the variation in species dominance, other unmeasured factors may influence species interactions in the field. For example, seed size (Gross & Werner 1982); phenology; rooting depth (McKane *et al.* 1990); ability to access atmospheric N; limitation by other resources such as phosphorus, light or water (Tsialtas *et al.* 2001); and interactions mediated by herbivores, pathogens or symbiotic fungi may all influence species abundance (Carson & Root 2000; Packer & Clay 2000; Klironomos 2002; Bever 2003). These factors, along with species differences in uptake capacity per unit root length and differences in N demand, may explain some of the residual variance in our data. For example, we found that *Koeleria cristata* had high RLD but low relative yield. This species is a short-statured, early season $C_3$ grass with relatively shallow roots. It is possible that the timing or depth of nutrient uptake, a high percentage of inactive roots, poorer water-use efficiency compared with the $C_4$ grasses, or shading due to short stature may explain why its relative yield is low despite the values of its *R\** and RLD.

## Conclusion

Soil nitrate in long-term monoculture, a measure of *R\**, was significantly correlated with relative yield and abundance. In addition, the traits that best predicted relative yield and abundance were RLD and biomass : N, respectively. Biomass : N, below-ground biomass and fine root allocation were correlated with lower *R\**. This suggests that the dominant species in this N-limited community were those that were most efficient at acquiring, using and retaining limiting resources. The dominant competitors in our system all have high biomass : N (use N efficiently), have thin roots, and maintain high RLD and low soil nitrate concentrations in monocultures. Our results suggest that species functional traits may provide a useful predictor of species dominance in N-limited systems. The generally positive correlation between traits associated with competitive ability and species abundance suggests a strong role for competition in community assembly (Harpole & Tilman 2006).

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