Species evenness and invasion resistance of experimental grassland communities

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Concern for biodiversity loss coupled with the accelerated rate of biological invasions has provoked much interest in assessing how native plant species diversity affects invasibility. Although experimental studies extensively document the effects of species richness on invader performance, the role of species evenness in such studies is rarely examined. Species evenness warrants more attention because the relative abundances of species can account for substantially more of the variance in plant community diversity and tend to change more rapidly and more frequently in response to disturbances than the absolute numbers of species. In this study, we experimentally manipulated species evenness within native prairie grassland mesocosms. We assessed how evenness affected primary productivity, light availability and the resistance of native communities to invasion. The primary productivity of native communities increased significantly with species evenness, and this increase in productivity was accompanied by significant decreases in light availability. However, evenness had no effect on native community resistance to invasion by three common exotic invasive species. In this study, niche complementarity provides a potential mechanism for the effects of evenness on productivity and light availability, but these effects apparently were not strong enough to alter the invasibility of the experimental communities. Our results suggest that species evenness enhances community productivity but provides no benefit to invasion resistance in otherwise functionally diverse communities.

The study of biological invasions has a long history in ecology as a key avenue of insight into the processes controlling species distributions and abundances and the structure and functioning of ecosystems (Elton 1958). Furthermore, biological invasions are occurring at an unprecedented rate and posing serious threats to natural and managed systems worldwide, often resulting in substantial ecological and economic losses (Vitousek et al. 1997). For example, invasive species can disrupt ecosystem processes by displacing native species, altering patterns of resource use and modifying disturbance regimes (Mack and D’Antonio 1998, Mack et al. 2000), and these ecological consequences of invasions jeopardize native biodiversity and associated ecosystem services (Chapin et al. 2000). Invasive species also cause substantial economic losses by reducing the output of plant and animal production of managed systems and warranting costs associated with management practices (Pimentel et al. 2000).

Focusing on attributes of invaded communities, a critical question addresses why communities vary in their resistance to invasions (Lonsdale 1999). Elton (1958) hypothesized that a community’s resistance to invasion increases with species diversity. Modern ecologists have provided a mechanistic basis for Elton’s idea, proposing that species-rich communities either offer fewer vacant niches (niche complementarity effect of native species richness) or a greater probability that an invader will be competitively excluded by a superior competitor (sampling effect of native species richness) (Tilman 1999, Wardle 2001, Fargione and Tilman 2005). Tilman (1999) demonstrated that both of these diversity effects operate through resource reduction, with higher native species richness leading to greater resource reduction, greater community productivity, and lower invasibility in both heterogeneous (for niche complementarity) and homogenous (for the sampling effect) environments.
A substantial body of work testing the relationship between species richness and invasibility has emerged, with variable results that are in part due to the differing scales and contexts of studies (reviewed by Levine and D’Antonio 1999). However, species richness is not the only measure of community diversity and does not always correlate positively with other measures (e.g. species evenness), which may be a contributing factor to the variable results of richness—invasibility studies (Wilsey et al. 2005). Species evenness, or the relative abundances of species in a community, has rarely been considered in studies of community invasibility but warrants more attention for a number of reasons. Experimental studies manipulating species richness typically impose high levels of initial species evenness (Schwartz et al. 2000), but natural plant communities often exhibit low evenness (Weiher and Keddy 1999). Furthermore, relative abundances of species can account for substantially more of the variance in plant community diversity than does species richness (Stirling and Wilsey 2001, Wilsey et al. 2005) and tend to change more rapidly and more frequently in response to human activities and other disturbances than the absolute numbers of species within a community (Chapin et al. 2000). In addition, theoretical work predicts that evenness affects the extent to which niche complementarity results in exploitation of resource-based niches, with greater evenness resulting in greater resource drawdown and thus greater productivity and less opportunity for successful invasion (Nijjs and Roy 2000), though empirical studies testing this prediction offer little support to date (Polley et al. 2003).

To our knowledge, only one study has used a manipulative experiment to explicitly test the relationship between species evenness and the susceptibility of a community to invasion. Wilsey and Polley (2002) demonstrated that increased evenness of grassland plots resulted in a decrease in the natural recruitment of native species, though the response depended largely on the identity (e.g. monocotyledonous vs dicotyledonous invaders) of the recruiting species. Here we report the results of a greenhouse experiment designed to test whether native grassland species evenness promotes greater native community productivity, reduced light availability, and greater resistance to exotic invaders.

**Methods**

**Study species**

We assembled native communities from a pool of 25 species common to mesic prairies of the mid-western United States. Species within this pool were divided among five functional groups: C₃ grasses (*Agropyron smithii*, *Bromus kalmii*, *Elymus canadensis*, *Elymus virginicus* and *Glyceria grandis*); C₄ grasses (*Andropogon gerardii*, *Panicum virgatum*, *Schizachyrium scoparium*, *Sorghastrum nutans* and *Spartina pectinata*); composites (*Aster novae-angliae*, *Coreopsis tripteris*, *Echinacea purpurea*, *Helianthus helianthoids* and *Rudbeckia hirta*); legumes (*Amorpha canescens*, *Astragalus canadensis*, *Chamaecrista fasciculata*, *Desmanthus illinoensis* and *Desmodium canadense*); and a miscellaneous category denoted as other (*Asclepias tuberosa*, *Monarda fistulosa*, *Penstemon digitalis*, *Prunella vulgaris* and *Pycnanthemum tenuifolium*). Our study also included three exotic plant species: *Lolium arundinaceum*, *Melilotus alba* and *Rumex crispus*, all of which are common invaders of mid-western grasslands. *Lolium arundinaceum* is a highly persistent, cool-season grass that can substantially reduce grassland biodiversity, especially when infected by the endophytic fungus *Neotyphodium coenophialum* (Clay and Holah 1999). The abundances of *Melilotus alba* (a nitrogen-fixing legume) and *Rumex crispus* (a non-leguminous forb) are also negatively correlated with the performance of native species within grassland communities (Parker et al. 1993, Tracy and Sanderson 2004). Seeds of native species were obtained commercially (J. F. New Native Plant Nursery, Walkerton, IN, USA), and seeds of exotics were collected from naturally growing populations in the mid-west.

**Experimental design**

Native functional group richness, species richness, and total planting density were all held constant at five functional groups, five species, and thirty individuals per pot, respectively. We established three levels of native species evenness, with species abundances in the ratios of 1:1:1:1:6 (low evenness), 1:1:3:5:5 (medium evenness), and 1:1:1:1:1 (high evenness). Within each community, individuals were equally spaced, and species positions were randomized. Each level of evenness was replicated 30 times, yielding a total of 90 communities. For each replicate, a species representing each functional group was randomly selected from the corresponding species pool, and then these five species were randomly assigned to the five species abundances for each evenness treatment (Fig. 1). Thus, while all replicate communities contained five species and five functional groups, the species composition of each replicate, including which species or functional group was dominant within a given low or medium evenness replicate, was an independent random draw. Our design automatically excluded the sampling effect associated with manipulations of species or functional group richness. Furthermore, by randomizing each of the 90 replicate communities with respect to species composition and functional group dominance, our design avoided compositional effects.
Our design therefore favored the detection of evenness effects per se, such as those that may operate through enhanced niche complementarity (Nijs and Roy 2000). To establish an invasion treatment, we transplanted two individuals of each of the three exotic invasive species into half of the native communities within each level of evenness. The pots lacking exotic species served as controls.

Seedlings of each study species were grown from cold stratified seed (4°C for 10 d in wet sand) sown into flats of pasteurized soil. Germinating seeds were maintained at 21–24°C with a photoperiod of 14 h using artificial lighting. Seedlings of both native and exotic species were transplanted into pots at approximately 4 weeks of age. Thus, our experiment simulated a restoration context in which native and exotic species enter plots simultaneously (e.g. from the intentional seeding of native species into bare soil with recruitment of exotic species from seed banks or seed dispersal).

The experimental communities were grown in 7.65-l pots (20 cm diameter × 32 cm depth) in an Indiana Univ. greenhouse facility during the autumn of 2002. We filled all pots with a 2:1 mixture of medium-fine sand and old field topsoil that was sieved through 1-cm mesh. This sand-soil mixture exposed plants to natural microbial associates and facilitated the harvest of root biomass. We replaced all seedlings that died within the first week of transplanting. Experimental pots were watered and weeded as needed. We positioned all pots within a complete randomized block design of fifteen blocks, each containing all six combinations of species evenness and invasion treatments.

After a 4-month growing season, we counted the number of surviving individuals of the invasive species in each pot and harvested the communities. Shoots were clipped at stem bases, sorted by species, dried to a constant weight at 65°C, and weighed (± 1 mg). Due to the difficulty in separating root tissue by species in mixtures, we only obtained measurements of total root biomass per pot. Roots were sieved from pots, washed in tap water, and processed in the manner described for the shoots. Prior to harvest of the uninvaded control pots, we used a ceptometer to measure the level of light penetration (photosynthetically active radiation, PAR, measured as μmol m⁻² s⁻¹) into the canopy two cm above the soil surface in replicate pots from each evenness level.

Data analysis

Data were analyzed using SYSTAT statistical software (ver. 10.2, SPSS Inc.). We used general linear models to test for treatment effects on shoot and total native biomass (univariate models) and shoot biomass and survivorship of exotic species (multivariate models), and to quantify the relationship between shoot biomass and light penetration in the uninvaded control pots (regression model). Because each community consisted of an independent random draw with respect to both species composition and functional dominance, we could test for an effect of species evenness but not of species composition or functional group dominance on our response variables (Loreau and Hector 2001, Mikola et al. 2002). We tested assumptions of homogeneity of variance and normality by inspecting residual plots (normal probability plots, histograms, and residual-estimate scatter plots). Data were log-transformed to meet the assumptions of ANOVA (normalized, homoscedastic residuals) and to remove any artifacts of scale from interaction terms. We used
the Tukey-Kramer HSD test (Bonferroni-protected) to make pairwise mean comparisons on significant effects.

Results

Native community productivity, measured as total aboveground plus belowground biomass produced over the growing season, increased significantly with evenness in the absence of exotic species ($F_{2,28} = 4.19$, $p = 0.026$; Fig. 2) and was inversely related to light availability ($y = -0.04x + 0.92$, $R^2 = 0.33$, $p < 0.001$). However, native species evenness had little effect on community invasibility in our experimental grassland communities. There was a tendency for invasion to reduce the final diversity of low but not higher evenness communities (Fig. 3), but this effect was not significant (evenness × invasion interaction: $F_{2,70} = 2.15$, $p = 0.124$). Furthermore, native aboveground biomass decreased significantly in the presence of exotic species ($F_{1,70} = 11.78$, $p = 0.001$) regardless of the level of evenness (evenness × invasion interaction: $F_{2,70} = 0.46$, $p = 0.633$; Fig. 4). Evenness had no effect on the aboveground biomass or survivorship of any exotic species (aboveground biomass: Wilks’ lambda $F_{6,52} = 0.09$, $p = 0.997$; survivorship: Wilks’ lambda $F_{6,52} = 1.511$, $P = 0.193$; Fig. 5). *Melilotus alba* was the dominant invader at all levels of evenness (Fig. 5).

Discussion

Our results suggest that species evenness promotes grassland productivity and reduces light availability but is a relatively unimportant determinant of community vulnerability to invasion by three common exotic species. The effects of diversity on community properties (e.g. productivity and invasibility) are often explained by sampling or niche complementarity effects (Naeem et al. 1994, Tilman et al. 1997, Tilman 1999, Wardle 2001), but distinguishing between these effects can be difficult in studies that manipulate species richness as a surrogate for diversity because the effects of species richness are easily confounded with those of species identity (Wilsey and Porvin 2000, Wilsey and Polley 2002). The design of our study maintained a constant level of both species and functional group richness across the evenness treatments and thus avoided the sampling effects associated with richness manipulations. Our design also excluded compositional effects because both species composition and the identity of the dominant species in each replicate were randomly selected. Thus, the positive relationship that

![Fig. 2. Response of native communities (aboveground plus belowground biomass) to evenness treatments. Data represent means +1 SE, and different letters indicate significant differences ($p < 0.05$) among evenness treatments.](image2.png)

![Fig. 4. Response of native communities (aboveground biomass) to invasion treatments at each level of evenness. Data represent means +1 SE.](image4.png)

![Fig. 3. Response of native species diversity (Shannon-Weiner H) to evenness treatments in both the presence and absence of exotic invaders. Data represent means +1 SE.](image3.png)
we observed between evenness and native community productivity may be best attributed to an effect of niche complementarity. Theoretical studies predict that complementarity in resource use increases with species evenness in functionally diverse communities (Nijs and Roy 2000). Our measurements of light depletion support this interpretation, suggesting that canopy space was more completely filled in more even, and thus more productive, communities. Of the few other empirical studies to date, Wilsey and Potvin (2000) also detected a positive relationship between evenness and aboveground productivity, but negative (Mulder et al. 2004) and neutral (Polley et al. 2003) relationships have been observed as well.

Although the patterns of productivity and light depletion in our study suggest that resource use by native species was more complete under conditions of high species evenness, this effect of evenness was apparently not strong enough to resist invasion. Our results contrast with studies that manipulate species richness. Such studies generally show that resistance to invasion increases with species richness and often implicate niche complementarity in above- and/or belowground resource use as a mechanism of invasion resistance (Naeem et al. 2000, Hector et al. 2001, Lindig-Cisneros and Zedler 2002). Our study manipulated species evenness while holding richness constant at five species from five different functional groups. Certainly, the presence vs absence of species and/or functional groups achieved via manipulations of richness might be expected to have stronger effects on niche complementarity and associated drawdown in resource use than would variation in the abundances of individuals (achieved via manipulations of evenness). Of course, our study was a single-season mesocosm study, and whereas most richness–invasibility experiments have also been single-season in length, they have been conducted at larger scales in the field (Hector et al. 2001). On the other hand, the diversity effects of resident vegetation are expected to play out at the localized scale of plant neighborhoods, an expectation that is supported by both field (Kennedy et al. 2002) and complementary field and mesocosm studies (Naeem et al. 2000). To date, however, richness versus evenness effects on invasibility have not been directly compared, and too few evenness studies exist to draw firm conclusions about the importance of evenness or how its effect might vary with a community’s level of functional diversity.

Our study was not designed to separate the effects of the three invaders, and it is possible that the nature of the evenness–invasibility relationship is contingent on the identity of the invading species. For example, Wilsey and Polley (2002) found that high species evenness decreased invasion of grassland by naturally-recruiting dicotyledonous species, whereas species evenness had a negligible effect on resistance to monocotyledonous invaders. Observational studies of evenness–invasibility relationships have also been variable, demonstrating both positive (Robinson et al. 1995) and negative (Tracy and Sanderson 2004, Tracy et al. 2004) results, and suggesting that invasibility can also be contingent upon the identity of the dominant native species within low-evenness communities (Crawley et al. 1999, Smith and Knapp 1999, van Ruijven et al. 2003, Smith et al. 2004). In our study, invader biomass was dominated by the nitrogen-fixer Melilotus alba. Nitrogen fixation can provide a strong competitive advantage to exotics in nutrient-poor soils, especially when the ability to fix nitrogen is absent in the native community (Vitousek et al. 1987, Vitousek and Walker 1989, Musil 1993). Although all of our native communities contained native nitrogen fixers, it is possible that nitrogen-fixing ability varied among species. Escape from herbivores or pathogens (Tilman
1999), for example, might allow exotic invasives to allocate relatively more resources than native species to nitrogen-fixing symbionts. Quantification of nitrogen-fixing associations and nitrogen content in plants and soil would be helpful in addressing these hypotheses. More generally, studies are needed to evaluate how the identities of both the invaders and the dominant residents affect the relationship between species evenness, above- and belowground resources, and community invasibility.

In conclusion, results of this study suggest that species evenness enhances community productivity but provides no benefit to invasion resistance in functionally diverse communities. However, additional studies are needed to determine whether the effect of evenness on invasion resistance is contingent upon the functional diversity of the invaded community as well as the traits of both the resident and invading species.

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References

Elton, C. S. 1958. The ecology of invasions by animals and plants. – Methuen.


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