

Persistence in the seed bank: The effects of fungi and invertebrates on seeds of native and exotic plants¹

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Abstract: Fungal pathogens and invertebrate seed predators both may limit the establishment of persistent seed banks. Exotic plants may be less subject to attack by these natural enemies than are native species; if so, this may provide exotics with an important advantage by reducing seed mortality and enhancing the persistence of colonizing populations. We investigated this idea with a field experiment in which we used fungicides and screening to protect seeds of 39 native and exotic old field plants. The recovery of germinable seeds was improved by fungicide additions, but not by the exclusion of macroinvertebrates. The effects of fungicide varied among species and sampling dates, but the recovery of natives was not consistently improved more than the recovery of exotics. These results suggest that soil fungi have important impacts on the seed banks of many species, but that exotics are not consistently more resistant than natives to attacks by either fungal pathogens or invertebrates. Consequently, it is unlikely that the majority of invaders benefit from reduced pest loads at the seed bank stage.

Keywords: seeds, biological invasions, dormancy, exotics, fungi, old fields, pathogens.

Résumé : Les organismes pathogènes fongiques et les invertébrés prédateurs de graines peuvent nuire à la formation de réservoirs de graines qui persistent dans le sol. Les plantes exotiques pourraient être moins sujettes aux attaques de ces ennemis naturels que les espèces indigènes. Si tel était le cas, les plantes exotiques auraient alors un avantage compétitif certain : la mortalité des graines serait moins élevée, tout en favorisant la pérennité des populations. Nous avons testé cette hypothèse de façon expérimentale dans un champ en friche. Nous avons protégé, à l'aide d'un fongicide et de treillis, les graines de 39 espèces indigènes et exotiques. Le réservoir s'est enrichi de graines viables à la suite de l'application du fongicide, mais le fait d'exclure les macroinvertébrés n'a pas eu d'influence sur le réservoir. Les effets du fongicide ont varié selon l'espèce et la date de l'échantillonnage; toutefois, la reconstitution du stock de graines dans le réservoir n'a pas été plus marquée chez les espèces indigènes que chez les espèces exotiques. Ces résultats indiquent que les champignons du sol ont un impact significatif sur le réservoir de graines de plusieurs espèces, mais que les plantes exotiques ne sont pas plus résistantes aux attaques des champignons ou des invertébrés. En conséquence, il est peu probable que le réservoir de graines de la majorité des plantes envahissantes soit moins affecté par les pathogènes ou les prédateurs que celui des plantes indigènes.

Mots-clés : graines, invasions biologiques, dormance, plantes exotiques, champignons, champs en friche, organismes pathogènes.

Introduction

Invasions by alien (exotic) plants have altered the structure and function of ecosystems in many regions of the world (Elton, 1958; Drake *et al.*, 1989; Williamson, 1996; Mack *et al.*, 2000). For example, aliens frequently make up 25% to 35% of the species of local floras in temperate North America (Whitney, 1994) and in some cases threaten to displace native vegetation (US Congress, 1993; Vitousek *et al.*, 1996; Mack *et al.*, 2000). Still, despite their prominence in many floras, the majority of potential invaders never become established or have no noticeable impacts if they do (Williamson & Fitter, 1996a,b; Mack *et al.*, 2000). Many researchers have attempted to explain which invasions succeed and which fail, but despite some successes (Rejmánek & Richardson, 1996; Reichard & Hamilton,

1997), few efforts to date have produced rules with strong predictive value (Mack, 1996; Williamson, 1996; Mack *et al.*, 2000).

One possibility is that invaders may benefit from low rates of attack by natural enemies such as invertebrate herbivores and microbial pathogens. This idea is important to biological control, which often attempts to reduce populations of exotic plants by introducing or supplementing populations of natural enemies, and successful efforts to control exotics such as *Opuntia* spp. in Australia (Mann, 1970) and South Africa (Zimmermann, Moran & Hoffmann, 1986) and *Hypericum perforatum* (nomenclature for North American species follows Morton & Venn, 1990) in California (Huffaker & Kennett, 1959) do suggest that some invasions may be attributable to low rates of attack. There are two distinct ways this could occur, which differ in the mechanism causing this low pest load. First, invaders may lose their natural enemies when they are transported to a new area (the "predator escape" hypothesis; Elton, 1958;

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Crawley, 1986; 1987). Alternatively, species with intrinsically low pest loads may make better invaders because they are less likely to be eliminated by natural enemies in their new habitat (the "predator filter" hypothesis; Blaney & Kotanen, 2001a,b). These two hypotheses are difficult to distinguish, but the escape hypothesis predicts that invaders should have lower pest loads in new habitats than in their native regions, while the filter hypothesis predicts that pest loads should be equally low in both native and new areas. Maron and Vilà (2001) and Keane and Crawley (2002) both reviewed evidence for the predator escape hypothesis, but found only inconsistent support.

Plants are likely to be especially vulnerable to enemies that attack critical life history stages. Seeds, which are subject to attack by a wide range of fungal pathogens, invertebrate granivores, and other enemies (Leck, Parker & Simpson, 1989; Baskin & Baskin, 1998), may be such a stage for invasive plants. Seeds represent the primary opportunity most plants have for dispersal, which is necessary for the invasion of new areas. Following dispersal, seed banks may be important in buffering against the stochastic hazards faced by small populations (Venable & Brown, 1988; Stöcklin & Fischer, 1999), though perhaps at the cost of reduced rates of population growth (Rees & Long, 1992) or reduced success at subsequent stages of invasion (Kolar & Lodge, 2001; Sakai *et al.*, 2001). Invading species generally face these hazards repeatedly, both at the original site of introduction and with range expansion, as small colonizing subpopulations are formed (Crawley, 1989). There also is empirical evidence that invasiveness may be related to seed biology. For example, invasiveness in *Pinus* and other woody plants is correlated with large seed crops and animal dispersal, as well as with small seed size, which is believed to be correlated with dispersability (Rejmánek, 1995; 1996; Rejmánek & Richardson, 1996). Many invaders, including almost all of the world's worst weeds listed in Holm *et al.* (1977), produce significant seed banks, and the presence of a persistent seed bank is cited as being one of the most important factors in limiting the success of efforts to biologically control exotic weeds (Holloway, 1964; Dahlsten, 1986). Finally, Crawley, Harvey, and Purvis (1996) found aliens were more likely than British natives to have long-term seed banks, though Thompson, Hodgson, and Rich (1995) found that this was actually less true for invasive aliens than for natives which recently have invaded new areas. In some cases, invaders have been shown to develop larger seed banks in new regions, suggesting escape from natural enemies. Numerous Australian *Acacia* spp. develop much larger seed banks in South Africa than in their native regions, primarily as a result of escape from predispersal seed predators and gall rusts (Dean, Holmes & Weiss, 1986). Similarly, seed banks of *Chrysanthemoides monilifera* (L.) T. Nord. are much larger in Australia than in its native South Africa, reflecting both greater seed production and increased persistence in the soil (Scott, 1996). Finally, seeds of *Echium plantagineum* L. have a higher probability of incorporation into the seed bank in Australia than in its native European range (Grigulis *et al.*, 2001).

In this paper, we experimentally examine the impacts of natural enemies on seeds in the seed bank, using a suite of 19 native and 20 exotic plant species. We focus on the effects of pathogenic fungi and invertebrate seed predators,

since these are believed to be important sources of mortality for buried seeds (Leck, Parker & Simpson, 1989; Baskin & Baskin, 1998). We use our results to examine the hypothesis that successful alien species are less susceptible than natives to resident predators and pathogens; if true, this should mean that the experimental exclusion of natural enemies would benefit native species more than exotics. Along with Blaney and Kotanen (2001a,b), this is the first study to examine post-dispersal seed mortality over a wide range of co-occurring native and alien species, and one of the few to experimentally test a hypothesis proposed to explain invasiveness (Karieva, 1996). As well, this is one of the relatively few experimental studies examining causes of seed mortality in natural soils (Kremer, 1993; Baskin & Baskin, 1998).

Methods

STUDY SITE

We conducted this study at the Dead Man's Curve meadow of the University of Toronto Joker's Hill Research Station (<http://www.erin.utoronto.ca/~w3pkota/jh.html>), Regional Municipality of York, Ontario (44° 02' 25" N, 79° 32' 00" W). Nomenclature follows Gleason and Cronquist (1991). The site is a dry-mesic old field on a south-facing slope that has been abandoned from any agricultural use for more than 30 years. This meadow shows little evidence of its agricultural past; instead, it has been completely colonized by a diverse mix of native and alien species, typical of southern Ontario old fields. Grasses dominate the site, with the *Bromus inermis* and *Poa pratensis* important in mesic areas and *Poa compressa* and *Danthonia spicata* dominant in drier sites. The dominant herbs include *Solidago canadensis*, *S. nemoralis*, *Aster urophyllus*, and *A. novae-angliae*, as well as *Melilotus alba* in some areas.

EXPERIMENTAL SPECIES

We chose seeds of 19 native and 20 alien forbs and graminoids from a pre-existing collection of southern Ontario seeds (Table I). There is a chance that apparent differences between natives and aliens may be products of phylogenetic confounding (Crawley, Harvey & Purvis, 1996; Kotanen, Bergelson & Hazlett, 1998); in response, we selected species so as to represent a taxonomically broad and overlapping range of families for both natives and aliens, thus reducing the risk of phylogenetic biases. This approach allowed us to investigate a very diverse sample of the local flora; in another paper, we report the results of a separate experiment in which we took the alternative approach of statistically controlling for phylogenetic trends in a taxonomically narrower dataset (Blaney & Kotanen, 2001b). All of our experimental species occur primarily or entirely in open, upland habitats and forest edges. Most occur naturally within the Joker's Hill property (Table I), and all have wild populations occurring in the Regional Municipality of York (Riley, 1989). With four exceptions, seeds had been bulk-collected from wild populations in southern Ontario between June 1996 and June 1997 and stored dry in a freezer until use. Seeds of *Andropogon gerardii*, *Bromus kalmii*, *Elymus trachycaulus*, and *Sorghastrum nutans* were purchased from the Pterophylla Farm, Walsingham, Ontario, where they had been grown in 1996 from plants originating from local, wild seed stock.

TABLE I. Experimental species. Presence on Joker's Hill research station property (JH) is indicated by "Y". Native or alien origin follows Morton and Venn (1990).

Family	Natives Species	JH	Family	Aliens Species	JH
Asclepiadaceae	<i>Asclepias syriaca</i>	Y	Apiaceae	<i>Daucus carota</i>	Y
Asteraceae	<i>Artemisia campestris</i>		Asteraceae	<i>Arctium minus</i>	Y
Asteraceae	<i>Aster ericoides</i>	Y	Asteraceae	<i>Chrysanthemum leucanthemum</i>	Y
Asteraceae	<i>Hieracium scabrum</i>		Asteraceae	<i>Hieracium aurantiacum</i>	Y
Asteraceae	<i>Solidago nemoralis</i>	Y	Boraginaceae	<i>Echium vulgare</i>	Y
Boraginaceae	<i>Hackelia virginiana</i>	Y	Brassicaceae	<i>Erysimum cheiranthoides</i>	Y
Fabaceae	<i>Desmodium canadense</i>	Y	Caryophyllaceae	<i>Silene vulgaris</i>	Y
Fabaceae	<i>Lepedeza capitata</i>		Chenopodiaceae	<i>Chenopodium album</i>	Y
Lamiaceae	<i>Monarda fistulosa</i>	Y	Dipsacaceae	<i>Dipsacus sylvestris</i>	Y
Onagraceae	<i>Oenothera biennis</i>	Y	Fabaceae	<i>Medicago lupulina</i>	Y
Plantaginaceae	<i>Plantago rugellii</i>	Y	Fabaceae	<i>Melilotus alba</i>	Y
Poaceae	<i>Andropogon gerardii</i>		Fabaceae	<i>Vicia cracca</i>	Y
Poaceae	<i>Bromus kalmii</i>		Lamiaceae	<i>Leonurus cardiaca</i>	Y
Poaceae	<i>Elymus trachycaulus</i>		Lamiaceae	<i>Nepeta cataria</i>	Y
Poaceae	<i>Sorghastrum nutans</i>		Plantaginaceae	<i>Plantago major</i>	Y
Ranunculaceae	<i>Anemone cylindrica</i>	Y	Poaceae	<i>Elymus repens</i>	Y
Rosaceae	<i>Geum aleppicum</i>	Y	Poaceae	<i>Phleum pratense</i>	Y
Rosaceae	<i>Potentilla arguta</i>	Y	Polygonaceae	<i>Rumex crispus</i>	Y
Scrophulariaceae	<i>Penstemon hirsutus</i>		Rosaceae	<i>Potentilla recta</i>	Y
			Scrophulariaceae	<i>Verbascum thapsus</i>	Y

Total: 39 species (19 native, 20 alien)

TREATMENTS

We established 18 experimental plots throughout the Dead Man's Curve site in June 1997. Plots measured 1.5 m x 1.5 m in size and were spaced at least 10 m apart in a stratified random array. Each contained 16 small, thin-walled peat pots (4 cm x 4 cm x 5 cm depth), buried just below the soil surface. In order to allow access by soil biota, we cut a 2-cm x 3-cm window in the lower half of each side and removed the upper 1 cm from one side; a 4-mm drainage hole was also added in the bottom. We filled each pot with a mixture of experimental seeds in 20 cm³ of local field soil. This mixture included 20 seeds per species of a random subset of 9-10 experimental species and 20 biologically inert markers (2-mm glass balls). Quantification of the pre-existing seed bank in the soil revealed that some of the experimental species were present, but at a very low density (< 1%) relative to the added seeds.

The four treatments we used were 1) control (untreated), 2) fungicide addition, 3) invertebrate exclusion, and 4) fungicide addition + invertebrate exclusion. Treatments were interspersed. We excluded invertebrates by pre-drying the soil used at 50°C for 48 hours and by enclosing the peat pot in 1-mm nylon window screening to restrict subsequent access by soil fauna. For the fungicide additions, we injected 5 mL of fungicide into the pot immediately before burial in June 1997 and added identical doses in October 1997, May 1998, and September 1998. The fungicide we used was a 1:100 aqueous solution of Maestro 75DF (Zeneca Corp., Stoney Creek, Ontario, Canada; active ingredient 75% Captan by weight); this concentration was recommended by the manufacturer for use as a dip for bulbs and tubers. Captan is a non-systemic heterocyclic nitrogen fungicide used against a wide range of oomycetes, ascomycetes, and basidiomycetes (Sharville, 1961; Torgeson, 1969; Neergaard, 1977) and is particularly effective against seed-rotting organisms (Neergaard, 1977). It has been shown to have minimal effects on endomycorrhizal fungi and both positive and negative effects on ectomycorrhizae (Vyas, 1988),

depending on plant species. Laboratory trials failed to demonstrate any significant effect of Captan on survival or germination of our study species.

SAMPLING

We randomly selected 6 plots for retrieval in November 1997, June 1998, and October 1998 (hereafter called the 4-month, 11-month, and 16-month trials). At each trial, we carefully dug up the peat pots with their contents intact and recorded any seedlings growing out of each pot. After recovery, we thinly (5 mm depth) spread the contents of each pot over potting mix in a 15-cm diameter plastic flower pot. These pots were kept moist for three months in a greenhouse; after 1.5 months, we disturbed the layer of field soil to allow buried seeds a better chance of germination. We counted and removed emerging seedlings monthly; interim samplings indicated few seedlings died between monthly counts. We recorded seeds as recovered if we found them as seedlings in the peat pots in the field or if we observed them as seedlings in the greenhouse. At the end of the three-month germination period, we removed the field soil from the greenhouse pots and passed it through a 1-mm sieve to recover glass balls.

ANALYSIS

Before analysis, we corrected greenhouse germination for soil loss during sampling; we did not correct field germination, as it was assessed before the peat pots were collected and therefore should be independent of physical sampling losses. The formula we used was

$$\text{Corrected proportion of seeds recovered} = \frac{\left[(\text{seeds germinated in the field}) + \left(\frac{\text{seeds germinated in greenhouse}}{\text{proportion of glass balls recovered}} \right) \right]}{\text{seeds initially added}}$$

We arcsin transformed these corrected values to improve normality (Kirk, 1982).

We analyzed each trial separately because trials were expected to differ seasonally, because different plots were destructively sampled on each date, precluding a repeated-measures design, and in order to keep the design tractable. The basic analyses we used were 3-factor randomized block factorial ANOVAs (Type III sums of squares), with blocking by plot and with fungicide addition, invertebrate exclusion, and species as factors. We used a non-interactive model, as recommended by Newman, Bergelson, and Grafen (1997): we treated treatments as fixed effects and plot as a random blocking effect and used the residual as the error term. Of the 936 potential values generated by each of the 3 trials, 31, 56, and 16, respectively, were lost because pots were not successfully recovered and scored; these cells were interpolated using treatment \times species means, adjusting degrees of freedom accordingly (Underwood, 1997). We also performed Split-plot Factorial ANOVAs to compare mean responses of natives and aliens, treating species as random blocks and treatments as fixed effects and using blocks and their interactions as error terms (Kirk, 1982). We have reported means \pm 1 standard error.

Results

Seed recovery varied substantially among trials, with the highest rates of recovery at 11 months (Figure 1). The relative importance of field and greenhouse recovery also differed among trials: the increase in recovery between 4 and 11 months resulted from an increase in numbers of seedlings in the greenhouse, while reduced recovery in the 16-month trial was a result of reductions in both field and greenhouse germination (Table II). Rates of glass ball recovery remained almost constant (Table II), strongly suggesting that differences in seed recovery among trials were not due to differing levels of mechanical loss or earthworm activity.

Fungicide improved seed recovery for aliens (Figure 1). Though these effects were modest and partly obscured by differences among species (Figure 2), they were significant at all three trials (Table III). At the first trial, the positive effects of fungicide were even stronger for natives than for aliens, but effects on natives were nonsignificant at subsequent trials (Table III; Figure 1). Invertebrate exclusion produced no significant effects on seed recovery for either natives or exotics, though it had a tendency to produce better recovery than controls (Table III; Figure 1). There were few significant interactions. Fungicide effects did differ among alien species at the third trial (Table III), but otherwise species \times treatment effects were nonsignificant. There was a marginally significant ($p = 0.050$) fungicide \times invertebrate exclusion \times species interaction for natives at the 4-month trial, indicating that different species had a slight tendency to respond differently to different treatment combinations. Plot (= block) effects were not initially detectable, but had developed by the final sampling date (Table III); this may reflect the gradual development of local variation (for example, if the relatively dry weather during this experiment gradually increased the importance of local variation in soil water) or may simply suggest one of the plots included in the final sampling differed from the others for some chance reason. In either case, the use of plot as a blocking factor should prevent this local variation from biasing estimates of treatment effects.

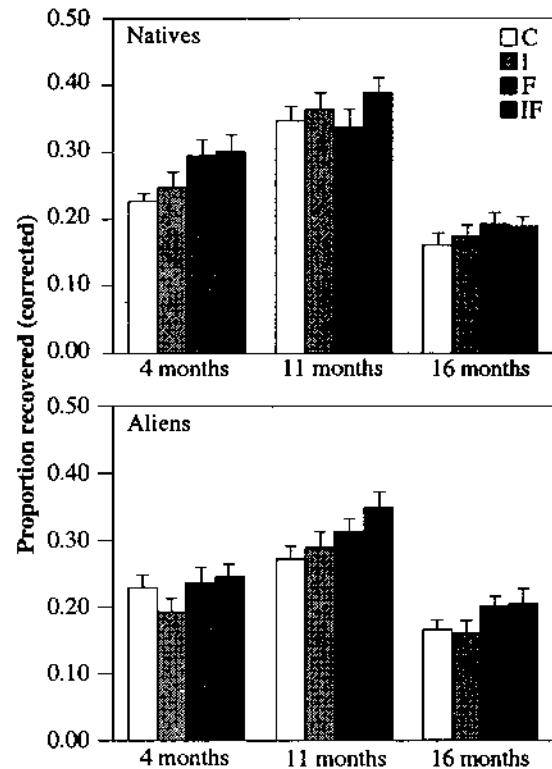


FIGURE 1. Overall results of each experimental trial. Proportion of seeds recovered (mean \pm SEM) are corrected for physical losses; see Tables III and IV for results of significance tests. Treatments are controls (C), invertebrate exclusion (I), fungicide addition (F), and invertebrate exclusion + fungicide addition (IF).

TABLE II. Uncorrected proportions of seeds in each trial recovered as seedlings in the field and by germination in the greenhouse, and proportion of neutral markers (glass balls) recovered by sieving soil.

Trial	Field proportion	Greenhouse proportion	Glass balls
4 months	0.067	0.149	0.874
11 months	0.068	0.219	0.870
16 months	0.036	0.112	0.884

When directly compared, natives and aliens did not clearly differ in their susceptibility to natural enemies. ANOVAs comparing mean recovery for native and alien species again found a significant fungicide effect (Table IV), but no significant invertebrate exclusion effects, native-exotic differences, or origin \times treatment interactions. However, at the 4-month trial, both the fungicide \times origin interaction and the fungicide \times invertebrate exclusion \times origin interaction approached significance ($p = 0.065$ and $p = 0.051$, respectively): fungicide tended to improve recovery more for natives than for exotics, while the invertebrate exclusion treatment tended to reduce recovery of exotics unless in the presence of fungicide (Figure 1). These results must be treated with suspicion, not only because of their (marginal) non-significance, small magnitude, and the confounding nature of the 3-way interaction (Kirk, 1982), but also because similar effects were detected at no other sampling (Table IV; Figure 1).

For both aliens and natives, recovery differed significantly among species on each date (Table III; Figure 2).

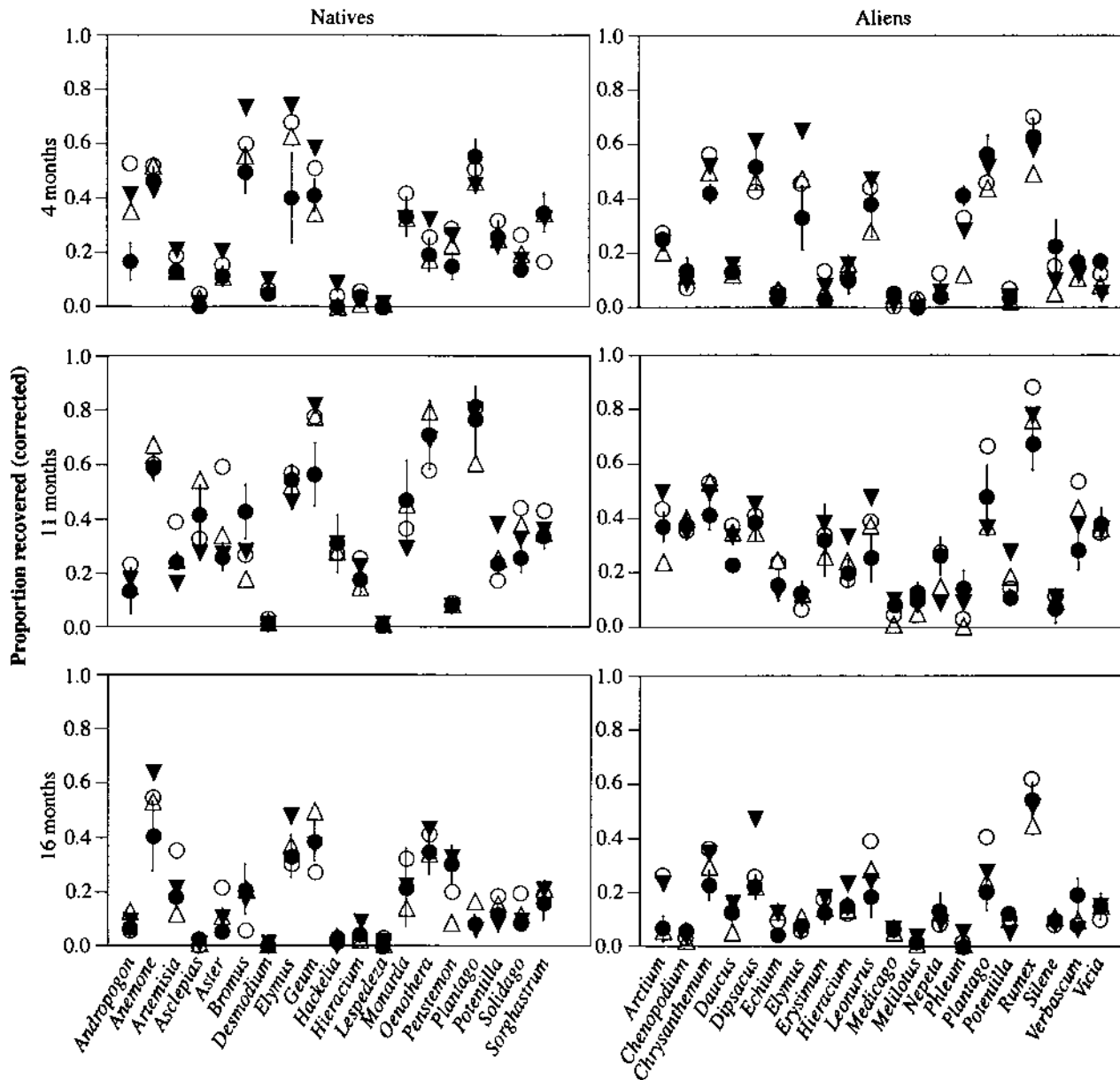


FIGURE 2. Results of each experimental trial, subdivided by species; see table I for complete species names. Points represent the proportion of seeds recovered, corrected for physical losses (mean \pm SEM for controls only); see tables III and IV for results of significance tests. Treatments are controls (full circles), invertebrate exclusion (empty triangles), fungicide addition (full triangles), and invertebrate exclusion + fungicide addition (empty circles).

Despite these large among-species differences, recovery of controls for each species generally scored lower than other treatments, explaining the significant fungicide effects (Figure 2). Patterns of recovery in legumes and grasses were distinct from the rest of the experimental species. Legumes generally germinated at very low rates compared to other species; as a result, *Lespedeza capitata* had the lowest recovery rates (< 1%) of any species in all three trials. Five of the six grass species showed maximal recovery in the 4-month trial, with declining recovery through the 11- and 16-month trials; most recovery occurred in the field. This strongly contrasted with the 33 remaining species: 26 of these species reached maximum recovery rates in the 11-month trial, and all were recovered primarily by germination in the greenhouse.

Discussion

FUNGICIDE ADDITION

Fungal pathogens are known to be a major source of mortality at the seed and seedling stages in crops, and there is evidence that this may be true of many natural systems as well (Dinoor & Eshed, 1984; Burdon, 1987; Blakeman & Williamson, 1994). Our results suggest that fungal mortality can be important to seeds of at least some species in old field habitats. Our estimates probably are conservative: although our fungicide treatment was sufficient to demonstrate significant impacts, applications were infrequent compared to many managed systems; this probably reduced effectiveness, diminishing differences between treatments and controls. Some other studies have found evidence of fungal impacts on seeds and seedlings in very different nat-

TABLE III. Results of 3-factor randomized block factorial ANOVAs on the recovery of experimental seeds after 4, 11, and 16 months. "Plot" was treated as the blocking factor and the residual was used as the error term.

Factor	4 months				11 months				16 months			
	Natives		Aliens		Natives		Aliens		Natives		Aliens	
	df	F	df	F	df	F	df	F	df	F	df	F
Plot	5	1.850	5	2.057	5	1.096	5	0.768	5	5.474***	5	4.264***
Fungicide	1	15.025***	1	4.336*	1	0.360	1	7.683**	1	1.155	1	5.885*
Invertebrate exclusion	1	1.563	1	0.499	1	2.463	1	1.256	1	0.017	1	0.390
Species	18	38.562***	19	42.813***	18	36.279***	19	29.827***	18	22.118***	19	19.453***
Fungicide × invertebrate exclusion	1	0.563	1	2.714	1	1.351	1	0.743	1	0.147	1	0.006
Fungicide × species	18	1.621	19	1.029	18	1.138	19	0.930	18	1.095	19	2.165**
Invertebrate exclusion × species	18	(p = 0.053) 0.851	19	0.997	18	1.309	19	1.551	18	0.783	19	0.739
Fungicide × invertebrate exclusion × species	18	0.643	19	1.616*	18	0.807	19	1.342	18	0.930	19	0.556
Error	360		379		345		369		370		384	

* = $P < 0.05$, ** = $P < 0.01$, *** = $P < 0.001$

TABLE IV. Results of split-plot factorial ANOVAs comparing mean recovery of seeds of native and exotic species. "Origin" refers to native versus exotic origins; "Species" was used as the blocking factor; blocks and their interactions were used as the error terms (df = 1,37 for all tests).

Factor	F		
	(4 months)	(11 months)	(16 months)
Origin	0.294	0.481	0.003
Fungicide	17.817***	6.502*	4.712*
Fungicide × origin	3.632	1.477	0.454
Invertebrate exclusion	(p = 0.065) 0.178	1.423	0.014
Invertebrate exclusion × origin	1.068	0.953	0.048
Fungicide × invertebrate exclusion	0.057	0.542	0.242
Fungicide × invertebrate exclusion × origin	4.065	0.006	0.107
Error	(p = 0.051)		

* $P < 0.05$, *** = $P < 0.001$

ural ecosystems. For example, Packer and Clay (2000) found *Pythium* caused significant mortality of seedlings of the temperate tree *Prunus serotina*. Crist and Friese (1993) found decomposition rates of up to 93.5% over 10 months in seeds of six shrub-steppe species, and isolated seven species of ascomycetes from inside their seeds. Lonsdale (1993) chemically excluded higher fungi and found 10% to 16% increases in seed survival over 7 months for the shrub *Mimosa pigra* L. in tropical Australia. Augspurger (1983) and Augspurger and Kelly (1984) demonstrated that damping-off disease killed up to 95% of seedlings of a wide range of tropical tree species. Finally, the presence of antimicrobial compounds in seed coats (Warr, Thompson & Kent, 1992; Hendry *et al.*, 1994) also suggests that fungi may be an important source of seed mortality in natural soils.

The "fungi" involved in seed decomposition in soil include oomycetes, zygomycetes, ascomycetes, and basidiomycetes (Kremer, 1993; Blakeman & Williamson, 1994). Oomycetes are especially important pathogens of germinating seeds, with genera such as *Pythium* and *Phytophthora* being the major cause of damping-off diseases and root-rot (Augspurger & Kelly, 1984; Paul, Ayres & Wyness, 1989; Packer & Clay, 2000). Due to physiological differences, few fungicides are effective against all groups (Paul, Ayres & Wyness, 1989). No assays of soil fungi were attempted in our study, but Captan is widely used in agriculture to control oomycetes, ascomycetes, and basidiomycetes in the soil

and on fruit, leaf, and seed surfaces (Sharville, 1961; Torgeson, 1969; Neergaard, 1977). Some taxa of pathogens may not have been controlled by the Captan used in this experiment (Sharville, 1961; Torgeson, 1969; Neergaard, 1977), but, again, this should render our results more conservative.

INVERTEBRATE EXCLUSION

A wide range of invertebrates may eat buried seeds, including millipedes, isopods, beetles, slugs, and termites (Baskin & Baskin, 1998), but ants and earthworms were the potential seed predators most commonly detected in our control pots. Ants can be significant seed predators in North American old fields (Mittelbach & Gross, 1984; Reader & Beisner, 1991), while seeds consumed by earthworms may be killed outright or suffer reduced dormancy, which can be fatal for buried seeds (McRill & Sagar, 1973; Grant, 1983; Thompson, Green & Jewels, 1994). We observed some very small earthworms and insects in our "invertebrate exclusion" pots, but the mesh was effective at excluding larger animals and was smaller than the size of many experimental seeds. Despite this, we failed to find significant effects of invertebrate exclusion in any trial. Our results suggest invertebrates were much less important as sources of mortality at our site than were soil fungi, at least for buried seeds; seeds on the soil surface may be more susceptible to predation by ants (Reader & Beisner, 1991).

TEMPORAL PATTERN IN SEED RECOVERY

The observed changes over time in recovery rates reflect an interaction between mortality and seasonal shifts in dormancy. The reduction in recovery rates between the 4-month trial and the 16-month trial is likely to have been a consequence of seed mortality, as seasonal dormancy effects should not differ between samples collected 12 months apart. In contrast, the fact that recovery was substantially lower in the 4-month trial (retrieved in fall 1997) as compared to the 11-month trial (retrieved in spring 1998) strongly suggests that seeds were more dormant in the fall than in the spring. This result is not unexpected given that dormancy cycles are likely of general occurrence among temperate zone herbs (Baskin & Baskin, 1998). Changes in dormancy through the three trials and the difficulty in dis-

tinguishing dead seeds from dormant seeds limit the extent to which our recovery values can be considered to represent seed survival; this does not, however, confound comparisons among treatments of recovery at any particular sampling date.

TAXONOMIC PATTERN

Both dormancy and vulnerability to enemies may be affected by characteristics of seeds including size, geometry, coat thickness, chemistry, and hardness (Harper, 1977; Kremer, 1993; Leck, Parker & Simpson, 1989; Baskin & Baskin, 1998). These characters varied markedly among our experimental species; as a result, many differences among taxa were apparent in our data. The responses of legumes and grasses were particularly striking. Legumes had very low rates of recovery, as might be expected given the hard, impermeable seed coats typical of the family (Baskin & Baskin, 1998). Sieving the soil after the germination period revealed that legume seeds still were present and were viable when the seed coats were scarified. Grasses had maximal recovery early in the experiment and were recovered largely as seedlings in the field. This pattern can be attributed to two factors. First, many grasses tend to have limited seed dormancy and seed banking abilities (Baskin & Baskin, 1998; Thompson *et al.*, 1998). Second, the relatively large seeds of most of the grasses in this study allowed seedlings to successfully push through several cm of soil to the surface, whereas seedlings of smaller seeded species may have perished below ground before they could be counted.

ALIENS VERSUS NATIVES

In a related study (Blaney & Kotanen, 2001a), we found no evidence that seeds of exotics enjoyed reduced rates of post-dispersal predation by vertebrates or surface-dwelling insects, compared to seeds of natives. The present study extends those results to the impacts of soil fungi and soil fauna. Predation by soil animals was equally unimportant for seeds of both natives and exotics. Results for fungi were more complex, but direct comparisons of natives versus exotics invariably failed to detect any significant differences in their responses to fungicide addition. When natives and exotics were considered separately, fungicide additions tended to be more beneficial for natives than exotics at the first trial, as might be expected if natives were more susceptible to fungal pathogens. However, the lack of any subsequent response by natives suggests that this trend was short-lived and did not contribute to long-term seed banking. In contrast, aliens always benefitted significantly from fungicide addition. Klironomos (2002) found rare native plants suffered more from soil fungal pathogens than did invasive exotics; however, he provided no evidence that aliens escape pathogens to a greater extent than equally abundant natives.

The most likely explanation for our results is that soil fungi were sufficiently generalist that they did not consistently discriminate between seeds of natives and exotics. Although host specificity is known among seed pathogens of agricultural plants (Neergaard, 1977; Agarwal & Sinclair, 1997), and there is evidence of soil seed pathogens having some degree of host-specificity (Kirkpatrick & Bazaz, 1979; Harman, 1983; Packer & Clay, 2000), many soil-

borne plant pathogens and decay organisms are broad generalists (Dix & Webster, 1995). It is also plausible that some species-specific seed pathogens inadvertently have been introduced to the New World on seeds or in soil or that pathogens of native species have become adapted to exotic relatives since the arrival of these aliens.

These results do not necessarily apply to other life history stages: a high proportion of the insects and pathogens that attack growing plants and developing seeds are specialists (Strong, Lawton & Southwood, 1984; Fritz & Simms, 1992) and therefore are likely initially to be absent in foreign habitats. Neither do these results preclude the possibility that the most seriously invasive species are problems precisely because they enjoy atypically low predator or pathogen levels (Klironomos, 2002). Successful biocontrol programs suggest that at least some problematic species may owe their success to low levels of attack; however, the fact that most biological control efforts fail (Crawley, 1986) also suggests that such examples may be exceptional. Comparing invasions in which low predator or pathogen loads are important and the probable majority in which they are not may improve efforts at biological control, as well as our general understanding of biological invasions.

Conclusion

Our results demonstrate that fungi have significant impacts on the recovery of viable seeds of a broad range of native and exotic species in old field soils; effects of invertebrates are small by comparison. Native and exotic species do not consistently differ in their susceptibility to these enemies. Although using such a broad taxonomic range of seeds captures much of the variation among old field species, a limitation of our approach is that relatively subtle native-alien differences may be lost in the "noise" created by the inclusion of very different species in the same dataset (Mack, 1996). In a separate experiment (Blaney & Kotanen, 2001b), we control for this problem by comparing congeneric natives and exotics and further demonstrate that our results hold true for fungal seed pathogens of closely related species.

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