LETTER

Plant–soil biota interactions and spatial distribution of black cherry in its native and invasive ranges

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Abstract

One explanation for the higher abundance of invasive species in their non-native than native ranges is the escape from natural enemies. But there are few experimental studies comparing the parallel impact of enemies (or competitors and mutualists) on a plant species in its native and invaded ranges, and release from soil pathogens has been rarely investigated. Here we present evidence showing that the invasion of black cherry (*Prunus serotina*) into north-western Europe is facilitated by the soil community. In the native range in the USA, the soil community that develops near black cherry inhibits the establishment of neighbouring conspecifics and reduces seedling performance in the greenhouse. In contrast, in the non-native range, black cherry readily establishes in close proximity to conspecifics, and the soil community enhances the growth of its seedlings. Understanding the effects of soil organisms on plant abundance will improve our ability to predict and counteract plant invasions.

Keywords

Biogeographical comparison, trans-Atlantic comparison, enemy release hypothesis, natural enemies hypothesis, facilitation, invasive tree, plant–soil community interactions, *Prunus serotina*, soil pathogens.

INTRODUCTION

Biogeographical experiments testing the relative effect of biotic interactions on organisms in their native and invaded ranges are necessary for testing our basic understanding of biological invasions. Some studies have compared herbivores (Memmott *et al.* 2000; Wolfe 2002), pathogens (Beckstead & Parker 2003; Mitchell & Power 2003) and competitors (Callaway & Aschehoug 2000) of plants in their native and non-native ranges. Half of these have used descriptive surveys to compare the above-ground enemies of plants in their native and invaded ranges (Memmott *et al.* 2000; Wolfe 2002; Mitchell & Power 2003), and the other half have used manipulative experiments to test the effect of pathogens (Beckstead & Parker 2003), interspecific competitors (Callaway & Aschehoug 2000), or soil communities (R.M. Callaway, G. Thelen, A. Rodriguez & B. Holben, unpublished data) on plants in their native and invaded ranges. Overall, these studies suggest that plants are less negatively effected by these interactions in their non-native than native ranges.

The interactions between plants and their soil community can result in dynamic feedback (e.g. Van der Putten *et al.* 1993; Packer & Clay 2000; Bever 2002, 2003; Klironomos 2002). The direction of feedback depends on the balance between negative effects of accumulating pathogens, parasites, and herbivores and positive effects of accumulating mycorrhizal fungi, N-fixing bacteria and other beneficial bacteria (Bever 2003). The net feedback affects plant establishment, growth and plant–plant interactions (Bever 2003). Several studies suggest that negative soil feedbacks are common in natural systems and exert distance- and/or density-dependent control, helping to maintain plant diversity (e.g. Packer & Clay 2000; Bever 2002, 2003; Klironomos 2002).

Previous research has shown that soil-borne *Pythium* spp. (Oomycota) alter survival, growth and abundance of black cherry (*Prunus serotina* Ehrh) in its native range (Packer & Clay 2000, 2002). We hypothesized that black cherry invading north-western Europe escaped from host-specific soil pathogens. Invasive plants are predicted to benefit most when they escape from host-specific enemies (Keane & Crawley 2002). *Pythium* spp. are often host-specific (Deacon

& Donaldson 1993; Mills & Bever 1998). However, little is known about the ecology of the *Pythium* spp. affecting black cherry in either range.

We conducted a trans-Atlantic comparison of nearest neighbour distances and plant–soil community interactions for black cherry in its native and invasive ranges. The enemy release hypothesis (Keane & Crawley 2002) predicts that black cherry would be more widely dispersed (low density) in its native range, where distance-dependent regulation by host-specific soil pathogens is stronger, relative to its non-native range. We tested the effect of soil origin (conspecific vs. heterospecific), seedling density and soil sterilization on the survival and growth of black cherry seedlings in their native and non-native ranges. Furthermore, we hypothesized that invading black cherry continues to benefit from mutualists (e.g. arbuscular mycorrhizal fungi) in its non-native range. Low-specificity of arbuscular mycorrhizal fungi would not limit the invasion of non-native plants (Richardson *et al.* 2000; Klironomos 2002 but see Bever 2002 and Klironomos 2003). Lastly, we related the nearest neighbour distribution of black cherry in the field to our experimental results.

MATERIALS AND METHODS

Nearest neighbour measurements

In the native range, we sampled nearest neighbour distances in the Indiana University Research and Teaching Preserve (N39°11.810′ W86°30.879′) and Griffy Lake Preserve (N39°11.913′ W086°30.763′) Indiana, USA. These sites were located within 5 km of one another and are composed of mesic deciduous forests dominated by *Fagus grandifolia*, *Acer sacchrum*, *Quercus* spp., *Fraxinus* spp., and *P. serotina*. The soils are moist and well-drained Alfisols. In the invaded range, *P. serotina* was sampled in The Leeren Doedel (N52°00.523′ E005°50.308′) and The Ossenbos (N52°08.020′ E005°48.008′), both in The Netherlands. The forests at these sites are dominated by *Pinus sylvestris* and *Quercus* spp., and occupy dry sandy soils.

Nearest neighbour distances were measured for seedlings (<1 m height), saplings (1–3 m) and trees (>3 m) using a modified pointed-centred quarter method (Cottam & Curtis 1956) with randomly selected mature black cherry trees ($n = 10–13$ focal trees per site) as centre points. Only seedlings ≤1 year of age were sampled as determined by examination of leaf scars and cotyledons. Nearest neighbour distances were measured from 0 to 50 m. If no neighbours were found within an individual quarter, then nearest neighbour distance was conservatively set at 50 m. We tested the effect of range and site (fixed factors) on each life-history stage using nested ANOVAs with site nested in range [PROC GLM with mean squares for site (range) as the error
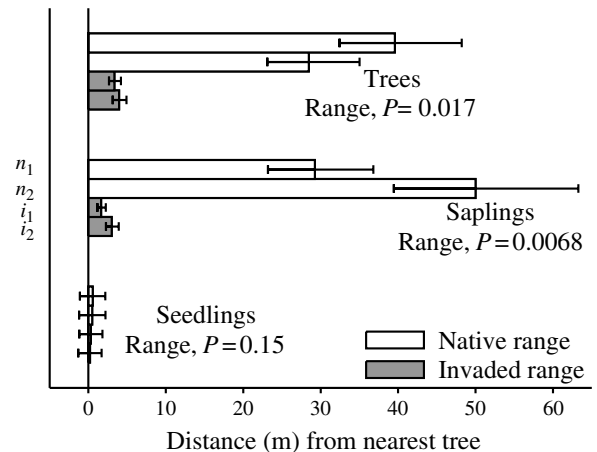

Figure 1 Distances from conspecifics of black cherry (*Prunus serotina*) in their native (white bars) and invaded ranges (grey bars). Bars represent back-transformed least squares mean distance from mature trees ($n = 10–13$ per site) to the nearest seedlings, saplings and trees ±95% CI. Individual sites are labelled n_1, Griffy Lake Nature Preserve; n_2, Indiana University Research and Teaching Preserve; i_1, The Leeren Doedel; and i_2, The Ossenbos. The effect of range on distance to nearest conspecifics was tested using nested ANOVAs with site nested within range.

term, SAS version 8.0; SAS Institute Inc., Cary, NC, USA]. Data were transformed to meet assumptions of homogeneity of variance. Least square means of nearest neighbour data were back transformed and used to calculate the 95% CI reported in Fig. 1.

Soil biota experiments

Greenhouse experiments tested the effect of soil sterilization (non-sterile vs. sterile soil), seedling density (1 vs. 3 seedlings planted per pot) and soil origin (collected near black cherry trees vs. near heterospecifics) on the survival and biomass of black cherry seedlings in their native and non-native ranges. Methods for the native range were previously reported by Packer & Clay (2000). Soil was collected in the Griffy Lake Nature Preserve. The greenhouse experiment conducted in the invaded range used soil and seedlings from Leeren Doedel. In The Netherlands experiment, the individual trees were the replicates ($n = 12$), the soil for the heterospecific soil origin treatment was collected 4–20 m away from the nearest conspecific and seedlings (germinated in 2002) from the field with surface-sterilized roots (5% bleach for 10 min then rinsed with DI water) were used. Soil inoculum was diluted with a 2 : 1 v/v sterile potting soil to avoid potential confounding effects of nutrient release by soil sterilization (see Troelstra *et al.* 2001); a foliar contact fungicide (Euparen-M; Bayer Inc.,

Leverkusen, Germany) was applied to leaves to reduce powdery mildew, while not impacting the soil biota; and plants were grown for 2 months.

For treatments with high seedling density, survival of one randomly selected seedling per pot was analysed. Similarly, biomass of one randomly selected living seedling was analysed. Log-linear analysis was used to test for an effect of factors on seedling survival (PROC CATMOD, SAS). Three-way ANOVAs tested for an effect of factors on seedling biomass (SPSS, version 10; SPSS Inc., Chicago, IL, USA).

RESULTS

Black cherry saplings and trees were 873 and 618%, respectively, farther from conspecific trees in forests in native range than in invaded range (Fig. 1; nested ANOVA, range, $F_1 = 56.58$, $P = 0.017$ and $F_1 = 145.66$, $P = 0.0068$, respectively). However, distance to nearest seedlings did not differ between ranges ($F_1 = 5.12$, $P = 0.15$).

There was no effect of treatments on seedling mortality in the invaded range ($\chi^2 < 1.41$, $P > 0.23$), while there was an interaction between soil sterilization, seedling density and soil origin on seedling mortality in the native range (Fig. 2a; log-linear analysis, $\chi^2 = 4.85$, $P = 0.028$). Seedling mortality ranged from 8 to 61% and mortality increased by $\geq 127\%$ when grown at high densities in non-sterile soil collected near black cherry trees relative to other treatments (Fig. 2a). There were no other significant effects of treatments on seedling survival in the native range ($\chi^2 < 1.92$, $P > 0.16$).

In both the native and invaded ranges, soil sterilization and seedling density had a significant effect on seedling biomass (Table 1). In the invaded range, mean biomass was 25% greater in non-sterile soils compared with sterilized soils (Fig. 2b, Table 1). In contrast, in the native range, mean biomass was 25% less in non-sterile soils compared

Table 1 Summary of ANOVA of effects of soil sterilization (non-sterile vs. sterile soil), initial seedling density (one vs. three) and soil origin (conspecific vs. heterospecific) on the seedling biomass of black cherry in their native and invaded ranges. *P*-value ≤ 0.05 in bold.

Effect	d.f.	MS	*F*	*P*-value
Native range				
Sterilization	1	0.128	11.17	**0.001**
Density	1	0.415	36.09	**<0.0005**
Origin	1	0.0003	0.03	0.87
Sterilization × density	1	0.101	8.80	**0.004**
Sterilization × origin	1	0.123	10.71	**0.001**
Density × origin	1	0.0101	0.88	0.35
Sterilization × density × origin	1	0.0883	7.67	**0.007**
Error	108	0.1151		
Invaded ranges				
Sterilization	1	1.657	11.35	**0.001**
Density	1	4.910	33.62	**<0.0005**
Origin	1	0.136	0.93	0.34
Sterilization × density	1	0.004	0.03	0.87
Sterilization × origin	1	0.015	0.10	0.75
Density × origin	1	0.035	0.24	0.63
Sterilization × density × origin	1	0.111	0.76	0.39
Error	85	0.146		

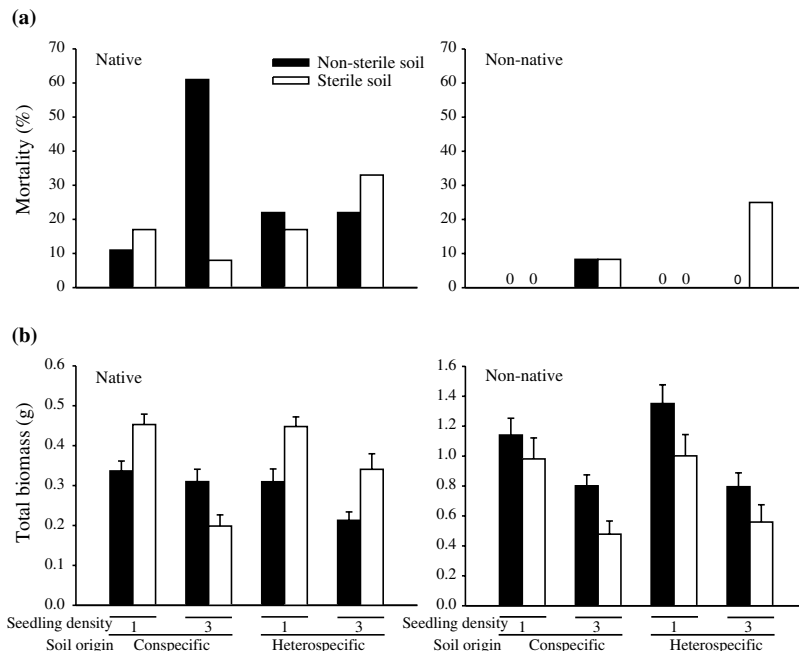Tests were performed using the type three sums of squares from SPSS version 10 (SPSS Inc.).

Figure 2 Effect of soil sterilization, initial seedling density, and soil origin on the mean a, mortality and b, biomass of black cherry (*Prunus serotina*) seedlings in their native (Indiana; $n = 12–17$) and non-native (The Netherlands; $n = 11–12$) ranges (+1SE). Means equal to zero are represented with '0'. The data for the native range were reported previously (Packer & Clay 2002). The survival data were analysed with log-linear analyses and the biomass data were analysed with three-way ANOVAs. Refer to Results section and Table 1 for the statistical output.

with sterilized soils (Fig. 2b, Table 1). There was also significant sterilization × density, sterilization × origin and sterilization × density × origin effects in the native range (Fig. 2b, Table 1).

DISCUSSION

Our study provides evidence that a tree species, whose abundance is limited by soil pathogens in its native range (Packer & Clay 2000), may become locally more abundant in its non-native range where control by the soil community is absent. Low density of black cherry saplings and trees in its native range corresponds with negative feedback from the soil community, while close aggregation in Europe corresponds with positive soil feedback. The possible cause of positive soil feedback has not been explicitly tested.

Our results, and a study on the invasive forb, *Centaurea maculosa* (R.M. Callaway, G. Thelen, A. Rodriguez & B. Holben, unpublished data) support the suggestion by Klironomos (2002) that escape from soil pathogens may enhance plant invasiveness. In contrast, studies on the invasive grass *Ammophila arenaria* found negative effects by soil pathogens in both its native and non-native ranges (Beckstead & Parker 2003; I.C. Knevel, T. Lans, F.B.J. Menting, U.M. Hertling & W.H. Van der Putten, unpublished data). There are other alternative hypotheses besides escape from soil-borne pathogens that cannot be excluded by our results. Across ranges, differences in other biotic interactions (e.g. interspecific competition) or abiotic factors (e.g. soil moisture) may also affect the distance to the nearest neighbour. However, results from the greenhouse experiments point to an important role of soil pathogens.

Pythium spp., which cause negative plant–soil feedback in Indiana (Packer & Clay 2000), depend upon soil moisture for spore dispersal and host detection (Hendrix & Campbell 1989), suggesting that variation in soil moisture may inhibit pathogens in the sandy soils in the non-native range. In its native range, black cherry occurs on a range of soil types including sand as long as growing conditions are cool and moist (Marquis 1990), suggesting that abiotic conditions necessary for seedling survival are also favourable for *Pythium* spp.

In the native range, a positive effect of soil community on biomass was observed only when seedlings were planted at high densities and in soil collected under black cherry trees (Fig. 2b). This may have resulted if the elimination of soil mutualists outweighed the beneficial effects of eliminating soil pathogens (Bowman & Panton 1993). However, we hypothesize that this singular positive effect of the soil community results from unequal densities within the high density treatment. Specifically, high mortality in pots planted at high densities in non-sterile soil collected near black cherry trees reduced initial planting density from three seedlings per pot to an average of 1.2 seedlings. Only 11% of pots maintained three seedlings per pot.

In contrast, pots planted at high densities in sterile soil collected near black cherry had an average final density of 2.7 seedlings with 67% of pots maintaining three seedlings per pot. The observed biomass differences are consistent with a negative effect of density on seedling biomass (Fig. 2b, Table 1). Thus, we conclude that the significant interactive effects for biomass are driven by the unequal densities of the high density treatment (Table 1).

In the invaded range, the high density of black cherry trees prevented us from collecting soil far away from conspecifics. This may have resulted in greater similarity between our conspecific and heterospecific treatments in the invaded range than the native range, but feedback responses were positive in all cases. Subsequent studies may include non-native forests that have been largely cleared and where distances between black cherry trees are much larger.

Little is understood about plant–soil community interactions associated with invasive plants in their native and invasive ranges. Our results suggest that black cherry has escaped the direct negative effect of host-specific soil pathogens and is facilitated by the soil community in its non-native range while being inhibited by the soil community in its native range. However, our experiment utilized 'whole soil' inoculum, which tested the net effect of the soil community on survival and growth of black cherry, while the direct effects of soil pathogens still needs to be verified.

Further understanding of the effect of soil communities on the abundance of native and non-native species will improve predictions of both the invasiveness of plant species and the invasibility of communities (Van der Putten 2003). Biogeographical comparisons are necessary to rigorously test our understanding of invasion dynamics and community assembly.

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