

Invasion status and mycorrhizal responsiveness jointly shape context-dependent plant-soil feedbacks in Northern Great Plains grasslands

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Abstract

Background and aims

Mycorrhizal responsiveness is often assumed to generate positive plant–soil feedback (PSF) through mutualist accumulation and pathogen suppression, but whether it does so independently of a species' invasion status is unclear. In a Saskatchewan mixed-grass prairie invaded by leafy spurge (*Euphorbia virgata* Waldst. & Kit.), we tested whether mycorrhizal-responsive species show more positive PSF, whether invasive species show more positive PSF, and whether feedback is weaker under community than conspecific conditioning.

Methods

In a two-phase glasshouse experiment, five focal species differing in mycorrhizal responsiveness and invasion status: leafy spurge, Lewis' blue flax (*Linum lewisii* Pursh), prairie sandreed (*Calamovilfa longifolia* (Hook.) Scribn.), smooth brome (*Bromus inermis* Leyss.), and western porcupine grass (*Hesperostipa curtiseta* (Hitchc.) Barkworth); plus a community treatment were grown in soils from four sites differing in texture.

Results

Mycorrhizal responsiveness alone did not predict PSF direction. Of the three responsive species, only leafy spurge, the invader, produced positive feedback; blue flax and prairie sandreed accumulated significant negative feedback, with blue flax the most negative in the experiment. Spurge's positive feedback occurred only under conspecific conditioning. Negative feedback was concentrated at the two sandy loam sites, where oomycete pathogens were detected but absent from the two sandy sites.

Conclusions

Mycorrhizal responsiveness translated into a positive soil legacy only when paired with invasion, suggesting leafy spurge's advantage reflects enemy release combined with mutualist accumulation rather than responsiveness alone.

Introduction

Understanding the mechanisms that allow non-native plants to establish and spread in native plant communities is a central question in invasion ecology. Plant–soil feedbacks (PSFs), whereby plants alter the microbial community in their rhizosphere, which in turn affects their own performance and that of other plants (Ehrenfeld et al. 2005), have emerged as a potentially important mechanism driving invasion success (Reinhart and Callaway 2006; Dawson and Schrama 2016). PSF can be positive, when a plant benefits from the microbial community it cultivates, or negative, when accumulated pathogens suppress its own growth (Kulmatiski et al. 2008). Invasive plants often generate more positive PSFs in their introduced ranges than native plants, potentially because they escape the soil pathogens that suppressed them in their native range and instead accumulate beneficial mutualists in the new environment (Reinhart and Callaway 2006; Dawson and Schrama 2016). This asymmetry between invasive and native species in PSF direction may therefore be a key mechanism contributing to invasion success (Suding et al. 2013).

The role of PSF in invasion may vary across stages of the invasion process, with different mechanisms contributing to initial establishment versus long-term persistence and dominance (Inderjit and Cahill 2015). Species that perform

better in heterospecific-conditioned soils than in their own conditioned soils may benefit from microbial communities that facilitate establishment in novel environments, potentially through reduced pathogen accumulation or exploitation of naïve mutualists (Kulmatiski et al. 2008; Inderjit and van der Putten 2010). In contrast, species that perform best in conspecific-conditioned soils may be reinforcing their dominance after establishment by cultivating soil microbial communities that preferentially benefit themselves or suppress competitors (Levine et al. 2006; Suding et al. 2013). Importantly, if PSF effects observed under single-species conditioning weaken when the soil is instead conditioned by a diverse community, this would suggest that soil-mediated advantages depend on invader-dominant conditions and may therefore contribute more to the reinforcement of dominance than to initial establishment. Because PSF strength and direction can shift substantially between single-species and community conditioning, comparing these two contexts is essential for determining whether feedback effects persist under competition (Kulmatiski 2019; Goossens et al. 2023).

Beyond invasion stage, PSF outcomes may also vary among species depending on how strongly they associate with and respond to soil mutualists, particularly arbuscular mycorrhizal fungi (AMF). Mycorrhizal-responsive species can increase AMF abundance in the soil they condition and may also suppress soil pathogens through mycorrhizal protection of roots (Newsham et al. 1995; Bennett and Klironomos 2019). Both of these effects should tend to make feedback more positive, by enhancing mutualist availability and reducing pathogen loads in conditioned soil (Klironomos 2002; Bever et al. 2010). The magnitude of any mutualist benefit to the conditioning species, however, depends on how its own mycorrhizal responsiveness compares with that of its neighbours. Because many AMF are not host-specific and can colonize and connect multiple co-occurring plant species (Van Der Heijden and Horton 2009), the mutualists a responsive species accumulates are also available to heterospecific plants; a positive conspecific feedback is therefore expected to emerge most strongly when the conditioning species benefits more from these shared mutualists than the species it is compared against, rather than whenever a species is mycorrhizal-responsive in absolute terms (Kadowaki et al. 2018). The net effect of mycorrhizal responsiveness on PSF may therefore also depend on whether a species is conditioning soil alone or as part of a diverse community. These mechanisms are especially relevant for understanding invasion in the Northern Great Plains, since leafy spurge (*Euphorbia virgata* Waldst. & Kit.) and smooth brome (*Bromus inermis* Leyss.) are among the most ecologically and economically damaging invasive plants (Haines et al. 2013; Piper et al. 2015), yet they differ markedly in their mycorrhizal associations: leafy spurge appears to rely heavily on AMF (Reinhart et al. 2017), whereas smooth brome has been shown to respond negatively to mycorrhizal inoculation (McHaffie and Maherali 2020).

To address these issues, we conducted a two-phase PSF experiment using soils from four invaded sites across Saskatchewan, Canada, and five plant treatments selected to span a gradient of mycorrhizal responsiveness within the Northern Great Plains flora: an invasive forb (leafy spurge - *Euphorbia virgata*), an invasive C3 grass (smooth brome - *Bromus inermis*), a native forb (Lewis' blue flax - *Linum lewisii*), a native C3 grass (western porcupine grass - *Hesperostipa curtiseta*), and a native C4 grass (prairie sandreed - *Calamovilfa longifolia*). Additionally, we included a mixed-species community to test whether PSF is diluted within a community context. Of these, leafy spurge, blue flax, and prairie sandreed are expected to be more responsive to AMF (Wilson and Hartnett 1998; Reinhart et al. 2012, 2017; Lekberg et al. 2013), while smooth brome and western porcupine grass are expected to be less responsive (Wilson and Hartnett 1998; Reinhart et al. 2012; McHaffie and Maherali 2020). During the conditioning phase, plant treatments shaped the soil microbial community; in the response phase, the same plant treatments were grown in soils inoculated with the conditioned soil from the previous phase in a fully factorial design to assess how plants respond to the microbial legacy established by each conditioning treatment. By including both single-species and community conditioning, this design allows us to distinguish between PSF operating during initial establishment and PSF operating within a competitive community context. We hypothesized that: (1) mycorrhizal-responsive species (leafy spurge, blue flax, prairie sandreed) would show more positive PSF than less responsive species (smooth brome,

western porcupine grass), because their accumulation of mycorrhizal mutualists and suppression of soil pathogens should make their conditioned soil more favourable, particularly where they are more responsive to AMF than the neighbours they are compared against; (2) invasive species (leafy spurge, smooth brome) would show more positive PSF than native species, consistent with invaders escaping host-specific pathogens or accumulating beneficial soil biota; and (3) PSF would be weaker under community than conspecific conditioning, suggesting that soil-mediated advantages depend on invader dominance and that PSF reinforces dominance rather than facilitating initial invasion.

Methods

Area of study

The field soils for the PSF experiment were collected from 12 blocks distributed among four sites (three blocks per site) spanning a 550 km linear range along the northern extent of the Great Plains in Saskatchewan, Canada (Fig. 1). These sites were initially selected to be part of another experiment focusing on leafy spurge control and were chosen based on municipal and provincial government data that reported invasion by leafy spurge. Soils were collected from both invaded and uninvaded areas within each block: 2 L from areas invaded by leafy spurge and 2 L from adjacent uninvaded areas. These were combined to form a single composite soil sample per block (4 L total), so that the inoculum captured soil microbial communities from both invaded and uninvaded portions of each block.

Despite the heavy presence of leafy spurge in the study sites, many native plant species are present, including grasses (e.g., *Hesperostipa curtiseta* (Hitchc.) Barkworth (western porcupine grass), *Calamovilfa longifolia* (Hook.) Scribn. (prairie sandreed), and *Festuca hallii* (Vasey) Piper); forbs (e.g., *Linum lewisii* Pursh (Lewis' blue flax), *Galium boreale* L., and *Thermopsis rhombifolia* (Pursh) Richardson); and shrubs (e.g., *Amelanchier alnifolia* (Nutt.) Nutt. ex M.Roem., *Rosa arkansana* Porter, and *Elaeagnus commutata* Bernh. ex Rydb). Other invasive plants include *Bromus inermis* (smooth brome), *Poa pratensis* L., and *Cirsium arvense* (L.) Scop.

Soil conditions at the four sites were highly variable (Table 1, Online Resource S1; see supplement for details on soil measurements), with two sites being more loamy and two more sandy. Consequently, both microbial abundances and nutrients differed between the two soil types, with the loamier sites having both more microbes and more nutrients.

Table 1

Soil and climate characteristics for the four study sites. Values are means (n = 3; one replicate per block) except for mean annual precipitation (MAP). MAP (mm) represents 1991–2020 climate normals (Environment and Climate Change Canada 2026) from the nearest available weather station to each site. Soil variables are: percentage sand, silt, and clay; bacteria, AMF, fungi, and oomycetes (log10 copies g⁻¹ soil); percentage carbon and nitrogen; and phosphorus (mg/kg). Phosphorus is not reported for North Battleford, as access to the site was lost in 2025 before phosphorus sampling was completed.

Site	MAP (mm)	Sand (%)	Silt (%)	Clay (%)	Bacteria (log10 copies g ⁻¹ soil)	AMF (log10 copies g ⁻¹ soil)	Fungi (log10 copies g ⁻¹ soil)	Oom. (log10 copies g ⁻¹ soil)	C (%)	N (%)	P (mg/kg)
St. Denis	357.1	61.00	30.00	9.00	7.12	4.50	6.09	2.39	3.44	0.32	426
North Battleford	383.2	89.33	8.00	3.00	6.60	4.00	5.46	0.00	1.61	0.06	-
Grenfell	488.8	63.67	30.00	5.67	7.21	4.87	6.02	1.21	7.23	0.68	1216
Manitou	424.2	96.67	1.67	1.33	6.22	3.95	4.73	0.00	0.98	0.08	233

Choice of species

A total of five species from different functional groups, reflecting different characteristics of the native and non-native flora of the Northern Great Plains, were selected for this study, and were chosen to span a gradient of expected mycorrhizal responsiveness. The invasive forb (leafy spurge) and the invasive grass (smooth brome) were chosen because of their severe ecological and economic impacts in the region (Liu et al. 2023), so we can better understand whether their invasion in grasslands is aided by PSF processes; the two also differ in their expected mycorrhizal responsiveness, as leafy spurge is strongly associated with AMF and can increase AMF abundance in invaded soils (Lekberg et al. 2013; Reinhart et al. 2017), whereas smooth brome, a C3 grass, responds negatively to mycorrhizal inoculation (Wilson and Hartnett 1998; McHaffie and Maherali 2020). The native forb (Lewis' blue flax) was chosen because it has been shown to have strong responses to mycorrhizal fungi (Reinhart et al. 2017), providing a mycorrhizal-responsive native counterpart to leafy spurge. The native C3 grass (western porcupine grass) was chosen because it is among the most common grass species in the study region (Fore et al. 2015) and, as a cool-season C3 grass, is expected to be relatively unresponsive to mycorrhizal fungi (Wilson and Hartnett 1998). The native C4 grass (prairie sandreed) was chosen because, even though the Northern Great Plains grasslands are dominated by C3 grasses (Fore et al. 2015), it is common in sandy areas such as our study sites, and because C4 grasses are more likely to be mycorrhizal-responsive than C3 grasses (Wilson and Hartnett 1998; Reinhart et al. 2012).

Plant-soil feedback experiment

For the first phase of the PSF experiment we used round pots of 2L volume filled with 25% field soil and 75% sterilized soil, the latter a 1:1 mix of sand and topsoil (nutrient analysis in Online Resource S2), reflecting the predominantly sandy to sandy loam textures observed across our study sites (Table 1). This 25% field soil to 75% sterilized soil ratio was used to introduce a live microbial inoculum from each block while standardizing the physical and chemical soil environment across pots through the sterilized fraction, so that conditioning effects would reflect the microbial communities cultivated by each plant treatment rather than pre-existing differences in soil properties among blocks (Brinkman et al. 2010). The pots were seeded with six different treatments, which include the five functional groups and a community treatment with all five species together. The 12 blocks from the field experiment multiplied by six treatments resulted in a total of 72 pots for the first phase, where the plants were grown for three months in the greenhouse to condition the soil from May to August 2024. Two pots showed zero germination and were subsequently excluded.

To understand the effects of the conditioned soils on the six different plant treatments, the second phase of the experiment used the soils from the previous phase as an inoculum. Each of the 70 retained conditioned soils were used to fill six pots filled with a mix of 10% conditioned soil and 90% sterilized soil (same mix as in phase one). This lower proportion of conditioned soil was used to transfer the microbial legacy established during the conditioning phase while minimizing the carry-over of abiotic differences, such as residual nutrients or altered soil chemistry, so that measured feedbacks would primarily reflect biotic rather than abiotic effects (Kulmatiski et al. 2008; Brinkman et al. 2010). In this phase, we used cone pots of approximately 800ml volume. These pots were then seeded with the six plant treatments, resulting in a total of 420 pots in the second phase. Each pot was sown with a pinch of seed of the relevant treatment rather than a fixed number of individuals, so the number of established plants was not standardized and could vary among pots. The plants were grown in the greenhouse for five months. At the end of the second phase in January 2025, the aboveground biomass was collected, dried, and measured, and this data was used to calculate the plant-soil feedback values. For the single-species pots, we calculated PSF as:

$$PSF = \ln \frac{\text{biomass conspecific}}{\text{biomass heterospecific}}$$

Where *biomass conspecific* refers to the biomass from pots where plants were conditioned by their own species; and *biomass heterospecific* refers to the biomass from pots where plants were conditioned by a different species, although heterospecific also refers to the community conditioned pots.

For the community pots, we calculated PSF as:

$$PSF = \ln \frac{\text{biomass in conspecific conditioned community}}{\text{biomass in heterospecific conditioned community}}$$

Where *biomass in conspecific conditioned community* refers to the biomass of an individual species within the response community when the soil was conditioned by that same species grown alone; and *biomass in heterospecific conditioned community* refers to the biomass of that same species within the response community when the soil was conditioned by any other treatment, including the other four single-species conditioning treatments and the community conditioning treatment. It should be noted that, during harvesting, western porcupine grass and prairie sandreed in community pots were collected together as “native grass” because, at the early growth stage at which biomass was harvested, the small immature shoots could not be reliably assigned to species. For this category, both sandreed conditioned and porcupine conditioned pots were treated as two separate conspecific conditioning treatments.

Statistical analysis

Statistical analysis was conducted using linear mixed models with R package lme4 (Bates et al. 2015), with significance tested using R package lmerTest (Kuznetsova et al. 2017). The analysis focused on two models: one used the PSF measured in the single-species pots, and the other used PSF measured in the community pots. Both had PSF as the dependent variable; conditioning species, response species, and site as the independent variables; and block as the random variable. Because PSF for each response species was calculated relative to its own conspecific-conditioned soil, the full three-way interaction (conditioning species * response species * site) and the interaction between conditioning and response species were not estimable. Consequently, we fit models containing the two-way interactions supported by the data (conditioning species * site and response species * site). Additionally, in the community pots, leafy spurge did not germinate in the community response pots at North Battleford and St. Denis, so no PSF values could be estimated for this species at these two sites in the community model. To assess differences between variable levels, estimated marginal means (EMMs) were extracted from the linear mixed model using the emmeans package (Lenth and Piaskowski 2017). To determine whether PSF values significantly differed from zero for all significant variables, Z-tests were performed on the EMMs against a null hypothesis (no feedback). In addition, pairwise comparisons of EMMs were conducted using Tukey's method to adjust for multiple comparisons.

Results

Single-species PSFs

Table 2
Mixed model results showing the effects of conditioning species,
response species, and site on PSF for the single-species pots.

Variable	NumDF	DenDF	F value	P
ConditioningSpecies	5	225.2	0.710	0.617
Site	3	8.2	1.167	0.379
ResponseSpecies	4	226.1	11.564	< 0.001
ConditioningSpecies:Site	15	225.0	1.661	0.060
Site:ResponseSpecies	12	225.6	6.283	< 0.001

The results of the single-species model (Table 2, Online Resources S3–S4, Fig. 2) showed a significant effect of response species and a significant response species * site interaction (both $p < 0.001$). In contrast, neither conditioning species ($p = 0.617$) nor the conditioning species * site interaction ($p = 0.060$) had a significant effect on PSF, indicating that the identity of the species that had conditioned the soil did not detectably alter the feedback experienced by the responding plant.

Because the response species * site interaction was significant, we examined how species differed from one another within each site (Online Resource S3). No pairwise differences among species were detected at Manitou, whereas the strongest and most numerous differences occurred at St. Denis, where leafy spurge differed significantly from every other species (spurge vs. flax: estimate = 2.715, $p < 0.001$; vs. sandreed: 2.318, $p < 0.001$; vs. brome: 1.635, $p < 0.001$; vs. porcupine: 1.416, $p = 0.002$), reflecting its markedly more positive PSF at this site. At North Battleford, leafy spurge again differed from the three species with negative or neutral feedbacks (flax, porcupine, and sandreed; all $p \leq 0.014$). At Grenfell, differences were driven mainly by the contrast between species with negative PSF (brome, sandreed) and those with neutral to positive PSF (flax, porcupine), for example flax vs. sandreed (estimate = 1.210, $p = 0.001$) and porcupine vs. sandreed (0.991, $p = 0.010$).

Testing each species against zero (Online Resource S4) clarified the direction of these feedbacks. Leafy spurge exhibited significant positive PSF at North Battleford (emmean = 0.85, $z = 3.70$, $p < 0.001$) and St. Denis (emmean = 1.60, $z = 5.11$, $p < 0.001$), while showing neutral PSF at Grenfell and Manitou. Smooth brome showed significant negative PSF at Grenfell (emmean = - 0.62, $z = - 2.92$, $p = 0.004$) and neutral PSF at all other sites. Among the native species, prairie sandreed exhibited significant negative PSF at Grenfell (emmean = - 0.82, $z = - 3.88$, $p < 0.001$) and St. Denis (emmean = - 0.72, $z = - 3.15$, $p = 0.002$), while blue flax showed significant negative PSF at North Battleford (emmean = - 0.63, $z = - 2.97$, $p = 0.003$) and St. Denis (emmean = - 1.12, $z = - 4.26$, $p < 0.001$), the latter representing the most strongly negative value observed across all species and sites. Western porcupine grass showed neutral PSF across all four sites.

Community PSFs

Table 3
Mixed model results showing the effects of conditioning species,
response species, and site on PSF for the community pots.

Variable	NumDF	DenDF	F value	P
ConditioningSpecies	5	110.4	0.228	0.950
Site	3	15.8	0.289	0.832
ResponseSpecies	3	112.9	0.069	0.976
ConditioningSpecies:Site	15	109.9	0.609	0.863
Site:ResponseSpecies	7	111.9	2.133	0.046

The results of the community model (Table 3, Online Resources S5–S6, Fig. 3) showed a significant response species * site interaction ($p = 0.046$), but no significant effect of conditioning species ($p = 0.950$), site ($p = 0.832$) or the conditioning species * site interaction ($p = 0.863$), indicating that conditioning species had no effect on PSF.

Despite the significant response species * site interaction, pairwise comparisons among response species within each site revealed no significant differences at any site (Online Resource S5; all $p > 0.09$), indicating that the interaction was not strong enough to resolve into detectable contrasts between individual species. Testing each species against zero (Online Resource S6) revealed a single significant feedback: native grass exhibited negative PSF at Grenfell (emmean = -0.702 , $z = -2.391$, $p = 0.017$), indicating that native grass performed worse in community-conditioned soil than in soil conditioned by other treatments at this site. No other response species showed significant PSF at any site (all $p > 0.08$).

These community results contrast with those of the single-species model. Whereas prairie sandreed (one of the two native grasses combined here as "native grass") also showed negative PSF at Grenfell when grown alone, the additional significant feedbacks detected when grown as single species (positive PSF for leafy spurge and negative PSF for smooth brome and blue flax across several sites) were absent under community conditioning.

Discussion

Our results provide only partial and uneven support for our three hypotheses. Neither invasion status nor mycorrhizal responsiveness cleanly predicted the direction of PSF. Instead, feedback varied strongly among species and sites, and the clearest signal was that leafy spurge, our one mycorrhizal-responsive invader, was the only species to generate consistent positive feedback, and did so only when grown alone. Consequently, PSF likely serves to reinforce the dominance of leafy spurge once it becomes established. More broadly, the feedbacks detected when species were grown as single species largely disappeared under community conditioning, so that almost no significant feedbacks remained once plants were grown together, suggesting that PSF is unlikely to be a significant driver of initial community assembly or invasion.

Our hypothesis that mycorrhizal-responsive species would show more positive PSF than less responsive species, through their accumulation of mycorrhizal mutualists and suppression of soil pathogens, was only partly supported and revealed an unexpected split. Of the three species expected to be responsive to arbuscular mycorrhizal fungi (leafy spurge, blue flax, and prairie sandreed), only leafy spurge produced the predicted positive feedback. Blue flax and prairie sandreed instead accumulated significant negative feedback at multiple sites, with blue flax showing the most strongly negative value in the entire experiment. Mycorrhizal responsiveness alone therefore did not predict the direction of PSF, suggesting that for the responsive native species any protective benefit of mycorrhizal association

was outweighed by the accumulation of host-specific antagonists in their own conditioned soil (Klironomos 2002; Bever et al. 2010). Critically, the one responsive species that did generate positive feedback was also the only invader among them, indicating that mycorrhizal responsiveness may translate into a positive, self-reinforcing soil legacy only when combined with the enemy release that can accompany invasion (Reinhart and Callaway 2006). This points to the role of soil microbes in invasion being contingent not on mycorrhizal association per se, but on whether a species can accumulate mutualists while escaping the pathogens that accumulate on responsive native species.

Our hypothesis that invasive species would show more positive PSF than natives was not supported, as only leafy spurge produced positive feedback, and only at two of four sites. Smooth brome, our other invader, accumulated neutral to negative feedback more closely resembling the native species than its fellow invader. Invasion status alone therefore does not predict PSF direction in this system, indicating that any release from soil antagonists is not a general property of invaders but depends on the individual species (Kulmatiski et al. 2008). Smooth brome's negative feedback is consistent with a growth-defense trade-off, in which fast-growing, acquisitive species invest less in belowground defense and accumulate host-specific antagonists more readily (Lemmermeyer et al. 2015).

Our hypothesis that PSF would be weaker under community than conspecific conditioning was supported at the level of overall feedback strength, though not through the specific mechanism we proposed. Almost all of the feedbacks detected in the single-species model, including the positive feedback of leafy spurge and the negative feedbacks of smooth brome, blue flax, and prairie sandreed, were absent in the community model, where only native grass at Grenfell departed significantly from zero. The poor correspondence between the two models echoes growing evidence that PSF measured on single species is a weak predictor of feedback in a community context (Kulmatiski 2019; Goossens et al. 2023). Importantly, conditioning species, including conditioning by the whole community, had no significant effect on PSF in either model. The weakening of feedback under community conditions therefore appears to reflect the diminished expression of PSF when responding plants grow together, rather than a change in the soil legacy created by different conditioning treatments (Grenzer et al. 2021). A further, striking pattern reinforces this interpretation: leafy spurge failed to germinate in the community pots at North Battleford and St. Denis, precisely the two sites where it had shown its strongest positive feedback when grown alone. Although this germination failure cannot be attributed to PSF with certainty, since competition and other factors may also have contributed, it is consistent with the possibility that spurge's positive soil legacy benefits the species chiefly when it is already dominant, in near-monospecific stands, rather than aiding its establishment within a diverse community. Taken together, these results suggest that the self-reinforcing feedback leafy spurge generates when grown alone may contribute more to the maintenance of dominance in established, invader-dominated patches than to invasion into intact, diverse communities (Kulmatiski 2019).

Feedbacks varied strongly among sites, as reflected in the significant response species $\times$ site interactions. The two finer-textured, sandy loam sites (St. Denis and Grenfell) contained oomycetes, a group that includes many root pathogens, whereas the two sandier sites (North Battleford and Manitou) had none; AMF abundance, by contrast, varied little among sites (Table 1). This difference likely reflects water availability, as finer-textured soils retain more moisture and oomycetes disperse via water-dependent zoospores that require moist conditions (Sterne et al. 1977; Walker and van West 2007). Because the significant negative feedbacks were concentrated at the two oomycete-containing sites, whereas Manitou, which lacked detectable oomycetes, produced no significant feedbacks in either model, these negative feedbacks appear to be driven by oomycete pathogens rather than by differences in mutualist availability. Notably, the two oomycete-containing sites were also the two highest in soil carbon (Table 1), so greater soil productivity and pathogen presence covaried; more carbon-rich soils can support larger and more active microbial communities, including pathogens (Prommer et al. 2020), which may reinforce the stronger negative

feedbacks observed at these sites. That such feedbacks emerged even in these relatively sandy soils is consistent with foundational work demonstrating pathogen-driven feedbacks in sandy dune systems (van der Putten et al. 1993).

Although we found no consistent community level feedback on native grasses, the fact that western porcupine grass and prairie sandreed were harvested together may limit the strength of this inference. Although these species can be distinguished by vegetative traits such as leaf venation and collar morphology, at the early growth stage at which biomass was harvested we were unable to assign the small, immature shoots to species with sufficient confidence to separate them reliably. Combining them is nonetheless a meaningful limitation, because the two grasses behaved differently when grown as single species: prairie sandreed showed significant negative PSF at Grenfell and St. Denis, whereas western porcupine grass showed no significant feedback at any site, and the two are expected to differ in mycorrhizal responsiveness (Wilson and Hartnett 1998; Reinhart et al. 2012). The significant negative feedback we detected for native grass at Grenfell in the community model could therefore be driven disproportionately by prairie sandreed, and we cannot resolve whether it reflects a shared response of the two native grasses or is attributable mainly to one of them.

Overall, our results indicate that neither invasion status nor mycorrhizal responsiveness alone reliably predicts the direction of plant–soil feedback in this system. The one species to generate consistent positive feedback, leafy spurge, was both mycorrhizal-responsive and invasive, suggesting that a self-reinforcing soil legacy may emerge only where responsiveness and invasion coincide; yet, even this feedback was confined to plants grown alone and did not persist under community conditioning, pointing to a role for PSF in reinforcing dominance rather than driving initial establishment. That feedbacks were largely undetectable once plants were grown together, and that the identity of the conditioning species had no significant effect, further cautions against inferring community-level dynamics from single-species feedbacks (Kulmatiski 2019; Goossens et al. 2023). Where feedbacks did occur, they were concentrated at the sites where oomycete pathogens were present and soil carbon was higher, suggesting that pathogen dynamics and soil productivity, rather than soil texture or mycorrhizal abundance, underlie the site-dependence of PSF. A more detailed study of soil microbial communities would help clarify this hypothesis.

Declarations

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Competing interests

The authors have no relevant financial or non-financial interests to disclose.

Author contributions

JAB and RRCT contributed to the study conception and design. The greenhouse experiment and data collection were performed by RRCT. Data analysis was conducted by RRCT and JAB. The dPCR work was performed by HNHP. Supplementary materials were written by HNHP and JAB, while the rest of the paper was written by RRCT and JAB. All authors read and approved the final manuscript.

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Data availability

The dataset and code that support the current study are available on FigShare at <https://doi.org/10.6084/m9.figshare.33098768>.

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Figures

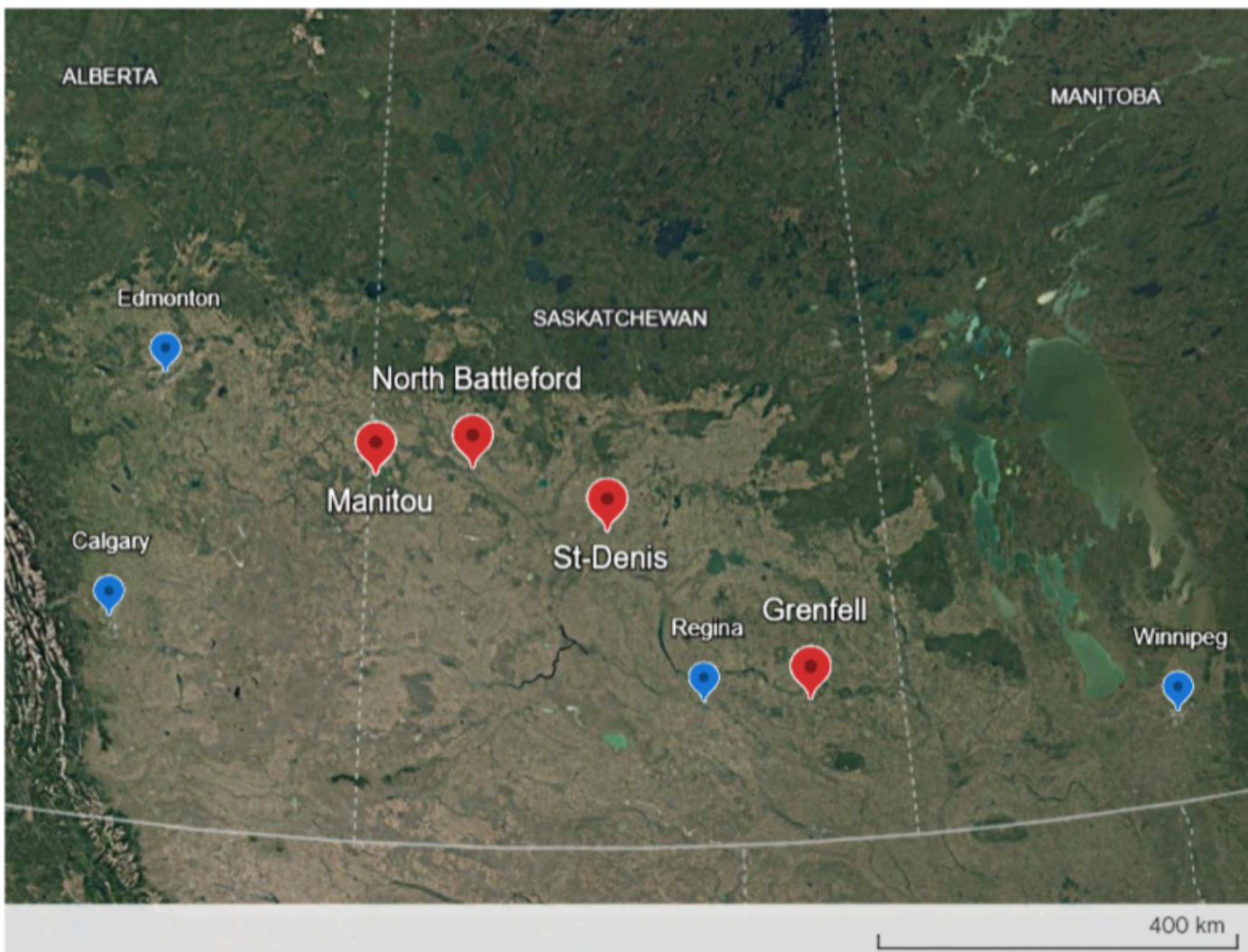


Figure 1

Location of the four study sites (red pins) across Saskatchewan, Canada, with reference Canadian Prairie cities shown for orientation (blue pins). Provincial boundaries are shown as dashed lines and international boundaries as solid lines

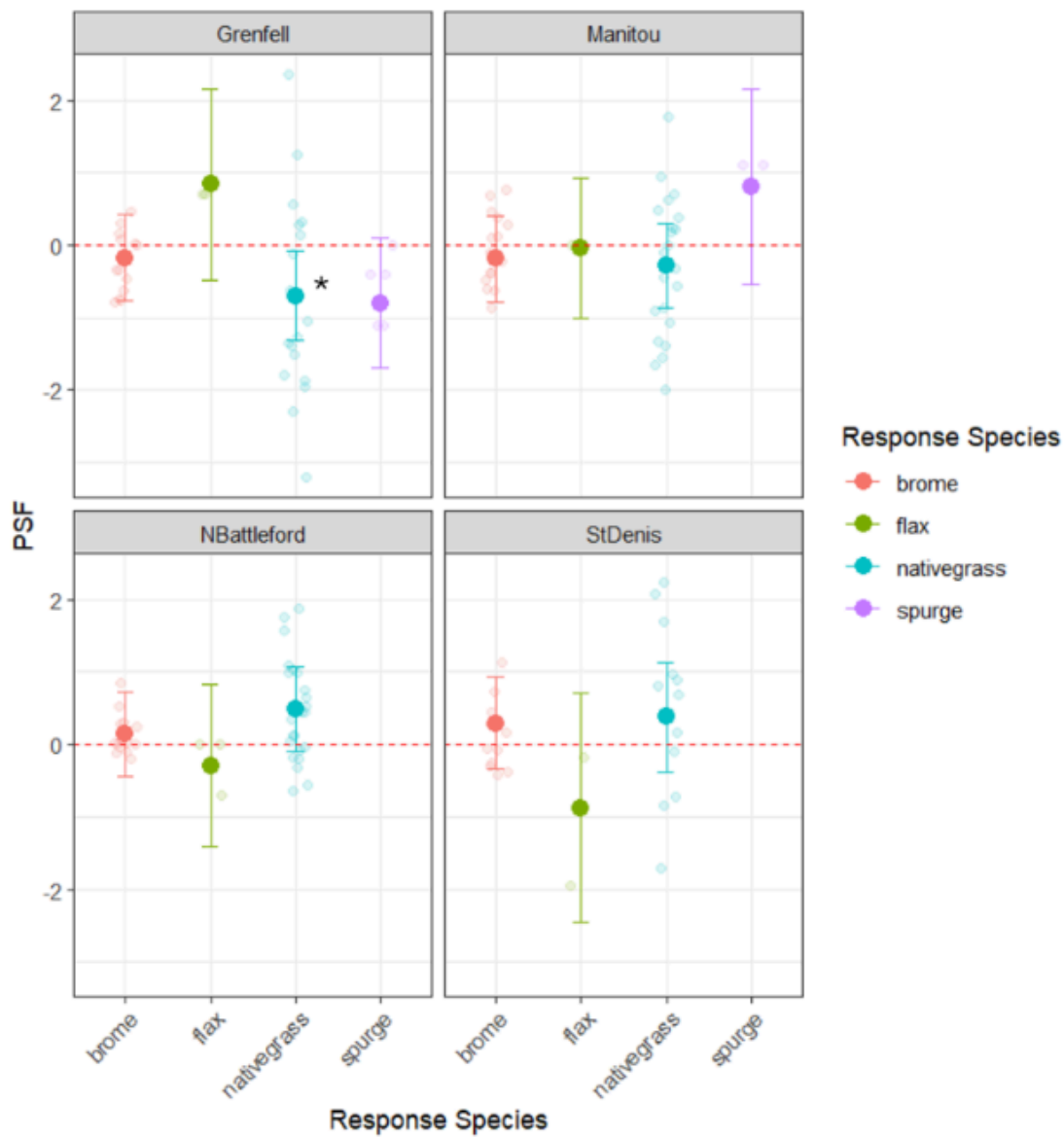


Figure 2

Estimated marginal means of PSF for each response species at each site based on the linear mixed-effects model for community pots. Leafy spurge is absent from North Battleford and St. Denis owing to germination failure in the response communities at these sites. Points represent model-estimated means, error bars show 95% confidence intervals, and asterisks denote PSF values significantly different from zero

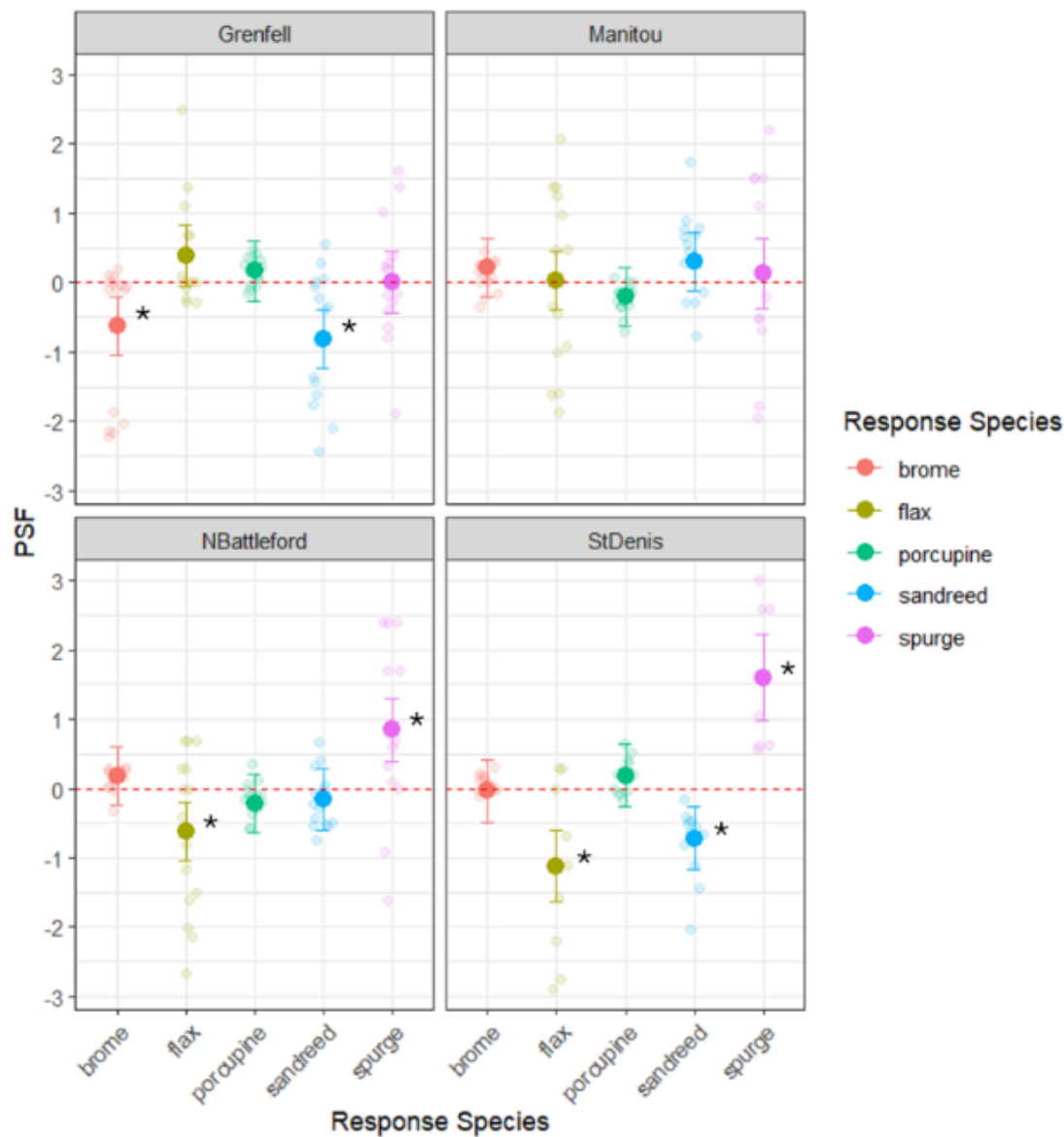


Figure 3

Estimated marginal means of PSF for each response species at each site based on the linear mixed-effects model for single-species pots. Points represent model-estimated means, error bars show 95% confidence intervals, and asterisks denote PSF values significantly different from zero

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