

The interaction between propagule pressure, habitat suitability and density-dependent reproduction in species invasion

Robert J. Warren II, Volker Bahn and Mark A. Bradford

R. J. Warren II (hexastylis@gmail.com) and M. A. Bradford, School of Forestry and Environmental Studies, Yale Univ., New Haven, CT 06511, USA. – V. Bahn, Dept of Biological Sciences, Wright State Univ., Dayton, OH 45435, USA.

Seedling recruitment limitations create a demographic bottleneck that largely determines the viability and structure of plant populations and communities, and pose a core restriction on the colonization of novel habitat. We use a shade-tolerant, invasive grass, *Microstegium vimineum*, to examine the interplay between seed and establishment limitations – phenomena that together determine recruitment success but usually are investigated individually. We add increasing amounts of seed to microhabitats containing variable levels of leaf litter thickness – with reduced leaf litter simulating disturbance – to investigate whether reduced seed limitation overcomes the establishment limitation posed by litter cover. We do this across gradients in understory light, moisture and temperature, and quantify germination, survival, and then per capita adult biomass and reproduction in order to understand the implications for invasion across the landscape. We find that the combined effects of seed and establishment limitation influence recruitment; however, propagule pressure overwhelms the inhibitory effects of leaf litter thickness. Leaf litter reduces germination by 22–57% and seedling survival by 13–15% from that observed on bare soil. However, density-dependent reproduction compensates as 1–3 plants can produce far more seeds (approx. 525) than are required for persistence. As such, just a few plants may establish in understory forest habitat and subsequently overwhelm establishment barriers with copious propagule production. These results, for a widespread, invasive plant, are consistent with the emerging perspective for native plants that seed and establishment limitation jointly influence recruitment. The ability for an exotic plant species to compensate for low population densities with high per capita seed production, that then overrides establishment limitations, makes its invasive potential daunting. Further work is required to test if this is a common mechanism underlying plant invasions.

Initial conditions often determine the trajectory of complex systems (Strogatz 2001). In plant communities, initial seedling establishment greatly determines subsequent population success or failure (Grubb 1977, Harper 1977, Fenner and Kitajima 1999, Moore and Elmendorf 2006, Albrecht and McCarthy 2009, Warren and Bradford 2011). Seedling recruitment initially depends on the arrival and density of colonizing propagules – such dispersal is recognized as a key component in population and community structuring (Pulliam 2000, Hubbell 2001, Tilman 2004). Following dispersal, the interaction between arriving propagules and local microsite suitability determines whether plant recruitment succeeds or fails (Grubb 1977, Eriksson and Ehrlén 1992, Houseman and Gross 2006, Clark et al. 2007, Moore 2009, Myers and Harms 2009, Klanderud 2010).

The interaction between seed limitation (propagule pressure) and establishment limitation (safe site, suitable habitat, microsite availability) influences the persistence of plant species and helps shape community structure (as noted by Clark et al. 2007, Poulsen et al. 2007). For example, high seed input falling on an asphalt highway illustrates an extreme establishment limitation, whereas a single seed falling on fertile ground can easily be eradicated by a stochastic

event, illustrating extreme seed limitation. Most natural systems likely fall somewhere between such extremes so that recruitment is a product of seed and establishment limitation (Clark et al. 2007, Eriksson and Ehrlén 1992) – although the relative importance of each component may be species- or system-specific (Eriksson and Ehrlén 1992, Turnbull et al. 2000, Lockwood et al. 2005, Clark et al. 2007, Duncan et al. 2009).

For invasive species, the interaction between seed and establishment limitation is a crucial step for invading novel communities (Lockwood et al. 2005, Colautti et al. 2006, Eschtruth and Battles 2009, Tanentzap and Bazely 2009). We evaluate the interplay between seed and establishment limitations on *Microstegium vimineum* recruitment (successful germination and seedling survival) by adding increasing amounts of *M. vimineum* seeds to microhabitats containing multiple, experimental levels of leaf litter thickness. *Microstegium vimineum* is an annual grass native to southeastern Asia, and its distribution now extends across 25 US states. Given that soil moisture, temperature and diffuse light also pose considerable influence on *M. vimineum* recruitment and reproduction (Warren et al. 2011a, b), we include microhabitat-specific measurements of these

variables to account for natural variation and spatial autocorrelation.

We investigate the degree of *M. vimineum* establishment limitation posed by leaf litter and the potential for increased seed pressure to overcome it. Previous work and theory suggests that plant populations are limited primarily by ($H1_a$) seed supply, so that recruitment is determined largely by stochastic processes (Eriksson and Ehrlén 1992, Hubbell 2001) or ($H1_b$) habitat resistance to establishment, so that recruitment success is dominated by establishment success, which depends on microsite conditions (Grubb 1977, Crawley 2000, Turnbull et al. 2000, Warren et al. 2011a). However, the relative contributions of seed pressure and establishment suitability on seedling recruitment may interact in a continuous manner (Eriksson and Ehrlén 1992, Lockwood et al. 2005, Clark et al. 2007, Poulsen et al. 2007, Tanentzap and Bazely 2009) so that ($H1_c$) the combined effects of seed- and establishment-limitation determine recruitment success.

At low densities with little intraspecific competition, many plants – particularly annuals (Clay and Shaw 1981, Smith 1983, Schmitt et al. 1987, Goldberg et al. 2001) – can effectively compensate for low initial numbers with high seed production. However, as the species density increases through recruitment success, intraspecific competition increases and seed production drops. Indeed, invasive plants generally are far more seed than establishment limited (Von Holle and Simberloff 2005), meaning that density-dependent effects progressively limit recruitment as propagule pressure increases (Lockwood et al. 2005, Duncan et al. 2009). We examine whether ($H2_a$) seed production in the experimentally established *M. vimineum* plants is density-dependent so that per capita seed production decreases with establishment density. Alternatively, given the high densities and fast spread of *M. vimineum* throughout the eastern US (Warren et al. 2011b), ($H2_b$) *M. vimineum* reproduction may be density-independent so that per capita seed production remains constant across a gradient in plant density. Given that *M. vimineum* is an invasive species that poses a considerable threat to understory plant communities (Warren et al. 2011b), we also assess the minimum amount of propagule pressure necessary for population establishment and persistence.

Methods

Study species

Since arriving in the US in the early 1900s (Fairbrothers and Gray 1972), *M. vimineum* has spread throughout the eastern US and midcontinental states (USDA 2010). It invades roadsides, trails and waterways, along with forest understories and wetlands, displacing native vegetation (Cole and Weltzin 2004, Oswalt et al. 2007, Flory and Clay 2009a, b, Warren et al. 2011b). *Microstegium vimineum* plants may be establishment limited by low light and leaf litter thickness in forest understories (Barden 1987, Redman 1995, Gibson et al. 2002, Glasgow and Matlack 2007, Oswalt and Oswalt 2007, Marshall and Buckley 2008, Christen and Matlack 2009, Knight et al. 2009, Schramm

and Ehrenfeld 2010), but copious *M. vimineum* seed production may overcome unsuitable habitat (i.e. establishment limitation, Eschtruth and Battles 2009).

Study design

The experimental plots were established during October 2009 at the Coweeta Long Term Ecological Research site (CWT) in Macon County, NC, USA (35°03'N, 83°25'W; 685 m elevation, 183 cm MAP, 13°C MAT). The experimental field site contains mesic deciduous forest edge proximate to modified (buildings and paved roads) landscape. Six plots were established within the 1-ha forest stand using a stratified random design to capture variation in ambient understory soil moisture, temperature and diffuse light created by forest edge and lowland stream drainage (Fig. 1). Site use permission restricted introduction of invasive species to novel habitat, and *M. vimineum*'s persistent seed bank made using previously invaded habitat undesirable. As a compromise, we translocated 5-cm deep soil 'plugs' from nearby (~100 m) uninvaded habitat with similar forested conditions to invaded habitat in PVC collars for seed introduction. Within each 1-m² plot, 12 'plugs' were created using 15-cm long × 10-cm dia. PVC collars embedded to a depth of 5 cm (to ensure transplanted soil was flush with the soil surface). *Microstegium vimineum* seeds were harvested at the study site in Sept. 2009, and added to the plugs at four levels: 0, 10 (Sd1), 40 (Sd2) and 160 (Sd3) seeds. The seed amount was based upon 0.25 ×, 1 × and 4 × mean seed output per plant when seeds were collected. The leaf litter treatment within the plugs occurred at three levels: bare soil (Lt1); the remaining, decomposed leaf litter from previous years that resided as a shallow (~1 cm depth) crust on top of the forest soil (Lt2); and additional to the previous year's litter, 5 cm (depth) of the current year's leaf litter (Lt3). The litter mostly consisted of deciduous tree leaves typical of mid-elevation forests (e.g. *Fraxinus* spp., *Acer* sp., *Liriodendron tulipifera*). To prevent contamination from natural *M. vimineum* seed dispersal, litter was collected with

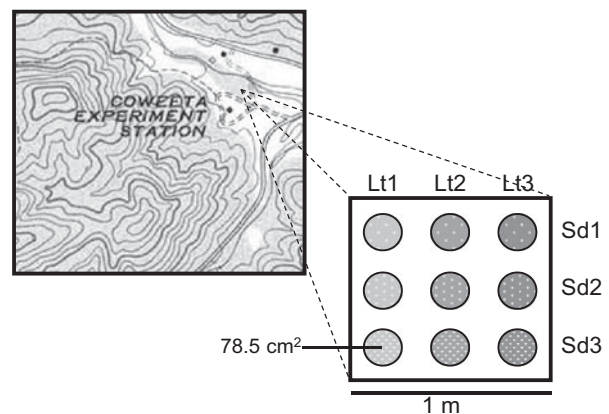


Figure 1. Study site diagram. Six experimental seed and establishment plots were placed at the Coweeta Long Term Ecological Research site in the southern Appalachian Mountains in Otto, NC, USA. Within each plot, 10 (Sd1), 40 (Sd2) and 160 (Sd3) *M. vimineum* seeds were added to bare soil (Lt1), the previous year's decomposed leaf litter (Lt2) and the previous year + 5 cm of new leaf litter (Lt3).

the soil plug at uninvaded sites. Seeds were added on top of Lt1 and Lt2 and between the previous and current year's litter in Lt3. This methodology mimicked *M. vimineum* falling on disturbed soil, undisturbed soil without concurrent leaf litter accumulation and undisturbed soil with concurrent leaf litter accumulation. The experimental plugs were covered with 1 mm² pore screen mesh (secured with a metal clamp) immediately after seed addition in October 2009 to prevent unintentional input from additional leaf litter and/or *M. vimineum* seed rain. The screens promptly were removed when biological activity began in spring May 2010 so there were no effects on plant germination or growth, and there were no impediments to ambient light, soil moisture and temperature exchange – particularly because the translocated soil plugs had been inserted into the soil. The inclusion of the no seed treatment verified there was no soil or litter contamination from *M. vimineum* seed other than the intended treatment additions. These units only served to ensure the method of seed introduction was robust and so were not evaluated in data analysis.

Demography

Germination was recorded as the number of seedlings appearing in May 2010; survival was calculated as the proportion that survived to October 2010; growth was calculated as per capita biomass (as annuals, growth only occurs during 1 season) and reproduction was recorded as the number of seeds produced in October 2010. The adult plants were harvested, the seeds removed and all plant matter was dried for one week at 60°C before weighing. *Microstegium vimineum* produces both cleistogamous (self-pollinated) and chasmogamous (outcrossed) seeds (Cheplick 2005, 2006), and these were separated for independent count and biomass analysis to analyze relative output, which may reflect an environmental response (Porras and Munoz 2000, Cheplick 2007). Cleistogamous seeds were identified as those contained within the plant stems and axils whereas chasmogamous seeds were exposed for pollination.

Abiotic monitoring

Soil moisture, temperature and diffuse light were measured at each plot every other week May–October 2010. Soil moisture and temperature measurements were taken at each corner of the 1-m² plots and averaged for data analysis. Two diffuse light measurements were taken for each plot and averaged. Volumetric soil moisture percentage was measured with a handheld measurement system. Ambient soil temperature was measured with a T-shaped digital thermometer inserted 8 cm into the soil. Percent photosynthetically active radiation (PPFD, diffuse light) was calculated as the difference between plot-level PAR readings and a fully exposed PAR reference site. The understory measurements were taken with a LI-191 line quantum sensor, which integrates light measurement along a 1-m wand, and the open reference measurements were taken with a spherical PAR sensor and logged with a datalogger. Measurements were taken on overcast days between 10 a.m. and 2 p.m. to minimize variation in diffuse light.

Data analysis

We used generalized linear mixed models (GLMM) to evaluate the fixed effects (litter thickness, seed addition and plant density) on proportional per capita and total *M. vimineum* germination, proportional seedling survival, and outcrossed seed production assuming binomial error distributions for the proportions and Poisson error distributions for the count data. For the germination models, we coded the treatments as 10, 40 and 160 seeds and 0, 1 and 5 cm leaf litter as these were biologically meaningful. We specifically designed the field experiment in six clusters to take advantage of the ability for GLMM models to detect and account for random effects (Bolker et al. 2009). We account for potential spatial autocorrelation, and the influence of ambient abiotic heterogeneity, by evaluating soil moisture, light and temperature as random effects in the GLMMs. This approach is similar to 'blocking' in a factorial design, but can detect variation in the abiotic gradients otherwise considered unobserved error in block effects. The mixed models were fit using the Laplace approximation in the 'lme4' package (Bates and Maechler 2009) for the R statistical programming environment (R Development Core Team 2009). We also used generalized linear models to examine correlations between plant growth and seed output, and seed number and seed biomass. In evaluating the impact of the treatments on *M. vimineum*, the inclusion or exclusion of the random effects was based on Akaike's information criterion (AIC) values (Akaike 1973). AIC measures the distance of models to truth; that is, the information loss in the data given that the model is a simplification of reality and cannot explain full variance in the data (Burnham and Anderson 2002). In effect AIC is a measure of goodness-of-fit, as the more familiar R² measure, with an added penalty for model complexity as measured by the number of fit parameters in the model.

Graphical analysis of total and per capita seed production as functions of plant density suggested the responses were density-dependent so we fit a nonlinear model $y = e^{(a + bx)}$ using the nls() package (Bates and Watts 1988) in the R statistical package (R Development Core Team 2009). Overdispersion did not occur in any of the Poisson or binomial models (< 1.1). The variance inflation factors for soil moisture and temperature were < 5, indicating that they independently explained variance in *M. vimineum* demography; however, diffuse light and temperature were collinear. These plot-level variables, along with a plot variable, were included in the mixed models as random factors and AIC was used to select the random factor with best fit so that collinearity did not influence statistical results. The relative differences in abiotic conditions across plots remained similar throughout the growing season, so mean temperature, light and soil moisture across May–September were used for analysis.

Results

Mean temperature ($\pm$ SD) at the study site during the *Microstegium vimineum* growing season (May–September 2010) was $18.3 \pm 0.4^\circ\text{C}$; mean diffuse light was $48.7 \pm 31.4\%$ (this ranged from $72.2 \pm 18.0\%$ in May to $24.7 \pm$

22.7% in September as a consequence of overstory canopy development), and mean soil moisture was $34.6 \pm 11.9\%$. Across plots, mean temperatures were relatively similar, ranging from 17.8–18.7°C; but there were large gradients in diffuse light (7–91%) and soil moisture (19.6–50.3%).

We introduced 3780 *M. vimineum* seeds of which 38% germinated across all plots and treatments. Gradients in diffuse light best explained increased *M. vimineum* germination across plots, and light was included as a random effect in the GLMM. The proportion of germinating seeds decreased significantly with leaf litter thickness (coeff. = -0.26 , SE = 0.04, z-value = -6.10 , $p < 0.001$) and seed addition (coeff. = -0.009 , SE = 0.0008, z-value = -10.58 , $p < 0.001$), and there was no significant interaction (Fig. 2a), though with 3×3 treatments the power to detect interactions was low. For total seeds germinating, leaf litter also had a significantly negative effect (coeff. = -0.14 , SE = 0.06, z-value = -2.22 , $p = 0.03$), but it was offset by a significantly positive effect of seed addition (coeff. = 0.91, SE = 0.06, z-value = 15.95, $p = 0.001$), and there was no significant interaction (Fig. 2b).

Once they germinated, most seedlings survived to the reproductive adult stage (93%). Gradients in diffuse light best explained increases in the proportion of *M. vimineum* seedlings surviving across plots. Leaf litter thickness had no effect on per capita seedling survival (coeff. = -0.33 , SE = 0.59, z-value = -0.55 , $p = 0.583$) but increased seed addition corresponded with significantly increased seedling survival (coeff. = 0.788, SE = 0.24, z-value = 2.40, $p < 0.017$). There was no interaction between seed input and litter layer thickness on seedling survival.

As an annual, per capita *M. vimineum* biomass equals growing season plant growth. Mean vegetative growth across all plots was 351.9 ± 391.0 mg per plant and mean reproductive growth (seeds) was 32.1 ± 38.6 mg per plant. Per capita plant growth was highly correlated with per capita seed output (coeff. = 4.09, SE = 0.12, t-value = 34.57, $p < 0.001$, $r^2 = 0.96$) (Fig. 3), as were seed number and seed

biomass (coeff. = 0.0004, SE = 0.0002, t-value = 23.45, $p < 0.001$, $r^2 = 0.91$). The strong correlation between growth and reproduction makes evaluating both redundant, and reproductive output is the functionally important vital rate for annual persistence, so we evaluated total and per capita seed production. Due to failed germination and seedling death, we used plant density (number of surviving plants) to evaluate reproduction. Because both the per capita and total seed production appeared curvilinear, we first determined the least-squares estimates of the model parameters assuming quadratic, power and exponential functions. A nonlinear approach did not improve model fit for total seed production, but it did for per capita seed production. A GLMM was used to evaluate total seed output per plant density. Gradients in diffuse light best explained increases in *M. vimineum* total seed production across plots, and total seed output increased significantly with plant density (coeff. = 0.004, SE = 0.002, z-value = 17.47, $p < 0.001$) but there was high variance associated with this relationship (Fig. 4). In contrast, per capita seed production decreased significantly with plant density in a negative exponential pattern (coeff.a = 6.23, SE = 0.18, t-value = 34.38, $p < 0.001$; coeff.b = -0.18 , SE = 0.04, t-value = -4.86 , $p < 0.001$) (Fig. 4). The proportion of cleistogamous (self-pollinated) to total seeds did not change with leaf litter thickness (coeff. = 0.02, SE = 0.003, z-value = 0.75, $p < 0.453$) or plant density (coeff. = -0.0002 , SE = 0.0009, z-value = -0.25 , $p = 0.81$), and there was no interaction effect between these variables.

Discussion

Our results demonstrate that the combined effects of seed and establishment limitation influence recruitment; however, the inhibitory effects of leaf litter thickness generally are overwhelmed by propagule pressure (Fig. 2). Leaf litter reduces germination by 22–57% and seedling survival

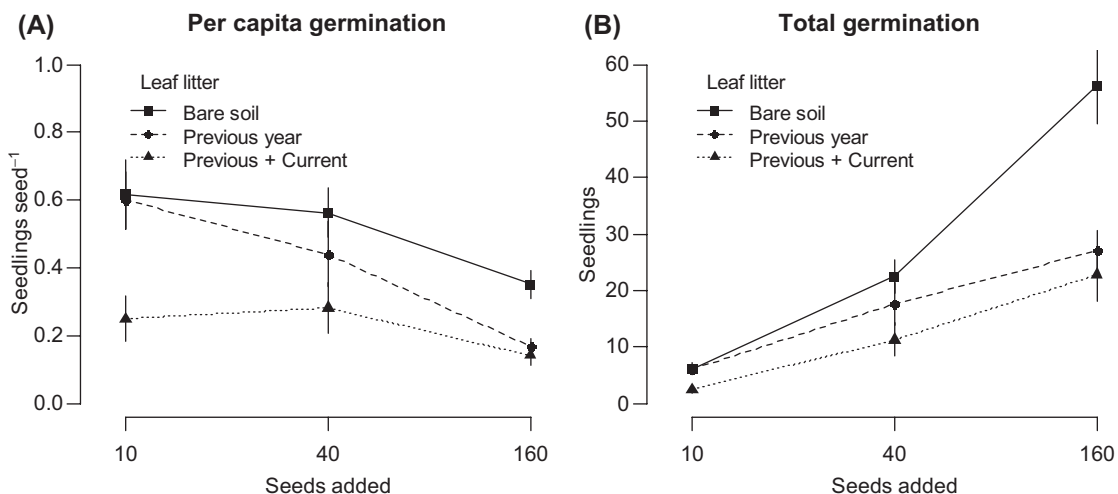


Figure 2. Interaction plots of the effect of seed input and leaf litter on seedling per capita (A) and total (B) germination ($\pm 95\%$ CI). The treatments were the addition of 10, 40 and 160 *Microstegium vimineum* seeds on substrates of bare soil, decomposed leaf litter from the previous year and the previous year's litter + 5 cm of the current year's leaf litter. Given robust germination (38%) and high seedling survival (94%), initial *Microstegium vimineum* seed input relates strongly to subsequent plant density and hence individual plant size.

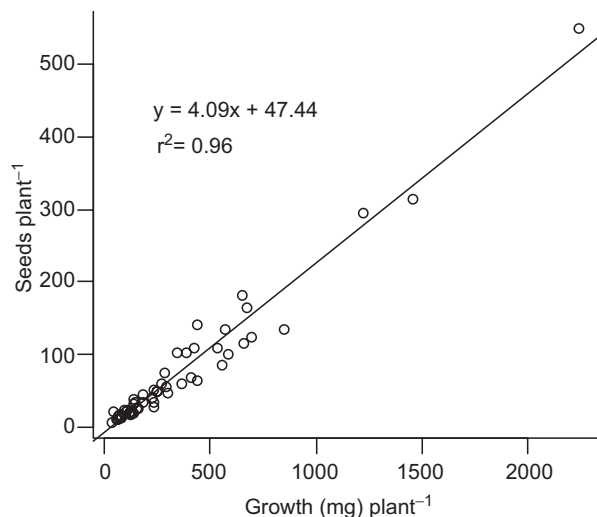


Figure 3. Linear regression of per capita *Microstegium vimineum* plant growth and seed production. The amount of seeds produced by each plant increases significantly with the size of the plant ($p < 0.001$).

by 13–15% from that observed on bare soil, suggesting the germination stage is very susceptible to this habitat limitation. At low seed input, greater leaf litter thickness can even prevent colonization. However, density-dependent reproduction and recruitment compensates for low plant density as 1–3 plants can produce far more seeds (approx. 525 seeds) than required for persistence (10–40 seeds) (Fig. 4) and more seeds germinate and survive where fewer seeds arrive (Fig. 2a). These results suggest that just a few plants may establish in understory forest habitat and subsequently overwhelm establishment barriers with copious propagule production.

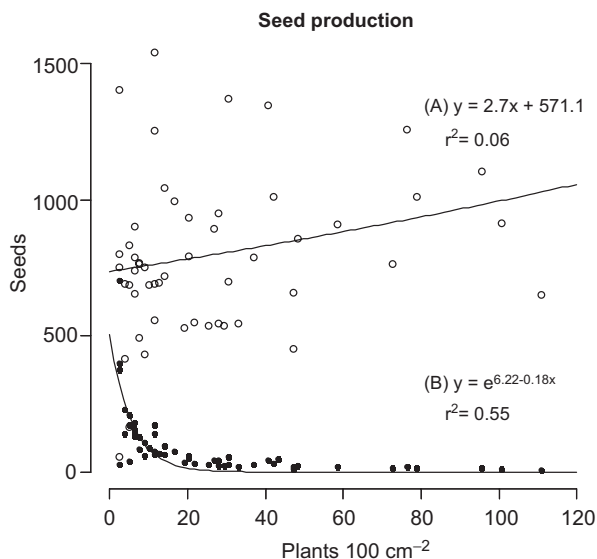


Figure 4. Scatter plots of the effect of plant density on (A, empty circles) total and (B, filled circles) per capita *Microstegium vimineum* seed output. Both the positive linear relationship for seeds per total plants, and the negative exponential relationship for per capita seeds, were significant ($p < 0.001$).

Seed versus establishment limitation

Progressively higher amounts of leaf litter cover proportionally lower the capacity for *Microstegium vimineum* germination, but seedling density still increases with seed addition (Fig. 2) – suggesting that propagule pressure is a greater influence on *M. vimineum* establishment than habitat suitability. That we rarely observe *M. vimineum* producing anything less than copious seed output suggests it is more limited by dispersal ability than suitable habitat or seed production. Indeed, empirical investigations reveal that *M. vimineum* seeds rarely travel > 2 m from source plants (Barden 1987, Gibson et al. 2002, Oswalt and Oswalt 2007, Christen and Matlack 2009, Huebner 2010a, b, Rauschert et al. 2010, Warren et al. 2011a), though greater distances are possible via roads and waterways (Gibson et al. 2002, Cole and Weltzin 2004, 2005, Christen and Matlack 2009, Huebner 2010a, b).

Forest leaf litter poses a considerable recruitment barrier for many shade-adapted understory species (Xiong and Nilsson 1999), including *M. vimineum* (Glasgow and Matlack 2007, Oswalt and Oswalt 2007, Miller and Matlack 2010, Schramm and Ehrenfeld 2010, Warren et al. 2011a); although not always (Marshall and Buckley 2008, Miller and Matlack 2010). Notably, current research on leaf litter effects has focused on the impacts of habitat manipulation without systematically manipulating seed input. Our results showing propagule pressure may overwhelm leaf litter limitations (Fig. 2) highlight the need for studies to manipulate both seed rain and leaf litter to understand fully *M. vimineum* recruitment. The need for studies to simultaneously consider both propagule pressure and establishment limitation seems applicable for understanding the population ecology of both exotic and native species (Clark et al. 2007).

Our results are concomitant with existing recruitment theory positing that the effects of seed and establishment limitation occur on a continuum with simultaneous, though not necessarily equal, control on plant populations (Eriksson and Ehrlén 1992, Clark et al. 2007, Poulsen et al. 2007). The greater influence of propagule pressure than habitat quality on *M. vimineum* recruitment fits with the theoretical expectations (Lonsdale 1999, Lockwood et al. 2005, Liu and Stiling 2006) and empirical results (Von Holle and Simberloff 2005, Eschtruth and Battles 2009, Tanentzap and Bazely 2009) for invasive species, including *M. vimineum* (Eschtruth and Battles 2009). We extend this investigation, however, by quantifying subsequent survival, growth and reproduction in the introduced *M. vimineum* plants and connecting recruitment success with density-dependent dynamics and population persistence.

Invasion and density-dependent recruitment and reproduction

A further consideration into postdispersal processes is whether propagule pressure causes density-dependent mortality between seed arrival and life stage progressions (Poulsen et al. 2007). Whereas we find more overall seedlings with more seeds added (Fig. 2b), we also find decreased germination with more seeds added (Fig. 2a). Intraspecific shading is the likely mechanism for this pattern as light is a

common plant germination cue (Boyd and Van Acker 2004), and we find that leaf litter thickness – which shades seeds (Xiong and Nilsson 1999) – also inhibits *M. vimineum* germination (Fig. 2a). *Microstegium vimineum* seeds can remain viable for at least three years, a strategy that allows the annual plant population to persist through temporal variability in habitat quality, e.g. drought (Warren et al. 2011b). Our results suggest that a viable seed bank also gives *M. vimineum* a buffer for density-dependent mortality.

Recruitment success is an essential, if not the essential, stage in a plant's life history (Grubb 1977, Goldberg et al. 2001, Howard and Goldberg 2001, Albrecht and McCarthy 2009). Yet the survival, growth and reproduction of established seedlings are obviously also necessary stages for population persistence (Harper 1977, Sakai et al. 2001). Given that our results suggest propagule pressure as the critical component in *M. vimineum* recruitment, we need to consider the subsequent propagule pressure generated by *M. vimineum* recruits. We find, as have others (Cheplick 2010, Warren et al. 2011b), that *M. vimineum* vegetative growth and reproduction (by seed number or biomass) are strongly correlated (Fig. 3). We also find that, regardless of the degree of habitat resistance to establishment, *M. vimineum* populations compensate for fewer plants with greater per capita seed production so that seed output is high regardless of population density (Fig. 4). For example, even when only 2–3 *M. vimineum* seedlings germinate, subsequent adult plant reproduction from these few plants always is at least 45 seeds per plant, and averages approx. 550 seeds, though it varies greatly across patches because of the density-dependent reproduction. As we demonstrate in a heterogeneous understory forest habitat (with measured variation in soil moisture, light and temperature), *M. vimineum* can establish approx. 95% of the time with only 10 seeds, and always establishes with >40 seeds. These results suggest that low seed input and poor habitat quality in terms of establishment increase the potential for stochastic colonization failure, but in most cases, *M. vimineum* mesic forest invasions may proceed from little seed input even where there is pronounced establishment limitation. Nord et al. (2009) did not find density-dependent reproduction when investigating the early life stages of *M. vimineum*, but also examined substantially larger 'patches' of plants. By limiting our experimental plots to ~80 cm², we observe density-dependent effects from saturation that may take many years to detect in larger plots. As such, our work complements rather than contradicts previous work with the species.

Whereas leaf litter thickness poses a barrier to *M. vimineum* colonization, other limitations (e.g. soil moisture, temperature and light) also limit *M. vimineum* germination, survival and reproduction (Warren et al. 2011a, b). We intentionally established study plots across ambient gradients to take advantage of environmental heterogeneity in forest understory habitats. We evaluate the influence of ambient abiotic gradients at 1-m² scales, and we find that *M. vimineum* germination and survival are highest in the sunnier plots, and reproduction is highest in the wetter, sunnier plots. These resource gradients are well known to influence *M. vimineum* populations (Warren et al. 2011a, b), and they correlate across forest edges with leaf litter thickness (Lang and Orndorff 1982, Boerner 1984, Facelli and Pickett

1991). Still, our sites generally contained soil moisture and light levels conducive for *M. vimineum* recruitment and reproduction (Warren et al. 2011a, b), and the plants may fail to overcome litter barriers when also confronted with harsher conditions. In addition, by measuring abiotic conditions at the plot rather than individual plant scale, we have low power to detect environmental influences. As such, future research should look beyond interactions between seed and establishment limitation to include multiple, interacting habitat limitations. Such limitations may include disease, herbivory and interspecific competition (Bradford et al. 2010, Kleczewski and Flory 2010), although these are presumably lesser influences on invasive species (Keane and Crawley 2002, Agrawal et al. 2005, Liu and Stiling 2006).

Microstegium vimineum's potential to form a 1–3 year seed bank (Barden 1987, Gibson et al. 2002, Judge 2008) means that failed germination may indicate dormancy or mortality. Whereas seed dormancy is an important component of population dynamics, it may have little impact on the results presented here. This is because seed dormancy is more integral for overcoming temporal than spatial variation in habitat suitability (or density dependent mortality); viable dormant seeds would likely only lessen the importance of leaf litter as a recruitment barrier. Furthermore, successful seed dormancy depends on seed quality (Evans and Etherington 1990, Gallagher and Fuerst 2006, Poorter et al. 2008), and seed biomass is a good indicator of seed fitness (Liu et al. 2000, Gallagher and Fuerst 2006, Padilla et al. 2007). We find that *M. vimineum* seed mass cm⁻² correlates linearly with seed number cm⁻², making our use of seed number for assessing reproduction a suitable proxy for seed quality and dormancy potential. A second potential indicator of habitat suitability is the ratio of cleistogamous (self-pollinated) to chasmogamous (outcrossed) seeds, with the proportion of chasmogamous seeds increasing with increased resources (Porras and Munoz 2000, Cheplick 2007). We find, however, no significant variation in chasmogamous seeds in response to the experimental treatments. We acknowledge that direct tests of seed viability would be preferred and suggest doing so with ungerminated seeds in future work.

Conclusions

Our results suggest that *M. vimineum* recruitment is a function – although not equally – of both seed and establishment limitation. These results, for a widespread invasive plant, are consistent with the emerging perspective for native plants that seed and establishment limitation jointly influence plant recruitment. Notably, *M. vimineum* has the ability to compensate at low population densities with high per capita seed production, which then overrides establishment limitation. This ability makes its invasive potential daunting, and further work is required to test if this is a common mechanism underlying plant invasions.

Acknowledgements – This research was supported by National Science Foundation grants DEB-0823293 and DEB-0218001 to the Coweeta LTER Program. We thank staff and administrators of

the Coweeta Hydrological Laboratory for access to the properties and for logistical support.

References

- Agrawal, A. A. et al. 2005. Enemy release? An experiment with congeneric plant pairs and diverse above- and below-ground enemies. – *Ecology* 86: 2979–2989.
- Akaike, H. 1973. Information theory as an extension of the maximum likelihood principle. – In: Petrov, B. N. and Csaki, F. (eds), 2nd Int. Symp. on Information Theory. Akademiai Kiado, pp. 267–281.
- Albrecht, M. A. and McCarthy, B. C. 2009. Seedling establishment shapes the distribution of shade-adapted forest herbs across a topographical moisture gradient. – *J. Ecol.* 97: 1037–1049.
- Barden, L. S. 1987. Invasion of *Microstegium vimineum* (Poaceae), an exotic, annual, shade-tolerant, C-4 grass, into a North Carolina floodplain. – *Am. Midl. Nat.* 118: 40–45.
- Bates, D. M. and Watts, D. G. 1988. Nonlinear regression analysis and its applications. – Wiley.
- Bates, D. and Maechler, M. 2009. Linear mixed-effects models using S4 classes. – R package ver. 0.999375-31.
- Boerner, R. E. J. 1984. Nutrient fluxes in litterfall and decomposition in four forests along a gradient of soil nutrient availability within a southern Ohio watershed. – *Can. J. For. Res.* 14: 794–802.
- Bolker, B. M. et al. 2009. Generalized linear mixed models: a practical guide for ecology and evolution. – *Trends Ecol. Evol.* 24: 127–135.
- Boyd, N. and Van Acker, R. 2004. Seed germination of common weed species as affected by oxygen concentration, light and osmotic potential. – *Weed Sci.* 52: 589–596.
- Bradford, M. A. et al. 2010. Native, insect herbivore communities derive a significant proportion of their carbon from a widespread invader of forest understories. – *Biol. Invas.* 12: 721–724.
- Burnham, K. P. and Anderson, D. R. 2002. Model selection and multimodel inference: a practical information-theoretic approach. – Springer.
- Cheplick, G. P. 2005. Biomass partitioning and reproductive allocation in the invasive, cleistogamous grass *Microstegium vimineum*: influence of the light environment. – *J. Torrey Bot. Soc.* 132: 214–224.
- Cheplick, G. P. 2006. A modular approach to biomass allocation in an invasive annual (*Microstegium vimineum*; Poaceae). – *Am. J. Bot.* 93: 539–545.
- Cheplick, G. P. 2007. Plasticity of chasmogamous and cleistogamous reproductive allocation in grasses. – *Aliso* 23: 286–294.
- Cheplick, G. P. 2010. Limits to local spatial spread in a highly invasive annual grass (*Microstegium vimineum*). – *Biol. Invas.* 12: 1759–1771.
- Christen, D. C. and Matlack, G. R. 2009. The habitat and conduit functions of roads in the spread of three invasive plant species. – *Biol. Invas.* 11: 453–465.
- Clark, C. J. et al. 2007. Are plant populations seed limited? A critique and meta-analysis of seed addition experiments. – *Am. Nat.* 170: 128–142.
- Clay, K. and Shaw, R. 1981. An experimental demonstration of density-dependent reproduction in a natural population of *Diamorpha smallii*, a rare annual. – *Oecologia* 51: 1–6.
- Colautti, R. I. et al. 2006. Propagule pressure: a null model for biological invasions. – *Biol. Invas.* 8: 1023–1037.
- Cole, P. G. and Weltzin, J. F. 2004. Environmental correlates of the distribution and abundance of *Microstegium vimineum*, in east Tennessee. – *Southeastern Nat.* 3: 545–562.
- Cole, P. G. and Weltzin, J. F. 2005. Light limitation creates patchy distribution of an invasive grass in eastern deciduous forests. – *Biol. Invas.* 7: 477–488.
- Crawley, M. J. 2000. Seed predators and plant population dynamics. – In: Fenner, M. (ed.), *Seeds: the ecology of regeneration in plant communities*. CABI, pp. 167–182.
- Duncan, R. P. et al. 2009. Safe sites, seed supply, and the recruitment function in plant populations. – *Ecology* 90: 2129–2138.
- Eriksson, O. and Ehrlén, J. 1992. Seed and microsite limitation of recruitment in plant-populations. – *Oecologia* 91: 360–364.
- Eschtruth, A. K. and Battles, J. J. 2009. Assessing the relative importance of disturbance, herbivory, diversity and propagule pressure in exotic plant invasion. – *Ecol. Monogr.* 79: 265–280.
- Evans, C. E. and Etherington, J. R. 1990. The effect of soil-water potential on seed-germination of some British plants. – *New Phytol.* 115: 539–548.
- Facelli, J. M. and Pickett, S. T. A. 1991. Light interception and effects on an old-field plant community. – *Ecology* 72: 1024–1031.
- Fairbrothers, D. E. and Gray, J. R. 1972. *Microstegium vimineum* (Trin) A. Camus (Gramineae) in the United States. – *Bull. Torrey Bot. Club* 99: 97–100.
- Fenner, M. and Kitajima, K. 1999. Seed and seedling ecology. – In: Pugnaire, F. I. and Valladares, F. (eds), *Handbook of functional plant ecology*. Marcel-Dekker, pp. 589–611.
- Flory, S. L. and Clay, K. 2009a. Invasive plant removal method determines native plant community responses. – *J. Appl. Ecol.* 46: 434–442.
- Flory, S. L. and Clay, K. 2009b. Non-native grass invasion alters native plant composition in experimental communities. – *Biol. Invas.* 12: 1285–1294.
- Gallagher, R. S. and Fuerst, E. P. 2006. The ecophysiological basis of weed seed longevity in the soil. – In: Basra, A. (ed.), *Handbook of seed science and technology*. Haworth Press, pp. 521–557.
- Gibson, D. J. et al. 2002. Life history of *Microstegium vimineum* (Poaceae), an invasive grass in southern Illinois. – *J. Torrey Bot. Soc.* 129: 207–219.
- Glasgow, L. S. and Matlack, G. R. 2007. The effects of prescribed burning and canopy openness on establishment of two non-native plant species in a deciduous forest, southeast Ohio, USA. – *For. Ecol. Manage.* 238: 319–329.
- Goldberg, D. E. et al. 2001. Density dependence in an annual plant community: variation among life history stages. – *Ecol. Monogr.* 71: 423–446.
- Grubb, P. J. 1977. Maintenance of species-richness in plant communities – importance of regeneration niche. – *Biol. Rev.* 52: 107–145.
- Harper, J. L. 1977. *Population biology of plants*. – Academic Press.
- Houseman, G. R. and Gross, K. L. 2006. Does ecological filtering across a productivity gradient explain variation in species pool-richness relationships? – *Oikos* 115: 148–154.
- Howard, T. G. and Goldberg, D. E. 2001. Competitive response hierarchies for germination, growth and survival and their influence on abundance. – *Ecology* 82: 979–990.
- Hubbell, S. P. 2001. *The unified neutral theory of biodiversity and biogeography*. – Princeton Univ. Press.
- Huebner, C. D. 2010a. Establishment of an invasive grass in closed-canopy deciduous forests across local and regional environmental gradients. – *Biol. Invas.* 12: 2069–2080.
- Huebner, C. D. 2010b. Spread of an invasive grass in closed-canopy deciduous forests across local and regional environmental gradients. – *Biol. Invas.* 12: 2081–2089.
- Judge, C. A. 2008. Japanese stiltgrass (*Microstegium vimineum*) management for restoration of native plant communities. – *Invasive Plant Sci. Manage.* 1: 111–119.
- Keane, R. M. and Crawley, M. J. 2002. Exotic plant invasions and the enemy release hypothesis. – *Trends Ecol. Evol.* 17: 164–170.

- Klanderud, K. 2010. Species recruitment in alpine plant communities: the role of species interactions and productivity. – *J. Ecol.* 98: 1128–1133.
- Kleczewski, N. M. and Flory, S. L. 2010. Leaf blight disease on the invasive grass *Microstegium vimineum* caused by a *Bipolaris* sp. – *Plant Disease* 94: 807–811.
- Knight, T. M. et al. 2009. Deer facilitate invasive plant success in a Pennsylvania forest understory. – *Nat. Areas J.* 29: 110–116.
- Lang, G. E. and Orndorff, K. A. 1982. Surface litter, soil organic matter, and the chemistry of mineral soils and foliar tissue: landscape patterns in forests located on mountainous terrain in West Virginia. – In: Muller, R. N. (ed.), *Proc. 4th Central Hardwood Forest Conference*. – Univ. of Kentucky.
- Liu, H. and Stiling, P. 2006. Testing the enemy release hypothesis: a review and meta-analysis. – *Biol. Invas.* 8: 1535–1545.
- Liu, Z. M. et al. 2000. A comparative study of morphological responses of seedling roots to drying soil in 20 species from different habitats. – *Acta Bot. Sin.* 42: 628–635.
- Lockwood, J. L. et al. 2005. The role of propagule pressure in explaining species invasions. – *Trends Ecol. Evol.* 20: 223–228.
- Lonsdale, W. M. 1999. Global patterns of plant invasions and the concept of invasibility. – *Ecology* 80: 1522–1536.
- Marshall, J. M. and Buckley, D. S. 2008. Influence of litter removal and mineral soil disturbance on the spread of an invasive grass in a Central Hardwood forest. – *Biol. Invas.* 10: 531–538.
- Miller, N. P. and Matlack, G. R. 2010. Population expansion in an invasive grass, *Microstegium vimineum*: a test of the channelled diffusion model. – *Divers. Distrib.* 16: 816–826.
- Moore, K. A. 2009. Fluctuating patch boundaries in a native annual forb: the roles of niche and dispersal limitation. – *Ecology* 90: 378–387.
- Moore, K. A. and Elmendorf, S. C. 2006. Propagule vs niche limitation: untangling the mechanisms behind plant species' distributions. – *Ecol. Lett.* 9: 797–804.
- Myers, J. A. and Harms, K. E. 2009. Seed arrival, ecological filters, and plant species richness: a meta-analysis. – *Ecol. Lett.* 12: 1250–1260.
- Nord, A. N. et al. 2009. Environmental factors influence early population growth of Japanese stiltgrass (*Microstegium vimineum*). – *Invas. Plant Sci. Manage.* 3: 17–25.
- Oswalt, C. M. and Oswalt, S. N. 2007. Winter litter disturbance facilitates the spread of the nonnative invasive grass *Microstegium vimineum* (Trin.) A. Camus. – *For. Ecol. Manage.* 249: 199–203.
- Oswalt, C. M. et al. 2007. Effects of *Microstegium Vimineum* (Trin.) A. Camus on native woody species density and diversity in a productive mixed-hardwood forest in Tennessee. – *For. Ecol. Manage.* 242: 727–732.
- Padilla, F. M. et al. 2007. Early root growth plasticity in seedlings of three Mediterranean woody species. – *Plant Soil* 296: 103–113.
- Poorter, L. et al. 2008. Are functional traits good predictors of demographic rates? Evidence from five Neotropical forests. – *Ecology* 89: 1908–1920.
- Porras, R. and Munoz, J. M. 2000. Cleistogamy in *Centaurea melitensis* (Asteraceae): capitulum variability and spatio-temporal development patterns. – *Plant Syst. Evol.* 223: 251–262.
- Poulsen, J. R. et al. 2007. Plants as reef fish: fitting the functional form of seedling recruitment. – *Am. Nat.* 170: 167–183.
- Pulliam, H. R. 2000. On the relationship between niche and distribution. – *Ecol. Lett.* 3: 349–361.
- Rauschert, E. S. J. et al. 2010. Slow spread of the aggressive invader, *Microstegium vimineum* (Japanese stiltgrass). – *Biol. Invas.* 12: 563–579.
- Redman, D. E. 1995. Distribution and habitat types for Nepal *Microstegium* [*Microstegium vimineum* (Trin.) Camus] in Maryland and the District of Columbia. – *Castanea* 60: 270–275.
- Sakai, A. K. et al. 2001. The population biology of invasive species. – *Annu. Rev. Ecol. Syst.* 32: 305–332.
- Schmitt, J. et al. 1987. Density-dependent flowering phenology, outcrossing, and reproduction in *Impatiens capensis*. – *Oecologia* 72: 341–347.
- Schramm, J. W. and Ehrenfeld, J. G. 2010. Leaf litter and understory canopy shade limit the establishment, growth and reproduction of *Microstegium vimineum*. – *Biol. Invas.* doi: 10.1007/s10530-010-9711-1.
- Smith, B. H. 1983. Demography of *Floerka proserpinacoides*, a forest-floor annual: II density-dependent reproduction. – *J. Ecol.* 71: 405–412.
- Strogatz, S. H. 2001. Nonlinear dynamics and chaos: with applications to physics, biology, chemistry and engineering. – Westview Press.
- Tanentzap, A. J. and Bazely, D. R. 2009. Propagule pressure and resource availability determine plant community invasibility in a temperate forest understory. – *Oikos* 118: 300–308.
- Tilman, D. 2004. Niche tradeoffs, neutrality, and community structure: a stochastic theory of resource competition, invasion and community assembly. – *Proc. Natl Acad. Sci. USA* 101: 10854–10861.
- Turnbull, L. A. et al. 2000. Are plant populations seed-limited? A review of seed sowing experiments. – *Oikos* 88: 225–238.
- USDA, NRCS 2010. The PLANTS Database. – NRCS, National Plant Data Center.
- Von Holle, B. and Simberloff, D. 2005. Ecological resistance to biological invasion overwhelmed by propagule pressure. – *Ecology* 86: 3212–3218.
- Warren, R. J. and Bradford, M. A. 2011. The shape of things to come: woodland herb niche contraction begins during recruitment in mesic forest microhabitat. – *Proc. R. Soc. B* 278: 1390–1398.
- Warren, R. J. et al. 2011a. Performance and reproduction of an exotic invader across temperate forest gradients. – *Ecosphere* 2: 1–19.
- Warren, R. J. et al. 2011b. The putative niche requirements and landscape dynamics of *Microstegium vimineum* – an invasive Asian grass. – *Biol. Invas.* 13: 471–483.
- Xiong, S. J. and Nilsson, C. 1999. The effects of plant litter on vegetation: a meta-analysis. – *J. Ecol.* 87: 984–994.