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ORIGINAL PAPER

Spatial variability of an invasive earthworm (*Amynthas agrestis*) population and potential impacts on soil characteristics and millipedes in the Great Smoky Mountains National Park, USA

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Abstract European and Asian earthworm invasions are widespread in North America. European earthworms especially are well-known to cause dramatic changes in ecosystems in northern, formerly glaciated portions of the continent, but less is known about the impacts of earthworm invasions in unglaciated areas inhabited by indigenous earthworms. We monitored fluctuations in the spatial extent of an *Amynthas agrestis* (Megascolecidae) population in the Great Smoky Mountains National Park in eastern Tennessee, USA. Two years of monthly growing-season observations revealed that the distribution of the earthworm population was dynamic, but overall distribution was closely linked to temperature and moisture with dramatic reductions of earthworm numbers associated with very dry conditions. In plots where *A. agrestis* were more often detected, we measured increased A-horizon soil aggregation and decreased thickness of the Oe/Oa-horizon. However, *A. agrestis* was not related to A-horizon microbial biomass, A-horizon C:N, Oi-horizon (litter) thickness, or mass of forest floor (O-horizon). Reductions in millipede species richness and density were associated with frequency of *A. agrestis* presence, possibly due to direct competition for food resources (Oe/Oa material). This evidence for potentially negative interactions between millipedes and *A. agrestis* suggests that expansion of the non-native earthworm into new habitats in the Park may alter soil physical properties and could pose a threat to native millipede diversity.

Keywords Earthworm · *Amynthas* · Invasive species · Millipede · Soil aggregation

Introduction

Invasive earthworms are a global problem and can cause considerable changes to ecosystems (Bohlen et al. 2004a, b; Hendrix 2006; Hendrix et al. 2008). In North America, the vast majority of what is known about earthworm invasions comes from studies conducted in previously glaciated regions where no native earthworms are present and where European earthworms (family Lumbricidae) have been widely

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introduced. Invasion by non-lumbricid earthworms is a more recently documented phenomenon (Burtelow et al. 1998; Callaham et al. 2003; Steinberg et al. 1997) and the ecological consequences of these invasions are poorly understood.

We studied a population of one such non-lumbricid earthworm in the unglaciated southeastern region of North America, where native Nearctic earthworms occur. This megascolecid earthworm was *Amynthas agrestis* (Goto and Hatai 1899), a native of east-central Asia. Several species of *Amynthas* were documented in the central (Illinois) and southeastern (Mississippi) US by the 1890's (Garman 1888; Gates 1937; National Museum of Natural History 2008). Approximately 15 species have established and spread widely across the eastern US (Reynolds and Wetzel 2004). *Amynthas agrestis* has been known in the US since 1939 (Gates 1982) and was documented in the study region in the 1970's (Reynolds 1978). Burtelow et al. (1998) stated that "little is known about *Amynthas* beyond the physical zoological description." Although some basic biological information has come to light recently, including temperature and moisture tolerances for certain species (Fragoso et al. 1999; Richardson et al. 2009), this assessment remains generally accurate. In fact, even the physical taxonomic description leaves much to be desired: each *Amynthas* species has several parthenogenetic morphs in which the male sexual system is eliminated or reduced to varying degrees (Gates 1956) and previous taxonomic works have based identification in part on male characters (e.g. Reynolds 1978). Thus far, only the parthenogenetic "R" morph of *A. agrestis* is known from North America (Gates 1958). Studies on the introduction and invasion biology of *Amynthas* spp. are rare, but occurrences of *A. agrestis* have been documented in the southern Appalachian Mountains (Callaham et al. 2003; Reynolds 1978). It is suspected that most *Amynthas* species, like many other earthworms, were transported by humans in soil of potted plants and continue to be dispersed with horticultural plants, through earthmoving activities, and by anglers using *Amynthas* as fishing bait (Callaham et al. 2003; Gates 1958). Once introduced, *Amynthas* spp. can have significant impacts on soil structure and processes. For example, in forests in New York, USA, *A. gracilis* increased soil N-mineralization and nitrification, reduced forest floor organic matter content, and increased microbial biomass (Burtelow et al. 1998; Steinberg et al. 1997).

Because there have been few field-based observations on *Amynthas agrestis*, we sought to use a population in Great Smoky Mountains National Park (GSMNP) to observe the expansion of the species into new habitat. We hypothesized that this population of *A. agrestis* would advance unidirectionally and at a steady pace. We expected that the non-native earthworms would reduce the thickness and mass of organic horizons (both Oi and Oe/Oa) through direct consumption, and increase soil aggregation in the A-horizon via casting. We also hypothesized that *A. agrestis* would increase soil microbial biomass and increase the proportion of total soil C contained in the A-horizon relative to soils without *A. agrestis*. Finally, we hypothesized that because of the expected effects on litter and microbial biomass, the presence of the non-native earthworm would also decrease the abundance and species richness of the native millipede community and decrease the abundance and species richness of the earthworm community through competitive interactions.

Methods

Site description and experimental design

GSMNP encompasses most of the highest peaks of the southern Appalachian Mountains, and spans the border between the states of Tennessee and North Carolina, USA. The west end of the Park is bordered by US Highway 129; for 3.8 km this road is flanked on the east by GSMNP and on the west by the Chilhowee Reservoir, a dammed portion of the Little Tennessee River. The Chilhowee Dam began operation in 1957 and the dam project relocated ~5 km of US Highway 129 to its present location. Recreational opportunities, including fishing with live bait, are permitted on this reservoir. Recreational fishing is also allowed in the Park, but live bait is not permitted.

Local topography drains to the reservoir and consists of alternating valleys and ridges with tall (70–120 m) bluffs that abut the road. During initial surveys, invasive earthworms (specifically *Amynthas* sp.) were found in every suitable habitat (i.e. areas that were not vertical rock faces) along this stretch of road. Population boundaries for *A. agrestis* were located and delineated in several small watersheds to find a suitable location to set up sampling plots. We delineated *A. agrestis* population boundaries during spring and summer of 2005 by searching through leaf litter and at the soil surface for *Amynthas* individuals, which were recognizable due to their thrashing defense behavior. Hand searching proceeded from the roadside into the forest, 5–10 m at a time, until no *Amynthas* could be found. The search then proceeded in 1 m increments from the last known *Amynthas* location for ~5 m. When a location was reached without *Amynthas*, it was electroshocked (see below) for 3 min to determine that no *Amynthas* individuals had burrowed into the soil. Several earthworms were taken during delineation and all were determined to be *Amynthas agrestis*.

Valley vegetation was dominated by *Acer* spp., *Quercus* spp., *Liquidambar styraciflua*, and *Liriodendron tulipifera*, while the more xeric ridges were dominated by white pine (*Pinus strobus*). In general, the area was classified as mesic-xeric oak-dominated climax forest (Whittaker 1956). Ridge soils were a complex of moderately deep Junaluska and deep Brasstown series soils classified as fine-loamy, mixed, subactive, mesic Typic Hapludults. Valley soils were a complex of shallow Cataska series and moderately deep Sylco series soils, which are loamy-skeltal, mixed, active (Sylco) or semiactive (Cataska), mesic Typic Dystrudepts (USDA 2007).

We selected two adjacent valleys with distinct earthworm population boundaries; five parallel transects were placed across the boundary at randomly chosen locations. Each transect consisted of nine 3 × 3 m plots placed 3 m apart (Fig. 1), centered on the boundary. In 2007 we anticipated the need for additional plots beyond the initial boundary location in the western valley and therefore three additional plots were placed on these three transects (Fig. 1b). Due to limitations in the number of leaf litter samples that could be processed we concurrently discontinued sampling the three sets of plots furthest behind the initial population boundary on these three transects.

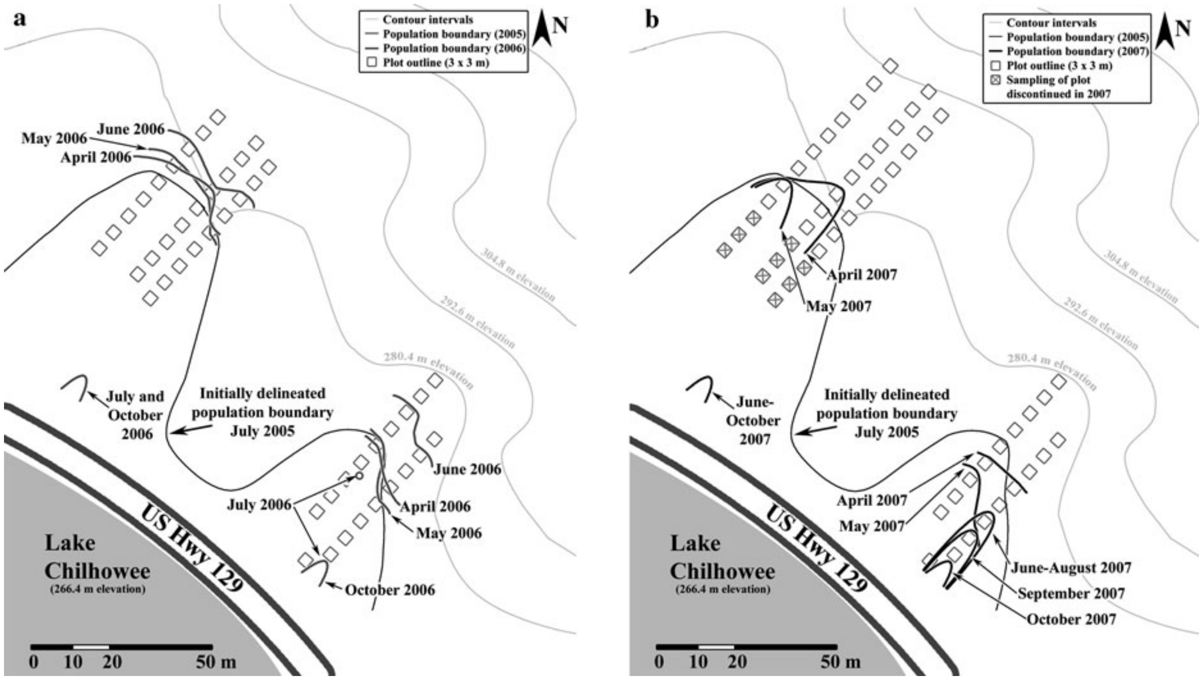

Fig. 1 Map of study site and changes in population boundary of *Amynthas agrestis*, as determined by a combination of bimonthly electroshocking within plots and monthly leaf-litter searches between plots in 2006 (**a**) and 2007 (**b**). No earthworms were found during August and September 2006 due to equipment failure and low soil moisture levels. Plot locations are based on field measurements and GPS locations, contour intervals based on the Calderwood Quadrangle USGS 7.5 min series topographic map

Soil sampling

Soil cores (6 cm diameter, 15 cm depth) and ~100 g additional A-horizon soil were collected from each plot at the end of the study (October 2007). Upon return to the lab, the O-horizon was separated into Oi- and Oe/Oa-horizons. The thickness and air-dry mass of these horizons were measured. Samples of air-dry A-horizon soil were analyzed for C and N content on a Carlo Erba NA1500 CHN Combustion Analyzer (Carlo Erba, Milan, Italy). To characterize the distribution of water-stable aggregate size classes in each plot, ~50 g of air-dry soil was wet-sieved (Six et al. 2000). Rocks larger than 2 mm were removed by hand before weighing the sample. Soil aggregate fractions were categorized by size-class into POM (floatable particulate organic material), >2,000 μm, 2,000–250 μm, and 250–53 μm. The <53 μm fraction contained very little material and was excluded from the analysis. Microbial biomass was measured by chloroform fumigation-extraction (Vance et al. 1987) using a K_{ec} of 0.38.

Sampling of fauna

We monitored the population boundary monthly by extensive hand searching near the plots and near the last known location of the boundary. We sampled earthworms and millipedes inside the plots bimonthly from April to October in 2006 and 2007. No sampling occurred during winter months as cold weather limited soil fauna activity. Furthermore, *A. agrestis* appears to have an annual life cycle (Callaham et al. 2003; Reynolds 1978) and is thought to overwinter in the cocoon life stage. Data from earthworm electroshocking were compiled to assess the extent of the area occupied by *A. agrestis* in sampling months.

Earthworms were sampled via the octet electroshocking technique (Schmidt 2001) at a randomly chosen location within each of the 3 × 3 m plots. The electroshocking method was used because it has been demonstrated to have high extraction efficiency and it requires minimal soil disturbance (Weyers et al. 2008); chemical extraction methods were not feasible because they could have altered soil moisture, and in turn affected earthworm movement. Through June 2006 we used a commercially acquired electroshocker (DEKA 4000 W, DEKA Gerätebau, Marsberg, Germany) at 300, 350, 400, 500, and 600 V for 2 min each. This machine failed during July 2006 and as a result we were unable to collect earthworms during August and October 2006. During 2007 we used a similar instrument for electoshocking collections (Weyers et al. 2008), and field observations indicated that this instrument produced a comparable sample. This second octet device was not automated and so opposing pairs of probes (four probes total) around the octet were activated manually at 300 V for 2.5 min each, then at maximum voltage (350–450 V) for 2.5 min each. All earthworms responding to electroshocking were preserved in 5% formalin and identified to the lowest taxonomic level possible.

Millipede abundance was measured by sampling with the leaf litter collection/Berlese extraction technique (Snyder et al. 2006). Litter was sampled at a randomly chosen location in each plot by placing a 30 cm diameter frame on the ground and collecting the organic soil horizons within the frame. This litter was placed into a canvas bag and kept cool until it was returned to the lab. Litter was transferred onto a Berlese funnel, where it was extracted for 72 h into 70% ethanol. Millipede species richness was estimated for extracted material in combination with a 0.5 person-hour timed hand collection (Snyder et al. 2006). For each sampling date, all earthworm and millipede sampling was completed within a 24 h period.

Environmental data collection

Soil environmental conditions were monitored on-site using two HOBO data loggers each equipped with two temperature and two dielectric soil moisture sensors, one set placed at the interface of the O- and A-horizons (mineral soil surface) and one set at 10 cm depth in the mineral soil. One data logger was placed in the western valley at 269 m elevation and one was located on the ridge top at 284 m elevation. Elevations of the data loggers and the center of each plot were measured with rod and transept from a nearby National Geodetic Survey benchmark. Additional precipitation and air temperature data were obtained from a National Park Service air quality monitoring station at Cades Cove, 16 km northeast of the study site, 561 m elevation.

Statistical analysis

The boundaries of the area occupied by *A. agrestis* fluctuated dramatically over the course of the study at the spatial scale of our transects. Therefore, it was necessary to base our analyses on what was known for certain about *A. agrestis* in each plot, i.e., a count of how often each plot was occupied by *A. agrestis* during monitoring. We considered this to be the best available means of evaluating the net effects of *A. agrestis* on various soil and biological properties. For example, if a plot fell within the boundary of the *A. agrestis* population at the time of a monitoring event, then this was scored as 1; plots outside the population boundary received a score of 0. These presence/absence scores were then summed across all eleven monitoring dates. In this way, every plot was assigned a score indicating the total number of times *A. agrestis* was detected over the course of the study. Because no earthworms were found at one monitoring event, these values ranged from 0 (never occupied by *A. agrestis*) to 10 (almost always occupied by *A. agrestis*).

The relationship between elevation and millipede species richness was investigated with Spearman correlation. Relationships between *A. agrestis* scores and soil properties, millipede and earthworm species richness and abundance, were tested with linear regression analysis (regressing total presence/absence values against soil aggregate distributions, litter depth, millipede richness, etc.). Soil environmental data, logged every 15 min, and air temperature, logged every hour, were averaged for the month preceding each date that monitoring took place. These averages were individually regressed against the net change in position (forward or backward from the original July 2005 position) of the *A. agrestis* population boundary at each transect for all 10 monitoring events (the initial delineation was used to calculate net change in position and therefore excluded from this analysis). Data analysis was performed with SAS (Version 9.1, SAS Institute, Cary, NC, USA).

Results

Population boundary fluctuation

The spatial extent of the *Amynthas agrestis* population was far more variable on a monthly basis than expected (Fig. 1). The boundary generally moved into the forest during early 2006. Using data from July 2005 and June 2006, we estimated that the boundary moved at a rate of 12 ± 1.9 (SE) m year^{-1} for this period. During late 2006 and all of 2007, the population receded to lower elevations (toward the road, Fig. 1b). The severe drought which impacted the region in late 2006 and 2007 (Table 1) may have contributed to a decline in earthworm activity and limited the ability of *A. agrestis* to advance further into the forest or even to maintain the initially observed position (Fig. 1a, July 2005 position). The fluctuations in position of the population boundary were significantly related to weather conditions (Table 2).

Due to equipment failure no earthworms were sampled during August and October 2006, but searches of the leaf litter in July and October 2006 revealed earthworms only in the portions of the site which had the lowest elevation and thus had highest surface soil moisture (Fig. 1a); during August and September no earthworms were found. *Amynthas agrestis* could also be found only in these wettest locations during July–October 2007 even via

Table 1 Observed temperature and precipitation at Cades Cove weather station (*) compared to long-term Gatlinburg climate averages (NOAA data, 1971–2000) and net change in population boundary position

		April	May	June	July	Aug	Sept	Oct
Mean air temperature (°C), month preceding sampling*	2006	11.9	15.5	20.5				
	2007	11.8	16.1	19.4	21.7	22.3	22.6	18.3
Long-term average air temperature (°C)		12.4	16.8	20.9	22.9	22.2	19.1	13.2
Precipitation, month preceding sampling (mm)*	2006	190.4	91.6	59.7				
	2007	81.3	127.7	46.7	90.3	98.2	63.3	6.6
Long-term average precipitation (mm)		110.7	141.7	147.8	154.2	116.6	117.3	75.2
Mean net change in position of population boundary (m)	2006	2.4	2.1	10.8				
	2007	−4.2	−15	−22.8	−22.8	−22.8	−24	−27

Table 2 Results of linear regressions of temperature and moisture data against net change in position of population boundary (relative to the position documented in 2005, $n = 10$ dates)

Independent variable	R^2	P
Monthly precipitation	0.1302	0.0100
Air temperature	0.2229	0.0005
Ridge soil surface temperature	0.2788	0.0002
Ridge soil surface moisture	0.1955	0.0043
Valley soil surface temperature	0.4409	<0.0001
Valley soil surface moisture	0.1863	0.0172

electroshocking, which has high extraction efficiency (Weyers et al. 2008). During June and August 2007 *A. agrestis* was collected in only one plot and October 2007 sampling produced no earthworm specimens. However, the position of the population boundary was determined in part by searches outside of the plots. Thus, although the western transects did not extend to the lowest reaches of this valley bottom, *Amynthas* individuals were visually confirmed in this location during these hot and dry periods (Fig 1a and b).

Soils

The number of times *Amynthas* was scored in plots was related to changes in soil structure, but not chemical or biological characteristics. Soil microbial biomass (1,442 ± 114 SE µg C g dry soil^{-1}) and soil C:N (16.68 ± 0.48 SE) were not related to *Amynthas* presence. Plots with higher scores showed increases in A-horizon soil aggregation (Fig. 2) and reductions in the thickness of some organic horizons (Fig. 3). Plots with higher *Amynthas* detection scores had a higher percentage of water-stable soil aggregates in the >2,000 µm size class ($P = 0.0035$), with a corresponding decrease of 250–253 µm aggregates ($P = 0.0011$, Fig. 2). No change was observed in particulate organic matter ($P = 0.0795$) or 2,000–250 µm soil aggregates ($P = 0.0836$). The O-horizon was divided into Oi (recently fallen litter) and Oe/Oa (partially decomposed litter) horizons. Oi-horizon thickness was not significantly related to *A. agrestis* presence ($P = 0.0785$). Higher *A. agrestis* scores were related to decreases in the thickness of the Oe/Oa-horizon ($P = 0.0010$), this relationship was not observed for the mass of this layer ($P = 0.1506$).

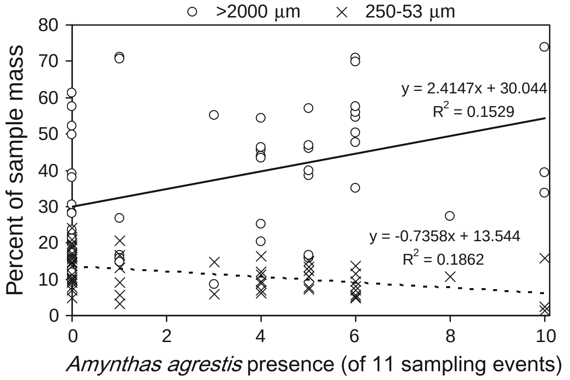

Fig. 2 Regression of percent water-stable aggregates in the >2,000 µm size class (*solid line*, *open circle*) ($P = 0.0035$) and 250–253 µm (*dotted line*, *times*) ($P = 0.0011$) against *Amynthas agrestis* presence/absence score

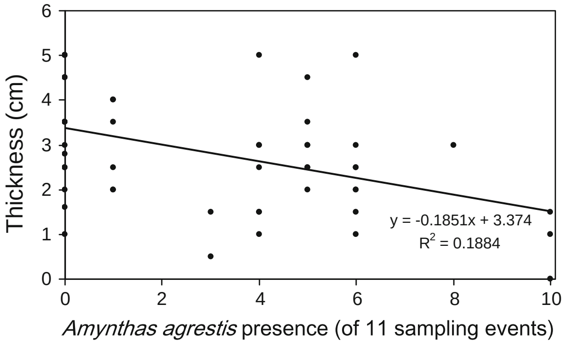

Fig. 3 Regression of F/H horizon thickness (cm) in October 2007 against *Amynthas agrestis* presence/absence score ($P = 0.0010$)

Earthworms

Several other species of earthworms were found at the study site, including native *Diplocardia* spp. and *Bimastos* spp., invasive Asian *Amynthas corticis*, and invasive European Lumbricidae (*Aporrectodea* sp., *Dendrobaena octaedra*, *Eiseniella tetraedra*, *Lumbricus rubellus*, and *Octolasion tyrtaeum*). Although lumbricid earthworms were found throughout the plot array, mean lumbricid abundance was statistically significantly postively related to *A. agrestis* presence ($P = 0.0267$), but the strength of this relationship was not particularly strong ($r^2 = 0.1363$, data not shown). *Amynthas corticis* was found almost exclusively in one moist valley bottom and therefore it was strongly associated with increased *A. agrestis* presence in the statistical analysis ($P = 0.0004$, data not shown). Mean native earthworm abundance was not associated with *A. agrestis* presence ($P = 0.7835$).

Millipedes

Over the course of the study, millipede species richness varied between 1 and 14 species per plot; 29 species were identified from the study site in total. Millipede species richness was significantly negatively related to presence of *A. agrestis* (Fig. 4a, $P < 0.0001$). Millipede abundance was similarly negatively related (Fig. 4b, $P < 0.0001$) to *A. agrestis* presence. Millipede species richness (cumulative for all sampling dates) was not significantly correlated with plot elevation ($P = 0.0709$).

Discussion

Population boundary fluctuations

We expected that the *A. agrestis* population would expand into new habitat unidirectionally and at a steady rate. However, this was not the pattern we observed. Instead, the position of the population boundary fluctuated over the course of the entire study. This variability seems to have been strongly related to weather conditions. Moisture was apparently an important factor in the distribution of these earthworms, but their environmental tolerances are only beginning to be understood in controlled laboratory studies (Richardson et al. 2009). It is not particularly surprising to find that these earthworms responded to changing moisture conditions in the field, in light of what is known about moisture requirements for most soil fauna (Coleman et al. 2004), but it is notable that these observations may have some utility when developing management approaches to control of earthworm invasions. Although few successful invasive earthworm management techniques have been documented (Callaham et al. 2006), the limitation of *A. agrestis* populations during particular conditions has important implications. Management recommendations can in part be based on weather conditions so that control techniques are only applied when the area occupied by *A. agrestis* earthworms is reduced, ultimately reducing the resources (time, personnel, chemicals, or funding) needed for control of invasive species. If populations of *A. agrestis* can be reliably predicted to contract during very dry conditions, it may be advantageous for managers interested in controlling these earthworms to wait for such conditions to occur, and thus limit the area of forest that must be treated (e.g. with chemical or other techniques).

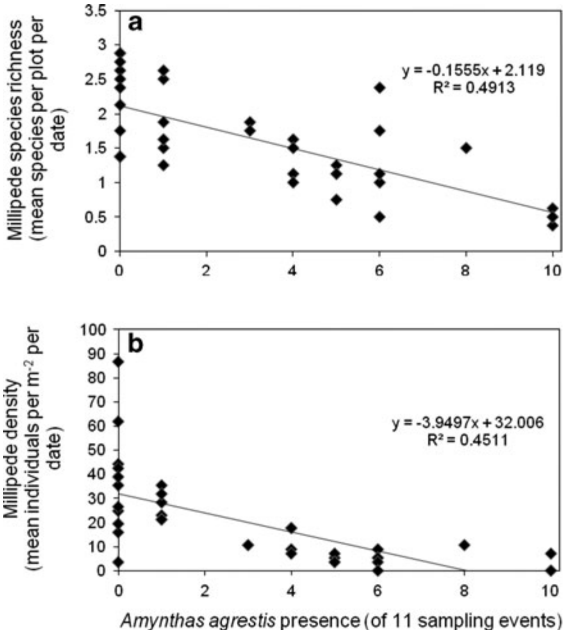

Fig. 4 Regression of mean millipede species richness (**a**) and mean density (**b**) against *Amynthas agrestis* presence/absence score ($P < 0.0001$). Mean density of millipedes across all dates and plots was 21.1 individuals m^{-2}

There are other possible explanations for the dynamic patterns we observed for this population of *A. agrestis*. In addition to weather-related moisture limitations, biotic factors may have contributed to slowing the advance of *A. agrestis* into new habitats at our site. For example, competition with millipedes may represent a form of biotic resistance to the spread of *A. agrestis*. Biotic resistance—a situation in which the organisms native to a particular habitat may be able to repel invading organisms (*sensu* Lake and O'Dowd 1991; Baltz and Moyle 1993)—has been proposed to operate in earthworm communities, but this hypothesis has not been well tested in earthworm invasions (Hendrix et al. 2006; Hendrix et al. 2008). Invasive species also frequently experience predator and parasite release when invading a new habitat (Torchin et al. 2003), but over time, native predators and parasites may also begin to take advantage of the new food or host (Carlsson et al. 2009), and this has been suggested for earthworm predators, such as salamanders (Maerz et al. 2005). Preliminary evidence of internal parasites in invasive earthworms from the study site (B. A. Snyder personal observations), increased carabid beetle activity in the vicinity of *Amynthas* population in 2006 (S. C. Rostkowski unpublished data), and unusually frequent encounters with eastern worm snakes (*Carphophis amoenus*) during sampling at the site (M. A. Callaham personal observations) could suggest that this is occurring.

Clearly, there are a number of unresolved issues pertaining to the fluctuation of the boundaries of the *Amynthas* population as we measured it. In addition to the possible mechanisms acting on the population discussed above, it is possible that *Amynthas* was, in fact, present in soils beyond our detected population boundary, that these were simply in the cocoon form, and that these cocoons would hatch when temperature and moisture requirements were met. Another potentially confounding factor is that, although very modest relative to the surrounding landscape, there were elevation changes along the transects we sampled, and these differences may have influenced the advance of this *A. agrestis* population deeper into the GSMNP. More work, including long-term monitoring of this and other *A. agrestis* populations, will be required to adequately address these issues, and we hope that the results presented here will provide a good base of information for these future studies.

Ecological implications of *Amynthas* invasion: soil characteristics

The dynamic nature of the population boundary observed at our site presented some challenges to interpretation of our results. We suspect that the introduction of *A. agrestis* to this site was fairly recent, but it is also possible that the dynamic behavior of the population may have been ongoing for a long time. Whatever the case, at least during the 2 years we observed this population, we did find relationships between *A. agrestis* presence and variables indicative of effects that might be expected in light of results from previous studies of earthworm invasions. Although it is not possible to use our observations from this field study to demonstrate direct causal relationships between the earthworm and the differences documented in soil characteristics, there is ample evidence (albeit using other earthworm species and in different soil types) from other studies that invasive earthworms can negatively affect leaf litter and forest floor characteristics (McLean and Parkinson 1997; Bohlen et al. 2004a; Hale et al. 2005; Suárez et al. 2006), as well as the type and distribution of soil aggregates (Bossuyt et al. 2006; Haynes et al. 2003).

Ecological implications of *Amynthas* invasion: biotic interactions

During the course of the study, both millipede abundance and species richness were found to be negatively related to *A. agrestis* presence. Like the invasive earthworm, millipedes are primarily epigeic (Hopkin and Read 1992), and this suggests that if both are present in the same soil an interaction is very likely. Again, although our results are not evidence for direct causal relationships, there are examples from other studies which show that earthworms and millipedes may directly compete for food resources (Snyder 2008a; Snyder et al. 2009). If this pattern holds across sites, invasion of *A. agrestis* across larger spatial scales may prove to be a formidable challenge for millipede conservation in the southern Appalachian Mountains where native millipede diversity is high (Snyder 2008b).

The potential for such competitive interactions does not preclude, however, other explanations for the observed distributions of millipedes and *A. agrestis* at our site, such as an environmental or topographic gradient to which both millipedes and *A. agrestis* responded. The initially observed (2005) *A. agrestis* population boundary coincided to a great degree with the topographic variability at the site, but we expect this was primarily due to weather-related factors. Although it is generally expected that millipede density and richness would be greatest in areas with higher moisture (Hopkin and Read 1992), we did not detect any effect of elevation on these measures of the millipede community. In fact, we observed millipede richness and density to be higher in upslope (drier) sites, and we suggest that this may be further evidence of negative interactions with *A. agrestis* in lower-lying (moist) areas of the study site.

Conclusions

The results of this study document correlative relationships between the number of times an invasive earthworm was detected in soils of the southern Appalachian Mountains, and several soil physical and biological characteristics. This study represents the only example (known to us) of a field-based assessment of how *A. agrestis* populations behave, and potentially interact with other organisms in forests of the southeastern USA. These findings may have implications for management of populations of invasive *A. agrestis* earthworms, as well as for conservation of native millipede diversity. However, we caution that this study reports observations from a single population of the invasive earthworm, and thus, it would be inappropriate to extend our interpretations beyond similar habitat types and locations.

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