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Linkage between exotic earthworms, understory vegetation and soil properties in sugar maple forests

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ABSTRACT

The comminuting and soil mixing activities of earthworms can affect soil physical, chemical and biological properties, which in turn can influence plant growth and survival. Accordingly, there is growing concern that the spread of exotic earthworms into northern temperate forests may compromise biodiversity and tree species recruitment. We report on a study where we sampled earthworms, soils, and understory plants in plots established in 40 mature sugar maple stands distributed over 3 areas in the Eastern Townships of Southern Québec (Canada). Earthworms were found in 19 of 40 plots, and earthworm frequency of occurrence (E_{fo}) as well as the complexity of earthworm communities reflected human accessibility to the plots. Plant species richness decreased, and species evenness increased, with E_{fo}. The E_{fo} was related to a decrease in the cover of 5 plant species, and to an increase in the cover of 2 other plant species or plant functional groups. Increasing E_{fo} also correlated with higher soil pH, lower forest floor thickness and lower soil C:N ratio. Among these 3 variables, redundancy analysis (RDA) revealed that soil pH and forest floor thickness correlated with plant community composition. Based on neutral lipid and phospholipid fatty acid profiles, we found that soil bacteria and fungi increased with a decrease in forest floor thickness, bacteria and arbuscular mycorrhizal fungi (AMF) increased with soil pH, whereas actinobacteria and AMF increased with E_{fo}. We discuss the possible mechanisms by which earthworms might directly or indirectly alter understory plant community composition. By considering the location and land use management of each study site, our study provides further evidence that the spread of exotic earthworms in sugar maple stands of Southern Québec may be linked to human activities, with implications for further research and conservation issues.

1. Introduction

In southern Québec (Canada), it has been established that native earthworm species did not survive the Wisconsin glaciation, which ended over 11,000 years ago (Gates, 1970; James, 1995). Accordingly, 17 of the 19 known earthworm species in southern Québec were introduced in recent centuries by European settlers (Addison, 2009). Given that their natural rate of spread is no more than 5–10 m yr^{-1} (Marinissen and van den Bosch, 1992), and given that they are predominantly found in agricultural fields, along roads and near fishing lakes, exotic earthworm dispersal throughout the landscape is thought to mainly be mediated by human activities (Gundale et al., 2005; Keller et al., 2005; Tiunov et al., 2006; Holdsworth et al., 2007a; Cameron and Bayne, 2009; Sackett et al., 2012). In northern temperate forest ecosystems similar to those found in southern Québec, studies have revealed negative effects of exotic earthworms on the recruitment of certain plant species (e.g., Hale et al., 2006; Corio et al., 2009), which has prompted others to reflect on the need for land management policies that could mitigate earthworm dispersal (e.g., Callaham et al., 2006; Hale, 2008). In order for policy makers in southern Québec to engage in such a dialogue, empirical data is required to show a correlation between earthworm abundance, and earthworm-mediated impacts on forest vegetation and soil properties in this region.

The introduction of exotic earthworm species into previously earthworm-free northern temperate forest soils, such as in Minnesota and Wisconsin, was shown to either increase or decrease the abundance of different plant species, resulting in an overall net reduction of understory plant diversity (Gundale, 2002; Hale et al., 2006; Holdsworth et al., 2007b; Corio et al., 2009; Gibson et al., 2013). Various mechanisms may explain a negative effect

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of earthworms on the recruitment of certain plants. A few manipulative studies have shown, for example, that comminuting and soil mixing activities of earthworms might have a direct effect on seed viability (McCormick et al., 2013), seed germination (Milcu et al., 2006; Aira and Piearce, 2009; Eisenhauer et al., 2010; Drouin et al., 2014) or seedling survival (Eisenhauer et al., 2010; Drouin et al., 2014). Earthworms could also have indirect effects on the performance of understory plant species by modifying the physico-chemical and biological properties of surface soil in which plant roots thrive (Sackett et al., 2013). For example, earthworms may reduce or eliminate organic forest floors (Hale et al., 2006; Gundale et al., 2005), thereby increasing soil moisture loss resulting in a negative impact on plants with shallow root systems (Corio et al., 2009). A second mechanism by which earthworms might indirectly affect understory plant species is by modifying the structure of soil microbial communities. For example, Lawrence et al. (2003) reported a negative effect of earthworms on the colonization of sugar maple (*Acer saccharum* Marsh.) roots by beneficial arbuscular mycorrhizal fungi (AMF). A third indirect mechanism is that earthworms might alter the availability of soil nutrients such as N and P (Hale et al., 2005a; Groffman et al., 2015; Resner et al., 2015), as well as certain groups of soil fauna (Eisenhauer et al., 2007; Snyder et al., 2011), all of which could reduce the performance of certain understory plants.

Sugar maple stands are found throughout the St. Lawrence Lowlands and in most of the geographic area between the St. Lawrence River and Vermont, which makes it one of the most abundant forest types in southern Québec. Moreover, sugar maple holds a high economic value because of its timber, its fibre for pulping, and for the production of maple syrup. Studies have reported negative effects of exotic earthworms on the recruitment of sugar maple in the understory (e.g. Hale et al., 2006; Holdsworth et al., 2007b; Corio et al., 2009), and these same studies also found a higher recruitment of various *Fraxinus* spp. and *Carex* spp., which are also abundant in sugar maple stands of southern Québec. Thus, mature sugar maple stands with similar understory species was a sensible choice of forest type for us to conduct the first regional survey on the ecological impacts of exotic earthworms in southern Québec.

Our first objective was to test whether earthworm abundance correlated with changes in the relative abundance and diversity of various understory plant species. Our second objective was to explore possible relationships between soil properties and changes in understory plant communities potentially caused by the presence of earthworms. Our longer-term objective was to provide data that might foster dialogue on land management policies to mitigate earthworm dispersal in the province of Québec.

2. Materials and methods

2.1. Study area, experimental design and field sampling

The Eastern Townships (10,508 km^2) in southern Québec (Canada) lie within the sugar maple–basswood biogeoclimatic zone (Gosselin, 2007). Mean annual rainfall and temperature for the region, based on 30-year running averages for the City of Sherbrooke, are respectively 1144 mm and 4.1 °C (Environment Canada, 2013). In September 2010, a total of 40 square sampling plots (9 m^2) were established across 5 sites located within a 25 km radius around Sherbrooke (Fig. 1). Two of these sites, each comprising 10 plots, were 11 km apart on land owned by Domtar Corporation (a pulp and paper company) and located northeast of the town of Windsor (Area #1). The area in which these sites are found is not freely accessible to the public but is traversed by secondary roads accessible only to logging trucks. Two other sites, each comprising 5 plots, were located 8 km apart within the Parc National du Mont-Orford, which is 5 km north of the City of Magog (Area #2). One of these sites is a conservation area that is inaccessible to the public whereas the second site is visited by thousands of campers and fishermen each year. The fifth site, comprising 10 plots, was Mont-Bellevue, a municipal recreational forest in the City of Sherbrooke with heavy human traffic (Area #3). The 5 different sites are thus associated with different levels of human activities. The distance between neighbouring plots within each site varied between 100 and >1000 m.

Each sampling plot was established in a mature sugar maple stand with common understory species. In September 2010, 3 parallel transects were established 1.5 m apart from one another. Fresh litter and woody debris were removed and 4 soil cores (7.5 cm dia., 30 cm depth) were sampled at 1 m intervals along each transect ($n = 12$ cores per plot). The thickness of the forest floor F horizon (Soil Classification Working Group, 1998) in each core was measured, and these values were used to calculate the average forest floor thickness in each plot. Each soil core was then placed in a plastic bucket and dissected by hand to ascertain the presence (i.e. at least 1 individual) of earthworms. The same earthworm sampling scheme was repeated in late June 2011, using 3 parallel transects that were established at 75 cm distance from the transects used in the previous year. The frequency of abundance of earthworms (E_{fo}) in each plot was based on the total number of cores, with the 2 sampling dates combined (i.e. $n = 24$ cores per plot), in which at least one earthworm was detected. In October 2012, a final visit was made to each plot to collect earthworm specimens for identification purposes. We applied a mustard solution (4 g L^{-1}) over the entire area of each plot to expel earthworms from the soil (Gunn, 1992; Lawrence and Bowers, 2002). The specimens were preserved in aqueous 90% ethanol and brought back to the laboratory. Sexually mature individuals were identified morphologically using Reynolds' (1976) identification key. Following the first earthworm survey in September 2010, all soil cores within each plot were pooled, earthworms and other visible fauna were removed, the soil was gently mixed by hand, and a 500 g subsample was kept under ice packs in a cooler. These 40 bulk soil samples were transported to the University of Sherbrooke, where they were sieved to pass a 5 mm mesh and stored at 4 °C until their physico-chemical properties could be analyzed. A soil subsample (ca. 20 g) from each plot was immediately frozen until analyzed for its lipid fatty acid profile. Other data that were gathered on this date included an estimate of tree basal area using a 2-factor wedge prism, and soil drainage class according to the criteria developed by the Expert Committee on Soil Survey (1982).

In early June 2011, the understory vegetation in each plot was surveyed using the line-intercept method (Kent and Coker, 1992). Stakes were driven into the soil at the 4 corners of each plot and at 1 m intervals between the 4 corner stakes. Twine was strung across opposing stakes and all understory plants, including juvenile trees <5 cm stem dia., which intersected the twine along its length were noted. To account for plant species that might only appear later in the summer, an identical survey was conducted in late August 2011. Most species were recorded on both dates, and their abundance values were thus averaged over both sampling dates. For the few species that were recorded on a single date, the abundance value for that single date was used.

2.2. Soil physico-chemical properties

For each of the 40 sampling plots, soil pH was measured electrometrically using 1:5 soil:water slurries. Total C and N were measured by high temperature combustion followed by thermo-conductometric detection, using a Vario Macro elemental analyzer (Elementar Analysensysteme GMbH, Hanau, Germany). A 100 g