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## How do campsites, forest fires, and entry point distance affect earthworm abundance in the Boundary Waters Canoe Area Wilderness?

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Factors controlling the spread of invasive earthworms in Minnesota's Boundary Waters Canoe Area Wilderness are poorly known. Believed to have been introduced by anglers who use them as bait, invasive earthworms can alter the physical and chemical properties of soil and modify forest plant communities. To examine factors influencing earthworm distribution and abundance, we sampled 38 islands across five lakes to assess the effects of campsites, fire, and entry point distance on earthworm density, biomass and species richness. We hypothesized that all three parameters would be greater on islands with campsites, lower on burned islands, and would decrease with distance from the wilderness entry point. In addition to sampling earthworms, we collected soil cores to examine soil organic matter and recorded ground and vegetation cover. Campsite presence was the single most important factor affecting sampled earthworm communities; density, biomass and species richness were all higher on islands having campsites. Fire was associated with reduced earthworm density, but had no direct effects on earthworm biomass or species richness. Fire influenced earthworm biomass primarily through its negative relationship to groundcover and through an interaction with entry point distance. Distance affected density but no other factor. For islands with campsites, however, distance from the entry point had a counterintuitive effect in that earthworm biomass, which increased with entry point distance.

| 1  | How do campsites, forest fires, and entry point distance                                     |
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| 2  | affect earthworm abundance in the Boundary Waters Canoe                                      |
| 3  | Area Wilderness?   |
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#### 20 Abstract

21 Factors controlling the spread of invasive earthworms in the forest ecosystems of 22 Minnesota's Boundary Waters Canoe Area Wilderness are poorly known. Believed to have been 23 introduced by anglers who use them as bait, invasive earthworms can alter the physical and 24 chemical properties of soil and modify forest plant communities. To examine factors influencing 25 earthworm distribution and abundance, we sampled 38 islands across five lakes to assess the 26 effects of campsites, fire, and entry point distance on earthworm density, biomass and species 27 richness. We hypothesized that all three parameters would be greater on islands with campsites, 28 lower on burned islands, and would decrease with distance from the wilderness entry point. In 29 addition to sampling earthworms, we collected soil cores to examine soil organic matter and 30 recorded ground and vegetation cover.

31 Campsite presence was the single most important factor affecting sampled earthworm 32 communities; density, biomass and species richness were all higher on islands having campsites. 33 Fire was associated with reduced earthworm density, but had no direct effects on earthworm 34 biomass or species richness. Fire influenced earthworm biomass primarily through its negative 35 relationship to groundcover and through an interaction with entry point distance. Entry point 36 distance itself affected density, and through an inaction with campsite presence, earthworm 37 biomass as well. For islands with campsites, earthworm biomass increased as distance from the 38 entry point increased.

39

#### 40 Introduction

41 Following the end of the last glaciation 12,000 years ago, northern forests of the present-42 day United States were believed to have been entirely free of earthworms (Tiunov et al., 2006). 43 Since the glacial retreat, native earthworm populations have largely remained below the glaciers' 44 southern terminus, but in recent times non-native earthworms from Europe have proliferated and 45 become increasingly common on old glaciated landscape (Callaham et al., 2006; Hopfensperger 46 & Hamilton, 2015). As non-native earthworms expanded their range north, they have profoundly 47 impacted the northern forest ecosystems that evolved in their absence (Hale, Frelich & Reich, 48 2005; Frelich et al., 2006). Many earthworms feed on leaf litter and decomposing plant material 49 that accumulates as a spongy layer of duff on the forest floor. The duff layer provides habitat for 50 ground-dwelling animals and understory vegetation and helps prevent soil erosion. Earthworms 51 consume the duff and excrete nutrient-rich castings, which in combination with their bioturbation 52 and mixing of soil layers, can stimulate microbial activity and greatly accelerate soil nutrient 53 cycling (Bohlen et al., 2004a). Although these processes benefit horticultural and agricultural 54 systems (Bertrand et al., 2015; Sharma, Tomar & Chakraborty, 2017), they can fundamentally 55 alter the functioning of northern forest ecosystems (Bohlen et al., 2004b). Among other 56 detrimental effects, earthworms can alter forest seed banks, kill plant roots, and increase soil 57 compaction, which can lead to increased erosion and nutrient leaching (Costello & Lamberti, 58 2009; Hopfensperger, Leighton & Fahey, 2011; Drouin et al., 2014; Nuzzo, Davalos & Blossey, 59 2015), and negatively affect native flora and fauna (Hale et al., 2008; Loss & Blair, 2011; 60 Fisichelli et al., 2013; Dobson & Blossey, 2015). 61 Non-native earthworms are common in the Boundary Waters, a vast wilderness region of 62 interconnected lakes straddling the Canada–United States border between Ontario and Minnesota

63 (Fig. 1). In northern Minnesota this region has been designated the Boundary Water Canoe Area

64 (BWCA) Wilderness. Known for its pristine lakes and excellent sports fishing, the BWCA has long been a destination for anglers (Heinselman, 1999), and it is probable that anglers played an 65 important role in spreading non-native earthworms into the area by using them as fish bait 66 67 (Cameron, Bayne & Clapperton, 2007; Hale, 2008; Kilian et al., 2012). Non-native earthworms 68 such as "night crawlers" (Lumbricus terrestris) and "red wigglers" (Lumbricus rubellus or 69 Dendrodrilus rubidus) have been sold as fishing bait for decades and are now abundant in 70 Boundary Waters forests. It is not unusual for anglers to dump unused bait (Keller et al., 2007; Frelich & Reich, 2009), and the bait containers contain not only adult earthworms, but also soil 71 72 that could potentially harbor earthworm cocoons. Campsites are likely dumping grounds for 73 unused bait and probable points of earthworm invasion because visitors to the BWCA are 74 required to stay at designated campsites.

75 While earthworms are novel to the Boundary Waters, fire has shaped the region for 76 millennia (Heinselman, 1999). Historically, fire disturbance occurred at 50-100-year intervals, but fire suppression practices over the past century have increased fire intervals to 700 years or 77 78 more (Heinselman, 1999; Frelich & Reich, 2009). Forest fire frequency could matter to 79 earthworms because burning away duff and litter layers has the potential for controlling their 80 numbers (Callaham et al., 2003). Fire kills adults that inhabit the leaf litter (i.e., epigeic species) 81 and may reduce the viability of earthworm cocoons in the soil (Ikeda et al., 2015). For soil-82 dwelling and deep-burrowing earthworms (i.e., endogenic and anecic species), burning away the 83 duff and litter layers would limit food resources, likely starve earthworms, and reduce their 84 fitness (Callaham & Blair, 1999; Coyle et al., 2017). Despite its potential for earthworm control, 85 only a handful of studies have examined how fire affects earthworm dynamics in North 86 American ecosystems (James, 1982; 1988; Callaham & Blair, 1999; Callaham et al., 2003; Ikeda

et al., 2015), and fewer still have examined earthworm responses to fire in northern forests(Frelich et al., 2006).

89 To assess the role of campsites and forest fires in influencing the distribution and 90 abundance of non-native earthworms in the BWCA, we examined lake islands that either had or 91 did not have campsites and were either burned or not burned by the Pagami Creek Fire of 2011. 92 Earthworm abundance was measured in terms of earthworm density, biomass and species 93 richness. We studied islands because they constitute discrete forest patches separated by water 94 that could be characterized as having one condition or the other for each variable (i.e., campsites 95 present/absent, burned/not burned). The islands sampled were located along a chain of 96 interconnected lakes that bordered the northern edge of the 2011 Pagami Creek Fire (Fig. 1). 97 Canoe access for this lake chain is largely limited to a single entry point on Lake One at the west 98 end of the chain. This isolated entry allowed us to examine how distance from the source pool – 99 i.e., arriving anglers carrying earthworm bait – affected earthworm abundance across the islands 100 examined.

101 We hypothesized that islands with campsites would have more earthworms than those 102 without because campsites would receive multiple earthworm introductions from anglers 103 dumping unused bait in or near the campsite (Novo et al., 2015). By contrast, islands that were 104 burned would have fewer earthworms than those not burned because decreased duff and litter on 105 the forest floor, reduced soil organic matter, and decreased leaf litter inputs from trees and 106 vegetation would decrease food availability, thereby limiting earthworm growth and fecundity. 107 Islands having the greatest number of earthworms, therefore, would be those that had campsites 108 and were not burned, whereas those having the least would be burned and without campsites. We 109 also hypothesized that earthworm density and biomass would decrease as distance from the entry

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point increased. We assumed campsites closest to the entry point would have higher visitation
rates because day and weekend anglers would be less likely to travel far into the wilderness
(Lime, 1971). Hypotheses about relationships between variables and earthworm biomass are
summarized in Fig. 2.

114

#### 115 Field-Site Description

116 The 4410 km<sup>2</sup> BWCA Wilderness is located in Minnesota's Superior National Forest (47°57' N, 91°48' W) and extends 185 km along the Minnesota/Ontario border (Fig. 1, inset). 117 118 Designated as a wilderness area in 1978, the area contains approximately 1175 glacial lakes and 119 hundreds of kilometers of rivers and streams (Heinselman, 1999). It is the largest Forest Service 120 Wilderness area east of the Rocky Mountains and the most heavily used wilderness in the United 121 States, with over 150,000 overnight and day-use visitors annually (Eagleston & Marion, 2017). 122 Lakes and waterways comprise approximately 20% of the Boundary Waters wilderness and 123 cross-country canoe travel is made possible by a network of portage trails that connect lakes. 124 Forests in the BWCA are a mixture of conifers and hardwoods that include various species of 125 pine (*Pinus banksiana*, *P. resinosa*, and *P. strobus*), white cedar (*Thuja occidentalis*), balsam fir 126 (Abies basamea), white spruce (Picea glauca), and hardwoods such as paper birch (Betula 127 papyrifera), big-toothed aspen (Populus grandidentata) and white oak (Quercus alba) (Dickens, 128 Gerhardt & Collinge, 2005) The climate of the BWCA region is hemiboreal with mild/cool summers and long winters 129 (Köppen classification Dfb). Average July and January temperatures are 20° and -11° C, 130 respectively, with temperature extremes of 32° and -40° C occurring (Dickens et al., 2005). 131 132 Annual precipitation ranges 66-78 cm and occurs mostly in winter as snow (ca. 40%). Soils of

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133 the area are thin and acidic and overlay Canadian Shield bedrock. Charcoal is almost universal in

- 134 upper soil layers, indicative of the key role fire has played in shaping this ecosystem
- 135 (Heinselman, 1999).

Recreational access to the BWCA is controlled through a permit-based quota system that
limits the number of people entering through each wilderness entry point. Visitors can obtain
permits from May thru September and group sizes are limited to nine. Camping is restricted to
approximately 2000 designated campsites established by the by U.S. Forest Service, that are
unambiguously indicated by cast iron fire grates located at each site (Eagleston & Marion,
2017a).

142

#### 143 Material & Methods

144 On 9 July 2016 we put in canoes at BWCA entry point 30 on Lake One and headed east 145 along a route that skirted the northern edge of the 2011 Pagami Creek Fire (Fig. 1). Over 7 d we 146 sampled 38 islands on lakes One, Two, Three, Four, Hudson and Insula. Islands were selected on 147 the basis of their fire history (burned/not burned) and the presence/absence of campsites (Table 148 1). Island selection was made using maps showing campsite locations (Fisher map F4) and the 149 area covered by the Pagami Creek Fire (USDA Pagami Creek Fire Map). Initial selection 150 followed a balanced design for each combination of characteristics (i.e., burned/campsite, 151 burned/no campsite, not burned/campsite, not burned/no campsite). However, field sampling 152 revealed islands marked as burned were not, and some campsites on burned islands could not be 153 found. Consequently, these islands were discarded from our sample set and resulted in an 154 unbalanced design.

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Distances between islands and the entry point were examined using straight-line distances instead of hypothetical canoe travel distances because the latter can be highly variable. Canoeists and anglers need not follow direct pathways between lakes and islands, and side trips to explore fishing spots and campsite locations are common. Also, multiple pathways may exist for reaching islands, especially in geographically complex lakes such as Insula. Thus, determining actual travel routes becomes a problematic exercise whereas straight-line distances are easily made and independent of human choice.

162 Sampling islands followed a standard procedure: Upon reaching an island, its burn 163 condition, campsite status and GPS coordinates were recorded. Following this, three sampling 164 sites spaced equidistantly around the island's perimeter and approximately 10 m inland were 165 chosen using three criteria: 1) site accessibility from shore, 2) availability of flat and open 166 ground, and 3) lack of dense underbrush allowing easy access to the ground for sampling. After 167 reaching a site, a center point was established and the percent of understory vegetation cover and 168 percent of exposed soil was estimated within a 1 m radius of the site center. Visual estimates of 169 cover and exposure were made with the aid of a chart depicting the Braun-Blanquet percent 170 cover classes. Percent exposed soil was converted into percent groundcover by subtracting 171 values from 100. To assess soil organic matter, three soil cores were collected within the 172 established 1 m radius. The upper 10 cm of each 2.5 cm diameter core was collected and 173 combined in a plastic bag for later organic matter analysis in the lab.

Earthworms were sampled following the procedures outlined in Hale (2013). A mustard water solution was prepared by combining 240 ml of powdered mustard with 4 L of water. This solution acts as an irritant that when poured on the ground and allowed to soak into the soil causes earthworms to emerge as they attempt to avoid the mustard. Half of this mustard solution

178 was poured inside a 0.2 m<sup>2</sup> quadrat placed at the site's center and emerging earthworms were 179 collected with forceps and placed in 50 ml plastic centrifuge tubes. After 2 minutes, the 180 remaining solution was poured into the quadrat and earthworms were collected until they 181 stopped emerging. This procedure was repeated at each site and earthworms from all three sites 182 were combined for each island and preserved in a solution of 70% isopropyl alcohol and 2% 183 formalin. Field conditions made it impractical to clear earthworm guts prior to preservation. 184 Back in the laboratory, earthworms were identified using Hale (2013) and counted. Worms from each site were then combined, dried and weighed to determine biomass. 185 186 We used the loss-on-ignition method to determined soil organic matter. Soil was dried in 187 an oven at 40° C for 7 days, then homogenized with mortar and pestle. Homogenized samples 188 were put into pre-weighed porcelain crucibles and weighed, then ignited in a muffle-furnace (Fisher Scientific Isotemp Muffle Furnace Model 550-126) at 550° C for 3 h. The samples were 189 190 subsequently cooled, placed in a desiccator overnight, and then weighed to the nearest 0.001 g so 191 biomass could be calculated.

192

#### 193 Statistical analyses

To examine the effect of campsite presence, fire history and entry point distance on earthworm richness, density and biomass, we used General Linear Models (GLM). A Structural Equation Modelling (SEM) was used to us examine relationships between earthworm biomass and the variables included in the metamodel (Fig. 2).

198 GLMs for each response variable were performed using JMP 8.0.1 (SAS Institute Inc.,
199 2009). Models used a Poison distribution to fit variables and a log link function to create linear

200 models. Chi-squared  $(X^2)$  probabilities ascertained significance levels of individual factors and 201 interactions. We started with a full model that included main factor effects and all interactions, 202 but three-way interactions were not significant and discarded from final models. We chose not to 203 use a nested design (i.e., islands nested within lakes) because BWCA lakes may be arbitrarily 204 defined. Lakes in the Boundary Waters are often interconnected and multiple lakes may be 205 combined under one name, or alternatively, a single lake may be divided into multiple lakes. For 206 example, Lake Insula is comprised on many small "lakes", whereas Lake Three and Lake Four are two sections of the same lake (Fig. 1). 207

208 The SEM was created in AMOS 16 (Amos Development Corporation, 2007) and used to 209 examine direct and indirect relationships between earthworm biomass and fire history, entry 210 point distance, understory plant cover, groundcover, and soil organic matter. We used four 211 indices to assess the fit of the SEM to avoid bias from any one index (Hintz & Lonzarich, 2018). 212 A chi-square test evaluated the consistency of the data with p-values greater than 0.05 specifying good fit (Grace et al., 2010). To evaluate model fit we looked at the goodness-of-fit index (GFI), 213 214 standardized root mean square residual (SRMR), and root mean square error of approximation (RMSEA). GFI values greater than 0.90 and SRMR and RMSEA values below 0.08 indicate 215 216 good fit (Hooper, Couglan & Mullen, 2008; Hu & Bentler, 1999).

217

#### 218 **Results**

We sampled 38 islands across the six connecting lakes, of these, 12 were burned and 13 had campsites (Fig. 1, Table 1). Island size ranged from 0.4 to 11.8 ha and straight-line distances from the entry point ranged between 3.4 and 26.3 km. Earthworms occurred on all but two of the

sampled islands. Mean ( $\pm$  1 SE) earthworm biomass and density across islands was 12.2  $\pm$  2.5 g m<sup>-2</sup> and 27.2  $\pm$  3.5 individuals m<sup>-2</sup>, respectively.

224 Eight earthworm species were identified (Table 2), of which Dendrobaena octaedra 225 (Savigny 1826) and Lumbricus rubellus (Hoffmeister 1843) were most common. Species 226 richness on islands averaged 1.95 species. Juvenile *Lumbricus* spp. were the most frequently 227 encountered taxonomic group and many of these juveniles were likely L. terrestris (Linnaeus 228 1758); however, their developmental stage precluded positive identification. Campsite presence was the sole factor affecting earthworm species richness ( $X^2 = 4.13$ , df = 1, 37; p = 0.04), with 229 230 campsite-bearing islands averaging 1.13 more species than those without. 231 Campsite presence and burn history had important effects on earthworm abundance. 232 Campsite presence was the only factor that affected both earthworm biomass and density (Table 233 3). The effect on biomass was greater, with campsite-bearing islands having 81% more

234 earthworm biomass as compared to islands lacking campsites. By comparison, earthworm

density on campsite-bearing islands increased by only 37%. Fire was associated with reductions
in earthworm density, but not biomass (Table 3, Fig. 3). Earthworm density also responded to a
Burned\*Campsite interaction such that burned, campsite-bearing islands had approximately half
the density of non-burned, campsite-bearing islands (Fig. 3A). Islands without campsites had
comparable earthworm densities regardless of burn history.

Distance from the entry point directly influenced earthworm density and had interactive effects with campsite presence and burn history on earthworm biomass (Table 3). Across islands, earthworm density showed a significant decrease as entry point distance increased. By contrast, earthworm biomass increased with entry point distance for campsite-bearing islands while remaining unchanged for islands without campsites (i.e., the Campsite\*Distance interaction

shown in Fig. 3A). There was also a Distance\*Burned interaction such that earthworm biomass
decreased as distance from the entry point increased on burned islands, whereas non-burned
islands showed the opposite trend (Fig. 3B).

- 248 Our SEM (Fig. 5) showed a good fit to the data with a high GFI (0.96), low SRMR (0.08)
- and RMSEA (0.00), and a non-significant chi-square ( $X^2 = 6.32$ , df = 9, p = 0.71). Overall, the

250 model explained 47% of the variation in earthworm biomass found on BWCA islands.

SEM analysis broadly corroborated the results of the GLM in that both models showed that campsite presence was the most important single factor affecting earthworm biomass. The SEM showed that entry point distance had a direct and positive relationship with earthworm biomass whereas fire exerted its influence through other factors. Fire was associated with less groundcover and soil organic matter, but more understory plant cover. Of these three factors, only groundcover affected earthworm biomass. Groundcover had a positive relationship to earthworm biomass whereas soil organic matter and understory plant cover showed none.

258 Understory plant cover was also positively correlated to earthworm biomass.

259

#### 260 Discussion

We hypothesized that earthworms would be more abundant on islands with campsites, less abundant on those that had burned, and would decrease as distance from the entry point increased. The data provide strong support for the first hypothesis (campsites), partial support for the second (burn history), and little support for the third (entry point distance).

265 Whether measured in terms of biomass or density, earthworms were consistently more 266 abundant on islands with campsites as compared to those without. This result corroborates the

267 findings of previous studies showing that human activity influences the distribution and 268 abundance of non-native earthworms (Hendrix et al., 2008). These include correlations with road 269 proximity (Cameron & Bayne, 2009) and boat launches (Cameron, Bayne & Clapperton, 2007), 270 and their spread through use as fish bait (Keller et al., 2007). That wilderness campsites may be 271 important points of invasion for non-native species is often overlooked in studies of campsite 272 impacts (e.g., Monz, Pickering & Hadwen, 2013; Eagleston & Marion, 2017a; Marion et al., 273 2016), despite the fact that campsites are where people leave behind food wastes (a potential 274 source of seeds), and may inadvertently transport seeds and other propagules on clothing, 275 footwear or camping equipment (Ansong & Pickering, 2013; Eagleston & Marion, 2017b). 276 Campsites might be construed, therefore, as nodes in a distribution network facilitating the 277 spread of exotic species across wilderness areas. This may be especially true in the BWCA 278 because nearly all travel is done by canoe, which makes established campsites and canoe 279 portages (Dickens et al., 2005) the main points of contact between visitors and the wilderness 280 landscape.

281 Fire effects were not as prominent as campsite effects. Although burned islands did have 282 lower densities of earthworms as predicted, earthworm biomass and richness were unaffected. 283 Fire can kill epigenic species that live in the litter (Ikeda et al., 2015), but is less likely to affect 284 endogenic and anecic species that burrow into the soil. Fire increases soil temperatures only a 285 few degrees near the surface, and just a few just centimeters below the temperatures remain 286 unchanged (Ikeda et al., 2015). Rather than killing worms directly, we expected fire to exert 287 influence through its effects on ground cover and soil organic matter, and through understory 288 vegetation, decreasing the former two and increasing the latter. Our SEM showed that fire had

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289 the predicted effects these three variables, but only groundcover had a significant effect on 290 earthworm biomass such that where litter was abundant, so was earthworm biomass. Less clear is why understory vegetation responded positively to earthworms. Numerous studies 291 292 have shown that earthworms invading northern forests typically cause understory vegetation to 293 decline (see Bohlen et al., 2004b for review). Earthworms can consume newly germinated 294 seedlings (Drouin et al., 2014), the fine roots of plants (Gilbert et al., 2014), and alter the soil 295 microflora to favor only certain species (Drouin, Bradley & Lapointe, 2016). Of course, 296 earthworms can have beneficial effects, as is seen in horticultural and agricultural systems (Bertrand et al., 2015; Sharma, Tomar & Chakraborty, 2017), but why these benefits would 297 298 occur on islands and not in other areas of the BWCA is outside the scope of our study. 299 The most perplexing result of our study was the positive relationship between entry point 300 distance and earthworm biomass. Our prediction was that earthworm biomass would *decrease* on 301 islands further away from the entry point, and this was based on the assumption that discarding 302 unused earthworm bait would be more frequent in areas of high human traffic closer to the entry 303 point. We reasoned that weekend anglers do not travel far from entry points and bait dumping 304 would become less frequent further away as angler traffic diminished. Of course, our 305 assumptions about human behavior may be wrong. We speculate that anglers may actually be 306 less likely to dispose of worms if they are fishing for only short time. In a survey of Wisconsin 307 and Michigan boat owners, Keller et al. (2007) reported that 65% of anglers saved leftover 308 earthworm bait for future fishing trips as opposed to 41% who disposed of them on land or in 309 trash. Similar results were found by Kilian et al. (2012) in their survey of Maryland anglers.

310 Earthworms are sold in soil-packed containers and can be kept alive for weeks under cool and

311 moist conditions (Sherman, 2003). If bait containers are exposed to long days in a sunlit canoe,

312 however, their viability may be compromised. Desiccating soil and temperatures exceeding 30° 313 C can kill earthworms (Berry & Jordan, 2001), and we speculate that multi-day canoe trips are 314 likely to have higher bait mortality than day or weekend trips. Rather than pack out dead and 315 dying worms, long-distance anglers may choose to dump bait containers enroute as they pass 316 through the wilderness. Even if adult earthworms in containers are dead, the soil in which they 317 were kept could contain earthworm cocoons. Whether or not this practice actually occurs would 318 require further study, yet it highlights the need for understanding human behavior for managing 319 the spread of earthworms in the BWCA (Eagleston & Marion, 2017a; 2017b; Marion et al., 320 2016).

321

#### 322 Conclusion

323 Earthworms are often called ecosystem engineers because of the multitudinous effects 324 they have on soils, nutrient cycling, and plants and animals (Holdsworth, Frelich & Reich, 2007; 325 Eisenhauer, 2010). Their presence can profoundly alter ecosystems and their invasion into 326 wilderness areas presents a challenge to resource managers charged with maintaining these 327 natural systems in a state similar to what they were pre-European settlement (Blouin et al., 328 2013). Our investigation of how campsites, fire, and entry point proximity influence earthworm 329 abundance in the BWCA may have management implications. Our data support the hypothesis 330 that campsites are key points of invasion, and suggest that fire may lower earthworm densities 331 via its influence on worm food resources (e.g., litter and duff). Our data also indicate that the 332 distance from a wilderness entry point can influence the distribution and abundance of invasive 333 worms, and these effects may be best understood in the context of campsite use behaviors and 334 bait use practices. We recommend that resource managers promote "leave no trace" principles

| 335   | emphasizing the environmental risks posed by invasive species; for example, by incorporating  |
|---|---|
| 336   | this message in videos visitors are required to watch before entering the BWCA (Guo et al.,   |
| 337   | 2017). As part of their public education efforts, managers might also consider bait container   |
| 338   | warning labels that include instructions on the proper earthworm disposal after use. These  |
| 339   | actions may slow the spread of earthworms in the BWCA and other wilderness areas; however,  |
| 340   | removing these ecosystem engineers from invaded regions is problematic at best, if not  |
| 341   | impossible (Shartell et al., 2015). Once introduced, earthworms become part of the ecosystem  |
| 342   | (Bohlen et al., 2004b), and efforts directed at learning ways to mitigate their impacts will likely   |
| 343   | become as important as trying to control their spread (Tammeorg et al., 2014).  |
| 344   |   |
| 345   |   |
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|   |   |
| 348   | assistance with the SEM analysis.   |
| 348<br>349<br>350   | assistance with the SEM analysis. References  |
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| <ul> <li>348</li> <li>349</li> <li>350</li> <li>351</li> <li>352</li> <li>353</li> <li>354</li> <li>355</li> </ul>              | assistance with the SEM analysis.  References  Ansong, M., and C. Pickering. 2013. Long-distance dispersal of Black Spear Grass ( <i>Heteropogon contortus</i> ) seed on socks and trouser legs by walkers in Kakadu National Park. Ecological Management and Restoration 14(1):1-4. Berry, E. C., and D. Jordan. 2001. Temperature and soil moisture content effects on the growth of <i>Lumbricus terrestris</i> (Oligochaeta: Lumbricidae) under laboratory conditions. Soil                     |
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## Table 1(on next page)

Physical characteristics of the 38 sampled islands.

"X"s indicate that islands were burned by the Pagami Creek Fire or had a campsite. Entry distance is the linear distance from BWCA entry point 30 on Lake One (see Fig. 1), Island Size is the area of the island.

| 1 <b>Table 1</b> . |  |
|--------------------|--|
|--------------------|--|

| Island # | Latitude   | Longitude  | Lake   | Burned | Campsite | Entry Distance<br>(km) | Island Size<br>(ha) |
|----------|------------|------------|--------|--------|----------|------------------------|---------------------|
| 1        | N 47 55.45 | W 91 29.10 | One    | -      | Х        | 3.4                    | 3.85                |
| 2        | N 47 55.31 | W 91 29.19 | One    | -      | -        | 3.7                    | 4.72                |
| 3        | N 47 55.17 | W 91 29.68 | One    | -      | -        | 4.1                    | 2.89                |
| 4        | N 47 55.27 | W 91 29.56 | One    | -      | Х        | 4.2                    | 0.94                |
| 5        | N 47 55.03 | W 91 29.21 | One    | -      | -        | 4.3                    | 0.52                |
| 6        | N 47 54.90 | W 91 29.62 | One    | -      | -        | 4.4                    | 2.12                |
| 7        | N 47 55.20 | W 91 29.71 | One    | -      | -        | 4.5                    | 2.16                |
| 8        | N 47 55.03 | W 91 28.99 | One    | -      | -        | 4.5                    | 0.72                |
| 9        | N 47 55.03 | W 91 29.21 | One    | -      | -        | 4.8                    | 0.83                |
| 10       | N 47 54.33 | W 91 27.79 | Two    | Х      | Х        | 6.7                    | 1.40                |
| 11       | N 47 54.43 | W 91 27.33 | Two    | -      | -        | 7.0                    | 0.60                |
| 12       | N 47 53.04 | W 91 26.69 | Three  | -      | -        | 7.2                    | 0.40                |
| 13       | N 47 53.81 | W 91 26.60 | Three  | -      | Х        | 8.8                    | 2.63                |
| 14       | N 47 53.58 | W 91 26.73 | Three  | -      | -        | 9.0                    | 0.53                |
| 15       | N 47 53.70 | W 91 26.56 | Three  | Х      | -        | 9.0                    | 0.75                |
| 16       | N 47 53.42 | W 91 27.09 | Three  | -      | -        | 9.1                    | 0.65                |
| 17       | N 47 53.02 | W 91 26.50 | Three  | Х      | Х        | 9.4                    | 6.58                |
| 18       | N 47 53.04 | W 91 26.94 | Three  | Х      | -        | 9.5                    | 11.81               |
| 19       | N 47 52.98 | W 91 27.11 | Three  | Х      | Х        | 10.0                   | 9.25                |
| 20       | N 47 53.75 | W 91 25.85 | Three  | -      | -        | 10.7                   | 5.96                |
| 21       | N 47 53.01 | W 91 26.52 | Three  | Х      | -        | 10.8                   | 1.24                |
| 22       | N 47 53.12 | W 91 26.53 | Three  | -      | Х        | 11.0                   | 0.73                |
| 23       | N 47 54.00 | W 91 23.11 | Four   | -      | -        | 14.0                   | 1.74                |
| 24       | N 47 53.73 | W 91 20.85 | Hudson | Х      | Х        | 17.4                   | 1.31                |
| 25       | N 47 54.24 | W 91 17.02 | Insula | Х      | -        | 23.0                   | 5.02                |
| 26       | N 47 54.22 | W 91 16.38 | Insula | Х      | Х        | 23.6                   | 1.21                |
| 27       | N 47 54.56 | W 91 16.82 | Insula | -      | Х        | 24.0                   | 2.28                |
| 28       | N 47 54.55 | W 91 17.06 | Insula | Х      | -        | 24.0                   | 0.64                |
| 29       | N 47 54.62 | W 91 17.41 | Insula | Х      | -        | 24.0                   | 0.52                |
| 30       | N 47 54.70 | W 91 17.23 | Insula | -      | Х        | 24.1                   | 4.17                |
| 31       | missing    | missing    | Insula | -      | -        | 24.1                   | 7.31                |
| 32       | N 47 54.71 | W 91 17.64 | Insula | Х      | -        | 24.3                   | 1.18                |
| 33       | N 47 54.79 | W 91 17.77 | Insula | -      | -        | 24.6                   | 0.45                |
| 34       | N 47 55.16 | W 91 18.05 | Insula | -      | -        | 25.4                   | 0.68                |
| 35       | N 47 55.65 | W 91 16.80 | Insula | Х      | Х        | 25.7                   | 0.90                |
| 36       | N 47 55.65 | W 91 17.02 | Insula | -      | -        | 25.9                   | 1.27                |
| 37       | N 47 55.79 | W 91 17.26 | Insula | -      | -        | 26.1                   | 0.71                |
| 38       | N 47 55.93 | W 91 17.11 | Insula | -      | Х        | 26.3                   | 1 28                |

3

## Table 2(on next page)

Earthworm species or juvenile genera encountered on sampled islands.

"X"s indicate that islands were burned by the Pagami Creek Fire or had a campsite.

|--|

|          |        |          | isenia<br>etida            | endrobaena<br>ctaedra | endrodrilus<br>errestris | umbricus<br>ubellus | llolobophora<br>hlorotice | porrectodea<br>osea | ivenile<br>I <i>porrectodea</i> | ctolasion<br>vrteum | umbricus<br>errestris | ivenile<br>umbricus |
|----------|--------|----------|----------------------------|-----------------------|--------------------------|---------------------|---------------------------|---------------------|---------------------------------|---------------------|-----------------------|---------------------|
| Island # | Burned | Campsite | $\mathcal{F}_{\mathbf{f}}$ | 00<br>0               | D 22                     | $\Gamma_{r}$        | $A_{c}$                   | $A_{i}$             | JL                              | 0 5                 | Lı<br>te              | Jt                  |
| 1        | -      | Х        | 5                          | -                     | -                        | -                   | -                         | -                   | -                               | -                   | -                     | -                   |
| 2        | -      | -        | -                          | 45                    | -                        | -                   | -                         | -                   | -                               | -                   | -                     | -                   |
| 3        | -      | -        | -                          | -                     | -                        | -                   | -                         | 5                   | -                               | -                   | -                     | 20                  |
| 4        | -      | Х        | -                          | -                     | -                        | -                   | -                         | -                   | -                               | -                   | -                     | 5                   |
| 5        | -      | -        | -                          | 30                    | -                        | -                   | -                         | -                   | -                               | -                   | -                     | -                   |
| 6        | -      | -        | -                          | 10                    | -                        | -                   | -                         | -                   | -                               | -                   | -                     | -                   |
| 7        | -      | -        | -                          | 30                    | -                        | -                   | -                         | -                   | -                               | -                   | -                     | -                   |
| 8        | -      | -        | -                          | 5                     | -                        | -                   | -                         | 5                   | -                               | -                   | -                     | 35                  |
| 9        | -      | -        | -                          | 5                     | -                        | -                   | -                         | -                   | -                               | -                   | -                     | -                   |
| 10       | Х      | Х        | -                          | -                     | -                        | -                   | -                         | -                   | 5                               | -                   | -                     | 20                  |
| 11       | -      | -        | -                          | 30                    | -                        | -                   | -                         | -                   | -                               | -                   | -                     | -                   |
| 12       | -      | -        | 5                          | 5                     | -                        | -                   | -                         | -                   | -                               | -                   | -                     | -                   |
| 13       | -      | Х        | -                          | -                     | -                        | 30                  | -                         | -                   | 10                              | -                   | -                     | 50                  |
| 14       | -      | -        | -                          | -                     | -                        | 15                  | -                         | -                   | 5                               | -                   | -                     | 35                  |
| 15       | Х      | -        | -                          | -                     | -                        | -                   | -                         | -                   | -                               | -                   | 5                     | 20                  |
| 16       | -      | -        | -                          | 10                    | -                        | 5                   | -                         | -                   | -                               | -                   | -                     | -                   |
| 17       | Х      | Х        | 10                         | 5                     | -                        | -                   | -                         | -                   | -                               | 5                   | 5                     | -                   |
| 18       | Х      | -        | -                          | -                     | -                        | 10                  | -                         | -                   | -                               | -                   | -                     | 10                  |
| 19       | Х      | Х        | -                          | -                     | -                        | 5                   | -                         | -                   | -                               | -                   | 5                     | 10                  |
| 20       | -      | -        | -                          | 25                    | -                        | -                   | -                         | -                   | -                               | -                   | -                     | -                   |
| 21       | Х      | -        | -                          | -                     | -                        | 10                  | -                         | -                   | -                               | -                   | -                     | 35                  |
| 22       | -      | Х        | 40                         | 5                     | -                        | -                   | 5                         | -                   | 5                               | 10                  | -                     | 10                  |
| 23       | -      | -        | -                          | 5                     | -                        | -                   | -                         | -                   | -                               | -                   | -                     | 5                   |
| 24       | Х      | Х        | -                          | -                     | -                        | -                   | -                         | -                   | -                               | -                   | 5                     | 10                  |
| 25       | Х      | -        | -                          | -                     | -                        | -                   | -                         | -                   | -                               | -                   | -                     | -                   |
| 26       | Х      | Х        | -                          | -                     | -                        | -                   | -                         | -                   | -                               | -                   | -                     | 10                  |
| 27       | -      | Х        | -                          | 5                     | -                        | -                   | -                         | -                   | -                               | -                   | 5                     | 20                  |
| 28       | Х      | -        | -                          | 10                    | -                        | -                   | -                         | -                   | -                               | -                   | -                     | -                   |
| 29       | Х      | -        | -                          | 5                     | -                        | -                   | -                         | -                   | -                               | -                   | -                     | 5                   |
| 30       | -      | Х        | -                          | -                     | 10                       | -                   | -                         | 5                   | 15                              | -                   | -                     | 30                  |
| 31       | -      | -        | -                          | 15                    | -                        | -                   | -                         | -                   | -                               | -                   | -                     | 10                  |
| 32       | Х      | -        | -                          | -                     | -                        | -                   | -                         | -                   | -                               | -                   | -                     | 15                  |
| 33       | -      | -        | -                          | -                     | -                        | -                   | -                         | -                   | -                               | -                   | -                     | -                   |
| 34       | -      | -        | -                          | 20                    | -                        | -                   | -                         | -                   | -                               | -                   | -                     | 10                  |
| 35       | Х      | Х        | -                          | -                     | -                        | -                   | -                         | -                   | 5                               | -                   | 5                     | 30                  |

1 Table 2.

2

|   | 36     | - | - | -  | -   | -  | -  | - | 10 | -  | -  | -  | -   |
|---|--------|---|---|----|-----|----|----|---|----|----|----|----|-----|
|   | 37     | - | - | -  | -   | -  | -  | - | -  | -  | -  | -  | 15  |
|   | 38     | - | Х | -  | -   | -  | 20 | - | -  | -  | -  | 5  | -   |
|   | Totals |   |   | 60 | 265 | 10 | 95 | 5 | 25 | 45 | 15 | 35 | 410 |
| 3 |        |   |   |    |     |    |    |   |    |    |    |    |     |

3 4

### Table 3(on next page)

Effects of campsites, fire and entry point distance on earthworm biomass and density.

Results of the two General Linear Models used to examine earthworm biomass (g m<sup>-2</sup>) and density (individuals m<sup>-2</sup>) in relation to burn history (Burned), campsite presence (Campsite) and linear distance from the BWCA entry point (Distance), as well as interactions among these factors (df= 1, 36). Bold font indicates significant p-values. Whole model statistics:  $X^2$  density = 45.30, df= 6, 31, p < 0.0001;  $X^2$  biomass = 84.23, df= 6, 31, p < 0.0001.

#### 

#### **Table 3.**

|                   | Bi                    | omass   | Density               |         |  |  |  |
|-------------------|-----------------------|---------|-----------------------|---------|--|--|--|
| Factor            | <i>X</i> <sup>2</sup> | P-value | <i>X</i> <sup>2</sup> | P-value |  |  |  |
| Burned            | 0.67                  | 0.413   | 8.51                  | 0.004   |  |  |  |
| Campsite          | 27.82                 | <0.001  | 4.92                  | 0.027   |  |  |  |
| Distance          | 0.45                  | 0.500   | 17.01                 | <0.001  |  |  |  |
| Burned*Campsite   | 0.19                  | 0.665   | 13.74                 | <0.001  |  |  |  |
| Campsite*Distance | 5.32                  | 0.021   | 1.25                  | 0.26    |  |  |  |
| Distance*Burned   | 6.41                  | 0.011   | 2.19                  | 0.14    |  |  |  |

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### Figure 1(on next page)

Study location.

Map shows the location of the Boundary Waters Canoe Area Wilderness in northern Minnesota (inset) and the study area (small white rectangle), which has been enlarged to show the six connecting lakes (One, Two, Three, Four, Hudson and Insula) and the 38 islands that were sampled (shown in black). The dotted line represents the northern extent of the 2011 Pagami Creek Fire; the area south of the line was burned by the fire.



**Figure 1.** Map showing the location of the Boundary Waters Canoe Area Wilderness in northern Minnesota (inset) and the study area (small white rectangle), which has been enlarged to show the six connecting lakes (One, Two, Three, Four, Hudson and Insula) and the 38 islands that were sampled (shown in black). The dotted line represents the northern extent of the 2011 Pagami Creek Fire; the area south of the line was burned by the fire.

## Figure 2(on next page)

Metamodel depicting hypothesized relationships.

Solid lines show positive relationships; dashed lines show negative relationships.

Figure 2



## Figure 3(on next page)

Interactive effects influencing earthworm biomass.

The effect of campsite presence and fire history on earthworm biomass and density. Values are mean ( $\pm$  1 SEM). GLM analyses indicated a significant campsite\*burn interaction for earthworm density (A), but not earthworm biomass (B).

Figure 3



## Figure 4(on next page)

Interactive effects influencing earthworm biomass.

The interaction between entry point distance and burn history (A), and entry point distance and campsite presence (B) on earthworm biomass.

Figure 4



## Figure 5(on next page)

Structural equation model representing the relationships between study variables and earthworm biomass.

Latent variables associated with endogenous factors are not shown. Solid arrows indicate positive effects; dashed arrows indicate negative effects. Dotted grey arrows labeled "ns" indicated non-significant relationships (P>0.10). Model parameters are as follows: chi-square = 6.32, df = 9, p = 0.71; GFI = 0.96; RMSEA = 0.00; SRMR = 0.081.

Figure 5

