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Alliaria petiolata (M.Bieb.) Cavara & Grande [Brassicaceae], an Invasive Herb in the Southern Ozark Plateaus: A Comparison of Species Composition and Richness, Soil Properties, and Earthworm Composition and Biomass in Invaded Versus Non-Invaded Sites

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Citation

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ABSTRACT

Invasive species are widely recognized as organisms that severely alter ecosystem processes in the habitats to which they are introduced. *Alliaria petiolata* is one of the most important invasive plants in forests of the northern United States. This study examined the geographic distribution of the plant in the southern Ozarks, as well as the effect that it may be having on natural processes within forests of the region by comparing plant species richness, plant cover, and soil properties in invaded and non-invaded plots. It was found that *A. petiolata* is not significantly altering species richness, cover, or soil properties in the region, and this may be due to the fact that infestations were only about 1% as dense as those in forests of the northern United States. Populations of *A. petiolata* in the Ozarks were sampled to determine survivorship, fecundity, and preferred habitat types of plants in these forests. This study found that overall mortality of *A. petiolata* was 93.0%, with the highest mortality occurring between seedling and rosette stage (92.3%). Winters in this region are sufficiently mild enough to allow for very low rosette mortality (0.7%) compared with forests in the northern United States, and this may point to future infestations that rival those in northern forests. Adult plants averaged 70.3 ± 20.5 cm tall and produced an average of 2.1 ± 1.7 stems, 33.5 ± 45.6 siliques and 406 ± 670.8 seeds. Populations sampled in floodplain forests had higher densities of plants at each life stage, experienced higher overall survival rates, and were more reproductively successful than populations sampled in upland forests. Plots were also sampled to determine whether or not non-native earthworms were correlated with increased *A. petiolata* density. Although earthworm biomass and leaf litter cover were not significantly different between invaded and non-invaded plots, overall non-native earthworm biomass comprised 95.5% of the total biomass sampled, which indicates that non-native earthworms may be displacing native earthworms in disturbed floodplain and upland hardwood forests of the southern Ozarks.

ACKNOWLEDGMENTS

Jim Henson had a wait and see attitude, and look what happened to him. Now we've got wrong-sounding Muppets.

-Stewart G. Griffin

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INTRODUCTION

Invasive alien species have become one of the major environmental and economic issues of our time. The monetary cost of biological invasions in the United States (U.S.) is astronomical, adding up to nearly $120 billion annually (Pimentel et al. 2005). The cost of invasive plants within agricultural systems alone in the U.S. is estimated at $26 billion annually (Pimentel 2009). Although much of the focus of the problem of invasive species has been placed on agricultural systems, the impact these organisms have on native ecosystems is also great. Edward O. Wilson (1999) wrote that competition from invasive species is one of the primary causes of the decline and extinction of native species. He referred to introduced species as one of "the mindless horsemen of the environmental apocalypse," and with good reason, as more than forty percent of the animals and plants on the Federal Endangered Species List are on that list largely because of their failure to compete with or survive predation by alien species (Bell et al. 2003). Understandably, an important and growing area of concern is the threat that alien species pose to biodiversity in the natural ecosystems they invade (Gerber et al. 2009). In Arkansas alone, for example, nearly one quarter of the natural flora has been introduced, and 94 of those plants have become invasive within natural areas. Not coincidentally, a quarter of native plants in the state are also classified as rare and are tracked as species of conservation concern by state agencies (Gentry et al. 2013).

The federal definition of an invasive species is "an alien species whose introduction does or is likely to cause economic or environmental harm" (EO 1999). One such successful invader that is causing harm to natural systems is *Alliaria petiolata* (M. Bieb) Cavara and Grande (Brassicaceae), an herbaceous biennial native to Europe (Clapham et al. 1987). This weed is one of the few invasive plants that has become an established and widespread member of understory forest communities in eastern North America, and it has become one of the most problematic and important weeds in those forests (Raghu and Post 2008).

STUDY OBJECTIVES

The present study was initiated because, although extensive research has been conducted on *A. petiolata* in the northeastern and midwestern regions of the U.S., almost nothing is known about the distribution, habitat, reproductive output, and mortality of this plant in the southern Ozark Plateaus Ecoregion of Arkansas, Missouri, and Oklahoma. This ecoregion represents the southwestern edge of its

introduced range in North America, which, to the author's knowledge, is also the lowest latitude at which *A. petiolata* has been studied on the continent. In order to characterize infestations within this ecoregion, the questions listed below were addressed during the course of the study.

- What is the current geographic distribution of the plant in the southern Ozarks?

- What forest types does the plant invade in the region?

- Does species richness differ between invaded and non-invaded habitats?

- What is the mortality rate of this plant from one life phase to the next - from seedling, to rosette, and finally to adult plant?

- What is the fecundity of the plant, and does rosette size determine reproductive output?

- Do soil characteristics such as pH and major nutrients differ in infested versus non-infested sites?

- How does density, mortality and fecundity of *A. petiolata* monitored in the southern Ozarks compare to similar data collected within the midwestern and northeastern U.S. by other investigators?

It is very likely that biological control agents will be released within the next few years to control the spread of *A. petiolata* in the U.S. Because of this, it is important to characterize infestations within the southern Ozarks prior to the release of these agents. The most favored biological control agents, petiole mining and root-crown mining weevils of the genus *Ceutorhynchus* are known to reduce the stem height and seed production in *A. petiolata*. Baseline data about the densities, height of adult plants, and reproductive output must be collected in advance of the release of these agents to ascertain whether they are effective at reducing the spread of the plant in the future. Additionally, if *A. petiolata* does prove to become as large of a problem in natural areas within the southern Ozarks as it currently is in the Midwest and northeastern U.S., the author hopes that the results of this study will be used to monitor its spread over time and to develop and implement a successful plan for management.

STUDY SPECIES

NOMENCLATURE AND LIFE CYCLE

Alliaria petiolata (M. Bieb) Cavara and Grande (Brassicaceae), a cruciferous cool-weather biennial, is known by several common names within its native Europe, including garlic treacle-mustard,

Jack-by-the-hedge, hedge garlic, sauce alone, and poor man's mustard (Pratt 1854), names which allude to its natural habitat and also its use as a food plant. In the U.S. it is simply known as garlic mustard, named for the strong garlic odor emitted through its roots and leaves (Grime et al. 1988, Clapham et al. 1987). Synonyms of the scientific name commonly encountered in older floras include *Erysimum alliaria* L., *Sisymbrium alliaria* (L.) Scop., and *Alliaria officinalis* Bieb. (Pratt 1854, Nuzzo 2002).

In its native Europe, the life cycle of *A. petiolata* has been described as an annual (Pratt 1854, Grime et al. 1988), but in the U.S., it has variously been described as an annual, biennial (Clapham et al. 1987), or even a polycarpic perennial (Grime et al. 1988), depending on the reference. Cavers et al. (1979) reported that in Canada, the plant always flowers in the second season (which by definition would require it to be called an obligate biennial) but then described it as a winter annual or biennial. Clapham et al. (1987) and Tutin et al. (1964) earlier reported that within its native range of Europe and Asia, the plant behaves strictly as a biennial. In North America, it is known only as an obligate biennial; seedlings emerge in the spring and spend the first summer and winter as a dark green basal rosette. The following spring, one stem or, less often, several stems are produced which attain a height of up to 1.2 meters (Clapham et al. 1987, Nuzzo 2000). All of the surviving basal rosettes of *A. petiolata* flower during the second spring, regardless of size (Cavers et al. 1979).

DESCRIPTION OF FIRST YEAR PLANTS

Alliaria petiolata seeds are dormant at maturity and require a long period of cold stratification to break this dormancy. The optimal temperatures for germination under laboratory conditions are between 1 and 5 °C for a period of 90-105 days (Baskin and Baskin 1992, Raghu and Post 2008). Studies conducted under natural conditions found that seeds required about eight months of cold stratification in Kentucky (Baskin and Baskin 1992) and more than 18 months in Canada (Cavers et al. 1979). This cold stratification requirement stands in contrast to most other members of the Brassicaceae, which need a period of warm temperatures to germinate. Only two other North American members of this family require cold stratification. These are *Cardamine concatenata* (Michx.) O. Schwarz (cutleaf toothwort) and *Boechera laevigata* (Muhl. ex Willd.) Al-Shehbaz (smooth rockcress) (Baskin and Baskin 1992). Most of the seed of *A. petiolata* germinates within two years but can remain viable in the seed bank for up to ten years (Nuzzo 1999, Pardini et al. 2009, Sosnoskie and Cardina 2009).

Seedlings emerge in late winter in Arkansas (personal observation) while the forest canopy is open and before native herbaceous plants begin growing. Cotyledon blades about 6 mm long (Evans 2006) are oblong in shape with rounded or slightly emarginate apices and cuneate bases (personal observation), with entire margins and simple hairs (Bryson and DeFelice 2010, Figure 1). True leaves, 6 to 10 cm in diameter, soon develop which are reniform in shape with crenate margins. Plants form a basal rosette by mid-summer. Leaves of first year plants are dark green and have pubescent petioles 1 to 5 cm long. Young leaves smell of garlic when crushed, an odor which dissipates by the fall (Nuzzo 2000). Plants overwinter as a dark green basal rosette (Cavers et al. 1979, Figure 2), maintaining green leaves throughout the season (Dhillion and Anderson 1999).

DESCRIPTION OF SECOND YEAR PLANTS

During the spring of the second year, before the forest canopy closes, rosettes send up one or more flowering stems (Figure 3), these glabrate distally and pubescent proximally. Cauline leaves are alternate and 3 to 8 cm in diameter; proximal leaves resemble basal rosette leaves of the previous year with petioles 1 to 5 cm long, and distal leaves are sharply toothed, deltoid in shape, truncate at the base, have shorter petioles, and gradually become smaller toward the top of the stem (Figure 4). All of the leaves are glabrous adaxially and sparsely pubescent abaxially (Nuzzo 2000).

Flowers develop in April to May on terminal, bractless racemes that elongate in fruit to 25 cm; some plants also produce a few axillary racemes in spring and summer (Nuzzo 2000, Figure 5). In the southern Ozarks, rosettes bolt and begin to develop flowers in early April (personal observation, Figure 6) Terete pedicels 3 to 10 mm long carry perfect, complete, actinomorphic flowers 6 to 7 mm in diameter with four white, spreading, clawed petals 3 to 6 mm long, and erect to ascending sepals 3 to 4 mm long. The androecium is composed of six tetradynamous stamens with yellow anthers, borne in two series: the outer series is of two stamens about as long as the sepals, the inner series is of four exserted stamens (Simpson 2010). The gynoecium is composed of a superior, 2-carpelled ovary with an unbranched style 2 to 3 mm long (Al-Shehbaz 2010). Flowers contain four nectaries at the base of the pistil and remain open for an average of 2 days (Cruden and McClain 1996). Flowers are visited by bees, midges and syrphid flies (Figure 7), but are facultatively xenogamous, since in the absence of pollinators the plants readily self-pollinate (Cavers et al. 1979, Cruden and McClain 1996).

Fruit set exceeds ninety percent in this species (Cruden and McClain 1996). Fruits of *A. petiolata* are linear siliques, subterete to 4-angled in cross-section, and are 2 to 8 cm long and 1.2 to 2.5 mm wide (Al-Shehbaz 2010) at maturity (Figure 8). Ovaries contain between 6 and 22 ovules arranged in one row (Al-Shehbaz 2010, Cavers et al. 1979, Figure 9). Mature seeds are dark brown to black, elliptic to oblong in shape, between 2 to 4.5 mm long and about 1 to 2 mm wide (Cavers et al. 1979), and are longitudinally ridged (Nuzzo 2000, Figure 10). Cavers et al. (1979), citing Klyyken (1937), stated that these ridges trap air and enable seeds of *A. petiolata* to float, though not "very well" (citing another study [Lhotská 1975]). Seeds are released from siliques in mid-summer to early fall. In the southern Ozarks, siliques begin to dehisce and release seeds in mid-June (personal observation). Plants senesce following the release of seeds (Cavers et al. 1979).

Seed set is also high, exceeding ninety percent (Cruden and McClain 1996). Seed production varies within this species; isolated plants produce more seeds than ones growing in high density populations (Trimbur 1973), and plants growing in mesic to wet sites have higher seed production rates than ones growing on drier sites (Byers and Quinn 1998). In Illinois plants produce an average of 350 seeds, but seed production on individual plants is as high as 7,900 (Nuzzo 1999). Cavers et al. (1979) found that in one heavily infested garden in partial shade in Canada, 107,508 seeds were produced in a single square meter. *Alliaria petiolata* reproduces entirely by seed (Nuzzo 2000), and because plants are capable of self-fertilization, only one seed is needed to start an infestation.

SIMILARITY TO OTHER SPECIES

Alliaria petiolata resembles several other plants that are common in the southern Ozark forests it invades. *Viola* spp. (violets), *Geum canadense* Jacq. (white avens), and some other mustards resemble *A. petiolata* when in basal rosette form. However, a combination of characters can help to distinguish it from other rosette-forming species. The rosette leaves of *A. petiolata* are somewhat thin with rugose venation and deeply crenate margins, and have a garlic odor that emanates from the leaves of young plants when crushed. In the fall, however, the intensity of this odor decreases, but plants can still be distinguished from other species by examining the thin white taproot, which is s-shaped at the top, immediately below the root crown (Nuzzo 2000, Figure 11). *Viola* spp. and *Geum* spp. have fibrous root systems. When in flower, *A. petiolata* does not resemble any other plant growing in the southern Ozarks.

Figure 1. Seedlings of *Alliaria petiolata* emerging in March 2013. Photograph taken by Jennifer Ogle near Decatur in Benton County, Arkansas.

Figure 2. Basal rosettes of *Alliaria petiolata*, dark green and photosynthetically active in early March 2012, before most native herbaceous plants have emerged. Photograph taken by Jennifer Ogle near Decatur in Benton County, Arkansas.

Figure 3. *Alliaria petiolata* sending up a young flowering stalk in mid-March 2012. Note the differences in the shape of the rosette and cauline leaves. Photograph taken by Jennifer Ogle near Spavinaw Creek in Delaware County, Oklahoma.

Figure 4. *Alliaria petiolata* in flower. The size of the cauline leaves decreases toward the apex of the flowering stalk. Photograph taken by Jennifer Ogle at Tyler Bend on the Buffalo National River in Searcy County, Arkansas.

Figure 5. A small colony of *Alliaria petiolata* in full flower, growing over 1 meter tall. Photograph taken by Jennifer Ogle at Mt. Hersey on the Buffalo National River in Newton County, Arkansas.

Figure 6. Flowers of *Alliaria petiolata*. Photograph taken by Jennifer Ogle at Tyler Bend on the Buffalo National River in Searcy County, Arkansas.

Figure 7. Flowers of *Alliaria petiolata* being visited by a member of the superfamily Apoidea. Photograph taken by Jennifer Ogle at Mt. Hersey on the Buffalo National River in Newton County, Arkansas.

Figure 8. Fruits of *Alliaria petiolata*, in elongated racemes. Photograph taken by Jennifer Ogle at Tyler Bend on the Buffalo National River in Searcy County, Arkansas.

Figure 9. Dehisced siliques, showing seeds of *Alliaria petiolata*. Photograph taken by Jennifer Ogle at Tyler Bend on the Buffalo National River in Searcy County, Arkansas.

Figure 10. Mature seeds of *Alliaria petiolata*, showing the characteristic longitudinal ridges. Photograph taken by Jennifer Ogle.

Figure 11. The s-shaped tap root of *Alliaria petiolata*, a feature that helps distinguish it from other rosette-forming plants in the region. Photograph taken by Jennifer Ogle near Decatur in Benton County, Arkansas.

FOOD AND MEDICINAL USES

Alliaria petiolata has several food and medicinal uses. It was traditionally used as an herb by Europeans to flavor sauces, as "greens" in salads and as an accompaniment to salted meats and bread (Pratt 1854, Grieve 1959). Pratt wrote that it was often used by "those who cannot afford more costly condiments" (1886). It is a nutritious plant; the leaves contain more vitamin A than spinach and at certain times of the year contain more vitamin C than oranges (Cavers et al. 1979). Historically, it was used as a medicine to induce sweating and to open pores (Grieve 1959). It was also used topically as an antiseptic to treat wounds (Pratt 1854). Juices extracted from the leaves were useful in treating dropsy (Grieve 1959). Linnaeus found that while sheep, cows, and poultry ate the leaves of A. petiolata when offered, horses and goats refused them (Pratt 1854), potentially useful information when considering grazing animals as a method of biological control of the plant.

NATIVE AND INTRODUCED RANGE

Alliaria petiolata is native to northern Europe and western Asia (Welk et al. 2002). Tutin et al. (1964) reported its native range in Europe is "from about 68° N. southwards, but less common in the extreme south." Northern India, North Africa, and West and Central Asia are also included within its native range (Clapham et al. 1987, Welk et al. 2002). The plant has been introduced to New Zealand, Sri Lanka, and North America (Tutin et al. 1964, Welk et al. 2002). The New Zealand report was from one site at Ashburton in 1893, where the plants persisted for only about 10 years (Webb et al. 1988). The Sri Lankan occurrences were reported by Cavers et al. (1979), but the plant is not reported in recent floras of the region (Welk et al. 2002).

The North American introduction had an altogether different result than the Sri Lankan and New Zealand introductions. The plant was likely brought to this continent by European immigrants and was first observed growing outside of cultivation in 1868 on Long Island, New York (Evans 2006). By 1913 it was reported in Quebec and Ontario and southern New York, New Jersey and Virginia (Britton and Brown 1913). By 1952 it was reported as far west as Kansas (Gleason 1952). By 1979 it had made its way to the west coast at Portland, Oregon (Cavers et al. 1979), and by 1991 it had spread to 29 states in the Midwest and Northeast, and two Canadian provinces (Scott 2000). Currently, Alliaria petiolata has extended its range throughout the Northeast and New England and as far north as Ontario, west to

Nebraska, Oklahoma and Kansas (Myers et al. 2005), and south to Arkansas and Georgia (USDA, NRCS 2013). In addition, several isolated populations have been found in Utah, Idaho, Oregon and Washington. The Pacific Northwest populations were reported in 1959 (USDA, NRCS 2013) and are spreading (Blossey et al. 2001). In all, it is reported from 37 states, including Alaska (USDA, NRCS 2013), and seven Canadian provinces (NatureServe 2013).

Prior to the present study, *A. petiolata* was known in the southern Ozark Plateaus Ecoregion from seven counties in Arkansas - Benton, Franklin, Izard, Marion, Newton, Searcy, and Washington. Specimens housed at the University of Arkansas Herbarium (UARK) show that it was first collected in the state in 1972 just northwest of Decatur in Benton County. Its discovery in Arkansas was later reported by E. B. Smith and B. L. Lipscomb (1975). Although the plant is widespread throughout northern Missouri, only two counties, Barry and Howell, which border Arkansas to the north, reported populations prior to this study (Yatskievych 2006). The plant was first documented in Missouri in 1972, in Taney County along Lake Taneycomo, but a search ten years later failed to relocate the population (Mike Skinner, personal communication, April 12, 2013). In Oklahoma, the plant was first documented in 1994 in Delaware County, and remained the only county with documented infestations prior to the current study (OBS 2009).

HABITAT

Within its native range of Europe, *A. petiolata* is common in lightly shaded sites along edges of woods, hedgerows, wall bases, trails, and riparian areas (Clapham et al. 1987). It prefers alkaline soils and is intolerant of highly acidic soils (Grime et al. 1988). In the more arid, Mediterranean reaches of its native range, it prefers shaded areas and north-facing slopes of mountains (Welk et al. 2002). In its introduced range, however, it grows in many different habitat types and covers large areas (Blossey et al. 2001). It is most commonly found in moist, partial to deeply shade habitats but has also been found growing in open woods and areas that receive full sun. This species benefits from disturbance, whether it is natural, as in floodplains and riparian areas, or anthropogenic, such as edges of woods, trails and roadsides (Cavers et al. 1979, Meekins and McCarthy 2001). It can grow along very wet riverbanks or on steep slopes in sandy, well-drained soil (Blossey et al. 2001). It has demonstrated the ability to invade mature, undisturbed forests (Stinson et al. 2006) and is considered to be a permanent member of the

community once it becomes well established (Nuzzo 2000).

REASONS FOR SUCCESS

Alliaria petiolata has been successful within its introduced range for a variety of reasons. It is adept at competing for light. The wintergreen basal rosette begins to photosynthesize very early in the second year while the forest canopy is still open and temperatures are just above freezing (Evans 2006). Its leaves reach maximum photosynthetic rates during this time (Meekins and McCarthy 2000), when light levels are high and other plant competitors are absent or are just beginning to emerge. As the canopy closes and native spring ephemerals become active, plants bolt and produce shade-tolerant leaves on tall, fast-growing shoots which easily cover native plants. Clearly, a very early growing season coupled with light-adapted leaves gives this species a great competitive advantage over indigenous species (Myers et al. 2005).

An absence of predators also contributes to the success of this invasive plant. Reduced predation is a necessary element for the success of many alien species, a concept known as the enemy release hypothesis (Blossey 2011). Having no natural predators in its introduced range, resources that had been used to overcome herbivory are reassigned to focus on growth and reproduction (Bossdorf et al. 2004, Wittenberg and Cock 2005). Within its native range this species is used by at least 70 insect herbivores, five of which are specialists, and seven fungi (Blossey et al. 2001, Cipollini et al. 2005). It is the preferred food of the butterfly *Pieris brassicae* L. (large white) in Europe (Cavers et al. 1979). None of the insects associated with the plant in Europe occur in North America, and only eleven herbivores eat the species on this continent, none of which are monophagous (Cipollini et al. 2005).

Herbivory by most insects in North America is discouraged by the presence of several secondary compounds found in the leaves of *A. petiolata*. Glucosinolates, cyanide, peroxidase, trypsin inhibitors, alliarinoside, and isovitexin-600-O-b-D-glucopyranoside are several defense compounds utilized to deter predators (Cipollini et al. 2005). Glucosinolates, a class of sulfur- and nitrogen-containing organic compounds, impart a bitter taste to the consumer, and as a result are a primary deterrent against generalist predators (Renwick et al. 2001). Sinigrin and glucotropaeolin are two glucosinolates found in both the roots and aboveground tissues of *A. petiolata* (Huang et al. 1995). Cyanide levels (a possible breakdown product of glucosinolates) in first-year leaves of *A. petiolata* are a high as 100 ppm, an

amount toxic to many vertebrates (Cipollini and Gruner 2007). Peroxidase and trypsin inhibitors are also known deterrents against herbivory by generalist insects and infection by pathogens in other members of the mustard family (Cipollini et al. 2005). Alliarinoside deters feeding by first instars and isovitexin-600-O-b-D-gluco-pyranoside deters feeding by later instars (Renwick et al. 2001, Haribal and Renwick 2001).

Some species of butterfly in North America are attracted by the chemical compounds in this plant because of its chemical similarity to other native members of the mustard family. Two butterflies, the introduced *Pieris rapae* L. (small white) and the indigenous *P. oleracea* Harris (mustard white), are attracted to the sinigrin in *A. petiolata*, but the deterrents in the plant ultimately prove detrimental to the overall health of these species. Eggs that are laid on this plant usually do not survive larval development (Huang et al. 1995). This situation would be a concern for any species, but has been especially detrimental to a third butterfly, *P. virginiensis* Edwards (West Virginia white), which is native to North America and though is common throughout much of its range, has declined or disappeared in other parts of its range (NatureServe 2014). The habitat of this butterfly is very restricted; it can live only in heavily-shaded mesic forests on rich soils and has one primary host, *Cardamine diphylla* (Michx.) A. W. Wood (crinkleroot) (Benson et al. 2003). Unfortunately, this butterfly now shares its habitat with *A. petiolata*, which contains the same chemical attractants as, is more abundant than, and has a longer growing season than *C. diphylla*. The butterflies are attracted to oviposit on *A. petiolata* and the larvae which feed on its leaves usually die by the fourth instar because of the toxicity of the plant (Nuzzo 2000), creating a population sink for this species. The long-term survival of *P. virginiensis* has become seriously threatened as a direct result of the spread of the presence of *A. petiolata* in the core part of its range (NatureServe 2009, 2014).

Yet another reason for the success of this invasive species is that it is able to disrupt the ecological interactions between native plants and mycorrhizal fungi. Ninety-five percent of plants form mycorrhizal associations (Rodgers et al. 2008), including most woody species, and are dependent on these associations for survival. Mycorrhizae increase nutrient and water uptake, thus improving growth in plants (Burke 2008). One recent study demonstrated that arbuscular mycorrhizal fungi (AMF) were unable to adequately colonize in the roots of *Acer saccharum* Marshall (sugar maple)*, A. rubrum* L. (red maple)*,* and *Fraxinus americana* L. (white ash) in the presence of *A. petiolata*, and the trees displayed slower

growth as a result (Stinson et al. 2006). In fact, soils that had been invaded by *A. petiolata* showed almost no colonization by AMF. Wolfe et al. (2008) examined the question of whether *A. petiolata* is effective at inhibiting ectomycorrhizal (EM) associations. Core samples were taken from invaded and uninvaded soils of three distinct wooded areas in New England. These were a *Pinus strobus* L. (eastern white pine) forest, a mixed deciduous conifer forest, and an urban arboretum where both native and nonnative trees had been planted. They found that EM was greatly inhibited in the plots that had been invaded, which in turn lead to reduced uptake of nutrients by the trees that grow there. Meekins and McCarthy (1999) found that *Quercus prinus* L. (chestnut oak), an upland oak species that also thrives on rich soils along streams (McQuilkin 1990), was unable to compete in the presence of *A. petiolata*. This is a concern, as *Quercus* species are already experiencing regeneration issues in many North American forests (Royse et al. 2010). It is important to note that in the Meekins and McCarthy study, competitive ability, not allelopathy, of *A. petiolata* was attributed to the negative impact on *Q. prinus* seedlings, which were unable to grow up through a tall understory layer.

Studies have also demonstrated that *A. petiolata* increases nutrient levels in the soil, creating a habitat suitable for its own reproduction and spread, at the expense of native plants. Rodgers et al. (2008) found that invaded soils had higher levels of available nitrogen, phosphorus, calcium, and magnesium than uninvaded soils. These soils were also higher in pH. The ability of *A. petiolata* to change the composition of the soil, combined with its ability to decrease mycorrhizal colonization, creates conditions that favor its own growth and expansion, further negatively impacting native plant communities.

MANAGEMENT TECHNIQUES

MECHANICAL AND CHEMICAL

Many management techniques have been and are still being developed in an effort to control the spread of *A. petiolata*. Control of this species is a high priority in small populations and in high quality forests, but a lower priority in low quality areas where populations are well established (Nuzzo 2000). The main objective in management is to prevent seed germination. In areas where a population is already well established, several methods can be utilized, including fire, herbicides, cutting, pulling and mowing.

To control larger populations, prescribed fires have been used. This method is a difficult management technique and has yielded inconsistent results. Repeated burnings are necessary and the

fires have to be of high intensity to successfully reduce plant populations. Low-intensity fires fail to kill the root crown and also clear the leaf litter from the area, leaving behind a seed bed suitable for further seed germination (Nuzzo 2000, Pardini et al. 2009). Even with repeated high-intensity fires, the plant is not eliminated completely.

Chemical controls have also been utilized in the effort to control *A. petiolata*. Application of glyphosate in early spring or late autumn was shown to decrease cover by 95 percent, but this method may not be practical over large areas. Also, native herbaceous plants can be negatively affected by the use of non-selective herbicides (Nuzzo 2000, Evans 2006). Bentazon also has been used with similar success rates (Evans 2006). Late fall is the optimal time to apply these chemicals, when most native plants have gone dormant for the season and when rosettes of *A. petiolata* are still active.

Mechanical methods for controlling *A. petiolata* include cutting, pulling, and mowing. Cutting and pulling are effective but very labor-intensive tasks. Plants should be cut at ground level to achieve 99 percent death rates; cutting at 10 cm above ground results in 71 percent death rates. Plants should be cut when they are flowering, as this is the time when they have the least amount of resources to rebound from cutting (Nuzzo 2000, Evans 2006). Fruiting stems must be removed from the site, as fruits of cut stems continue to mature (Solis 1998).

Pulling is a labor-intensive activity and as such is self-limiting to small populations. But it is a very effective method if enough of the root is removed to prevent growth from buds in the root crown (Evans 2006). It can also be done at any time during the year. Care must be taken when using this method; soil should be replaced and tamped down to minimize the chances of an infestation of other weedy plants. Because seeds are viable in the seed bank for up to 10 years, pulling must be done every year until seedlings stop emerging.

Bauer et al. (2010) found that repeated pulling of second-year plants was effective in reducing numbers of first-year rosettes. They demonstrated that pulling late in the year (mid-May) was more effective than pulling early in the year (early March), because of increased intraspecific competition, which causes a larger reduction of first-year plants.

Mowing is just as effective as cutting, but the equipment used is likely to cause damage to native plants. Also, mowing may spread seeds if they are present. Mowing may be a good option along

roadsides, but care must be taken to clean equipment between sites (Evans 2006).

BIOLOGICAL

Alliaria petiolata has largely proven resistant to chemical and mechanical efforts to control its spread; these methods are effective on small populations, but the positive results are usually short-lived and have done very little in reversing the spread of this plant on a large scale. Because of this, it has become necessary to explore long-term and large-scale solutions to curb the spread of *A. petiolata*, and the most promising methods toward accomplishing this are biological agents (Evans 2006).

Of the 900 biological agents that have been used to control weeds in the U.S., about 35 percent have been successful (Perrings et al. 2005). If protocols are followed carefully (i.e., risk assessment studies), biological control can be a safe and very effective way of curbing an infestation. The initial costs are very high for this method, but subsequent costs are low and the result is effective and efficient control of the target weed (Wittenberg and Cock 2005).

Several biological agents have been identified that may prove successful in controlling *A. petiolata*. Of the 70 or more species that feed on this plant in its native range, five species of weevil in the genus *Ceutorhynchus* and a flea beetle in the genus *Phyllotreta* have been considered (Davis et al. 2006, Evans 2006). The flea beetle did not show strong enough host specificity, and one of the weevils could not be readily obtained (CABI 2013). The four remaining *Ceutorhynchus* weevils are the most appropriate candidates for control of this weed (Katovich and Becker 2014).

Ceutorhynchus constrictus Marsh is a seed feeder, and in its native range is the most widespread and common weevil of the species under consideration. Females feed on leaves and lay more than 150 eggs in one season. The larvae eat two to three developing seeds each before leaving plants and pupating in the soil (Davis et al. 2006). Adults emerge the following spring to feed on leaves and to mate. In trials, *C. constrictus* preferred *A. petiolata* over 54 other host plants offered (Evans 2006). Of the four weevils under consideration, this one shows the most specificity for *A. petiolata*; it is the only one which attacked only *A. petiolata* in field trials (Davis et al. 2006).

Ceutorhynchus alliariae Brisout and *C. roberti* Gyllenhal are shoot-borers that feed on the leaves of *A. petiolata* as adults but whose larvae mine the shoots and leaf petioles of host plants from early to late spring, and then move to the soil to pupate (Gerber et al. 2012). Females of *C. alliariae* lay eggs

individually, and *C. roberti* lay clusters of as many as eight eggs (Gerber et al. 2007). Larvae of both species were mostly observed mining on second year plants but also have been seen mining on rosettes. Both species reduced plant height by about 10 cm at most densities. When these two species are released together, they are able to reduce seed production by 25 percent and cause wilting of shoots (Davis et al. 2006, Gerber et al. 2007). These last two results were only seen when weevils were present at high densities; *A. petiolata* has demonstrated the ability to compensate for weevil attack at low densities by producing more inflorescences (Gerber et al. 2007). It should be noted that in the Gerber experiment, plants were grown in the absence of competition from other plant species, and also were given fertilizer throughout the trial; both factors may have contributed to the ability of *A. petiolata* to compensate for low densities of predation. Because host specificity tests are more complete for *C. alliariae*, it is likely that this weevil would be released before *C. roberti*.

Alliaria petiolata is the most susceptible to mortality during late winter, when plants bolt and begin flowering (Katovich and Becker 2014). Because this is also the time when *Ceutorhynchus scrobicollis* Nerensheimer and Wagner is active, it is considered to be the most effective biological control agent. A monophagous root-crown mining weevil that is dormant in the summer, it oviposits in leaf petioles or on leaf surfaces in mid-fall and continues through spring of the following year. Larvae are petiole miners and later instars feed on the root crown and lower shoots. Plants that are infested with this species have reduced seed production and if infestations are sufficiently high, will die without producing seed (Davis et al. 2006). Gerber et al. (2009) found that *C. scrobicollis* was the most effective at killing rosettes (up to 43 percent mortality) and reducing seed production (up to 49 percent reduction). In response to attack, *A. petiolata* sent up several secondary shoots, but they were not as tall or strong as the main shoot; overall plant height was reduced by 54 percent.

It is evident to those who are studying these four weevils that *C. scrobicollis* is the strongest candidate for release, and for several reasons. It demonstrated strong host specificity. It has the largest impact on basal rosette mortality. Unlike *C. alliariae* and *C. roberti,* which can stimulate seed production at low densities, *C. scrobicollis* elicited no defensive response from its host at any density (Davis et al. 2006). Gerber et al. (2009) reported that some *C. scrobicollis* females were able to survive for three years and they oviposited each season. Females of this species can lay over 400 eggs during their lifetime,

which would contribute to a "rapid population build-up" (Gerber et al. 2009), resulting in fast and effective control of *A. petiolata*. A petition was submitted in 2008 for release of this weevil in the field. As of 2014, work was continuing with all four of these weevils, and various tests, such as open-field and no-choice oviposition and larval development tests, were being conducted on them (CABI 2013).

DISCUSSION

Alliaria petiolata is devastating to native forest understory plant assemblages in many regions of the U.S. and Canada, reducing biodiversity of the native herbaceous layer (Anderson et al. 1996, Godwin 2012) and inhibiting the regeneration of important native tree species (McCarthy 1999, Stinson et al. 2006). The situation is quite bleak, with investigators using words such as "dominant" and "permanent" to describe its residence within North American forests (Nuzzo 2002, Godwin 2012). *Alliaria petiolata* employs several methods for outcompeting native plants, including the use of several secondary compounds (Huang et al. 1995, Cipollini et al. 2005, Cipollini and Gruner 2007), self-compatibility (Cavers et al. 1979, Cruden and McClain 1996), high seed output and germination (Cavers et al. 1979, Nuzzo 1999), the ability to change soil pH and nutrient levels to suit its needs (Rodgers et al. 2008), and the ability to adapt to different light regimes (Meekins and McCarthy 2000, Myers et al. 2005). Many methods for control of the plant have been tested (Nuzzo 2000, Evans 2006), but nothing as of yet has proven to be effective at quelling the spread of this plant. It appears that the last hope for controlling this plant is through the use of biological agents (Gerber et al. 2009). It is the sincere hope of the author that the present study will contribute to the growing body of work being conducted on *A. petiolata*, and that it might provide a meaningful contribution to its eventual control.

Chapter 2

Alliaria petiolata (M.Bieb.) Cavara & Grande [Brassicaceae]:

A Comparison of Invaded and Non-Invaded Sites, Survivorship, and Reproductive Success

STUDY AREA - DISSECTED SPRINGFIELD PLATEAU-ELK RIVER HILLS ECOREGION

GEOLOGY AND SOILS

The study area encompasses two subsections within the Springfield Plateau Subdivision of the Ozark Plateaus Physiographic Province (Woods et al. 2004, Nunn 2013): the Elk River Hills and the Dissected Springfield Plateau (Foti et al. 1999). The Elk River Hills Subsection is a small region of rugged hills cut by tributaries of the Neosho River (Foti et al. 1999, Nigh and Schroeder 2002), and is situated in extreme northwestern Arkansas, northeastern Oklahoma and southwestern Missouri (Chapman et al. 2002, Woods et al. 2004, Woods et al. 2005). The Dissected Springfield Plateau Subsection is topographically similar and lies to the east, stretching southeast nearly to Batesville in northern Arkansas, bordering the Boston Mountains to the south for much of its length (Chapman et al. 2002, Woods et al. 2004, Woods et al. 2005). Together, these rugged, hilly regions comprise the Dissected Springfield Plateau-Elk River Hills Ecoregion (Figure 1) described by Woods et al. (2004) and features steep slopes, narrow valleys, rock outcrops, and karst features. This ecoregion is associated with the many streams that cut through its surface, such as the Elk and Buffalo rivers and Spavinaw Creek (Nigh and Schroeder 2002, Woods et al. 2004, Woods et al. 2005).

During much of the Paleozoic Era, the entire Ozark Plateaus Physiographic Province was buried beneath shallow seas, resulting in the deposition of carbonate minerals (Guccione 1993) and ash layered on top of Precambrian igneous bedrock (Nunn 2013), but the entire region was lifted upward near the end of the Pennsylvanian Period with the collision of the North and South American continental plates, forming the Ozark Dome (Foti 2008, Nunn 2013). Once above sea level, the dome was subjected to hundreds of millions of years of weathering and erosion (Nigh and Schroeder 2002), forming the deeply dissected, flat-topped plateaus characteristic of the region as we know it today (McFarland 1998, Foti 2008). The exposed, uppermost layers of the Springfield Plateau uplands are Mississippian sedimentary limestone interbedded with chert (Nunn 2013), known as the Boone Formation (McFarland 1998), and the exposed rock types which occur along rivers are Chattanooga Shale and Cotter Dolomite (Woods et al. 2004).

Soils are of the order Ultisols and suborder Udults (Smith 1989, NRCS 1999). Ultisols are the common soils of the warm, humid, forested southeastern United States, and are typical of such

environments worldwide (NRCS 1999). These soils are products of the heavy weathering of parent material and tend to be acidic and infertile (Smith 1989). Udults are the most common suborder of Ultisols and are humus-poor soils (NRCS 1999). Soils in the Dissected Springfield Plateau-Elk River Hills Ecoregion formed from limestone and chert parent materials, and the soils examined in the present study occurring on slopes are typically of well-drained, thin colluvium heavy with fragments of chert, while in the narrow floodplains soils are deeper gravelly silt loams. Commonly occurring soil series within the ecoregion are Clarksville, Moko, and Portia (Woods et al. 2004).

CLIMATE

The region encompassing the Dissected Springfield Plateau-Elk River Hills Ecoregion has a humid subtropical climate, a temperate region characterized by hot, humid summers, relatively mild winters, and no true dry season (Peel et al. 2007). Annual precipitation in the area ranges from 112 to 122 cm (Johnson and Duchon 1995, Woods et al. 2004, Tyrl et al. 2007). Though this precipitation is generally well-distributed throughout the year, much of it falls as rain in the spring when air from Canada and the Gulf of Mexico collide and produce heavy storms which cause localized flooding (Skiles 1981, Smith 1989). Occasional periods of drought are not uncommon during the summer months, but are usually of short duration (Skiles 1981, Smith 1989). Mean annual temperature is 15 degrees Celsius (°C) (Skiles 1981), with average temperatures as low as -4 °C in the winter and as high as 34 °C in the summer (Woods et al. 2004).

The present study was conducted during some of the worst droughts in the southern United States in recent memory. In 2013, the National Weather Service in Little Rock noted that "drought has been a common theme in Arkansas since 2010." Beginning in late August 2010 the southern Ozarks experienced abnormally dry weather which progressed to moderate drought by late winter of 2011. Following the spring rains in late April of 2011, conditions improved until mid-summer, when abnormally dry to moderate drought conditions again prevailed and continued into late fall of 2011. By August 2011, conditions had deteriorated to severe drought. Beginning in May 2012 and continuing into fall of the same year (the termination of the study), the southern Ozarks was experiencing abnormally dry weather that steadily progressed to exceptional drought (Figure 2).

VEGETATION

The Ozark Plateaus are part of a large ecotone between the Great Plains prairies and the Eastern Deciduous Forest (Stambaugh and Guyette 2006), and as such, contain elements of both biomes. During the Late Glacial period, about 12,000 BP (Sauer 1988, Foti et al. 1999), the region's climate became warmer and drier and the Dissected Springfield Plateau-Elk River Hills Ecoregion was dominated by oak-hickory savannas, woodlands, prairies, and glades (Nigh and Schroeder 2002, Andre et al. 2009). As climate again changed, around 4,000 years ago, the region was also able to support open-canopied upland hardwood forests (Foti et al. 1999) with densities of about 50 trees per acre (Foti 2004). These areas were maintained in those states by low-intensity natural and, more commonly, anthropogenic fires (Pyne 1997, Batek 1999) as well as by large grazers such as elk and bison (Andre et al. 2009). That tree-covered areas of the Ozarks supported such large grazing animals speaks to the fact that the canopy was more open than it is today, and was described by some early observers as "park-like" in appearance (Hicks 2000).

With the advent of European settlement in the region, however, the landscape that had sustained itself for thousands of years changed very quickly. Permanent settlements were established in the early 19th century and by the early 20th century, most of the virgin trees in the Ozark forests had been cut and cleared (Nigh and Schroeder 2002). Subsequent fire suppression efforts which began in the early 1900s and became the norm by the 1930s (Koen 1939) resulted in a regrowth of dense stands of completely closed canopy forests (Stambaugh and Guyette 2006) with densities of 300 to 1,000 trees per acre (Andre et al. 2009). In some areas of the forest a shift from oak species to shade- and fire-intolerant species, such as *Acer rubrum* L. var. *rubrum* (red maple), *A. saccharum* Marshall (sugar maple), and *Fagus grandifolia* Ehrh. (American beech), occurred (Soucy et al. 2005; Andre et al. 2009). Areas once dominated by woodlands, savannas, prairies, and glades transitioned into havens for *Juniperus virginiana* L. var. *virginiana* (eastern red-cedar) thickets or even closed canopy forests (Foti et al. 1999). Indeed, many of the historic vegetation types that occurred widely in the Ozarks have almost disappeared completely - prairies have been replaced by cultivated fields, and barrens and open woodlands have become overrun with dense woody vegetation (Foti et al. 1999). The herbaceous layer of grasses and forbs that once thickly blanketed the forests, woodlands, and savannas of the region have been shaded out as a result of increasing densities of canopy cover (Foti et al. 1999, Stambaugh and Guyette 2006).

Today, the upland forests of the Dissected Springfield Plateau-Elk River Hills Ecoregion are largely composed of dense second-growth *Quercus-Carya* (oak-hickory) forests (Bryant et al. 1993). Native mesic forests of *A. saccharum*, *Quercus alba* L. (white oak), *Q. rubra* L. (northern red oak)*,* and *F. grandifolia* still occur in ravines and along north-facing slopes (Woods et al. 2004). Many of the riparian forests and woodlands, also known as "bottoms" or "bottomland", of the ecoregion were long ago cleared for pastureland and agriculture (Nigh and Schroeder 2002). In eastern Oklahoma, for example, as much as 85 percent of the riparian forests that stood prior to European settlement have been destroyed (Pritchard 1993). In some areas of the Ozarks these forests have been allowed to grow back and can be described as dense, second-growth mixed hardwood forests (personal observation). As these forests become more and more fragmented with increasing human use and development, opportunity for invasion by non-native species also increases (Yates et al. 2004).

All three of the aforementioned forest types are susceptible to infestation by *A. petiolata* (Nuzzo 2000), lotic riparian communities in particular (Welk et al. 2002). Riparian vegetation provides many benefits to the surrounding environment by filtering nonpoint source pollution, stabilizing stream banks (Fallon 1993), and controlling input of sediments into streams (Hood and Naiman 2000). Riparian corridors also provide habitat, either temporarily or permanently, for the majority of vertebrate species occupying a region (Naiman et al. 1993). Invasions by non-native species into this sensitive ecosystem type is a growing area of concern in ecology (Planty-Tabacchi et al. 1996). Invasibility of riparian forests by non-native species is high largely because of the characteristics of riparian corridors themselves; they are highly dynamic transition areas between aquatic and terrestrial systems (Pritchard 1993, Planty-Tabacchi et al. 1996), pathways across regions that serve "as both conduit and barrier" to species movement (Malanson 1993).

Forests of the region have changed dramatically over the past 150 years due to fire suppression, fragmentation, and the introduction of invasive species. The present study attempts to quantify the current extent of invasion and effect on native species composition by one invasive species in particular, *A. petiolata*, within upland and riparian forests of the Ozarks.

GEOGRAPHIC DISTRIBUTION, BOTANICAL INVENTORY, AND ENVIRONMENTAL FACTORS

INTRODUCTION

Because *Alliaria petiolata* (M.Bieb.) Cavara & Grande (garlic mustard) has not been studied in the southern Ozark Plateaus, which represents the lowest latitude at which a study of this plant has been conducted in North America, it is important to characterize infestations of the plant within the region, and to determine if *A. petiolata* is demonstrating the ability to alter pH and soil nutrients, as it has in the northeastern United States (Rodgers et al. 2008). The objectives of the present study are to (1) determine the current geographic distribution of *A. petiolata* within the southern Ozarks, (2) to characterize habitats and forest types invaded by the plant, (3) to measure species richness and diversity between the herbaceous layers of invaded and non-invaded plots, and (4) to determine if there are significant differences in soil pH and the levels of important soil nutrients between invaded and non-invaded study sites in the southern Ozarks.

METHODS AND MATERIALS

DISTRIBUTION OF ALLIARIA PETIOLATA

For the purposes of this study, the southern Ozark Plateaus region is defined as the region of the Ozark Highlands which occurs in Arkansas, Oklahoma, and the eight southernmost counties in Missouri that are located within the ecoregion - Barry, Howell, McDonald, Oregon, Ozark, Ripley, Stone, and Taney. In order to better understand the distribution and preferred habitat of *Alliaria petiolata* in the southern Ozark Plateaus, a specimen search was conducted at the University of Arkansas Herbarium (UARK), the largest herbarium in the state, and which also maintains a database of the vascular plant records of every other herbarium in the state; the Oklahoma Vascular Plants Database (OVPD), an online source of roughly 60 percent of the specimens housed in that state's herbaria; and the Missouri Botanical Garden's virtual herbarium, Tropicos.org. Fourteen records from eleven counties in Arkansas, southern Missouri and Oklahoma were found from these herbarium and database searches, and specimens represented collections that were made from 1972 to 2009. The farthest south it had been collected was 35.662426° N latitude, along the Mulberry River near Cass in Franklin County, Arkansas.

In addition to herbarium specimens, data compiled by botanists who conducted botanical surveys at the Buffalo National River in 2006 and 2007 were studied to find additional sites where *A. petiolata* had been observed. The author was also informed of the locations of two sites where *A. petiolata* was growing by a botanist at the Arkansas Natural Heritage Commission (Figure 3).

The sites of all of these records were then visited to determine if *A. petiolata* still occurred in those areas. When a previously recorded site was relocated, GPS coordinates of the site were recorded. After the search of known sites, a thorough search of new locations for the plant was conducted at public access points, trails, and, where permitted, on private property along and near major stream systems and upland areas within the southern Ozarks. These included the Buffalo, Current, Elk, Illinois, Jacks Fork, Kings, Mulberry, Spring, War Eagle, and White rivers, along with Lee and Spavinaw creeks. All six of the Level IV Ecoregions within the Boston Mountains and Ozark Highlands ecoregions (Woods et al. 2004) were surveyed. Additionally, public access points along the Arkansas River from Van Buren east to its confluence with the Mulberry River in the Arkansas Valley Level III Ecoregion (Woods et al. 2004), and public access points along the Current River in the Mississippi Alluvial Plain Level III Ecoregion (Woods et al. 2004) in Arkansas were surveyed. When *A. petiolata* was discovered at a new location, its coordinates were recorded with a GPS unit. A voucher specimen was also collected at each site that contained at least one adult plant, and an herbarium specimen was prepared and deposited at UARK.

STUDY SITES

Field work in study sites was conducted between March 2011 and November 2012. To be accepted for sampling, a site under consideration contained at least five adult plants, one hundred seedlings, and/or 15 rosettes of *A. petiolata* at the time of the initial survey but also contained an area of similar habitat that appeared to be completely free from infestation. Based upon these guidelines, only ten of the 60 surveyed sites were found to be suitable for study (Figure 4). At each site, two permanent 100 m² monitoring plots (10 m x 10 m or 5 m x 20 m, depending on the configuration of the site) were established at least 50 m apart, one within an area of *A. petiolata* infestation and one free from infestation, with two sides parallel to the hill slope (if present). Four 1 m² subplots were established within each quadrant of the 100 m² plot. Each pair of 100 m² plots was located on similar landform types determined using the 9-unit slope model developed by Dalrymple et al. (1968), shared the same aspect and topographic position (Table 1), and were known to possess similar soil types based on published records (Soil Survey Staff 2011, Table 2).

Each study site was described based primarily on the identities and frequencies of the most abundant canopy and sub-canopy trees, while also taking into account identifications of the dominant

herbaceous vegetation in the subplots and the physical features of the site. Each site was then assigned a classification code using a database maintained by the United States National Vegetation Classification (USNVC) Partnership, the USNVC Hierarchy Explorer (USNVC 2014).

BOTANICAL INVENTORY

Botanical inventories were conducted in the spring, summer and fall seasons of 2011 and 2012 to determine species richness, and cover classes of vegetation in the herbaceous layer were recorded in May of both years, using Daubenmire (1959) cover classes of trace-5%, 6-25%, 26-50%, 51-75%, and 96-100%. In the 100 m² plots species composition of the canopy, sub-canopy, shrub (1-5 m), and woody vine strata were documented, and cover classes were determined for each stratum. The diversity of the herbaceous layer within each study site in May of 2011 and 2012 was calculated using the Shannon-Wiener Diversity Index.

Species identifications were made in the field when possible. Unknown plants were collected, pressed and dried, then stored at UARK and later determined using *Keys to the Flora of Arkansas* (Smith 1994), *Flora of North America* (Flora of North America Editorial Committee 1993+), *Steyermark's Flora of Missouri* (Yatskievych 1999, 2006), and *Flora of Missouri* (Steyermark 1963). Nomenclature follows the *Atlas of the Vascular Plants of Arkansas* (Gentry et al. 2013).

A voucher specimen of *A. petiolata* was collected outside of each invaded plot and deposited at UARK. When *Alliaria petiolata* was encountered in the subplots, they were counted and recorded for each category (seedling, rosette, adult). When an adult plant was encountered, its height and number of siliques was counted and recorded.

ENVIRONMENTAL DATA

Five soil samples were collected randomly from each quadrant and the center of the plot, below the leaf and duff layer, to a depth of 10 cm. The samples were thoroughly homogenized and air dried, then sent to the Brookside Laboratories, Incorporated in New Knoxville, Ohio, for analysis. The 1:1 in H_2O method was used to measure soil pH levels of each sample. Sulfur, phosphorus, calcium, magnesium, potassium, sodium, boron, iron, manganese, copper, zinc, and aluminum were measured using the Mehlich 3 extraction method. Organic matter and estimated nitrogen release in each sample were measured using the loss on ignition method at 360 degrees Celsius (°C).

Slope and landform type were estimated and recorded, aspect was measured using a Silva Trekker 420 compass, and topographic position was determined using a Garmin GPSmap 60 CSx unit. Nonvegetative ground cover was recorded as percent bedrock, large rocks (>10 cm), small rocks, litter/duff, coarse woody debris (>10 cm diameter), and bare soil. At the center of each plot, leaf litter depth and soil depth to bedrock (or to 40 cm if bedrock was not reached) was measured. Photographs were taken in each cardinal direction from the center of each plot.

STATISTICAL ANALYSES

In the present study, the mean, minimum, maximum, standard error, and standard deviation were calculated using Microsoft Excel 2013, version 15. JMP version 11 statistical software (SAS Institute 2013) was used to calculate Pearson's correlation coefficient, construct simple linear regressions, and to perform t tests and repeated measures analyses. Variables were transformed when necessary to better fit the normality assumption.

RESULTS

DISTRIBUTION OF ALLIARIA PETIOLATA

The collection sites of the 14 *Alliaria petiolata* herbarium specimens were visited to ascertain the extent of current invasions. Only eight of the 12 original herbarium records could be relocated in the field. The four remaining sites were visited repeatedly during the course of the study to determine if plants had regenerated from the seed bank. At the end of the study, no plants had been found at those four locations (Appendix A).

All of the new occurrences of *A. petiolata* within the areas surveyed were located in the Ozark Highlands and the Boston Mountains ecoregions, in elevations ranging from 89 to 447 meters. The farthest south it was found during the surveys was 35.91391° N latitude, but this site was determined to be unsuitable for study. No plants were located in areas surveyed in the Arkansas Valley to the south or in the Mississippi Alluvial Plain to the east. In Oklahoma, the Ozark Plateaus were surveyed to the southwest along the Illinois River to Tahlequah, north to the town of Disney along Grand Lake, and west to Lake Hudson in Mayes County. No plants were found farther west than Disney. At the termination of the study, 62 occurrences of *A. petiolata* in 17 counties had been documented, either for the first time or relocated from previous records (Figure 5, Appendix A).

Prior to the present study, *A. petiolata* was known to occur in 11 counties within the southern Ozark Plateaus. These were Benton, Franklin, Izard, Marion, Newton, Searcy, Van Buren, and Washington counties in Arkansas, Barry and Howell counties in Missouri, and Delaware County in Oklahoma. As a result of this survey, six new counties were added to that list. These were Boone, Carroll, Madison, and Stone counties in Arkansas, McDonald County in Missouri, and Mayes County in Oklahoma (Table 3).

The sites of herbarium records taken for Izard and Franklin counties in Arkansas could not be relocated during the survey, despite repeated visits to search for plants in those areas. The location on the Franklin County herbarium specimen label reads, "South bank of the Mulberry River east of Fisher Ford near Rob Harmon cabin." I examined topographic maps and was not able to find that area. I then spoke with river guides at Turner Bend, and they were unaware of a place called Fisher Ford on the Mulberry River. The locality of the specimen may be an error, instead referring to Fisher Ford on the Illinois River in Benton County, a location where plants were found during the present study.

BOTANICAL INVENTORY

All of the monitoring plots were located in upland or riparian areas within the rugged and hilly Dissected Springfield Plateau-Elk River Hills Ecoregion, and were located at elevations ranging from 181 to 373 meters (Table 1). A total of 165 taxa of vascular plants were identified in the monitoring plots, representing 68 families (Appendix A). Forty taxa were trees, 13 were shrubs, 14 were vines, and 98 were herbs. Twenty-one of the taxa were identifiable only to genus, and three were identifiable only to family. Thirteen species (7.8%) are not native to the region, and 5 of those (3.0% of the total taxa) are classified as invasive species (Gentry et al. 2013). As the target species, *A. petiolata* was present in half of the study plots. *Euonymus fortunei* (Turcz.) Hand.-Mazz. (winter-creeper), *Microstegium vimineum* (Trin.) A. Camus (Japanese stilt grass), and *Perilla frutescens* (L.) Britton (beefsteak-plant) were present in a few of the invaded and non-invaded plots, and *Lonicera japonica* Thunb. ex Murray (Japanese honeysuckle), was only present in two invaded plots. A total of 128 taxa, 12 of which were introduced, from 51 families were associated with plots invaded by *A. petiolata*. A total of 115 taxa, 8 of which are introduced, from 55 families were associated with the plots that were not invaded by *A. petiolata*. No rare plants were identified in any of the monitoring plots.

Average species richness in invaded plots was 28.5 ± 4.8, and in non-invaded plots it was 24.5 ± 8.7. In nearly every case, species richness was greater in the invaded plots than in the non-invaded plots (Table 4, Figure 6). The exceptions to this were the Mt. Hersey and Tyler Bend sites, where species richness was greater in the non-invaded plots. The most commonly encountered species, with at least one individual present in more than half of the study plots, were, in order of descending dominance, *Parthenocissus quinquefolia* (L.) Planch (Virginia-creeper), *Stellaria media* (L.) Vill. (common chickweed), *Lindera benzoin* (L.) Blume (spicebush), *Acer rubrum* var. *rubrum*, *Lamium purpureum* L. (purple dead-nettle), *Acer negundo* L. (box-elder), *Symphoricarpos orbiculatus* Moench (coral-berry), and *Chaerophyllum procumbens* (L.) Crantz (spreading chervil).

Separate t tests were used to compare plant species richness and non-native plant cover in invaded and non-invaded plots. Log transformed non-native plant cover was significantly higher in invaded plots (mean = 3.04% ± 0.64) than it was in non-invaded plots (mean = 1.74% ± 0.73; p = 0.0349, Figure 7). The t test used to determine whether plant species richness was different between plots approached significance (p = 0.2223, Figure 8).

FOREST CLASSIFICATION, SPECIES RICHNESS, AND CANOPY COVER OF EACH STUDY SITE

The most common USNVC forest types encountered in the study sites were White Oak – Red Oak – Sugar Maple Mesic Forest (Faber-Langendoen 2006) and early-successional Box-Elder (American Sycamore, Eastern Cottonwood) Forest (Southeast Ecology Group 2004). Sites ranged from floodplains to mesic or dry-mesic forests (Table 5). The upland sites were largely composed of forests with an average of 55.0% cover, a sub-canopy with an average cover of 35.0%, and a shrub layer which contributed an average of 31.7% cover. The riparian sites had an average canopy, sub-canopy, and shrub cover of 65.0%, 61.0%, and 29.0%, respectively.

Lake Eucha I: Lake Eucha I, on the City of Tulsa's property in Delaware County, Oklahoma, is an Ozark Successional Honey-locust – Black Walnut Woodland (CEGL004345, Foti and Pyne 2010), a closed-canopy ruderal forest with 80 percent cover on a level floodplain situated at the mouth of Spavinaw Creek at Lake Eucha. The site is cut in half by an access road and turnabout to the creek, and a well-traveled four-wheel drive trail lies just to the east of the invaded plot. *Juglans nigra* L. (black walnut) and *Celtis laevigata* Willd. (sugarberry) dominate in the canopy, with lesser amounts of *Carya*

cordiformis (Wangenh.) K. Koch (bitternut hickory), and *Acer negundo*, *Ulmus alata* Michx. (winged elm), *Aesculus glabra* Willd. (Ohio buckeye), *Morus rubra* L. (red mulberry), and *Maclura pomifera* (Raf.) C. K. Schneid. (Osage-orange) in the sub-canopy. The shrub layer varies from completely absent in the non-invaded plot to about 25% cover in the invaded plot and is composed of *Symphoricarpos orbiculatus* with lesser amounts of *Lindera benzoin*. The thick (nearly 82% percent cover) herbaceous layer in the non-invaded plot is dominated by the grasses *Chasmanthium latifolium* (Michx.) H. O. Yates (river-oats) and *Elymus virginicus* L. var. *virginicus* (Virginia wild rye) and by a more sparse and diverse assemblage of herbs and tree seedlings, notably *Tradescantia ernestiana* E. S. Anderson & Woodson (Ernest's spiderwort), *Lamium purpureum*, *Viola pubescens* Aiton (downy yellow violet), and *Alliaria petiolata* in the invaded plot. Overall plant species richness is 46 at this site, 39 of which are native. In the invaded plot, species richness was 39, whereas in the non-invaded plot, species richness was 19.

Lake Eucha II: The Lake Eucha II site is also on the City of Tulsa's property but represents an upland Ozark forest with well-drained, shallow, rocky soil. The slope is somewhat steep with a northwest aspect and is classified as a White Oak - Red Oak - Sugar Maple Mesic Forest (CEGL002058). Primary canopy trees are *Quercus rubra*, *Q. alba, Carya cordiformis*, and *C. tomentosa*, which provide 65 percent cover. The sub-canopy is 50 percent and is dominated by *Acer saccharum*, with lesser amounts of *Q. alba* and *Fraxinus americana*. *Lindera benzoin*, *Symphoricarpos orbiculatus*, *Acer saccharum*, and *Ostrya virginiana* are present in the shrub layer. In the invaded plot, the herbaceous layer in the early spring was completely covered with *Erythronium rostratum* W. Wolf (yellow trout-lily), *Lamium purpureum*, *Chaerophyllum procumbens*, *Stellaria media*, and *Podophyllum peltatum* L. (May-apple), and later with *Alliaria petiolata*, and *Parthenocissus quinquefolia*. The herbaceous layer of the non-invaded plot was much sparser, with the only notable taxa being *Carex* sp. and *Parthenocissus quinquefolia*, with only a few individuals of *Enemion biternatum* Raf. (false rue-anemone), *Erythronium rostratum*, and *Cardamine concatenata* (Michx.) O.Schwarz (toothwort). This site has low species richness with only 35 species, 30 of which are native. The invaded plot contained 24 species, and the non-invaded plot had 16.

Spavinaw Creek: The Spavinaw Creek site in Delaware County, Oklahoma is Box-Elder (American Sycamore, Eastern Cottonwood) Forest (CEGL004690), a mature closed-canopy floodplain forest with 70 percent canopy cover along the creek for which the site is named. This site is owned by the

City of Tulsa and lies between Lake Eucha and Spavinaw Lake, which together serve at the city's water supply. The site is also frequented by off-road vehicle users who ride along the river on trails through the woods. The canopy dominant is *Acer negundo*, followed in abundance by *Platanus occidentalis* L. (sycamore), *Celtis laevigata*, and *Juglans nigra*. The sub-canopy is composed of *Acer negundo* and *Celtis laevigata*. The sparse shrub layer is of *Lindera benzoin*. Throughout the site, a mix of the grasses *Chasmanthium latifolium*, *Diarrhena americana* P. Beauv (American beakgrain), and *Elymus virginicus* var. *virginicus* dominate the herbaceous layer, but in the invaded plot *Tradescantia ohiensis* Raf. (Ohio spiderwort), *A. petiolata*, *Chaerophyllum procumbens*, *Stellaria media*, and *Enemion biternatum* are also common. Species richness at this site was intermediate when compared with the other sites, with 38 total species, 32 of which are native. Species richness in the invaded plot was 29 and 23 in the non-invaded plot.

   Gravette: The Gravette site is on private property in Benton County, Arkansas and is situated on a lower, southeast-facing, moderately steep to steep slope in a narrow valley along Rocky Dell Hollow Road. The area is deeply incised into several narrow ravine, and *Alliaria petiolata* is well-established throughout, mainly along the lower slopes of the ravines, but is also advancing in some areas into the upland parts of the site. The forest is a mature White Oak - Red Oak - Sugar Maple Mesic Forest (CEGL002058). The canopy provides about 60 percent cover and is composed mainly of *Quercus alba* L. (white oak), with lesser amounts of *Quercus rubra* L. (northern red oak). The sub-canopy is composed of *Fraxinus americana* L. (white ash), *Cornus florida* L. (flowering dogwood), *Ulmus rubra* Muhl. (slippery elm), and *Carya cordiformis*. The shrub layer is dense and includes *Castanea pumila* (L.) Mill. var. *ozarkensis* (Ashe) G.E. Tucker (Ozark chinquapin), *Lindera benzoin*, *Ostrya virginiana* (Mill.) K. Koch (hop-hornbeam), and *Sassifras albidum*. The herbaceous layer is very sparse, with *Alliaria petiolata*, *Galium tricornutum* Dandy (rough corn bedstraw), and *Polystichum acrostichoides* (Michx.) Schott (Christmas fern) the most notable species present in the invaded plot and *Desmodium nudiflorum* (L.) DC*. (naked-flower tick-trefoil), *Parthenocissus quinquefolia* dominating the non-invaded plot. Species richness was intermediate related to other sites, with 32 of the 35 species native. Species richness was comparable in the invaded and non-invaded plots, with 23 and 20 species identified, respectively.

Baker Ford: The forest at the Baker Ford site, an access point to the Buffalo National River in Newton County, Arkansas, is best identified as an Interior Mid- to Late-Successional Sweetgum-Oak Forest (CEGL007217, Evans and Nordman 2005), a transitional forest type that is typical following disturbance of mixed oak-hickory forests. This east- to southeast-facing moderately steep slope has 55 percent canopy cover, and is principally composed of *Liquidambar styraciflua* L. (sweet-gum), with *Quercus falcata* Michx. (southern red oak) also present in the invaded plot. The sub-canopy, which has about 50 percent cover, is mainly composed of *Ulmus americana* L. (American elm) and *Cornus florida* in the invaded plot, and *Tilia americana* L. var. *americana* (American basswood) and *Sassafras albidum* (Nutt.) Nees (sassafras) in the non-invaded plot. This was the only site in which *Ptelea trifoliata* L. (wafer-ash) was identified. The most dominant species of herbaceous layer of the invaded plot were *A. petiolata*, *Podophyllum peltatum*, and *Symphoricarpos orbiculatus*. In the non-invaded plot, the most common species were *Desmodium glutinosum* (Muhl. ex Willd.) A.W. Wood (tick-trefoil), *Toxicodendron radicans* (L.) Kuntze (poison-ivy), and *Phegopteris hexagonoptera* (Michx.) Fée (broad beech fern). Overall species richness at the site was 48, with 42 of those native. Species richness in the invaded plot was 34, and 26 in the non-invaded plot.

Decatur: The Decatur site is located on private property in Benton County, Arkansas on heavily forested upland midslopes of the Spavinaw Creek valley, and is classified as a mature White Oak - Red Oak - Sugar Maple Mesic Forest (CEGL002058). The slopes are moderate with a northwest-facing aspect. The principal canopy trees are mature *Quercus rubra* with *Q. falcata*, which provide 60 percent cover, and sub-canopy is composed of *Carya cordiformis*, *Acer rubrum*, and *Ostrya virginiana*. *Fraxinus americana* is also widespread in the shrub layer and sub-canopy throughout the site, but in the study plots occurs only in the shrub layer. *Acer saccharum* is also present, but only as seedlings in the herbaceous layer. Other members of the shrub layer, which covers about 30 percent of the plots, are *Cornus florida*, *Lindera benzoin*, and *Quercus muehlenbergii* Engelm. (chinquapin oak). Herbaceous layer dominants include *Alliaria petiolata* and *Parthenocissus quinquefolia* in the invaded plot, and *P. quinquefolia, Geranium maculatum* L. (wild geranium), *Stellaria media*, and *Desmodium nudiflorum* in the non-invaded plot. Overall species richness at the site is 38, four of which are non-native. Species richness in the invaded plot was 26, and 21 in the non-invaded plot.

Pineville: The Pineville site is located in a flat, frequently flooded valley along an unnamed tributary of the Elk River near Pineville, Missouri. The forest is best classified as a River Birch - American Sycamore Small River Floodplain Forest (CEGL002086, Landaal and Gawler 2008), despite the fact that neither species was recorded in the study plots. They were likely present in the forest, but were not recorded because they were outside of the study plots. Other defining characteristics of this ecological association are deep, alluvial soils, frequent flooding, and the presence of both bottomland and upland terrace tree species which were identified at the site (NatureServe 2014). The forest is dense, with 65 percent canopy cover from the dominant species *Quercus alba*, *Juglans nigra*, and *Celtis laevigata*. *Nyssa sylvatica* Marshall (black-gum), *Acer negundo*, *Carya cordiformis*, and *Morus rubra* are present in the understory and provide 70 percent cover. The shrub layer is composed solely of *Symphoricarpos orbiculatus*, and the herbaceous layer is dominated by *Stellaria media* and *Lamium purpureum* in early spring, and by *Podophyllum peltatum*, *Alliaria petiolata*, and *Parthenocissus quinquefolia* in the invaded plot, and *Elymus virginicus* var. *virginicus* and *Glechoma hederacea* L. (ground-ivy) in the non-invaded plot in the summer and into fall. Overall species richness at the site was 30, with 26 native species, and 22 in the invaded plot and 15 in the non-invaded plot.

Mt. Hersey: The Mt. Hersey site is located in mesic forest within the boundaries of the Buffalo National River in Searcy County, Arkansas. The two plots established at the site were determined to have two separate forest type classifications. The invaded plot, close in proximity to the Buffalo River on a high, gently sloping, northwest-facing terrace, is Sycamore – Silver Maple Calcareous Floodplain Forest (CEGL007334, Faber-Langendoen and Patterson 2004). The overstory here is *Platanus occidentalis* providing 45 percent cover, with a sparse understory of *Ulmus americana* and shrub layer of *Lindera benzoin* and *Symphoricarpos orbiculatus*. The non-invaded plot, farther away from the river on a west-facing, moderately steep upland slope, is a White Oak - Mixed Oak Dry-Mesic Alkaline Forest (CEGL002070, Guetersloh et al. 2005). The overstory is composed of *Quercus rubra* and *Q. muehlenbergii* with 65 percent cover, an understory of *Carya cordiformis*, *Juniperus virginiana* var. *virginiana*, and *Ulmus americana* with 30 percent cover, and a shrub layer of *Viburnum prunifolium*, *Cercis canadensis* L. var. *canadensis* (eastern redbud) and *Lindera benzoin*. The herbaceous layers of both plots were similar, except for the presence of *Alliaria petiolata* in the invaded plot. This site was very

botanically diverse, but the main herbaceous layer dominants were *Boehmeria cylindrica* (L.) Sw. (false nettle), *Impatiens capensis* Meerb. (spotted jewelweed), *Parthenocissus quinquefolia*, and *Alliaria petiolata* in the invaded plot, and *Stellaria media* and *Ranunculus micranthus* Nutt. in Torr. & A. Gray (rock buttercup). *Sanguinaria canadensis* L. (bloodroot) and *Cypripedium* L. sp. were also present in smaller numbers. This site was the most species-rich, with 61 species recorded, 58 of which are native. This site was also an exception in that the invaded plot contained less species than the non-invaded plot. In the invaded plot, 30 species were recorded, whereas 43 species were identified in the non-invaded plot.

Hasty: The Hasty site is located on the Buffalo National River in Newton County, Arkansas, and also represents two distinct forest types. The invaded plot is on a highly disturbed gently east-facing terrace slope of the Buffalo River, with deep, alluvial soils. The classification of this plot is Box-Elder (American Sycamore, Eastern Cottonwood) Forest (CEGL004690). The canopy is composed of *Acer negundo* and *Quercus muehlenbergii* and provides about 45 percent cover. The sub-canopy, which is denser at 75 percent cover, is composed of *Nyssa sylvatica* and *Morus rubra*, and the shrub layer is of thick patches of *Arundinaria gigantea* (Walter) Muhl. (river cane) with *Celtis laevigata* and *Asimina triloba* (L.) Dunal (pawpaw). The non-invaded plot, also in the floodplain but further upslope, is classified as Sweetgum (White Oak, Sugar Maple)/American Hornbeam/Northern Spicebush Forest (CEGL007826). *Liquidambar styraciflua* dominates in both the canopy and sub-canopy with 55 and 25 percent cover, respectively, but *Juniperus virginiana* var. *virginiana* and *Pinus echinata* are also present in small amounts in the sub-canopy. Shrubs include *Acer negundo*, *Lindera benzoin*, *Ulmus alata*, and *Quercus muehlenbergii*. *Laportea canadensis* (L.) Wedd. (wood-nettle), *Verbesina alternifolia* (L.) Britton ex Kearney (yellow ironweed), and *Parthenocissus quinquefolia* were notable in the herb layer of the invaded plot, and *Carex jamesii* Schwein. (James' sedge) and *Toxicodendron radicans* are prominent in the herbaceous layer, with *Microstegium vimineum*, *Parthenocissus quinquefolia*, *Poa sylvestris* A. Gray (woodland blue grass), and *Chaerophyllum procumbens* also present. Species richness was intermediate to high at this site, with 47 species recorded, 41 of which are native. The invaded plot contained 30 species and the non-invaded plot contained 28 species.

Tyler Bend: The Tyler Bend site is situated along the north side of a seasonally wet creek at the Buffalo National River in Searcy County, Arkansas. The narrow, flat to gently sloping site is bordered on one side by the creek and on the other by a substantial dry-stack stone wall that spans nearly one half of a mile along a park-maintained trail and the creek. The forest is classified as Box-Elder (American Sycamore, Eastern Cottonwood) Forest (CEGL004690) and is dominated by a canopy and sub-canopy of Acer negundo with 70 percent cover, and a shrub layer of *Ulmus rubra*, *Viburnum prunifolium*, and *Lindera benzoin*. The herbaceous layer is sparse with much bare ground, but notable species are *Laportea canadensis*, *Alliaria petiolata*, *Sanicula canadensis* L. (Canadian black-snakeroot), and *Chaerophyllum procumbens*, in the invaded plot, and *Cryptotaenia canadensis* (L.) DC. (honewort), *Laportea canadensis*, *Elymus virginicus* var. *virginicus*, and *Parthenocissus quinquefolia* in the non-invaded plot, and *Stellaria media* and *Lamium purpureum* in both plots in the early spring. Total species richness at the site was 43, with 37 of those native. The invaded plot was less species rich than the non-invaded plot at this site, with 30 and 34 species recorded, respectively.

VEGETATION COVER

The cover of canopy, sub-canopy, shrub, woody vine, and herbaceous strata in invaded and non-invaded plots in spring of 2011 are given in Table 6. Average cover was determined by using the midpoint of each Daubenmire (1959) class's range of cover (1 = 2.5%; 2 = 15.0%; 3 = 37.5%; 4 = 62.5%; 5 = 85%; 6 = 97.5%). In the canopy, invaded plots had an average cover of 60.0%, and non-invaded plots had an average cover of 62.0%. The composition of the riparian canopy was very different from the upland canopy. The most common tree in riparian plots was *Acer negundo* (n = 4), followed by *Celtis laevigata* (n = 3), and *Juglans nigra* (n = 3). The most common trees in the upland plots were *Quercus rubra* (n = 4), followed by *Q. alba* (n = 2), *Q. falcata* (n = 2), and *Q. muehlenbergii* (n = 2). Average cover in the invaded sub-canopy was 50.0%, and 41.0% in non-invaded sub-canopy. *Acer negundo* was the most common tree in the riparian sub-canopy (n = 6), followed by *C. laevigata* (n = 2) and *Ulmus alata* (n = 2). Upland sub-canopy dominants were *Carya cordiformis* (n = 3), *U. americana* (n = 3), and *A. saccharum* (n = 2). In the shrub layer, invaded plots averaged 31.0% cover, and non-invaded plots averaged 19.5% cover. The most frequently encountered shrub was *Lindera benzoin* (n = 12), followed by *Symphoricarpos orbiculatus* (n = 5) and *Viburnum prunifolium* (n = 2). The rest of the plants in the shrub layer were young

trees that had not yet reached the sub-canopy or canopy. Average cover in the vine layer of invaded plots was 8.0%, and 7.75% in the non-invaded plots. *Parthenocissus quinquefolia* was the most common vine (n = 8), followed by *Toxicodendron radicans* (n = 5) and *Vitis* spp. (n = 5).

Cover in the herbaceous layer was nearly equal between invaded (62.2%) and non-invaded (63.2%) plots. The site with the highest cover in the herbaceous layer was Mt. Hersey, an upland forest site which had an average of 93.5% cover in the spring of 2011. The total non-native vegetation cover in the invaded plots averaged 24.7%, and averaged only 2.0% in non-invaded plots. In May 2011, *A. petiolata* ranged in cover from 15.5% to 90.5%, with an average cover of 53.9%, and in May 2012 it ranged in cover from 7.5% to 65.0%, with an average cover of 31.0%. In 2011, it was the fourth most common plant in the herbaceous layer in the invaded study plots, and in 2012 it was the sixth most common in those plots. Table 7 shows the rank of *A. petiolata* in relation to nine of the other most common plants occupying the herbaceous layer in 2011 and 2012.

The Shannon-Wiener Diversity Index (H) for the herbaceous layer in each plot in 2011 and 2012 is shown in Table 8. The diversity of the herbaceous layer in invaded plots was higher in 2011 (H = 2.12) and 2012 (H = 1.92) than it was in non-invaded plots in 2011 (H = 2.05) and 2012 (H = 1.74).

SOIL CHARACTERISTICS

The properties measured on soil samples in invaded and non-invaded plots in 2011 and 2012 are presented in Tables 9 and 10, respectively, along with minimum, maximum, mean, standard deviation, and standard error of measurements taken for pH, organic matter, and each nutrient. Table 11 shows the same descriptive statistics of invaded and non-invaded plots for both years combined. Soil pH values for both years in invaded and non-invaded plots were slightly acid to neutral, ranging from 6.2 to 6.9 with an average value of 6.6 in invaded plots and 6.1 to 7.1 with an average value of 6.5 in non-invaded plots. Soil organic matter was nearly the same between invaded and non-invaded plots, with an average of 8.5% and 8.6%, respectively.

A repeated measures design was utilized to compare the response variables (pH, cation exchange capacity, organic matter, and nutrients) over time in invaded and non-invaded plots. The data for calcium, magnesium, and sodium were log transformed, and the data for boron were log 10 transformed, to better fit the normality assumption. P values for each nutrient by status (invaded or non-

invaded), year (2011 and 2012), and the interaction of status and year are listed in Table 12. Cation exchange capacity was significantly higher in invaded plots than it was in non-invaded plots (p = 0.017). Calcium levels (p = 0.0346) were higher in invaded plots than they were in non-invaded plots. Boron levels (p = 0.001) in both invaded and non-invaded plots were significantly higher in 2012 than they were in 2011. No other significant differences were found for the remaining variables. Graphs showing the interaction of status and time on pH, cation exchange capacity, organic matter, and each nutrient are shown in Appendix B.
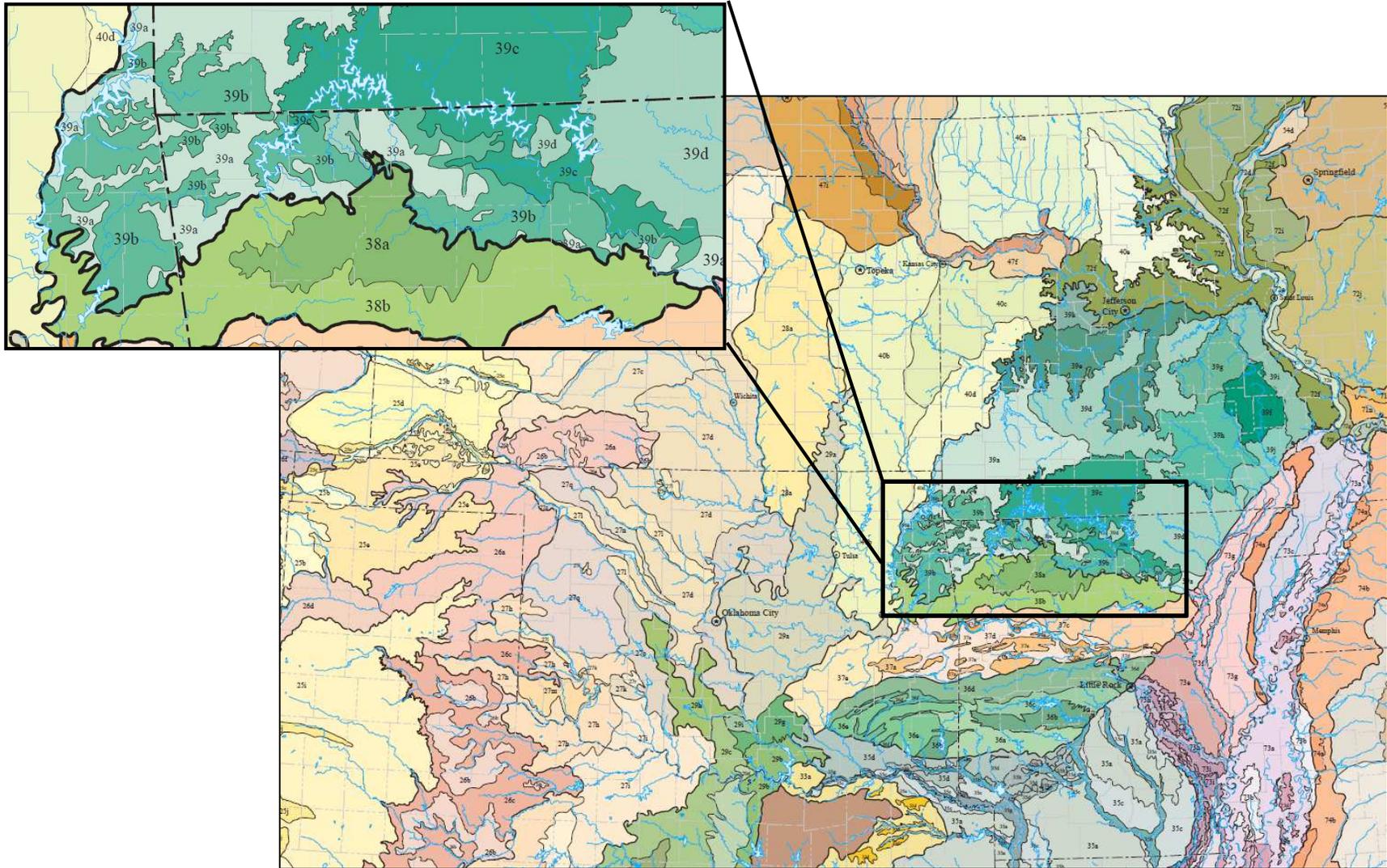
Figure 1. The Dissected Springfield Plateau-Elk River Hills Ecoregion (labeled as 39b on inset map). Modified from *Level III and IV Ecoregions of the Conterminous United States* (www.epa.gov/wed/pages/ecoregions.htm).
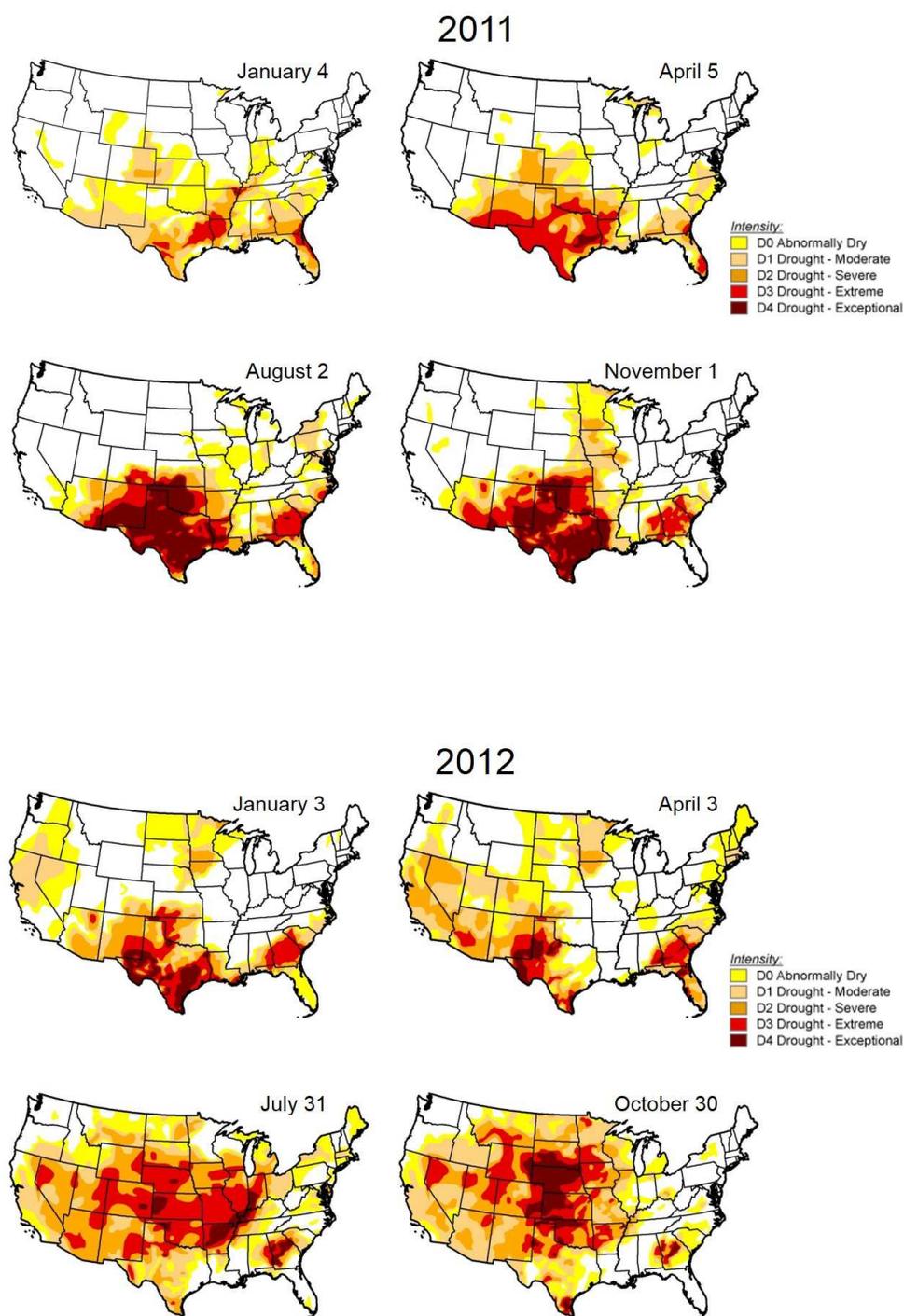
Figure 2. Drought conditions in the United States in 2011 and 2012. The U.S. Drought Monitor is produced in partnership between the National Drought Mitigation Center at the University of Nebraska-Lincoln, the United States Department of Agriculture, and the National Oceanic and Atmospheric Administration. Maps courtesy of NDMC-UNL.
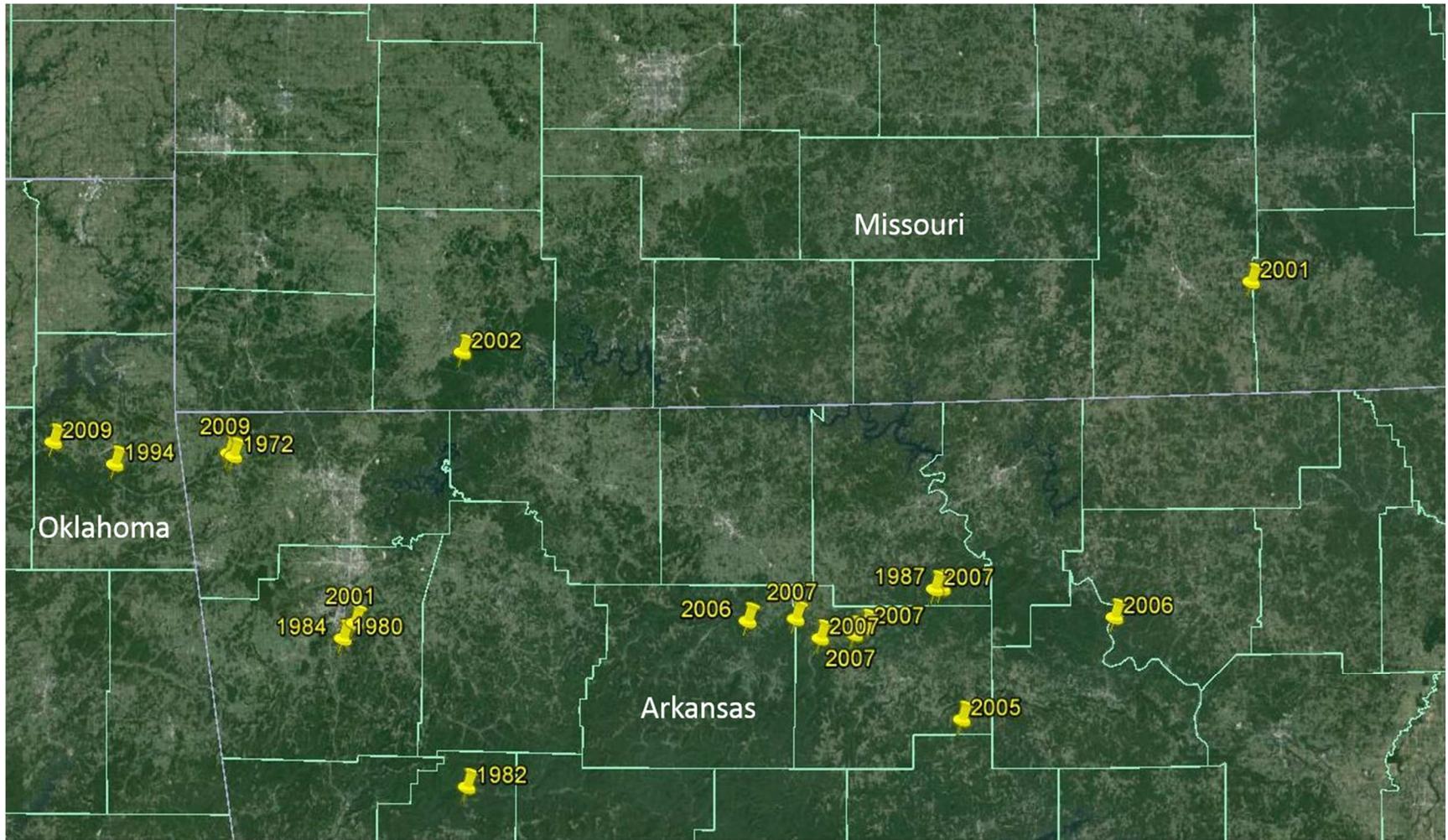
Figure 3. Location and year of each occurrence of *Alliaria petiolata* known to occur in the southern Ozark Plateaus prior to the current study. Information was derived from herbarium specimens, databases, data sheets, and first-hand accounts.
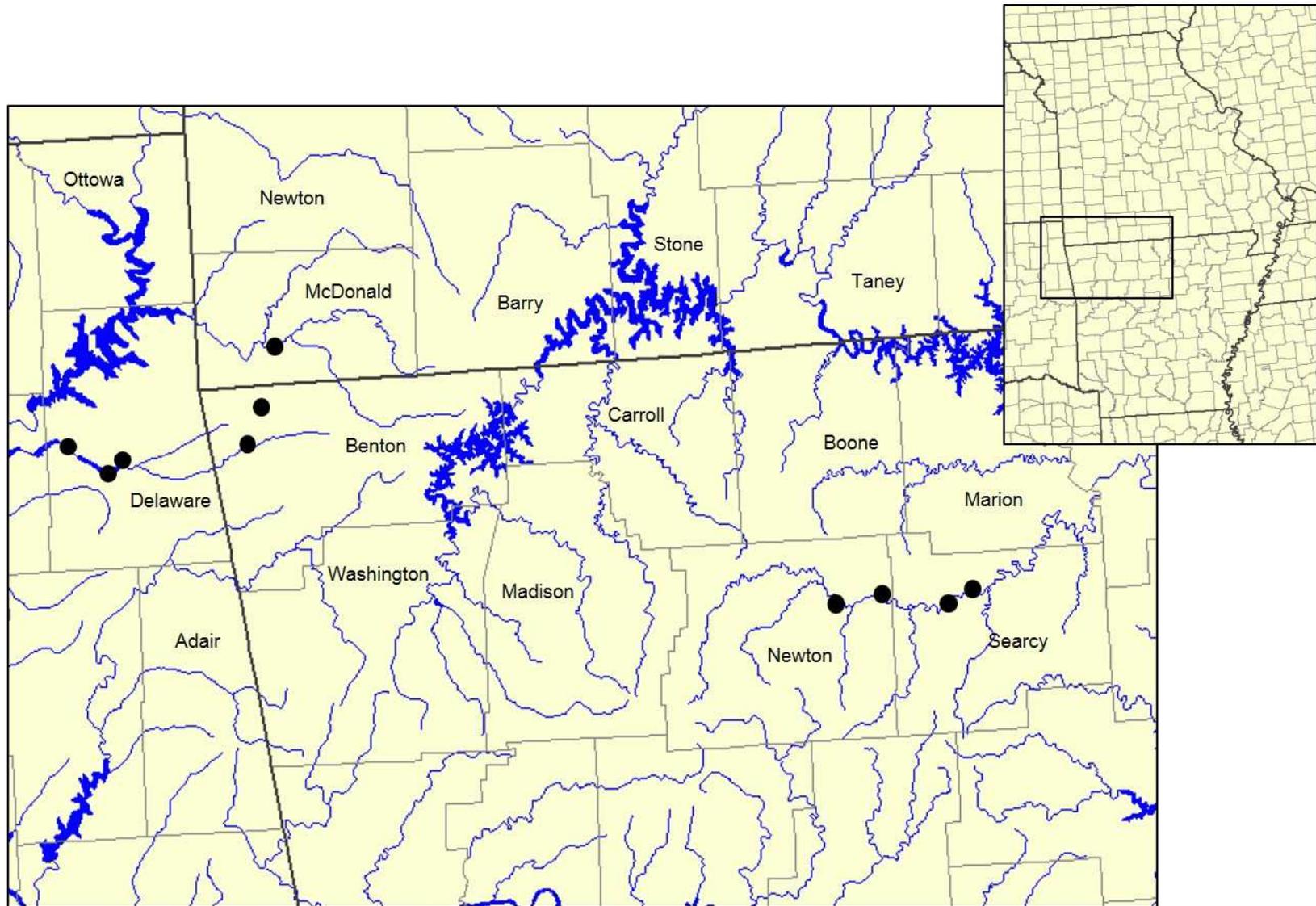
Figure 4. Study sites in Benton, Newton, and Searcy counties in Arkansas, McDonald County in Missouri, and Delaware County in Oklahoma. Base maps created using Map Maker at www.nationalatlas.gov.

Table 1. Location and topographic position of each 100 m² plot, showing similarities in elevation, slope and aspect between each paired plot. Note: A = invaded plot. B = non-invaded plot.

ID	Site Name	County, ST	Plot	Latitude	Longitude	Elevation (m)	Slope (%)	Aspect (º)	Habitat Type
E1	Lake Eucha 1	Delaware, OK	A	36.35544	-94.79408	244	Flat	Flat	Riparian Forest
			B	36.35500	-94.79491	228	Flat	Flat	
E2	Lake Eucha 2	Delaware, OK	A	36.34692	-94.82281	269	26-49	310	Upland Forest
			B	36.34661	-94.82835	261	26-49	315	
SP	Spavinaw Creek	Delaware, OK	A	36.40146	-94.96426	221	Flat	Flat	Riparian Forest
			B	36.40233	-94.96515	212	Flat	Flat	
GR	Gravette	Benton, AR	A	36.43073	-94.43856	373	26-49	130	Upland Forest
			B	36.43103	-94.43842	356	10-25	100	
BF	Baker Ford	Searcy, AR	A	35.98087	-92.81424	193	10-25	80	Upland Forest
			B	35.98146	-92.81401	221	10-25	140	
DE	Decatur	Benton, AR	A	36.37910	-94.48449	350	10-25	280	Upland Forest
			B	36.37951	-94.48460	342	10-25	320	
PI	Pineville	McDonald, MO	A	36.57127	-94.40910	279	Flat	Flat	Riparian Forest
			B	36.57315	-94.41288	282	Flat	Flat	
MH	Mt. Hersey	Newton, AR	A	36.01035	-92.95161	228	1-9	300	Upland Forest
			B	36.01068	-92.95125	239	1-9	310	
HA	Hasty	Newton, AR	A	36.00752	-93.08160	245	1-9	70	Riparian Forest
			B	36.00736	-93.08198	246	1-9	70	
TB	Tyler Bend	Searcy, AR	A	35.98126	-92.75320	181	1-9	110	Riparian Forest
			B	35.98093	-92.75362	181	1-9	Flat	

Table 2. Soil series codes and descriptions of each study sites. Note: A = invaded plot. B = non-invaded plot. Soil Survey Staff, Natural Resources Conservation Service, United States Department of Agriculture. Web Soil Survey. Available online at http://websoilsurvey.nrcs.usda.gov/.

ID	Plot	County, ST	Soil Series Code	Soil Series Description	Soil Texture
E1	A/B	Delaware, OK	Es	Elsah very gravelly loam, 0-3% slopes, frequently flooded	Very gravelly loam
E2	A/B	Delaware, OK	ClF	Clarksville stony silt loam, 20-50% slopes	Very stony silt loam
SP	A/B	Delaware, OK	Es	Elsah very gravelly loam, 0-3% slopes, frequently flooded	Very gravelly loam
GR	A/B	Benton, AR	NoF	Noark very gravelly silt loam, 20-45% slopes	Very gravelly silt loam
BF	A/B	Searcy, AR	38EF	Moko-Rock outcrop complex, 15-50% slopes	Very stony silt loam
DE	A B	Benton, AR	CvF Btc	Clarksville extremely gravelly silt loam, 12-50% slopes Britwater gravelly silt loam, 3-8% slopes	Extremely gravelly silt loam Gravelly silt loam
PI	A B	McDonald, MO	71754 76755	Waben-Cedargap, occasionally flooded complex, 2-5% slopes Cedargap, frequently flooded-Gladden, occasionally flooded complex, 0-2% slopes	Extremely gravelly silt loam Extremely gravelly loam
MH	A/B	Newton, AR	18	Estate-Lily-Portia complex, 20-40% slopes	Stony fine sandy loam
HA	A/B	Newton, AR	17	Estate-Lily-Portia complex, 8-20% slopes	Stony fine sandy loam
TB	A/B	Searcy, AR	30	Healing silt loam, occasionally flooded	Silt loam

Table 3. Counties in which *Alliaria petiolata* is known to occur in the southern Ozark Plateaus. Counties appearing in bold represent new records found during the course of the present study.

Arkansas	Missouri	Oklahoma
Benton	Barry	Delaware
Boone	Howell	**Mayes**
Carroll	**McDonald**	
Franklin		
Izard		
Madison		
Marion		
Newton		
Searcy		
Stone		
Van Buren		
Washington		
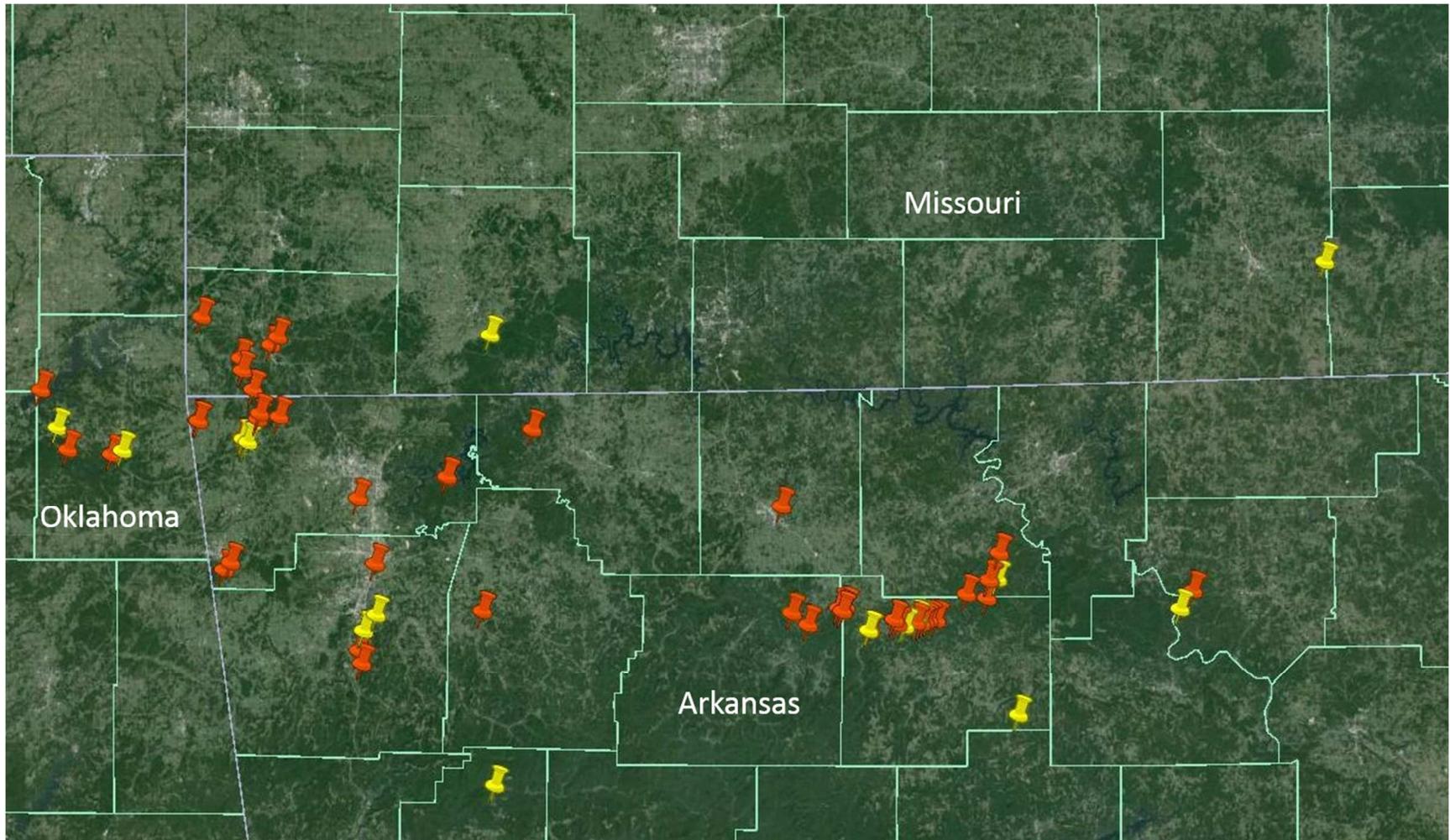
Figure 5. Locations of *Alliaria petiolata* documented prior to the current study (yellow pins) and during the course of the current study (orange pins). A list of the GPS coordinates for each pin is in Appendix A.

Table 4. Species richness of each study site.

ID	Habitat	Total Species	Native Species	Non-Native Species	Invaded Plots	Non-Invaded Plots
E1	Riparian Forest	46	39	7	37	19
E2	Upland Forest	35	30	5	24	16
SP	Riparian Forest	38	32	6	29	23
GR	Upland Forest	35	32	3	23	20
BF	Upland Forest	48	42	6	34	26
DE	Upland Forest	38	34	4	26	21
PI	Riparian Forest	30	26	4	22	15
MH	Upland Forest	61	58	3	30	43
HA	Riparian Forest	47	41	6	30	28
TB	Riparian Forest	43	37	6	30	34
Minimum		30	26	3	22	15
Maximum		61	58	6	34	43
Mean		42.1	37.1	5	28.5	24.5
Standard Deviation		8.9	8.9	1.4	4.8	8.7
Standard Error		2.8	2.8	0.4	1.5	2.7
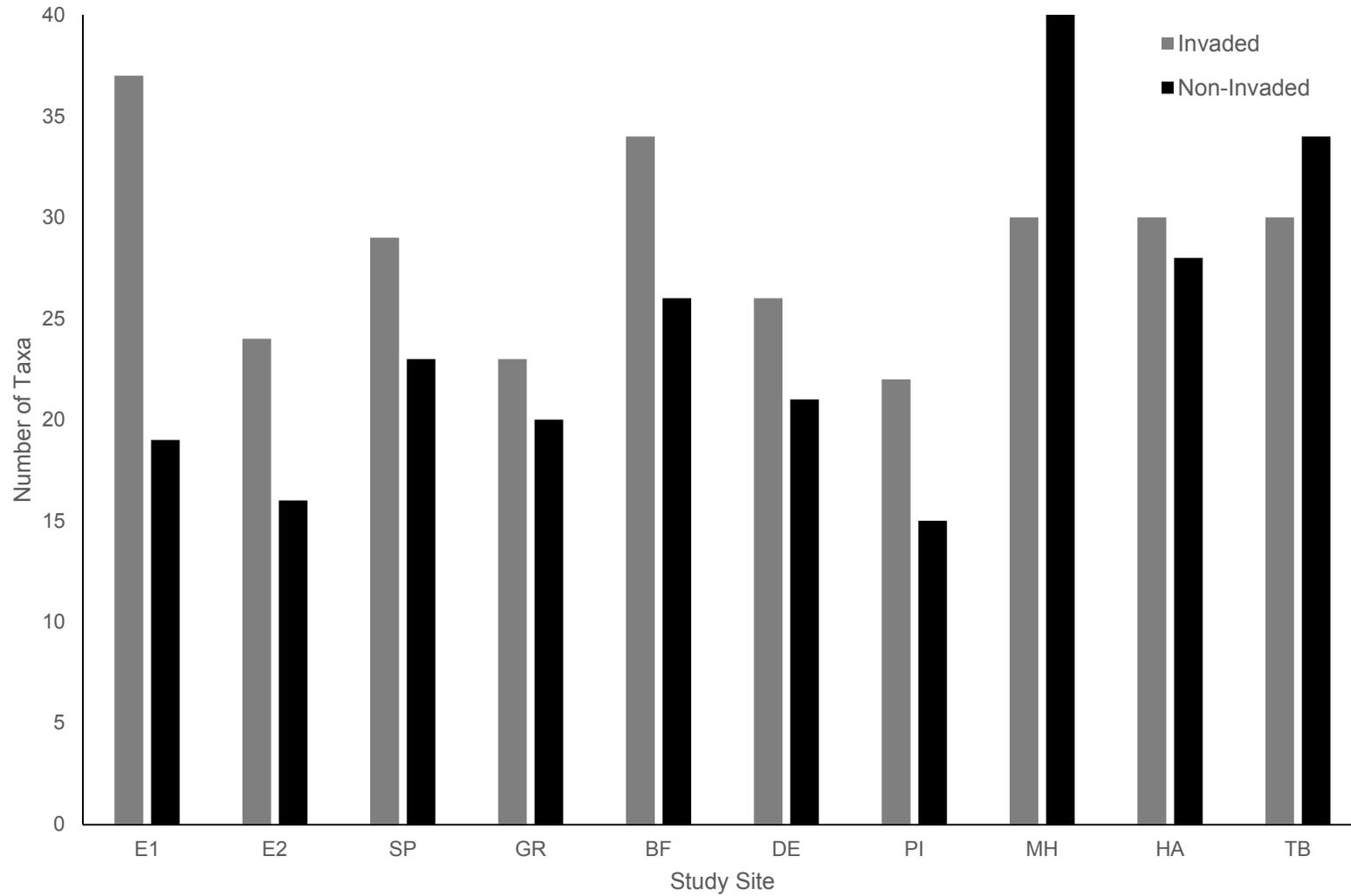
Figure 6. Species richness in invaded and non-invaded plots.
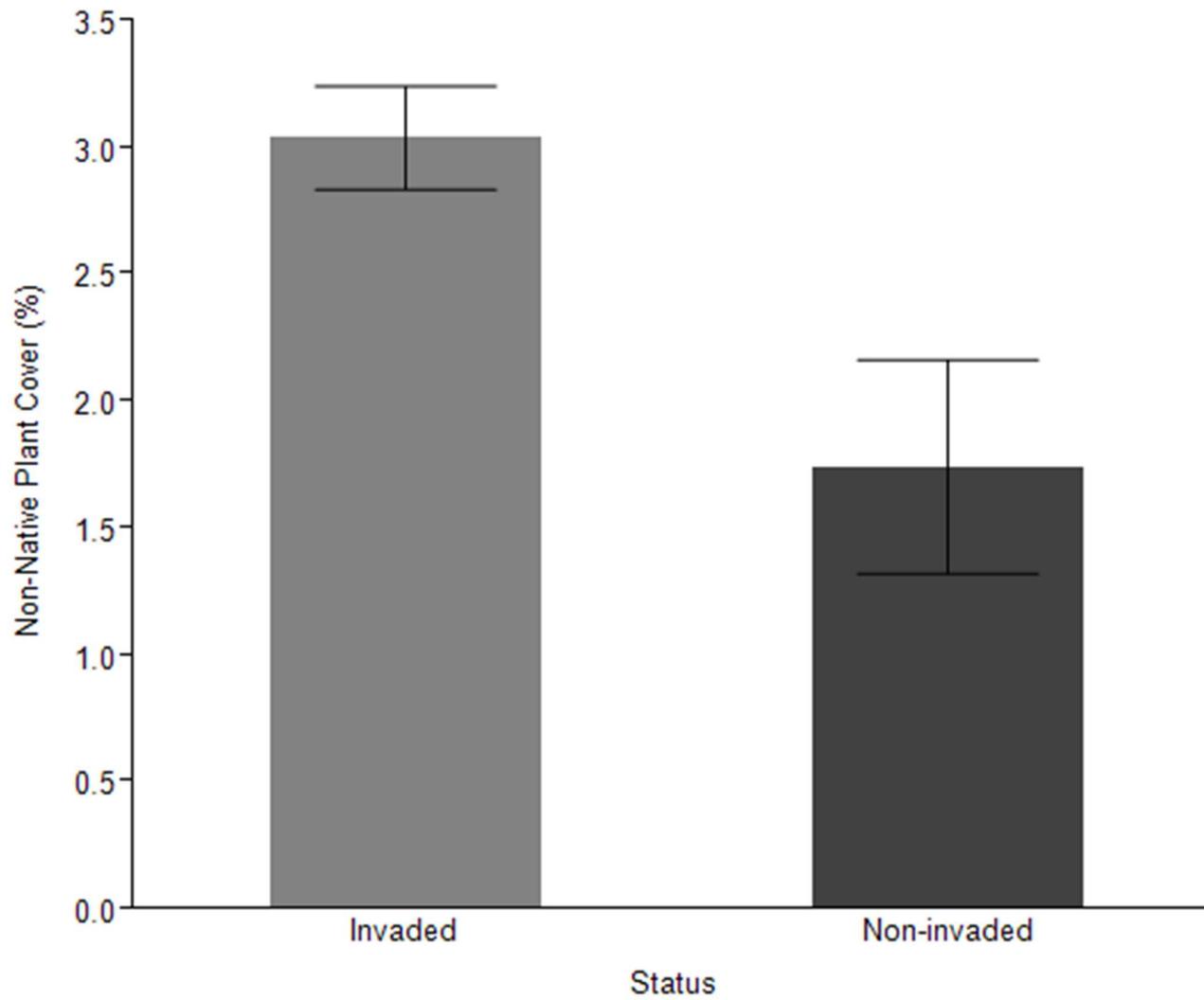
Figure 7. Cover of non-native plants (log transformed) in invaded and non-invaded plots.
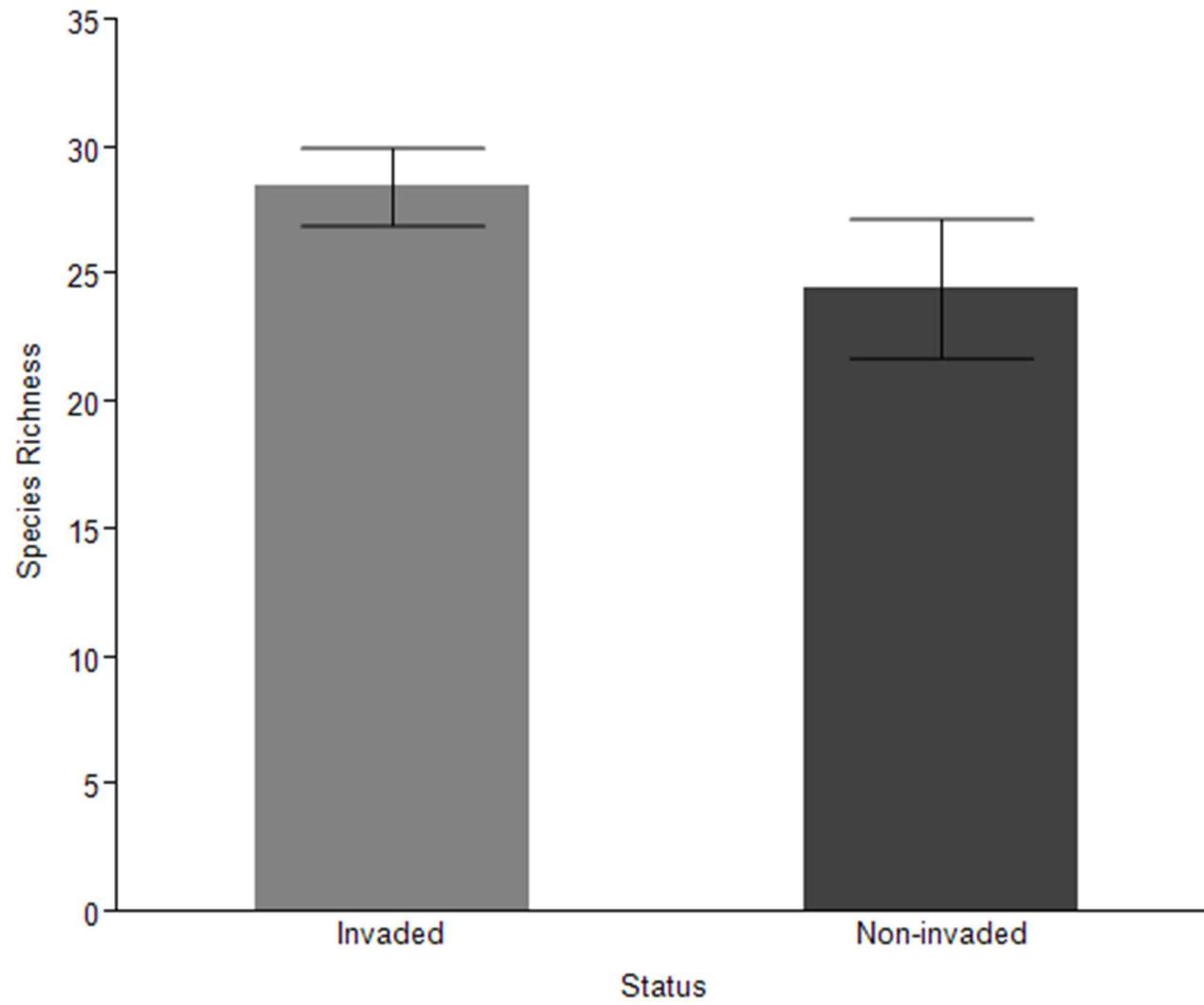
Figure 8. Species richness in invaded and non-invaded plots.

Table 5. Forest types of study sites using United States National Vegetation Classification (USNVC) codes and descriptions.

Site	USNVC Code	Common Name
E1	CEGL004345	Ozark Successional Honey-locust – Black Walnut Woodland
E2	CEGL002058	White Oak - Red Oak - Sugar Maple Mesic Forest
SP	CEGL004690	Box-Elder (American Sycamore, Eastern Cottonwood) Forest
GR	CEGL002058	White Oak - Red Oak - Sugar Maple Mesic Forest
BF	CEGL007217	Interior Mid- to Late-Successional Sweetgum-Oak Forest
DE	CEGL002058	White Oak - Red Oak - Sugar Maple Mesic Forest
PI	CEGL002086	River Birch - American Sycamore Small River Floodplain Forest
MH (Invaded)	CEGL007334	Sycamore – Silver Maple Calcareous Floodplain Forest
MH (Non-invaded)	CEGL002070	White Oak – Mixed Oak Dry-Mesic Alkaline Forest
HA (Invaded)	CEGL004690	Box-Elder (American Sycamore, Eastern Cottonwood) Forest
HA (Non-invaded)	CEGL007826	Sweetgum - (White Oak, Sugar Maple)/American Hornbeam/Northern Spicebush Forest
TB	CEGL004690	Box-Elder (American Sycamore, Eastern Cottonwood) Forest

Table 6. The estimated cover of plants in each stratum in the invaded (Inv) and non-invaded (Non) study plots in spring of 2011.

Site	Habitat	Canopy		Sub-canopy		Shrub		Vine		Herbaceous	
		Inv	Non	Inv	Non	Inv	Non	Inv	Non	Inv	Non
E1	Riparian Forest	85	75	55	25	25	0	15	0	46	20
E2	Upland Forest	55	75	35	65	15	25	0	0	41	84
SP	Riparian Forest	75	65	85	15	35	15	0	0	68	76
GR	Upland Forest	55	65	35	35	55	25	0	0	47	67
BF	Upland Forest	45	65	65	35	35	15	15	15	72	96
DE	Upland Forest	75	45	45	75	25	35	2.5	15	69	35
PI	Riparian Forest	65	65	75	65	35	15	2.5	15	51	44
MH	Upland Forest	45	55	15	45	35	35	15	15	94	93
HA	Riparian Forest	45	55	75	25	25	15	15	2.5	77	43
TB	Riparian Forest	55	55	15	25	25	15	15	15	56	75
Mean		60.0	62.0	50.0	41.0	31.0	19.5	8.0	7.8	62.2	63.2
Minimum		45	45	15	15	15	0	0	0	41	20
Maximum		85	75	85	75	55	35	15	15	94	96
Riparian Mean		65.0	63.0	61.0	31.0	29.0	12.0	9.5	6.5	59.7	51.5
Upland Mean		55.0	60.0	35.0	46.7	31.7	25.0	7.9	10.0	63.2	75.1

Table 7. The 10 most common herbaceous plants in invaded study plots in May 2011 and May 2012, with cover (%) and relative cover (%) of each species. Note: ** denotes an invasive species.

	May 2011			May 2012		
Rank	Name	Cover	Relative Cover	Name	Cover	Relative Cover
1	*Laportea canadensis*	68.0	6.34	*Chasmanthium latifolium*	46.3	5.31
2	*Podophyllum peltatum*	61.5	5.74	*Podophyllum peltatum*	36.3	4.16
3	*Tradescantia ohiensis*	55.5	5.18	*Toxicodendron radicans*	35.0	4.02
4	**Alliaria petiolata***	**53.2**	**4.96**	*Diarrhena americana*	33.8	3.88
5	*Impatiens capensis*	50.0	4.66	*Laportea canadensis*	33.8	3.88
6	*Polystichum acrostichoides*	50.0	4.66	**Alliaria petiolata***	**31.0**	**3.56**
7	*Parthenocissus quinquefolia*	28.6	2.67	*Tradescantia ohiensis*	30.0	3.45
8	*Carex* sp.	25.0	2.33	*Parthenocissus quinquefolia*	27.8	3.19
9	*Chaerophyllum procumbens*	25.0	2.33	*Euonymus fortunei***	25.0	2.87
10	*Boehmeria cylindrica*	24.4	2.27	*Polystichum acrostichoides*	25.0	2.87

Table 8. Values for the Shannon-Wiener Diversity Index of the herbaceous layer in each plot in 2011 and 2012, with the mean values of invaded and non-invaded plots for each year.

Site	Habitat	2011		2012	
		Invaded	Non-Invaded	Invaded	Non-Invaded
E1	Riparian Forest	2.44	1.90	2.78	1.29
E2	Upland Forest	1.58	1.32	1.81	1.40
SP	Riparian Forest	2.53	2.20	2.12	2.20
GR	Upland Forest	2.19	1.50	1.92	1.61
BF	Upland Forest	2.33	2.58	2.29	1.58
DE	Upland Forest	1.71	1.55	1.91	2.12
PI	Riparian Forest	2.10	1.28	1.74	1.10
MH	Upland Forest	2.42	2.70	2.37	2.26
HA	Riparian Forest	2.21	2.39	2.03	2.30
TB	Riparian Forest	1.68	1.81	1.54	1.51
Mean		2.12	1.92	2.05	1.74

Table 9. Nutrient levels, cation exchange capacity (CEC), pH, and percent organic matter in soils of invaded and non-invaded plots in 2011, with minimum, maximum, mean, standard deviation, and standard error. CEC was measured in meq/100g, estimated nitrogen release was measured in pounds/acre; sulfur was measured in ppm; all other nutrients were measured in mg/kg.

Invaded Plot	pH	CEC	OM	N	P	K	Ca	Mg	S	Fe	B	Cu	Mn	Na	Zn	Al
E1	6.8	21.81	8.5	118	53	201	3666	147	14	116	1.2	4.6	369	18	13.8	555
SP	6.9	15.73	3.7	87	19	106	2405	290	11	115	1.1	2.7	330	16	7.9	375
GR	6.4	17.52	12.0	126	37	127	2723	125	13	139	0.6	4.5	246	20	7.2	477
BF	6.5	17.32	6.2	106	44	159	2630	182	12	193	0.9	4.6	259	22	9.6	450
DE	6.2	14.41	11.9	126	44	155	1964	195	15	146	0.6	2.9	244	20	7.3	565
PI	6.2	22.45	9.8	124	68	207	3338	152	15	115	1.2	3.1	342	23	9.2	476
MH	6.4	22.87	7.0	110	33	141	3364	289	12	230	1.2	5.1	310	18	9.8	460
HA	6.7	27.01	3.2	82	25	90	4624	116	10	102	1.0	2.0	200	47	7.3	169
TB	6.7	40.28	11.9	126	48	185	7003	117	16	118	1.2	4.2	298	26	12.1	194
E2	6.5	18.22	8.2	116	104	258	2777	160	15	117	0.6	4.3	480	18	8.3	807
Minimum	6.2	14.4	3.2	82	19	90	1964	116	10	102	0.6	2.0	200	16	7.2	169
Maximum	6.9	40.3	12.0	126	104	258	7003	290	16	230	1.2	5.1	480	47	13.8	807
Mean	6.5	21.8	8.2	112.1	48	163	3449	177	13	139	1.0	3.8	308	23	9.2	453
Standard Deviation	0.2	7.5	3.2	16.2	24	51	1454	65	2	41	0.3	1.0	80	9	2.2	184
Standard Error	0.1	2.4	1.0	5.1	7.7	16.1	459.7	20.4	0.6	13.0	0.1	0.3	25.2	2.8	0.7	58.0

Table 9. (Cont.)

Non-Invaded Plot	pH	CEC	OM	N	P	K	Ca	Mg	S	Fe	B	Cu	Mn	Na	Zn	Al
E1	6.8	15.8	5.8	104	27	135	2677	92	14	199	0.7	7.3	248	24	9.3	513
SP	7.1	15.09	4.7	97	64	99	2375	265	14	166	1.1	7.6	449	23	11.6	414
GR	6.1	13.62	14.4	127	48	129	1915	130	13	148	0.7	4.6	149	19	6.0	551
BF	6.4	17.05	8.6	118	103	158	2481	208	12	192	0.7	4.8	226	27	5.6	625
DE	6.4	15.62	10.0	125	39	212	2169	235	15	106	0.8	3.3	322	19	4.5	814
PI	6.5	25.2	9.8	124	61	265	3906	214	17	89	1.5	4.1	400	19	13.1	443
MH	6.5	12.19	4.0	90	43	118	1768	173	12	120	0.8	2.7	301	22	5.4	571
HA	6.5	13.37	5.9	105	42	120	1922	194	11	148	0.8	2.3	377	40	3.3	544
TB	6.7	35.28	14.5	127	38	176	6075	133	18	112	1.3	5.0	284	22	12.2	228
E2	6.6	15.42	8.0	115	59	252	2391	129	13	111	0.8	2.8	385	18	4.9	757
Minimum	6.1	12.2	4.0	90	27	99	1768	92	11	89	0.7	2.3	149	18	3.3	228
Maximum	7.1	35.3	14.5	127	103	265	6075	265	18	199	1.5	7.6	449	40	13.1	814
Mean	6.6	17.9	8.6	113.2	52	166	2768	177	14	139	0.9	4.4	314	23	7.6	546
Standard Deviation	0.3	7.1	3.7	13.4	21	58	1309	55	2	38	0.3	1.9	91	6	3.6	168
Standard Error	0.1	2.2	1.2	4.2	6.7	18.5	413.9	17.5	0.7	11.9	0.1	0.6	28.9	2.1	1.1	53.1

Table 10. Nutrient levels, cation exchange capacity (CEC), pH, and percent organic matter in soils of invaded and non-invaded plots in 2012, with minimum, maximum, mean, standard deviation, and standard error. CEC was measured in meq/100g, estimated nitrogen release was measured in pounds/acre; sulfur was measured in ppm; all other nutrients were measured in mg/kg.

Invaded Plot	pH	CEC	OM	N	P	K	Ca	Mg	S	Fe	B	Cu	Mn	Na	Zn	Al
E1	6.6	23.86	9.7	124	69	186	3929	130	14	136	1.3	3.3	314	18	12.3	599
SP	6.8	18.54	5.0	100	20	167	2860	277	11	115	1.5	3.2	363	22	8.3	363
GR	6.5	17.7	13.1	127	23	116	2850	105	12	131	1.0	2.6	232	20	6.5	321
BF	6.6	23.94	8.0	115	27	159	3677	294	12	230	1.9	5.9	323	25	11.2	436
DE	6.3	12.9	11.0	126	35	124	1845	151	17	146	1.1	1.9	181	19	5.3	547
PI	6.8	32.49	4.7	97	18	97	5752	113	9	108	1.5	2.4	241	16	8.0	170
MH	6.9	37.36	9.0	120	50	224	6667	133	14	143	2.8	7.1	386	23	15.0	351
HA	6.6	16.91	5.6	103	55	178	2563	205	11	221	1.1	4.6	232	23	8.8	520
TB	6.4	19.03	11.8	126	58	257	2837	170	14	142	0.8	2.0	266	24	9.1	560
E2	6.5	19.82	8.9	119	93	312	3025	164	13	111	0.8	3.4	430	16	9.2	792
Minimum	6.3	12.9	4.7	97	18	97	1845	105	9	108	0.8	1.9	181	16	5.3	170
Maximum	6.9	37.4	13.1	127	93	312	6667	294	17	230	2.8	7.1	430	25	15.0	792
Mean	6.6	22.3	8.7	115.7	45	182	3601	174	13	148	1.4	3.6	297	21	9.4	466
Standard Deviation	0.2	7.5	2.9	11.5	25	67	1503	66	2	43	0.60	1.73	80	3	2.83	175
Standard Error	0.1	2.4	0.9	3.6	7.8	21.1	475.3	20.7	0.7	13.6	0.2	0.5	25.2	1.0	0.9	55.3

Table 10. (Cont.)

Non-Invaded Plot	pH	CEC	OM	N	P	K	Ca	Mg	S	Fe	B	Cu	Mn	Na	Zn	Al
E1	6.6	18.16	6.3	106	36	234	2929	97	15	201	1.1	8.6	271	34	10.4	511
SP	6.7	19.48	6.3	107	63	167	2953	287	16	151	1.5	5.3	444	24	11.9	375
GR	6.3	8.57	18.3	129	47	121	1170	114	14	213	0.8	2.9	99	28	6.5	663
BF	6.3	8.74	2.8	75	35	96	1201	125	10	141	1.0	2.2	271	19	3.9	620
DE	6.4	18.31	10.9	125	35	179	2623	251	14	95	1.1	4.4	379	18	4.7	801
PI	6.5	12.35	4.8	98	40	131	1746	198	11	144	1.3	2.2	304	23	3.0	599
MH	6.7	29.35	11.6	126	24	209	4988	129	14	136	2.3	6.4	327	23	11.4	401
HA	6.4	13.15	8.5	117	100	150	1890	167	13	185	1.1	2.8	201	20	3.6	736
TB	6.7	23.37	9.7	123	66	408	3655	214	15	98	1.3	4.7	385	26	14.6	453
E2	6.5	12.63	7.9	115	49	221	1875	127	11	115	0.8	2.1	318	15	4.6	785
Minimum	6.3	8.6	2.8	75	24	96	1170	97	10	95	0.8	2.1	99	15	3.0	375
Maximum	6.7	29.4	18.3	129	100	408	4988	287	16	213	2.3	8.6	444	34	14.6	801
Mean	6.5	16.4	8.7	112.1	50	192	2503	171	13	148	1.2	4.2	300	23	7.5	594
Standard Deviation	0.2	6.6	4.3	16.4	22	88	1189	64	2	41	0.43	2.16	98	5	4.21	155
Standard Error	0.05	2.09	1.37	5.20	6.96	27.91	375.88	20.27	0.63	12.92	0.14	0.68	31.12	1.73	1.33	49.10

Table 11. The minimum, maximum, mean, standard deviation, and standard error of nutrient levels, cation exchange capacity (CEC), percent organic matter, and pH for both years combined in soils of invaded and non-invaded plots. CEC was measured in meq/100g, estimated nitrogen release was measured in pounds/acre; sulfur was measured in ppm; all other nutrients were measured in mg/kg.

Invaded Plots	pH	CEC	OM	N	P	K	Ca	Mg	S	Fe	B	Cu	Mn	Na	Zn	Al
Minimum	6.2	12.9	3.2	82.0	18	90	1845	105	9	102	0.6	1.9	181	16	5.3	169
Maximum	6.9	40.3	13.1	127.0	104	312	7003	294	17	230	2.8	7.1	480	47	15.0	807
Mean	6.6	22.0	8.5	113.9	46.2	172.5	3525.0	175.8	13.0	143.7	1.2	3.7	302.3	21.7	9.3	459.4
Standard Deviation	0.2	7.3	3.0	13.8	23.8	58.6	1441.3	63.4	2.1	41.2	0.5	1.4	77.7	6.7	2.5	174.7
Standard Error	0.1	2.3	0.9	4.4	7.5	18.5	455.8	20.0	0.7	13.0	0.2	0.4	24.6	2.1	0.8	55.2
Non-Invaded Plots	**pH**	**CEC**	**OM**	**N**	**P**	**K**	**Ca**	**Mg**	**S**	**Fe**	**B**	**Cu**	**Mn**	**Na**	**Zn**	**Al**
Minimum	6.1	8.6	2.8	75.0	24	96	1170	92	10	89	0.7	2.1	99	15	3.0	228
Maximum	7.1	35.3	18.3	129.0	103	408	6075	287	18	213	2.3	8.6	449	40	14.6	814
Mean	6.5	17.1	8.6	112.7	51.0	179.0	2635.5	174.1	13.6	143.5	1.1	4.3	307.0	23.2	7.5	570.2
Standard Deviation	0.2	6.7	3.9	14.6	21.1	74.0	1224.5	58.3	2.1	38.5	0.4	2.0	92.7	5.9	3.8	159.3
Standard Error	0.1	2.1	1.2	4.6	6.7	23.4	387.2	18.4	0.7	12.2	0.1	0.6	29.3	1.9	1.2	50.4

Table 12. Calculated p values of pH, cation exchange capacity, organic matter, and nutrients, showing significance levels of differences in invaded and non-invaded plots (status), year, and the interaction of status and year. Significant p values appear in bold. The data for calcium, magnesium, and sodium were log transformed, and the data for boron were log 10 transformed.

Independent Variable	pH	CEC	OM	N	P	K	Ca	Mg	S	Fe	B	Cu	Mn	Na	Zn	Al
Status	0.718	**0.017**	0.905	0.812	0.564	0.783	**0.035**	0.951	0.440	0.992	0.608	0.387	0.897	0.951	0.179	0.111
Year	0.854	0.699	0.706	0.753	0.663	0.261	0.710	0.642	0.302	0.374	**0.001**	0.588	0.445	0.642	0.995	0.429
Status*Year	0.276	0.565	0.838	0.556	0.988	0.875	0.488	0.859	1.000	0.988	0.754	0.875	0.922	0.859	0.868	0.648

SURVIVORSHIP AND REPRODUCTIVE SUCCESS OF *ALLIARIA PETIOLATA* IN THE SOUTHERN OZARKS

INTRODUCTION

Within its introduced range, *Alliaria petiolata* behaves as an obligate biennial with three distinct life stages: seedling, rosette, and adult (Nuzzo 2000). Seeds fall from adult plants beginning in May (personal observation) and are dormant at maturity, requiring a period of cold-stratification to germinate (Cavers et al. 1979). In the southern Ozarks, seedlings emerge in late winter and develop into rosettes by early spring (personal observation). Plants overwinter as rosettes and in early spring of the second year send up a reproductive stalk, developing flowers in April and fruits by May. Siliques begin to shatter in mid-May (personal observation) and seeds fall within a few feet of the parent plant. Adult plants die after the release of mature seeds (Cavers et al. 1979).

Many studies have been conducted in the Midwest and northeastern United States to determine survivorship and reproductive success of *A. petiolata* (Cruden and McClain 1996, Byers and Quinn 1998, Evans and Landis 2007). In the southern Ozarks, the lowest latitude at which the plant has been studied in North America, such data have not been collected. The following questions were addressed during the course of this study in an attempt to quantify survivorship and reproductive success of this invasive species in the region.

- What is the survivorship of *A. petiolata* from seedling to adult?
- How tall are plants in the southern Ozark Plateaus, and how many siliques are produced by each plant?
- What is the reproductive output of *A. petiolata*?
- Is reproductive output related to rosette size?
- Does the reproductive success of *A. petiolata* vary between riparian and upland habitats?
- How do these data compare to studies conducted in other regions of the United States?

METHODS

To estimate survivorship of *Alliaria petiolata* in the southern Ozarks, all individuals at each life stage (seedling, rosette, and adult plant) were counted each time a plant was encountered within a subplot. Seedlings were counted in spring of 2011, rosettes were counted in fall of 2011 and spring of

2012, and adults were counted in summer of 2012. When an adult plant was encountered in the subplots, its height was measured and the number of siliques produced were counted and recorded.

To estimate the reproductive success of *A. petiolata* and to determine whether or not a relationship exists between rosette size or habitat to this success, 50 rosettes were tagged and several variables were measured during the spring and summer of 2012 at three upland and two floodplain sites. A 50 m transect was established near each of five of the invaded permanent plots, perpendicular to the slope (if present) and groups of ten rosettes were delimited at 5 m intervals on alternating sides of the transect. The diameter of each rosette was measured, the number of leaves was counted, the width of each leaf was measured, and the length of the longest petiole was measured. After the plants bolted and attained full height but before siliques released their seeds, plant height was measured and stems were counted. Plants were then collected from the site and taken to the laboratory where siliques were counted, then the seeds were removed from siliques and counted. Maturity of seeds was determined visually by size, color, and the characteristic longitudinal ridges that mature seeds of *A. petiolata* possess. Seeds that appeared to be immature were not included in the seed count.

In the statistical analyses performed on the data, values for mature seeds were log transformed, numbers of stems were 2nd root transformed, and numbers of siliques were 4th root transformed to better fit the normality assumption. The variables height and rosette diameter were not transformed. A correlation matrix using Pearson's coefficient (r) was used to determine which variables were dependent on rosette diameter. A value above 0.70 indicated a strong correlation between variables and was considered to show a strong positive relationship. Simple linear regression was calculated to determine whether seed production can be predicted based on rosette size. A t test was used to determine whether or not rosette size varies in different habitat types.

RESULTS

The results of the survivorship study are presented in Table 13, which shows the number of seedlings counted in 2011, the number of rosettes that survived the winter, and the number of those rosettes that reached reproductive maturity in 2012. A total of 1,939 seedlings were counted in the invaded study plots in 2011. Only 136 of those, or 7.0%, survived to reproductive age. The range of mortality was between 73.3% and 99.8%. The plots that experienced the highest mortality were Lake

Eucha 1, Decatur, and Tyler Bend, which had 99.8%, 99.5%, and 98.8% mortality, respectively. The sites that experienced the lowest mortality were Hasty, Baker Ford, and Lake Eucha 2, which had 18.6%, 73.3%, and 75.0% mortality, respectively. The riparian sites contained a higher average number of seedlings than the upland sites, and overall mortality was also slightly lower in that habitat type.

The average density of all plants at each life stage is also presented in Table 13. In spring of 2011, seedlings averaged 48.5 ± 231.6 plants/m^2. The following spring, in 2012, rosettes averaged 3.7 ± 17.9 plants/m^2. By fall of 2012, adults averaged 3.4 ± 18.7 plants/m^2. In riparian habitats the average number of plants/m^2 at each life stage was greater than in upland habitats, with 58.8 ± 252.8 seedlings, 5.3 ± 24.7 rosettes, and 5.0 ± 25.7 adult plants/m^2 in those plots versus 38.2 ± 229.2 seedlings, 2.2 ± 4.4 rosettes, and 1.9 ± 5.2 adult plants/m^2 in upland plots.

In 2011, 32 adult plants were counted and measured in the subplots. In 2012, 136 adult plants were counted and measured. The average height of all plants in both years was 66.2 cm, and the average number of siliques produced was 20.6. The tallest plants were at the Hasty site, where three plants measured 109 cm tall. The shortest plant was 18 cm tall, at the Gravette site. The most siliques present on any plant was 127, on a plant at the Mt. Hersey site. Several plants produced only 1 silique.

The data collected for the reproductive success study are presented in Table 14. Of the 50 rosettes that were tagged and measured, 48 survived to adulthood. Two additional plants were excluded because height and/or leaf width data were not recorded. All plants contained at least one silique (but two plants produced siliques that contained only immature seeds). A total of 1,607 siliques were counted, containing a total of 19,491 seeds. Each plant produced an average of 33.5 ± 45.6 siliques, and each silique contained an average of 12.1 mature seeds. Plants produced an average of 406 ± 670.8 mature seeds each. The highest number of seeds produced by an individual was 3301, on a plant occurring in a riparian habitat with a rosette diameter of 36 cm, an adult height of 109 cm, 6 stems, and 211 siliques. This plant was the largest in terms of rosette diameter, produced the second highest number of stems, and produced the most siliques of any plant sampled. The lowest number of mature seeds produced was zero, on a single-stemmed plant in an upland habitat with a rosette diameter of 14 cm and an adult height of 17 cm, with one stem and one silique.

Plants in riparian habitats were taller, produced more stems, and developed an average of three times more seeds (79.4 ± 20.5 cm, 2.8 ± 0.7 stems, and 680.5 ± 984.3 seeds) than plants in upland habitats (64.4 ± 19.9 cm tall, 1.5 ± 1.0 stems, and ± 226.1 319.0 seeds). Rosette diameter was very similar between habitats, with riparian plants averaging 16.6 cm wide and upland plants averaging 15.3 cm wide, but the number of leaves produced by plants was greater in riparian plots (36.3, versus 19.1 in upland habitats). Plants frequently produced only one stem (n = 29), but several produced 4 or more stems (n = 10). Ten of the 19 adult plants sampled in riparian habitats produced two or more stems (53.6%), whereas only eight of the 30 plants in upland habitats produced two or more stems (26.7%).

All of the variables measured in the field were positively correlated at 0.67 or above (Table 15). A simple linear regression calculated to predict reproductive output based on rosette size proved to be significant (F = 76.33, p = 0.0001), with an R^2 of 0.634 (Figure 9). A t test performed to compare reproductive success in each habitat resulted in a fairly small p value (p = 0.048). Plants produce more seeds in riparian habitats (mean seed production = 5.6) than in upland habitats (mean seed production = 4.8, Figure 10).

DISCUSSION

Since its discovery in the Ozark Plateaus Physiographic Province in the early 1970s, *Alliaria petiolata* has become an established member of the forest understory in many areas within the region. Herbarium specimens and other reliable data sources inform us that its spread was initially slow but has increased over time. It was documented from the region only seven times in eight counties from 1972 to 2000. Then from 2001 to 2009, *A. petiolata* was documented 13 times from three new counties within the southern Ozarks.

The present study was initiated in 2010, and at the conclusion, *A. petiolata* had been documented more than 60 times in 17 counties. The recent increase in reports of the plant is largely due to the work I conducted to find it in as many locations as possible in the region, but is also due to new reports by botanists who work regularly in the southern Ozarks of Arkansas and have documented its presence in areas where it had not previously been reported. Although the Welk et al. (2002) bioclimatic model predicted that the southern Ozarks was outside of the optimal invasion range of *A. petiolata*, the plant has clearly demonstrated an ability to successfully colonize this region over the past 43 years. However, it is

not likely that it will become established south of the Ozarks because the significant cold period needed for seeds to break dormancy may not be met in regions to the south.

Prior to the current study, the habitats in which *A. petiolata* was documented in the southern Ozarks were primarily riparian forests. It is now known that the plant occurs in upland forests almost as commonly as it does in riparian ones, and it was even documented growing in full sun at two locations – along a roadside at Baptist Ford on the White River in Washington County, Arkansas and in a cleared fencerow near Mount Pleasant in Benton County, Arkansas (see Appendix A). However, plants were more frequently found in forested areas, and individuals were more robust in riparian forests than they were in upland forests within the region. Plants growing in the riparian study plots were taller (79.4 ± 20.5 cm) and produced more stems (2.8 ± 2.3), siliques (54.1 ± 66.3), and seeds (680.5 ± 984.3) than plants growing in upland plots (height = 64.4 ± 19.9 cm; stems = 1.5 ± 1.0; siliques = 20 ± 20.4; seeds = 226.1 ± 319.0; see Table 14). A study conducted in southeastern Ohio found that plants showed similar differences in upland and lowland habitats (Meekins and McCarthy 2001), with plants in lowland plots growing an average of 20 cm taller than those in upland plots, as well as producing higher root, fruit, and total biomass, and more mature fruits and seeds per plant.

Population densities of *A. petiolata* were also higher at each life stage in riparian plots than they were in upland plots (see Table 13). Similar findings have been reported by other investigators. *Alliaria petiolata* benefits from natural or anthropogenic disturbances (Cavers et al. 1979, Meekins and McCarthy 2001), and one explanation for finding higher densities in riparian areas is that in areas of low disturbance, such as a more stable upland forest that does not experience regular flooding events, *A. petiolata* populations keep "a low profile" (Nuzzo 1999), maintaining small population sizes, but are capable of quickly increasing in size following a disturbance. Overall, population densities of *A. petiolata* were much lower in the study plots than they have been reported in other regions of the U.S. In Illinois, seedling density reached 5,080/m^2 (Nuzzo 2000), whereas in the present study, the most seedlings occurring in any study plot was 625 in a single square meter, with an overall average of 48.5/m^2.

Alliaria petiolata experiences a very high rate of natural seedling mortality (Byers and Quinn 1998, see Table 13). In the present study, 92.3% of seedlings died before reaching the rosette stage, and 0.7% of the rosettes counted in the fall died before reaching the adult stage, for an overall mortality rate of

93.0% from seedling to adult. Plants are much more stable once they reach the rosette stage in the fall, having survived summer droughts. Winter rosette mortality was extremely low in the study sites when compared with a study conducted in northern Illinois, where an average of 78.6% of rosettes were reported to have died during the winter (Nuzzo 2000).

It is important to note that during the present study, seedling counts were taken only once each spring, and in 2011 they were recorded quite late in the season. To truly know how many seeds germinate and accurately estimate their survival rate, seedling counts would need to be conducted on a more regular basis, as frequently as once per week, as seeds germinate beginning in February and continue into June (personal observation). Because counts were made only once per season for each life stage, I would expect that density numbers would increase greatly, and mortality figures would also increase, if sites had been visited more frequently throughout the growing season.

When compared with studies conducted in other regions of the U.S., the reproductive success of *A. petiolata* is very similar. A study conducted in Michigan (Evans 2006) reported up to 3,864 seeds on a single plant, and in the present study, the most seeds counted on a plant was 3301, with an average of 406. This average is higher than has been reported in other studies, such as in New Jersey, where plants averaged as many as 195 seeds per plant (Byers and Quinn 1998). The most siliques counted on a plant in the New Jersey study was 266, compared with 211 in the present study. Nuzzo (2000) reported the average number of siliques as 4-16, whereas in the southern Ozarks, the average number of siliques produced was much higher at 33.5. The height of *Alliaria petiolata* has been reported in the range of 0.05 m to 1.9 m, with an average of 1.0 m at flowering (Nuzzo 2000). In my reproductive success study, plants ranged from 0.17 m to 1.13 m, with an average of 0.7 m.

Adult plants averaged $3.4/m^2$, which is a very low number when compared with invasions in other regions of the U.S., where adult plants can reach densities of $500/m^2$ (Byers and Quinn 1998). It is clear that, while *A. petiolata* is increasing its distribution within the southern Ozarks, population densities have not yet reached levels similar to those observed in the Midwest and northeastern U.S. This is the reason I only had 10 study sites for the study. I simply could not find more habitats than that with densities large enough to study. However, although it was discovered too late to be included in the study, a large, dense infestation of *A. petiolata* was found at Hobbs State Park in Benton County, Arkansas, in spring of 2012

that rivaled Midwest and northeastern U.S. infestations. The population was the dominant vascular plant growing in a forested valley along a small creek, and the density of adult plants was estimated to be 100/m². In an ongoing effort, employees at the park are working to eradicate the infestation, and during an event to remove plants from the area in 2012, volunteers pulled and removed about 700 pounds of the plant.

The most common forest types of the 10 study sites in which *A. petiolata* was sampled were early-successional floodplain forests and mesic oak-maple forests (Southeast Ecology Group 2004, Faber-Langendoen 2006), but it also occurred in ruderal woodlands and successional sweetgum–oak forests (Foti and Pyne 2010, Evans and Nordman 2005, see Table 5). By comparison, a study conducted in Michigan (Evans 2006) found that *A. petiolata* occurs in similar forest types, but suitable habitat in that region also included the Black Oak - White Oak – Hickory Forest (CEGL002076, Guetersloh 1995) type, a dry to dry-mesic forest. In the southern Ozarks, it was never found growing in association with pine or oak-pine forest assemblages, although it has been reported in coniferous forests in Canada (Cavers et al. 1979) and Connecticut (Rodgers et al. 2008).

Of the 165 species identified in the study plots, 7.8% were introduced to the region. Three of the non-invaded plots contained only native species (Lake Eucha 2, Gravette, and Baker Ford). No more than six introduced species were found in any of the study plots, and these were all invaded plots. An average of 4.4 introduced species occurred in the invaded plots, contrasted with an average of 2.2 in the non-invaded plots. Cover of non-native vegetation was significantly higher in invaded plots in both years (2011 = 31.1%, 2012 = 24.7%) than it was in non-invaded plots (2011 = 7.1%, 2012 = 2.0%). A likely explanation for the higher number of introduced species and a higher amount of non-native species cover in the invaded study plots is that non-native species exploit disturbed areas (Larson 2003), and this is probably why *A. petiolata* was able to colonize these areas to begin with. Also of note is the fact that at eight of the study sites, species diversity and richness were higher in invaded plots than in non-invaded plots. This is likely due to the fact that richness and diversity can increase following a disturbance, especially intermediate-level disturbances such as seasonal flooding (Pollock et al. 1998).

In 2011, *Alliaria petiolata* was the most abundant herbaceous plant in three of the 10 plots in which it was sampled. In 2012, it was the most abundant plant in four of the 10 plots. Average cover of *A.*

petiolata was lower in spring of 2012 (31.0%) than it was in spring of 2011 (53.2%), even though the number of adult plants in 2012 was four times more what it was in 2011. This may be because counts were made a few weeks later in 2011 than they were in 2012, and adults and rosette plants were larger, as they had more time to develop aboveground biomass.

Alliaria petiolata has demonstrated an ability to alter soil pH, increase the levels of certain nutrients, and increase the decomposition rate of native tree leaves, creating conditions that favor its own growth (Rodgers et al. 2008). In the present study, soil characteristics such as pH, organic matter, and most nutrients were not significantly different between invaded and non-invaded plots (see Table 12). The only nutrient that was significantly higher in invaded plots across years was calcium. A secondary macronutrient that is a component of cell walls and is used in nitrogen metabolism, calcium is normally abundant in soils and contributes to their alkalinity (Kelling and Schulte 2004). However, invaded soils were not appreciably higher in pH than non-invaded soils. In fact, they were slightly acidic to neutral. If *A. petiolata* was increasing levels of calcium in the soil through soil exudates or decomposition of leaves, it would follow that nitrogen, phosphorus, and pH would also be increasing. Soil texture may explain why calcium levels were higher in the invaded plots, but texture was not determined from the soil samples. Published reports show that the composition of soils present in the area of each study site were primarily silt loams and sandy loams (see Table 2), with less than 10% clay in all but two sites. However, cation exchange capacity was significantly higher in the invaded plots than in non-invaded plots, indicating that the samples collected from invaded plots were high in clay (Espinoza et al. 2007). Clay is negatively charged and attracts cations such as calcium (Schlesinger 1997). This, and not the presence of *A. petiolata*, could explain why calcium levels were higher in the invaded plots.

Boron increased significantly from 2011 to 2012, from 0.9 ppm to 1.3 ppm, and these levels are within the normal range for the micronutrient (Kelling 1999). Although boron levels differed significantly between years, they were not significantly different between invaded and non-invaded sites, so the cause of the increase cannot be attributed to the presence of *A. petiolata*. Organic matter was slightly higher in 2012 than it was in 2011, and this may explain why boron was also higher, since boron is largely derived from organic matter (Kelling 1999).

Cover of herbaceous vegetation was similar in 2011 between invaded (62.4%) and non-invaded (63.2%) plots, and in 2012 overall cover dropped significantly, and it was higher in invaded plots (46.0%) than it was in non-invaded plots (42.9%). The drop in overall cover can be attributed to the drought in 2012, which had intensified greatly since it began in 2010 (see Figure 2). Also, cover of *A. petiolata* was lower in 2012, as the dominant life stages in the second year of the study were seedlings and rosettes, with fewer adult plants than in 2011.

During the course of conducting field work, vegetation cover was measured in spring of 2011 and 2012, and species that were assigned a cover class in the spring were not reassigned cover classes when botanical inventories were conducted again in summer and fall of each year. Cover classes should have been assigned to each species in each season, regardless of whether or not they were already identified previously, so that comparisons of cover could be made between seasons and at each life stage of *A. petiolata*. Also, I was not consistent in sampling certain variables in both years, such as coarse woody debris and leaf litter cover and depth, and so was not able to draw comparisons between years based on those variables.

Table 13. Density of *Alliaria petiolata* at each life stage and mortality from seedling in 2011 to adult in 2012 within study plots.

Site	Habitat Type	Seedlings	Rosettes	Adults	% Survivorship by Plot	% Mortality by Plot
E1	Riparian Forest	625	5	1	0.2	99.8
E2	Upland Forest	24	7	6	25	75.0
SP	Riparian Forest	64	6	4	6.3	93.8
GR	Upland Forest	110	16	16	14.5	85.5
BF	Upland Forest	30	8	8	26.7	73.3
DE	Upland Forest	558	9	3	0.5	99.5
PI	Riparian Forest	358	58	58	16.2	83.8
MH	Upland Forest	41	4	4	9.8	90.2
HA	Riparian Forest	43	35	35	81.4	18.6
TB	Riparian Forest	86	1	1	1.2	98.8
Total counted at each life stage		1939	149	136		
Total % Survivorship			7.7	7.0		
Total % Mortality			92.3	93.0		
Plants/m² - All plots		48.5	3.7	3.4		
Plants/m² - Riparian plots		58.8	5.3	5.0		
Plants/m² - Upland plots		38.2	2.2	1.9		

Table 14. Data recorded to determine reproductive success of *Alliaria petiolata* in the southern Ozarks. Length measurements are reported in cm.

Site-#	Habitat	Rosettes				Adults			
		Rosette diameter	Longest petiole	# of leaves	Average width of each leaf	Height	Stems	Siliques	Mature Seeds
E2-1	Upland	16.5	9.0	14	3.4	53	1	14	56
E2-2	Upland	30.0	13.0	50	4.0	95	4	61	760
E2-3	Upland	7.0	7.0	6	2.5	43	1	6	0
E2-4	Upland	15.5	8.0	13	4.2	91	1	26	356
E2-5	Upland	19.0	7.5	28	3.5	56	1	12	149
E2-6	Upland	8.0	6.0	5	2.8	36	1	6	51
E2-7	Upland	18.0	9.0	24	3.0	54	2	15	114
E2-8	Upland	10.0	8.0	10	2.8	56	1	8	101
E2-9	Upland	13.0	8.5	14	2.4	69	1	23	184
E2-10	Upland	18.0	9.0	22	3.2	83	1	22	317
SP-1	Riparian	20.5	9.5	73	2.3	88	7	80	789
SP-2	Riparian	17.5	7.5	68	1.9	83	4	38	583
SP-3	Riparian	15.5	8.0	17	4.2	67	2	19	267
SP-4	Riparian	13.0	8.5	16	2.8	53	1	7	36
SP-5	Riparian	24.0	9.5	56	2.9	96	5	80	745
SP-6	Riparian	22.5	10.0	58	2.9	103	1	37	516
SP-7	Riparian	12.0	7.0	22	2.4	60	1	7	57
SP-8	Riparian	14.5	7.0	27	1.8	63	2	22	264
SP-9	Riparian	13.0	13.0	20	3.0	52	1	1	9
SP-10	Riparian	10.5	13.0	no data	no data	56	1	6	67

Table 14. (Cont.)

Site-#	Habitat	Rosettes				Adults			
		Rosette diameter	Longest petiole	# of leaves	Average width of each leaf	Height	Stems	Siliques	Mature Seeds
DE-1	Upland	18.0	11.0	16	3.3	68	1	26	278
DE-2	Upland	16.5	10.0	8	3.3	50	1	8	83
DE-3	Upland	14.0	8.0	41	2.4	17	1	1	0
DE-4	Upland	20.0	12.0	34	3.5	(plant died before reaching maturity)			
DE-5	Upland	17.0	13.5	33	2.9	87	2	80	1225
DE-6	Upland	15.5	6.2	53	2.1	84	4	60	887
DE-7	Upland	13.0	8.0	17	2.6	61	3	22	230
DE-8	Upland	11.0	10.0	10	2.3	63	2	17	39
DE-9	Upland	8.0	6.0	10	2.7	52	1	4	28
DE-10	Upland	26.5	9.0	50	2.5	88	4	65	941
GR-1	Upland	6.5	4.0	6	1.3	no data	1	1	5
GR-2	Upland	20.0	10.0	10	3.7	86	1	14	145
GR-3	Upland	17.0	13.0	18	4.0	83	2	11	127
GR-4	Upland	14.0	9.5	5	3.6	60	1	10	85
GR-5	Upland	13.5	7.5	12	3.5	64	1	9	75
GR-6	Upland	9.0	9.5	7	1.9	45	1	3	11
GR-7	Upland	15.6	11.0	12	3.0	70	1	13	85
GR-8	Upland	19.0	10.0	10	3.3	93	1	18	142
GR-9	Upland	18.0	11.5	18	3.3	96	2	18	47
GR-10	Upland	10.5	9.0	16	2.5	65	1	7	40

Table 14. (Cont.)

Site-#	Habitat	Rosettes				Adults			
		Rosette diameter	Longest petiole	# of leaves	Average width of each leaf	Height	Stems	Siliques	Mature Seeds
PI-1	Riparian	21.0	10.5	65	3.3	98	5	81	646
PI-2	Riparian	15.5	8.0	40	2.2	80	1	35	245
PI-3	Riparian	36.0	12.0	60	4.2	109	6	211	3301
PI-4	Riparian	9.0	10.5	18	2.3	66	1	7	82
PI-5	Riparian	27.5	11.0	76	3.8	101	7	196	2700
PI-6	Riparian	26.5	9.0	62	2.6	113	5	138	2002
PI-7	Riparian	13.0	7.0	11	3.0	79	1	14	153
PI-8	Riparian	8.0	6.0	6	2.7	(plant died before reaching maturity)			
PI-9	Riparian	11.0	8.0	15	2.5	68	1	9	98
PI-10	Riparian	19.0	11.5	15	4.5	73	2	39	370
Total						3376	99	1607	19491
Mean		16.1	9.2	26.5	3.0	70.3	2.1	33.5	406.1
Minimum		6.5	4.0	5	1.3	17	1	1	0
Maximum		36.0	13.5	76	4.5	113	7	211	3301
Riparian Mean		16.6	8.9	36.3	2.8	79.4	2.8	54.1	680.5
Upland Mean		15.3	9.1	19.1	3.0	64.4	1.5	20.0	226.1

Table 15. Pearson's correlation coefficient between rosette diameter, height, stems, siliques, and mature seeds.

	Rosette Diameter	Height	Stems	Siliques	Mature Seeds
Rosette Diameter	1				
Height	0.7547	1			
Stems	0.7238	0.6776	1		
Siliques	0.8276	0.8302	0.8474	1	
Mature Seeds	0.7807	0.8346	0.7729	0.9468	1
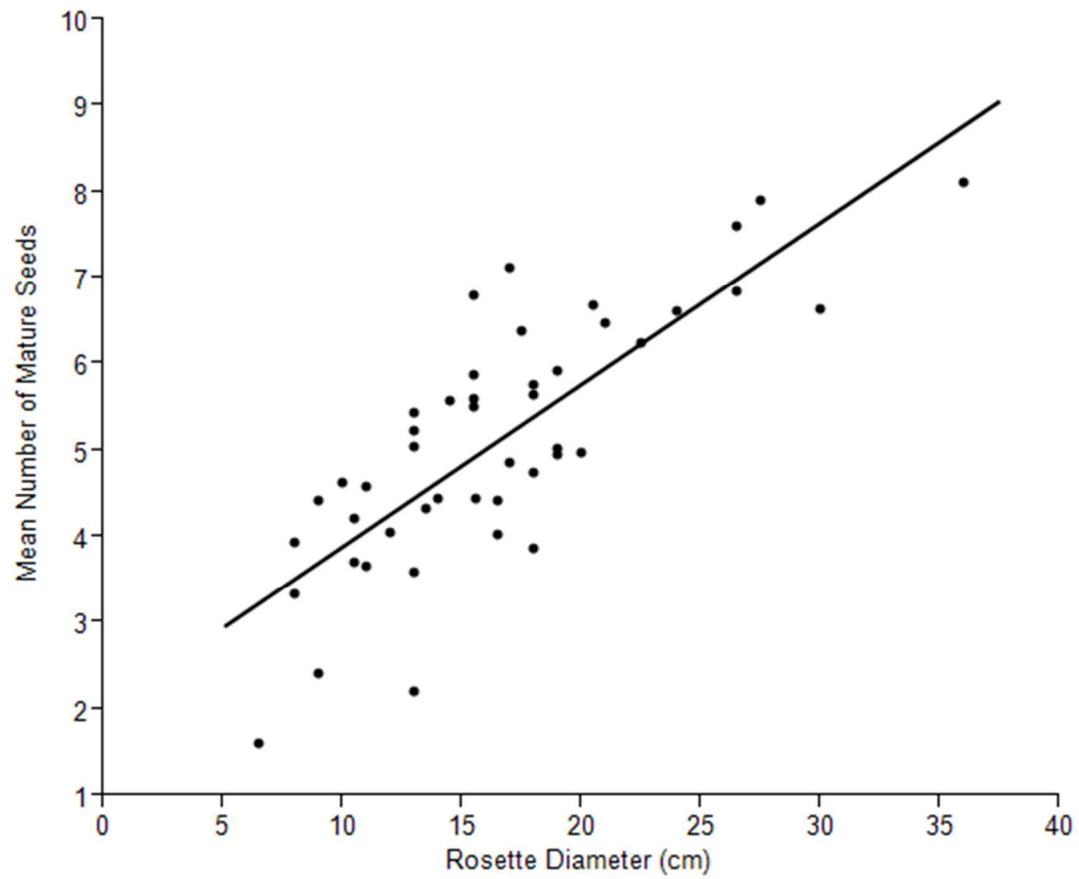
Figure 9. Reproductive success as a function of rosette diameter in *Alliaria petiolata* ($R^2 = 0.634$, F = 76.33, p value = 0.0001, N = 46). Mature seed data were log transformed.
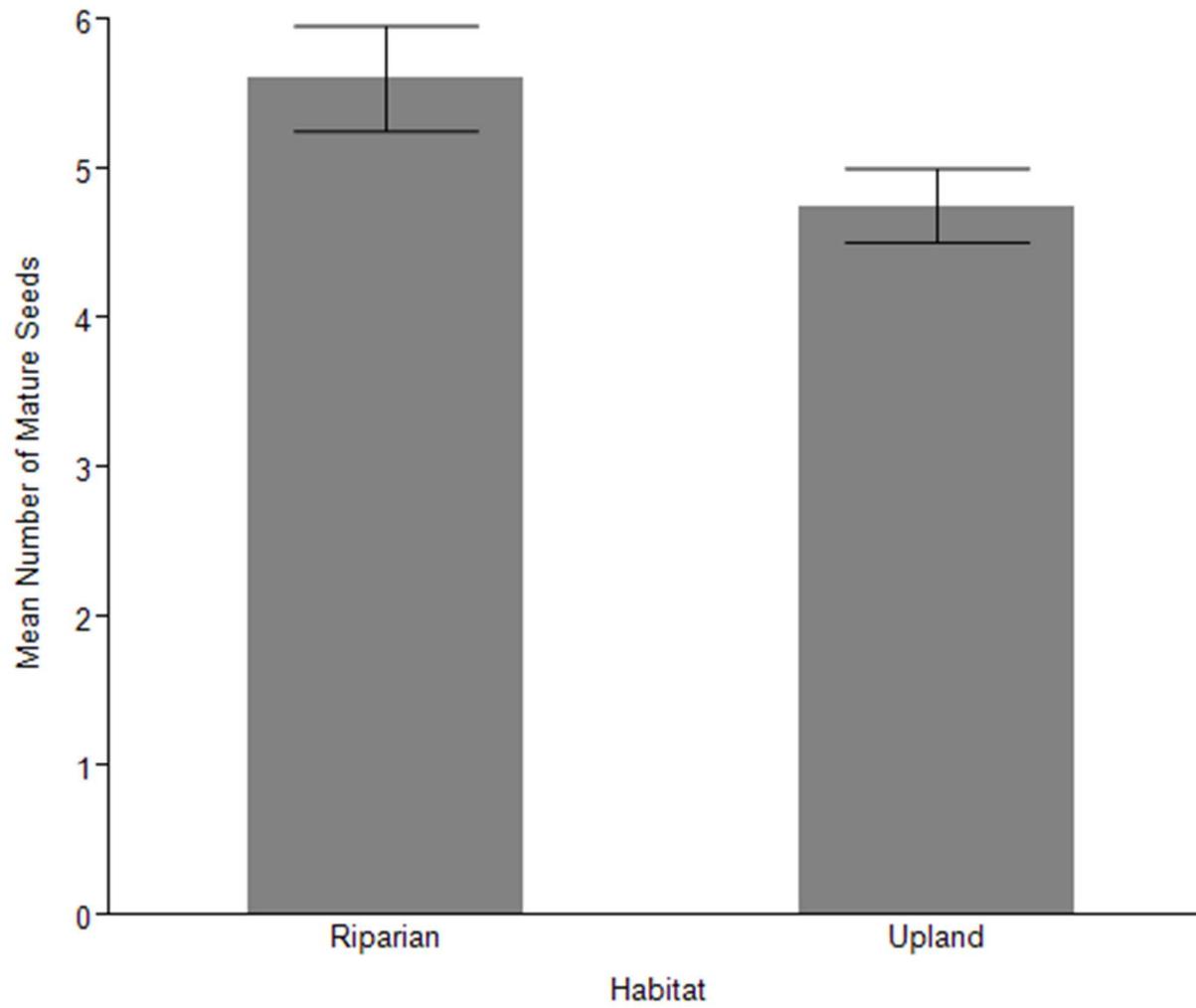
Figure 10. Reproductive success of *Alliaria petiolata* in riparian and upland habitats. Mature seed data were log transformed.

Chapter 3

A Comparison of Earthworm Composition and Biomass in Invaded Versus Non-Invaded Sites

INTRODUCTION

The beneficial role of earthworms in soil processes is well-documented. In regions of the Earth where they are abundant, earthworms play a vital role in improving soil structure, aeration, and drainage (Tomlin et al. 1995), as well as decomposition and nutrient cycling (Blair et al. 1995). While they are considered in many regions to be the most important and beneficial macrofauna inhabiting the soil (Hendrix and Bohlen 2002), recent research has focused on the negative role earthworms play in areas where they have been introduced. They have been implicated in the decline, not only of native earthworms in areas where their populations overlap (Kalisz and Wood 1995), but also of some vertebrate species (Loss et al. 2012, Maerz et al. 2009). Additionally, colonization by non-native earthworms into previously worm-free regions of North America – areas covered by ice sheets during Pleistocene glaciation - has been implicated as the cause of invasions by certain alien plant species (Fender 1995, Nuzzo et al. 2009).

EARTHWORMS IN HUMAN HISTORY, 4 B.C. - PRESENT

Earthworms have sparked our curiosities and imaginations for many centuries. The word "worm" carries several different connotations, most of which are negative. It is derived from the Old English "wyrm", which describes a legless serpent or dragon (Worm 2015). Today, the word is used to describe a person who is devious and not well liked, as well as a type of malicious software that sneaks around the internet searching for vulnerable host computers to infect (Worm 2015). When prefaced with "book", it is both an insect universally despised by librarians and a colorful way to describe a bibliophile (O'Connor 1898). The word has also appeared many times in modern popular culture. The subject matter of the children's book "How to Eat Fried Worms" was so disturbing to some parents that it made the American Library Association's list of the most frequently challenged books of the 1990s (Foerstel 2002). Several modern films have featured subterranean man-eating worms as antagonists designed to make audiences squirm in their seats (LaRue 2012). Children growing up in in the 1970s listened to "There's A New Sound" (Burrello 1953), a catchy, creepy song about earthworms, frequently played on the Dr. Demento Show: "There's a new sound, the newest sound around, the strangest sound that you have ever heard… it's deep down in the ground, and everyone who listens to it squirms, because this new sound so deep down in the ground is the sound that's made by worms."

Though frequently misunderstood and often demonized in popular culture, the importance of earthworms in the development and fertility of soil has been recognized since ancient times. In the 4th Century B.C., the Greek philosopher Aristotle called them "ges entera," which translates to English as "earth's intestines" (Rota 2011). Although this shows that he recognized, to a very limited extent, their ability to affect soil processes, Aristotle and his contemporaries knew almost nothing about the biology, ecology and reproduction of earthworms. In fact, at that time it was believed that earthworms arose spontaneously through abiogenesis and that through xenogenesis they gave birth to eels; a preposterous notion now, but a reasonable assumption at the time since no one had reported observing earthworms mating (or eels, for that matter). Because of the prevalent belief in spontaneous generation, no one at that time considered that soil fauna may actually play a role in the production of soil (Rota 2011). A few hundred years later, during the 1st Century B.C., the Egyptian pharaoh Cleopatra demonstrated a somewhat clearer understanding of the importance of earthworms in soil fertility and agriculture along the Nile when she created laws declaring them sacred and prohibited, under penalty of death, the removal of the lowly creatures from the land (Minnich 1977, Blanchart et al. 2010).

Many centuries later, in 1881 A.D., Charles Darwin published the book, *The Formation of Vegetable Mould through the Action of Worms,* and for the first time, the importance of earthworms was brought to the attention of scientists and the public at large (Edwards 2004). Darwin described in detail the manner in which earthworms "ploughed" the land long before humans developed agricultural tools, breaking down plant material and turning the soil, a phenomenon named "bioturbation" by scientists more than half a century later (Darwin 1881, Edwards 2004, Johnson et al. 2005). He thoughtfully wrote that even "archaeologists ought to be grateful to worms," because they bury and preserve objects dropped long ago by man (Darwin 1881). After 40 years of research on the "lowly organised creatures," he concluded that no other animal had "played so important a part in the history of the world" as did earthworms (Darwin 1881).

Darwin's research started a surge in research on earthworm taxonomy, biology, and ecology (Edwards 2004). Modern researchers agree with his assessment of the importance of earthworms in shaping the soil, calling them "ecosystem engineers" (Lavelle et al. 1997, Stewart 2004) and "keystone detritivores" (Frelich et al. 2006) because these macrofauna alter resource availability to other organisms

in the soil and litter (Lavelle et al. 1997, Chapin et al. 2002) and influence the soil profile, soil structure,

and even contribute to pedogenesis (Lee 1985).

EARTHWORM TAXONOMY AND ECOLOGY

Earthworms are tube-shaped invertebrates classified in phylum Annelida - the segmented worms;

class Clitellata - hermaphroditic aquatic or terrestrial worms and leeches featuring a clitellum which

functions in cocoon production; subclass Oligochaeta – a large group of between 6,000 (Brusca and

Brusca 2003) and 8,000 species (Edwards 2004) of worms with few setae, or bristles, and lacking

external outgrowths; and order Haplotaxida, a group of about 25 families of earthworms which occur in

nearly every terrestrial ecosystem on Earth, except those with the most extreme climates (Edwards and

Bohlen 1996, Hendrix and Bohlen 2002, Brusca and Brusca 2003). Nearly 100 described species from

five families are considered native to North America, and at least 45 species from two families have been

introduced to the continent (James and Hendrix 2004, Hendrix and Bohlen 2002).

Terrestrial earthworms live either on or in soil and consume organic matter, living organisms such

as bacteria and fungi, and soil (Lee 1985, Domínguez 2004). They are placed into three major ecological

groups based largely on their physical positions within the soil and/or litter layer - epigeic, endogeic, and

anecic (James and Hendrix 2004). Epigeic species inhabit the leaf litter above the soil surface where they

feed on decaying organic matter (Edwards 2004). Members of this group grow and move quickly and

have high rates of reproduction (Edwards and Bohlen 1996). They tend to be smaller and darker in color

than members of the other two ecological groups (James and Hendrix 2004). Endogeic species form non-

permanent burrows in the soil, occupying and feeding on mineral soil enriched with organic matter, and

have low rates of reproduction (Lee 1995). Though members of both of these ecological groups consume

large amounts of both inorganic and organic material (up to 30 times their weight each day), they are not

very efficient at assimilation, so much of that which passes through their systems remains undigested and

is deposited as organic-rich casts in or on the soil (Brown and Doube 2004). Microbes inhabiting the soil,

such as bacteria and fungi, are also consumed, and depending on the species of earthworm consuming

them, may either be activated by the availability of carbon and other nutrients in the earthworm's system,

or if they are consumed by a larger species, may even have time to reproduce before being deposited

back into the soil. Casts provide an ideal habitat and energy source for these microbes, which in turn

contribute to nutrient mineralization, facilitating the release of usable nitrogen, phosphorus and potassium for uptake by surrounding plant roots (Brown and Doube 2004). Epigeic and endogeic earthworms also contribute to soil structure and organization by aerating and incorporating organic matter from the surface into the soil (Parkinson et al. 2004). Anecic species produce permanent vertical burrows which extend from the surface into the deep mineral horizon, where they pull dead and decaying organic matter into their deep burrows (Edwards and Bohlen 1996). This ecological group contributes to pedogenesis by mixing organic matter from the surface and inorganic matter from deep within the soil profile (Edwards 2004).

EARTHWORMS AS FACILITATORS OF ALIEN PLANT INVASIONS

Throughout much of our modern history, earthworms have been largely regarded as beneficial soil organisms, and as a result can be found in nearly every region colonized by humans (James and Hendrix 2004). However, scientists have also recognized the potential for damage to native ecosystems by anthropogenic introductions of these non-native species (Kalisz and Wood 1995). Non-native earthworms are capable of severely altering soil processes within natural systems, especially in northern temperate forests, which were largely uninhabited by earthworms due to Pleistocene glaciation and permafrost (Callaham et al. 2006, Hendrix et al. 2006). These detritivores change ecosystem processes by consuming the previously thick litter layer on the forest floor, eliminating suitable habitat for plant growth and reproduction (Hale et al. 2005, Loss et al. 2012). They also negatively impact the seed bank by burying seeds deep into the soil, removing the possibility of germination (Hale et al. 2005). Nuzzo et al. (2009) demonstrated that invasion by *Alliaria petiolata* (M. Bieb.) Cavara & Grande (garlic mustard), *Berberis thunbergii* DC. (Japanese barberry), and *Microstegium vimineum* (Trin.) A. Camus (Japanese stiltgrass) was positively correlated with non-native earthworm invasions and negatively correlated with native woody plant cover.

Research conducted in northern hardwood forests of the United States has shown that non-native earthworms are severely altering native plant communities in areas that were previously free of them (Gundale 2002, Hale et al. 2006, Nuzzo et al. 2009), which in turn negatively impacts animals that rely on these communities for food, shelter, and reproduction. One recent study found that non-native earthworms caused a reduction in the density of *Seiurus aurocapilla* L. (Ovenbird) and *Catharus guttatus*

Pallas (Hermit Thrush) in Minnesota forests by reducing the cover of herbaceous plants, thus reducing suitable nesting sites for these ground-nesting birds (Loss et al. 2012). Another study found that the abundance of *Plethodon* ssp. (woodland salamanders) declined in the presence of non-native earthworms (Maerz et al. 2009). As earthworms stripped the forest floor of leaf litter, invertebrate soil fauna declined, which in turn led to a decline in juvenile salamanders, which rely on such fauna as a major component of their diets (Maerz et al. 2009).

TAXA OF TERRESTRIAL EARTHWORMS IN THE SOUTHERN OZARK PLATEAUS PHYSIOGRAPHIC REGION

Several surveys have been conducted to learn of the distribution of terrestrial earthworms in areas of the state that include those of the present study (Reynolds 2008a), principally in Arkansas. In the 1950s, David Causey, a professor in the Department of Zoology at the University of Arkansas, published papers on the distributions of earthworms in the state (Causey 1952, 1953). His collections were taken from 22 counties and, using current nomenclature, included 21 species from 11 genera. He regarded nine of those species to be native to North America and the rest as introductions from Europe and Asia. Several of the specimens were brought to him by students at the university, while others were collected during the course of collecting trips for other purposes (Causey 1952). Although some collections were made in natural areas, many were from cultivated regions. Some were donated by an earthworm farmer in the town of Manila (Mississippi County) and it can be assumed that these specimens were taken directly from cultivation or were recent escapes. He also notes that some of the records were from lawns, gardens, and formerly cultivated areas (Causey 1952). In 1977 Gordon E. Gates described a new endemic species of earthworm, *Diplocardia meansi* Gates (Means' giant earthworm) from Arkansas, named in honor of Dr. Bruce Means, who discovered its existence on Rich Mountain in Polk County (Gates 1977). Gates also reported eight more earthworm species in Arkansas in papers published from the 1950s to the 1980s (Gates 1955, 1959, 1972a, 1972b, 1973, 1982). In 2008, Dr. John W. Reynolds summarized the published reports of Causey, Gates, and others in a paper printed in the Canadian journal *Megadrilogica* (2008a). He included his own collections to the list (2008a), which added two species previously unreported for the state. More recently, surveys of earthworms were conducted in tall fescue (*Schedonorus arundinaceus* (Schreb.) Dumort.) fields at the Agricultural Experiment Station in

Fayetteville and in septic fields in Washington County (Hawkins et al. 2008, Rashé 2008, Tomlinson 2011). From those studies, two new species were reported for the state (Hawkins et al. 2008, Rashé 2008). Prior to the present study, 36 species of earthworms from 49 counties had been documented in Arkansas.

In Missouri, surveys of earthworms were conducted in 1936 by Henry Olson (Olson 1936), a professor based in Pennsylvania. He made personal collections in 25 counties and accepted specimens from friends and students from three other counties. In all, he identified 15 species of earthworms from four genera, four of which were native to North America. His study area focused largely on the eastern and northern regions of Missouri, and no collections were taken from the southern Ozarks. Stebbings (1962a, 1962b) discovered six species during investigations in Shannon County. Gates (1973) identified one new species not previously reported for the state. Reynolds (2008a) reported six additional species for the state, including one that had previously been reported by Stebbings. In all, 26 species from 37 counties were reported for Missouri prior to the present study.

The first published reports of earthworms in Oklahoma were by Harman (1954), who reported five new species, and Murchie (1962, 1963), who described two species new to science. Gates (1977, Reynolds 2010) later reported three species of earthworms as new records for Oklahoma. Reynolds (1980, 2010) reported 19 new species. Prior to the present study, a total of 34 terrestrial earthworms from 44 counties had been reported in Oklahoma.

STUDY OBJECTIVES

In the southern Ozark Plateaus Ecoregion of Arkansas, Missouri, and Oklahoma, a region that did not experience glaciation and is therefore currently inhabited by native earthworms, several questions arise when invasions by non-native earthworms occur. The present study addresses the following in an attempt to understand the relationship between native and non-native earthworms in areas where they now co-occur in the southern Ozarks, and whether non-natives have played a role in invasion by *Alliaria petiolata* by providing suitable habitat for its successful colonization.

- What is the current known distribution of earthworms in the southern Ozarks, and what percentage of the species are native to North America?

- Are non-native earthworms more abundant in sites invaded by *A. petiolata* than in sites free of the

plant?

- Is there a difference in the abundance of non-native earthworms in riparian forests versus upland forests?

- Are other factors, such as leaf litter volume and non-native plant cover, effected by earthworm abundance?

EARTHWORM ABUNDANCE, BIOMASS, AND FUNCTIONAL GROUPS IN INVADED VERSUS NON-INVADED SITES

METHODS AND MATERIALS

Earthworm sampling was conducted to compare abundance, biomass, and functional groups in the presence and absence of *Alliaria petiolata*. The liquid extraction method was used to collect earthworms from 40 plots, each plot measuring 33 cm x 33 cm. Collections were made on March 29-31, 2012, when soil temperatures (at 10 cm depth) at UAPB Point Remove, Arkansas (a NRCS Soil Climate Analysis Network site) were 17-19 °C. At each of the twenty established 100 m² plots used for *A. petiolata* research, two earthworm extraction plots were established using a random numbers table to determine at which of the five possible points (north, south, east, west or center of the 100 m² plot) they would be placed. The sampling square was constructed of welded 10 cm high steel, designed to sink into the ground to a depth of 2.5 cm to contain the liquid. The square was placed on the ground and leaf litter depth and cover of bare ground were recorded. Vegetation was removed and earthworms in the leaf litter were counted and recorded. Leaf litter was then cleared, exposing bare soil, and the number of middens was counted and recorded. To extract endogeic and anecic earthworms, 40 g of Frontier Herbs brand non-organic ground yellow mustard powder was mixed with four liters of water and poured into the plot form, one-third of the mixture at a time at 0, 5, and 10 minutes. Collection continued for a total of 15 minutes. All earthworms that surfaced during this time were collected with forceps, rinsed in water to remove debris, and anesthetized individually in a bowl containing 70 percent isopropyl alcohol (Figure 1). Samples were then transferred to polypropylene vials filled with 70 percent isopropyl alcohol for transport to the UARK laboratory where they were placed in 10 percent formalin for permanent preservation within 24 hours of collection. Samples were sent to the Natural Resources Research Institute's Great Lakes Worm Watch (GLWW) at the University of Minnesota for identification and to record biomass. Mature

earthworms were identified to species and the juveniles were identified to genus. Biomass of each earthworm was estimated using an allometric equation developed by researchers at GLWW (see Hale et al. 2004) which uses the natural log of the earthworm length to calculate the ash-free dry biomass of each earthworm collected. This equation was developed for several reasons. It is often desirable to preserve earthworms as voucher specimens, and using traditional drying methods to calculate biomass destroys the specimens. Also, earthworm biomass can fluctuate depending on moisture conditions in the soil and the gut contents of earthworms at the time of collection. This equation presents a simple alternative to dissection, ashing, or waiting 48 hours for worms to empty their guts naturally (GLWW 2013).

STATISTICAL ANALYSES

Statistical analyses were conducted to (1) compare earthworm abundance, leaf litter volume, and non-native plant cover in plots invaded by and not invaded by *A. petiolata*, (2) to compare earthworm abundance in upland and riparian habitats, and (3) to determine whether or not plant species richness is different in plots with higher earthworm biomass. JMP version 11 statistical software (SAS Institute 2013) was used to calculate Pearson's correlation coefficient, construct simple linear regressions, and to perform t tests. Variables were transformed when necessary to better fit the normality assumption.

RESULTS

Data derived from published papers ranging in time from 1936 to the present were used to compile a checklist of species documented in Arkansas, Missouri, and Oklahoma, with their status of native or introduced to North America (Table 1). A total of 48 species, as well as four taxa identified only to genus, have been documented from the three states. Species collected during the present study were also included in the list.

The results of data collection are presented in Table 2. A total of 467 earthworms were collected during the course of the study. Nine individuals were broken during the process of collecting and were excluded from analysis, leaving 458 individuals. In plots invaded by *A. petiolata*, a total of 233 earthworms were collected. In plots not invaded by *A. petiolata*, a total of 225 earthworms were collected (Figure 2). Eleven native earthworms were collected from the non-invaded Lake Eucha 2 and Mt. Hersey plots and the invaded Hasty plot, and had a total combined biomass of 0.5282 g. The remaining 447 individuals were introduced to North America from Asia and Europe, and had a combined biomass of

11.123 g (6.0625 g in invaded plots, and 5.510 g in non-invaded plots). The plot with the most earthworms by count was the invaded Gravette plot, an upland forest, with a total of 63 individuals. The plot with the least earthworms was the non-invaded Tyler Bend plot, a riparian forest, with no earthworms collected.

Two Oklahoma state records and several county records from each state were documented during the course of the present study (Table 3). A list of the species collected from each study plot is presented in Table 4. All three major ecological groups were represented from the individuals identified to species – two from the anecic group, 140 from the endogeic group, and 47 from the epigeic group (Table 5). Although only two adult individuals were identified from the anecic group, 19 middens were counted in the field. Because middens serve as a reliable indicator of adult *Lumbricus terrestris* L. (nightcrawler) presence, and they are the only earthworm in the southern Ozarks known to create such structures, it is assumed that 17 additional earthworms were *L. terrestris* that had not reach full maturity, but which were capable of burrowing. The remaining 261 earthworms not placed into an ecological group were identified as juveniles of the genus *Lumbricus*, and because the two known representatives of the genus in North America belong to two different ecological groups, and because the two species may occupy the same soil region until juvenile *L. terrestris* reach the 12 weeks of age (Lowe and Butt 2002), the unknown *Lumbricus* were not placed into either group.

A correlation matrix using Pearson's coefficient (r) was used to determine which variables were dependent on earthworm biomass (Table 6). The only variables that were very strongly correlated were earthworm abundance (log transformed) and earthworm biomass. Non-native plant cover (log transformed) and leaf litter volume (2nd root transformed) showed no relationship or a weak positive correlation, respectively, with biomass. Non-native plant cover and leaf litter volume were moderately negatively correlated. Plant species richness was not correlated with any other variable. Leaf litter volume was calculated by multiplying the depth of the leaf litter in the sampling square by the length and width of the square (33 x 33 x depth), and the product was multiplied by the area covered by leaf litter within the plot.

A t test was used to determine if non-native earthworms were more abundant in plots invaded by *A. petiolata* than they were in plots not invaded by the plant (Figure 3). Because the variables abundance

and biomass were positively correlated at 0.9124, biomass was chosen for the t tests. The p value was large (p = 0.776), indicating that biomass was not significantly higher in invaded plots (mean biomass = 0.6062 ± 0.4414 g) than it was in non-invaded plots (mean biomass = 0.551 ± 0.4417 g).

A t test was used to determine if non-native earthworms were more abundant in riparian plots than they were in upland plots (Figure 4). The p value was large (p = 0.9485), indicating that biomass was not significantly higher in riparian plots (mean biomass = 0.5849 ± 0.4560 g) than it was in upland plots (mean biomass = 0.5724 ± 0.3977 g).

Overall, leaf litter volume and biomass had a weak positive relationship. When the variables were analyzed by invaded status, the relationship changed. In invaded plots, the relationship was strongly positive (r = .5745), and in non-invaded plots the relationship was weakly negative (r = -24.11). Simple linear regression (Figure 5) calculated to predict leaf litter volume based on earthworm biomass approached significance in invaded plots (p = 0.0824), but was not significant in non-invaded plots (p = 0.5022).

In invaded plots, earthworm biomass had a weak negative relationship with species richness (r = -0.1856), but in non-invaded plots, the negative relationship was much stronger (r = -0.7209). A simple linear regression (Figure 6) calculated to predict plant species richness based on earthworm biomass was not significant in invaded (p = 0.6061) or non-invaded plots (p = 0.2537).

DISCUSSION

Analyses of data collected in plots within the southern Ozarks did not indicate that the presence of non-native earthworms was causing a decrease in plant species richness or in leaf litter volume. The data collected also do not support the hypothesis that non-native earthworms would be more abundant in plots inhabited by *Alliaria petiolata*, and therefore may be facilitating invasions by the invasive plant. Non-native plant cover was significantly higher in invaded plots (mean = 24.7%) than it was in non-invaded plots (mean = 2.0%), but earthworms do not appear to be the cause of this difference.

In a 2009 study conducted in New York and Pennsylvania, leaf litter volume and earthworm biomass were negatively correlated (r = -0.579), and earthworm biomass was consistently higher and leaf litter volume was consistently lower in invaded plots than they were in non-invaded plots (Nuzzo et al. 2009). This was not the case in the present study, where the relationship was positive in both invaded

and non-invaded plots, and where no differences were found in either variable between plots.

The sample number in this study was very low. Two plots were sampled at each *A. petiolata* study site, and these were combined for the statistical analyses, for a total of 10 invaded and 10 non-invade study sites. In each case, the statistical test used resulted in a large p value, and this was probably due to an inadequate sample size. To perform meaningful statistical analyses on the data collected, the sample size would need to be much larger than it was.

The devastating effects non-native earthworms have on hardwood forests are well documented in regions of the U.S. that were previously free of the animals (Hale et al. 2005, Hale et al. 2006, Nuzzo et al. 2009), such as Minnesota, New York, and Pennsylvania. These regions were largely devoid of native earthworms prior to the introduction of European and Asian species, and it is believed that they were extirpated by ice sheets during the Last Glacial Maximum (Tuinov et al. 2006). But because the southern Ozarks did not experience glaciation (Clark et al. 2009), the forests that developed after the last glacial period did so in the presence of earthworms, which are undoubtedly vital to the proper functioning of the hardwood forest ecosystem (Hendrix et al. 2006). Prior to this study, only one study had been conducted in the Ozarks documenting the relative abundances of non-native and native earthworms in hardwood forests (Stebbings 1962b). That study, conducted near St. Louis, found that native earthworms were more abundant than non-natives, even in floodplain sites. Because of lack of data on relative abundances of the two groups, it does not seem possible to conclude that native earthworms are in decline or that if they are, that non-native earthworms are the cause of their decline. However, non-natives (n = 447, or 97.5% of the total collected) were overwhelmingly more abundant than natives (n = 11, or 2.5% of the total) were in the study plots. Is it possible that a shift is occurring in their relative abundances, especially when considering that in Arkansas alone, 21 of the 36 known species of earthworms documented for the state are native?

Hendrix et al. (2006) wrote that "by considering the currently observed state of any particular earthworm assemblage, we can trace several possible series of events that may have led to that state from a presumed indigenous community in a pristine ecosystem". In other words, it may be possible to work backwards from an observation of relative abundances of non-native and native earthworms, and by determining what the condition of the habitat was prior to a disturbance, can speculate on the composition

of the native earthworm community prior to the disturbance. In the present study, disturbances were moderate to severe, depending on the site, and included logging, fire suppression, non-native vegetation, forest fragmentation, flooding, and evidence of feral hog activity. In the Stebbings (1962b) study, mentioned previously, disturbance was minimal in the floodplains with nearly equal abundances of native and non-native earthworms, and moderate in the mixed forests on hillslopes where natives were more abundant (Hendrix et al. 2006).

Hendrix et al. (2006) hypothesized that several sequences of invasion were occurring based on degree of disturbance and current earthworm abundances (Figure 7), and the pathways that fit best based on observed earthworm abundances and known disturbances in the study plots are pathways A and B1. Based on this, it seems reasonable to conclude that native earthworms were once more abundant in hardwood forests of the southern Ozarks, but due to significant disturbances to the ecosystem over a period of many decades, non-native earthworms have invaded and currently dominate in this ecosystem.

The one taxon of native earthworm identified in three plots by GLWW was a member of the Nearctic genus *Sparganophilus* belonging to the monotypic family Sparganophilidae. I question this determination, as *Sparganophilus* are limicolous (mud-dwelling) species that occur in semi-aquatic habitats such as in mud near streams and lakes, and in aquatic habitats, in mud beneath water up to 1 m deep (Reynolds 2008b). The family and generic names of this group were assigned to them by oligochaetologists because of their association with the genus *Sparganium*, aquatic plants commonly known as bur-reeds (Reynolds 1980). None of the habitats visited in the present study included semi-aquatic or aquatic habitats, so it is unlikely that the 11 species identified as belonging to the genus *Sparganophilus* actually belong to that group.

Figure 1. The method used to extract earthworms from the soil. **A**: A 33 cm x 33 cm sampling square was placed at a pre-determined point and cover of bare ground and leaf litter depth were recorded. **B**: Leaf litter was cleared after sorting through the material for epigeic species, then middens were counted. **C**: Hot mustard solution was poured into the sampling square at 0, 5, and 10 minutes. **D**: An earthworm emerging from the soil in response to the mustard solution coming into contact with its skin. Photographs taken by Jennifer Ogle near Mt. Hersey on the Buffalo National River in Newton County, Arkansas.

Table 1. Checklist of species occurring in Arkansas, Missouri, and Oklahoma. Species with an asterisk (*) are introduced to North America.

Family	Species	Arkansas	Missouri	Oklahoma
Acanthodrilidae	*Diplocardia biprostatica* Gates			x
Acanthodrilidae	*Diplocardia caroliniana* Eisen	x		x
Acanthodrilidae	*Diplocardia communis* Garman	x	x	
Acanthodrilidae	*Diplocardia conoyeri* Murchie	x	x	x
Acanthodrilidae	*Diplocardia hulberti* James			x
Acanthodrilidae	*Diplocardia invecta* Gates			x
Acanthodrilidae	*Diplocardia kansensis* James	x		
Acanthodrilidae	*Diplocardia keyesi* Eisen			x
Acanthodrilidae	*Diplocardia longa* Moore	x		x
Acanthodrilidae	*Diplocardia longiseta* Murchie	x		x
Acanthodrilidae	*Diplocardia meansi* Gates	x		
Acanthodrilidae	*Diplocardia nova* Gates	x		
Acanthodrilidae	*Diplocardia ornata* Gates	x	x	x
Acanthodrilidae	*Diplocardia riparia* Smith	x	x	x
Acanthodrilidae	*Diplocardia rugosa* James			x
Acanthodrilidae	*Diplocardia singularis* Ude	x	x	x

Table 1. (Cont.)

Family	Species	Arkansas	Missouri	Oklahoma
Acanthodrilidae	*Diplocardia verrucosa* Ude		x	x
Lumbricidae	*Allolobophora chlorotica* Savigny*		x	x
Lumbricidae	*Allolobophoridella eiseni* Levinsen*		x	
Lumbricidae	*Aporrectodea caliginosa* Savigny*	x		x
Lumbricidae	*Aporrectodea rosea* Savigny*	x	x	x
Lumbricidae	*Aporrectodea trapezoides* Dugès*	x	x	x
Lumbricidae	*Aporrectodea tuberculata* Eisen*	x		
Lumbricidae	*Aporrectodea turgida* Eisen*		x	
Lumbricidae	*Aporrectodea* sp.*			x
Lumbricidae	*Bimastos beddardi* Michaelsen	x		
Lumbricidae	*Bimastos gieseleri* Ude	x		x
Lumbricidae	*Bimastos heimburgeri* Smith		x	
Lumbricidae	*Bimastos longicinctus* Smith & Gittins	x	x	
Lumbricidae	*Bimastos parvus* Eisen	x		x
Lumbricidae	*Bimastos timidus* Eisen		x	x
Lumbricidae	*Bimastos zeteki* Smith & Gittins	x	x	
Lumbricidae	*Dendrobaena octaedra* Savigny*	x	x	
Lumbricidae	*Dendrodrilus rubidus* Savigny*	x	x	x

Table 1. (Cont.)

Family	Species	Arkansas	Missouri	Oklahoma
Lumbricidae	*Eisenia fetida* Savigny*	x	x	x
Lumbricidae	*Eisenia hortensis* Michaelsen*	x		
Lumbricidae	*Eiseniella tetraedra* Savigny*	x	x	
Lumbricidae	*Eisenoides carolinensis* Michaelsen	x		
Lumbricidae	*Lumbricus rubellus* Hoffmeister*	x	x	x
Lumbricidae	*Lumbricus terrestris* Linnaeus*	x	x	x
Lumbricidae	*Octolasion tyrtaeum* Savigny*	x	x	x
Lumbricidae	*Octolasion* sp.*			x
Megascolecidae	*Amynthas agrestis* Goto & Hatai*		x	x
Megascolecidae	*Amynthas diffringens* Baird*	x	x	x
Megascolecidae	*Amynthas hilgendorfi* Michaelsen*	x		x
Megascolecidae	*Amynthas hupeiensis* Michaelsen*	x		x
Megascolecidae	*Amynthas* sp.*			x
Megascolecidae	*Metaphire californica* Kinberg*	x		
Sparganophilidae	*Sparganophilus eiseni* Smith	x	x	x
Sparganophilidae	*Sparganophilus* sp.	x		x
Total (Total Native)		36 (21)	26 (12)	36 (20)

Table 2. Data collected in the field. Non-native plant cover was collected from 100 m² plots in May of 2012 and is included here for purposes of comparison.

Site	Habitat	Status	Non-Native Earthworm Abundance	Non-Native Earthworm Biomass (g)	Native Earthworm Abundance	Native Earthworm Biomass (g)	Leaf Litter Volume (cm³)	Non-Native Plant Cover (%)
E1	Riparian	A	52	1.1475	0	0	2382.2	16.25
		B	45	0.7581	0	0	1.1	10.00
E2	Upland	A	14	0.3345	0	0	2613.6	18.50
		B	13	0.6292	5	0.1098	8167.5	0
SP	Riparian	A	9	0.2645	0	0	1633.5	7.50
		B	14	0.1894	0	0	367.5	0
GR	Upland	A	63	1.3774	0	0	2178.0	25.00
		B	18	0.5939	0	0	2722.5	0
BF	Upland	A	17	0.3856	0	0	544.5	33.75
		B	7	0.2427	0	0	2178.0	0
DE	Upland	A	12	0.3392	0	0	2722.5	23.75
		B	50	1.096	0	0	3267.0	7.50
PI	Riparian	A	39	1.1205	0	0	1089.0	47.50
		B	38	1.1806	0	0	1633.5	0
MH	Upland	A	11	0.6411	0	0	0.5	22.50
		B	4	0.084	5	0.5038	1293.2	0

Table 2. (Cont.)

Site	Habitat	Status	Non-Native Earthworm Abundance	Non-Native Earthworm Biomass (g)	Native Earthworm Abundance	Native Earthworm Biomass (g)	Leaf Litter Volume (cm³)	Non-Native Plant Cover (%)
HA	Riparian	A	9	0.2016	1	0.0244	0.5	45.00
		B	26	0.7361	0	0	2178.0	2.50
TB	Riparian	A	6	0.2506	0	0	544.5	7.50
		B	0	0	0	0	0	0
Invaded Plots Total			232	6.0625	1	0.0244	13708.9	247.3
Mean			23.2	0.6063	0.1	0.0024	1370.9	24.7
Standard Deviation			20.4	0.4	0.3	0.0	1070.6	13.8
Minimum			6	0.2016	0	0	0.5445	7.5
Maximum			63	1.3774	1	0.0244	2722.5	47.5
Non-Invaded Plots Total			215	5.510	10	0.6136	21808.3	20.0
Mean			21.5	0.551	1	0.0614	2180.8	2
Standard Deviation			69.4	1.8	1.6	0.0	4443.1	72.4
Minimum			0	0	0	0	0	0
Maximum			50	1.1806	5	0.5038	8167.5	10

Table 3. Earthworms identified in each study plot. A = plot was invaded by *Alliaria petiolata*. B = plot was not invaded by *A. petiolata*. Note: An asterisk (*) denotes a native taxon.

Species	E1		E2		SP		GR		BF		DE		PI		MH		HA		TB	
	A	B	A	B	A	B	A	B	A	B	A	B	A	B	A	B	A	B	A	B
Allolobophora chlorotica		x																		
Amynthas sp.	x	x			x	x									x					
Aporrectodea caliginosa complex	x	x					x	x			x	x						x		
Aporrectodea sp.	x	x					x	x			x	x								
Lumbricus rubellus	x			x			x	x	x			x	x	x	x	x			x	x
Lumbricus terrestris				x											x					
Lumbricus sp.	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x
Octolasion tyrtaeum		x	x	x	x		x	x		x	x	x								
Octolasion sp.	x	x	x	x	x	x			x	x	x	x	x				x	x		
Sparganophilus sp.*				x													x	x		

Table 4. County records for earthworms documented during the course of the present study. First state records are marked with an asterisk (*).

Taxon	Arkansas	Missouri	Oklahoma
Allolobophora chlorotica			Delaware*
Amynthas sp.			Delaware
Aporrectodea caliginosa complex			Delaware
Aporrectodea sp.			Delaware
Octolasion tyrtaeum	Searcy	McDonald	
Octolasion sp.			Delaware
Sparganophilus sp.	Newton		Delaware
Lumbricus rubellus	Benton, Newton, Searcy	McDonald	Delaware
Lumbricus terrestris	Newton		Delaware*

Table 5. The species and abundance of earthworms collected from each ecological group. Note: an asterisk (*) denotes a species that is native to North America.

Ecological Group	Species	Abundance
Anecic	*Lumbricus terrestris*	19
Endogeic	*Allolobophora chlorotica*	1
	Amynthas sp.	14
	Aporrectodea caliginosa complex	16
	Aporrectodea sp.	43
	Octolasion sp.	41
	Octolasion tyrtaeum	14
	Sparganophilus sp.*	11
Epigeic	*Lumbricus rubellus*	47
Unknown	*Lumbricus* sp.	261
Total		467

Table 6. Pearson's correlation coefficients between earthworm biomass, earthworm abundance (log transformed), leaf litter volume (2nd root transformed), and non-native plant cover (log transformed).

	Earthworm Biomass (g)	Earthworm Abundance	Leaf Litter Volume (cm³)	Non-Native Plant Cover (%)	Plant Species Richness
Earthworm Biomass (g)	1				
Earthworm Abundance	0.9124	1			
Leaf Litter Volume (cm³)	0.3186	0.213	1		
Non-Native Plant Cover (%)	-0.0306	0.0226	-0.4591	1	
Plant Species Richness	0.5075	0.3363	0.1109	0.2193	1
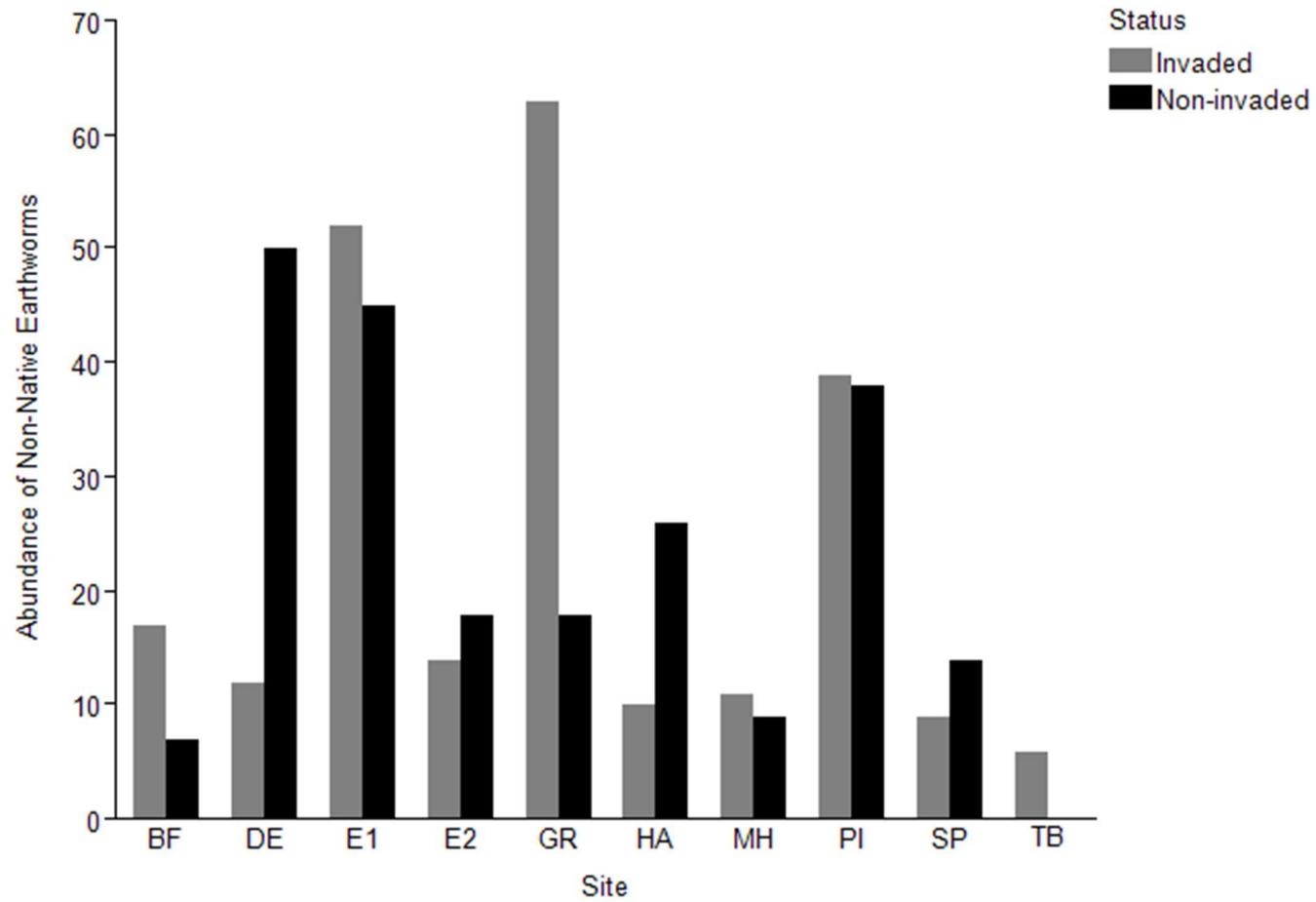
Figure 2. Abundance of non-native earthworms in each study site.
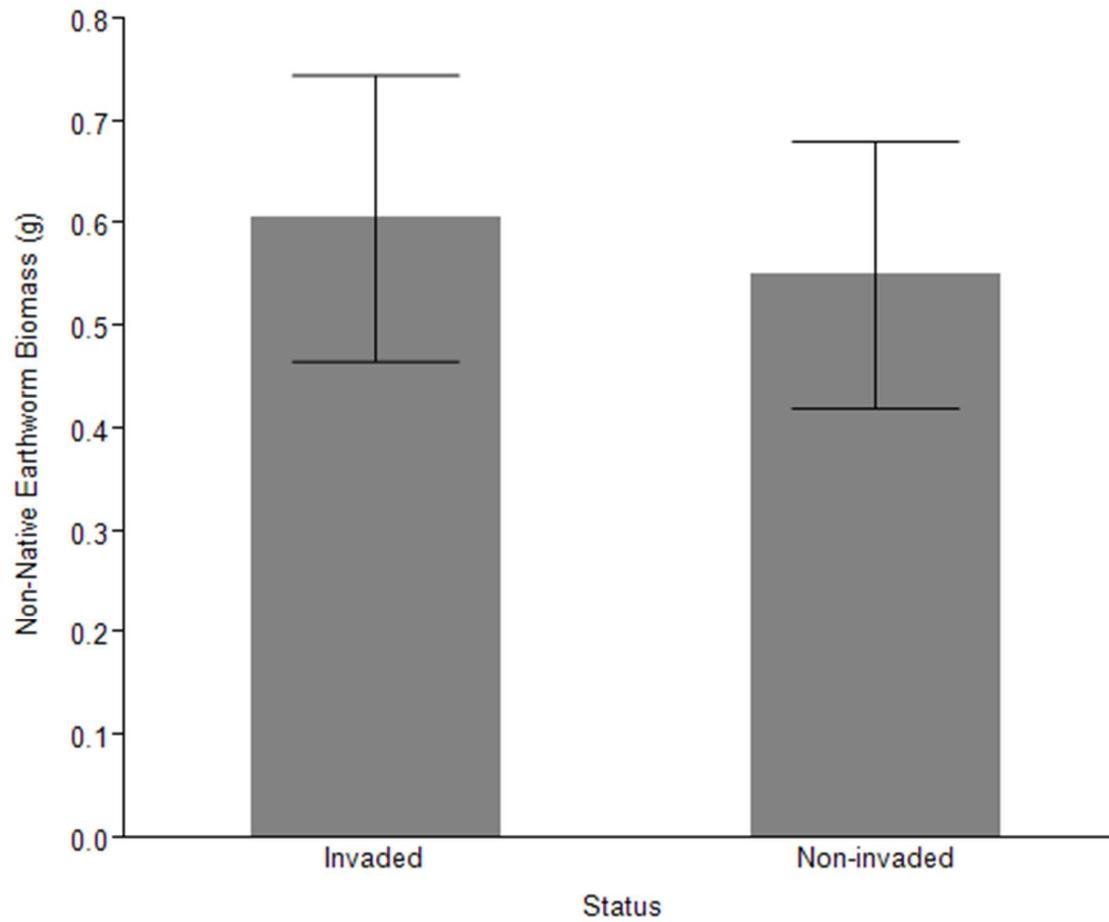
Figure 3. Mean non-native earthworm biomass in plots invaded by *Alliaria petiolata* and plots not invaded by the plant. Biomass data were 2nd root transformed. T = -0.2178, p = 0.776, n = 20.
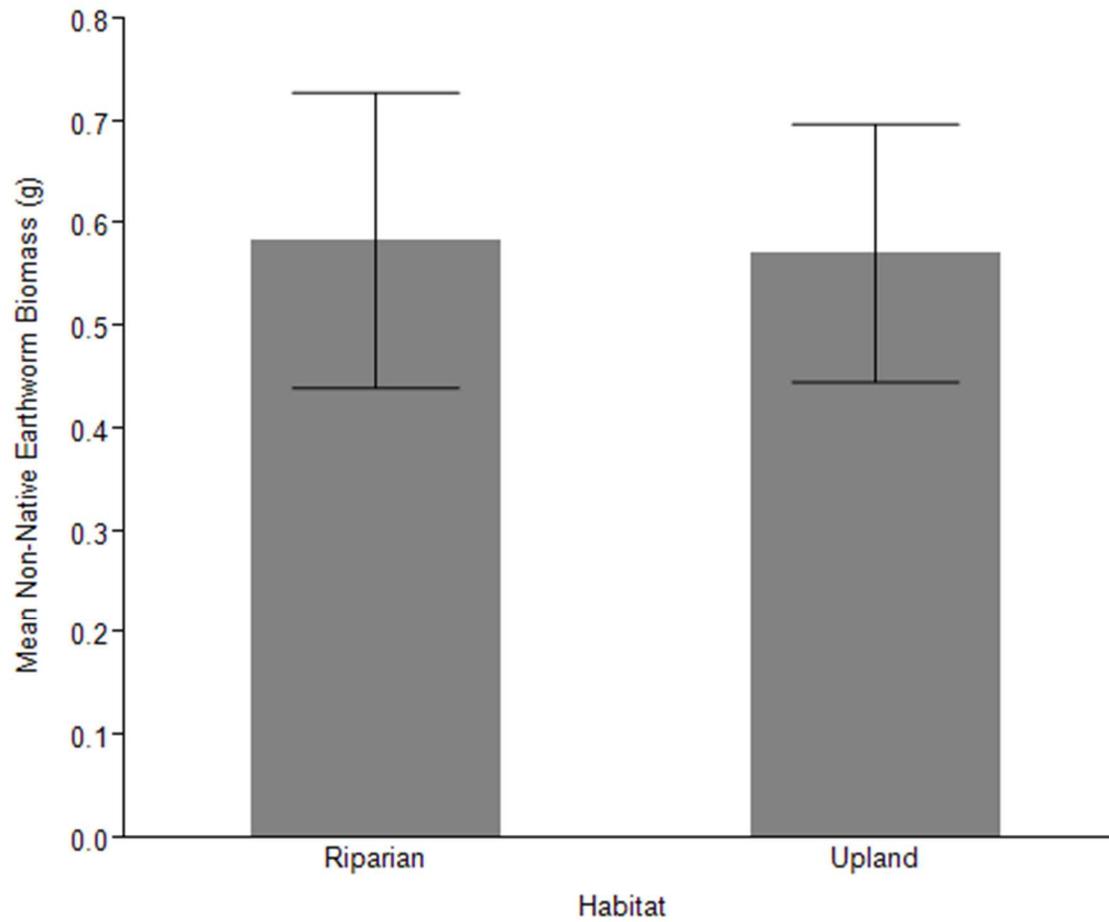
Figure 4. Mean non-native earthworm biomass in upland and riparian plots. T = -0.0655, p = 0.9485, n = 20.
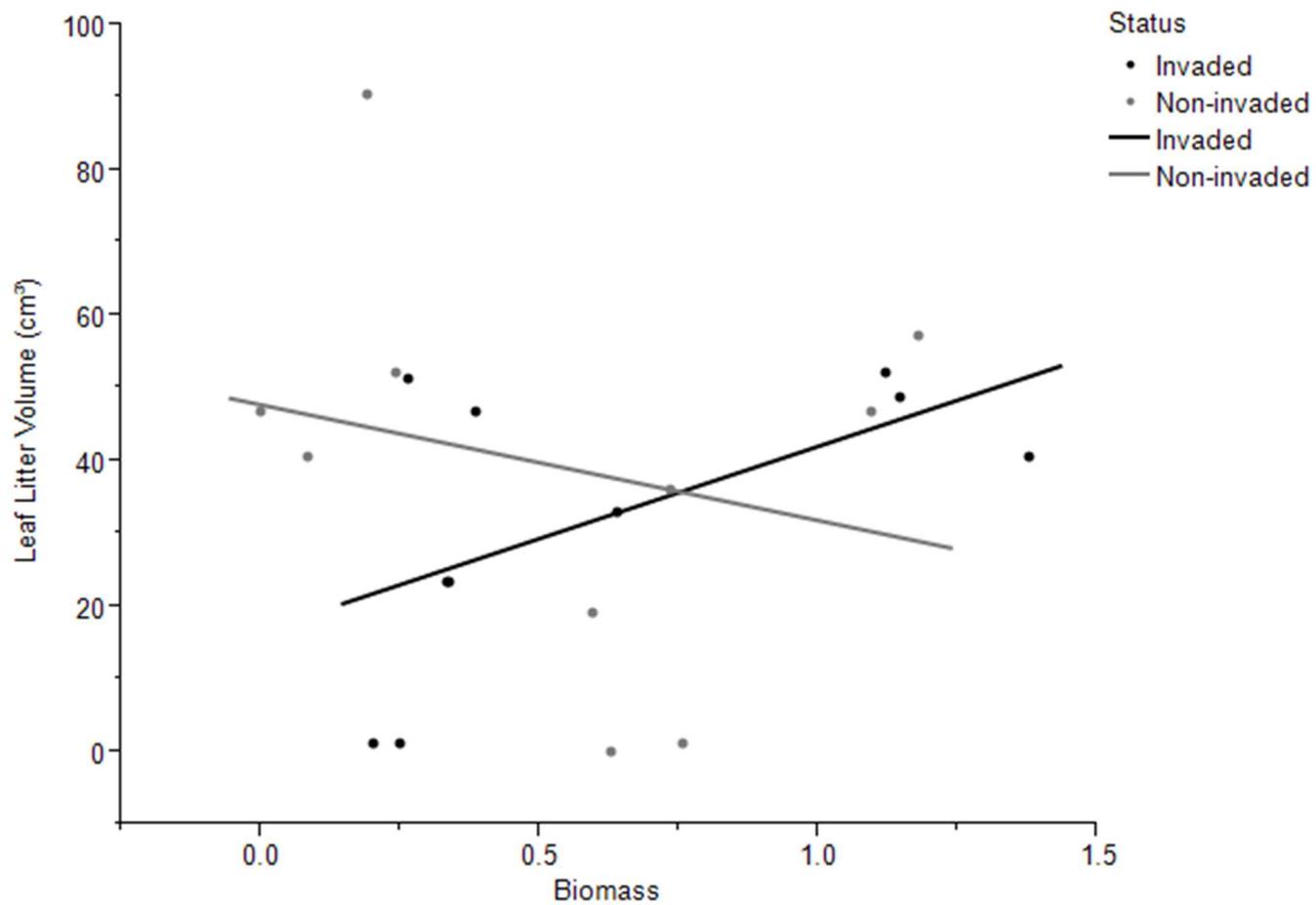
Figure 5. Leaf litter volume as a function of earthworm biomass in invaded (R^2 = 0.33, F = 3.9418, p = 0.0824, n = 10) and non-invaded plots (R^2 = 0.0581, F = 0.4398, p = 0.5022, n = 10).
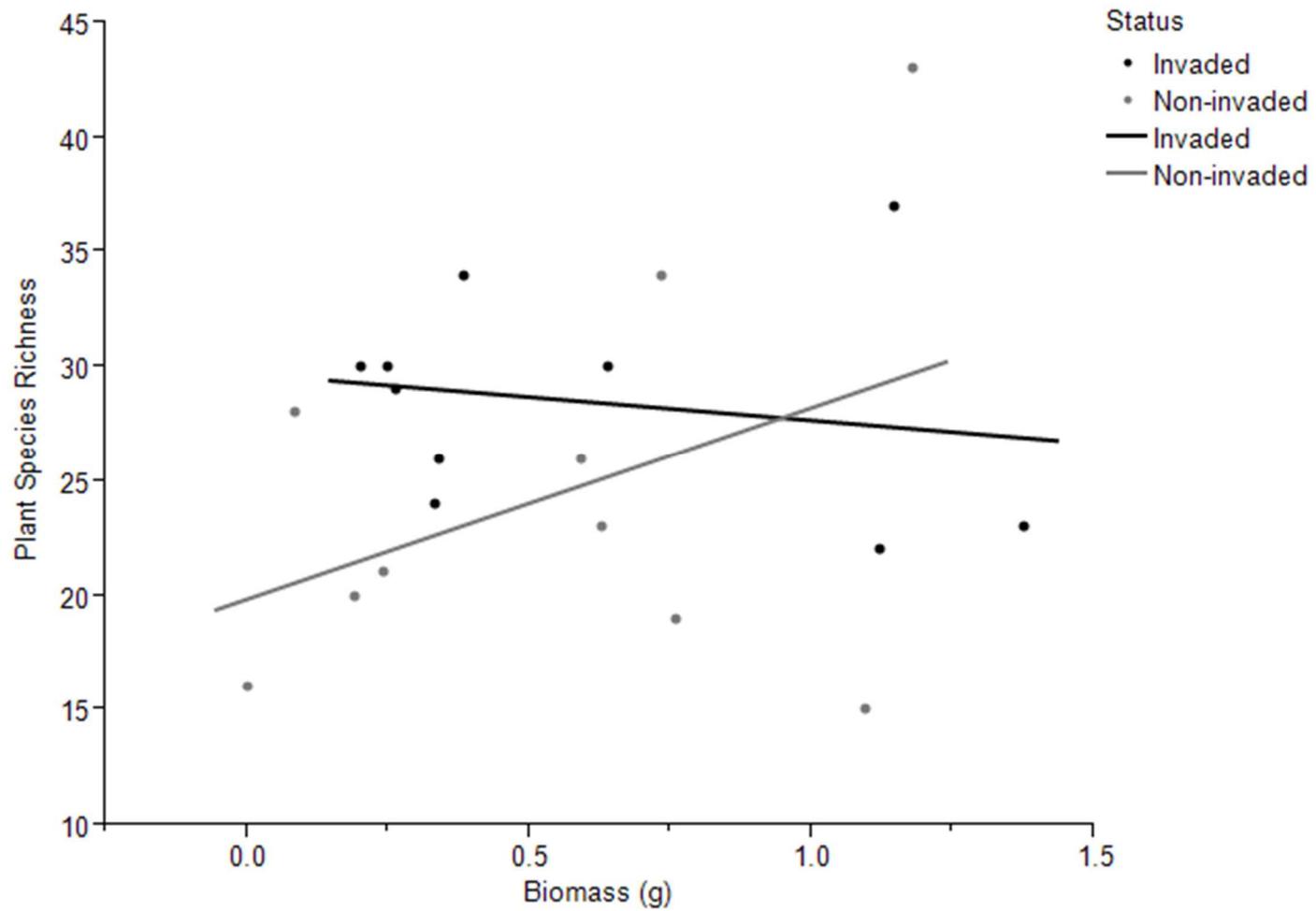
Figure 6. Plant species richness as a function of earthworm biomass in invaded (R^2 = 0.0347, F = 0.2881, p = 0.6061, n = 10) and non-invaded (R^2 = 0.159, F = 1.5122, p = 0.2537, n = 10) plots.
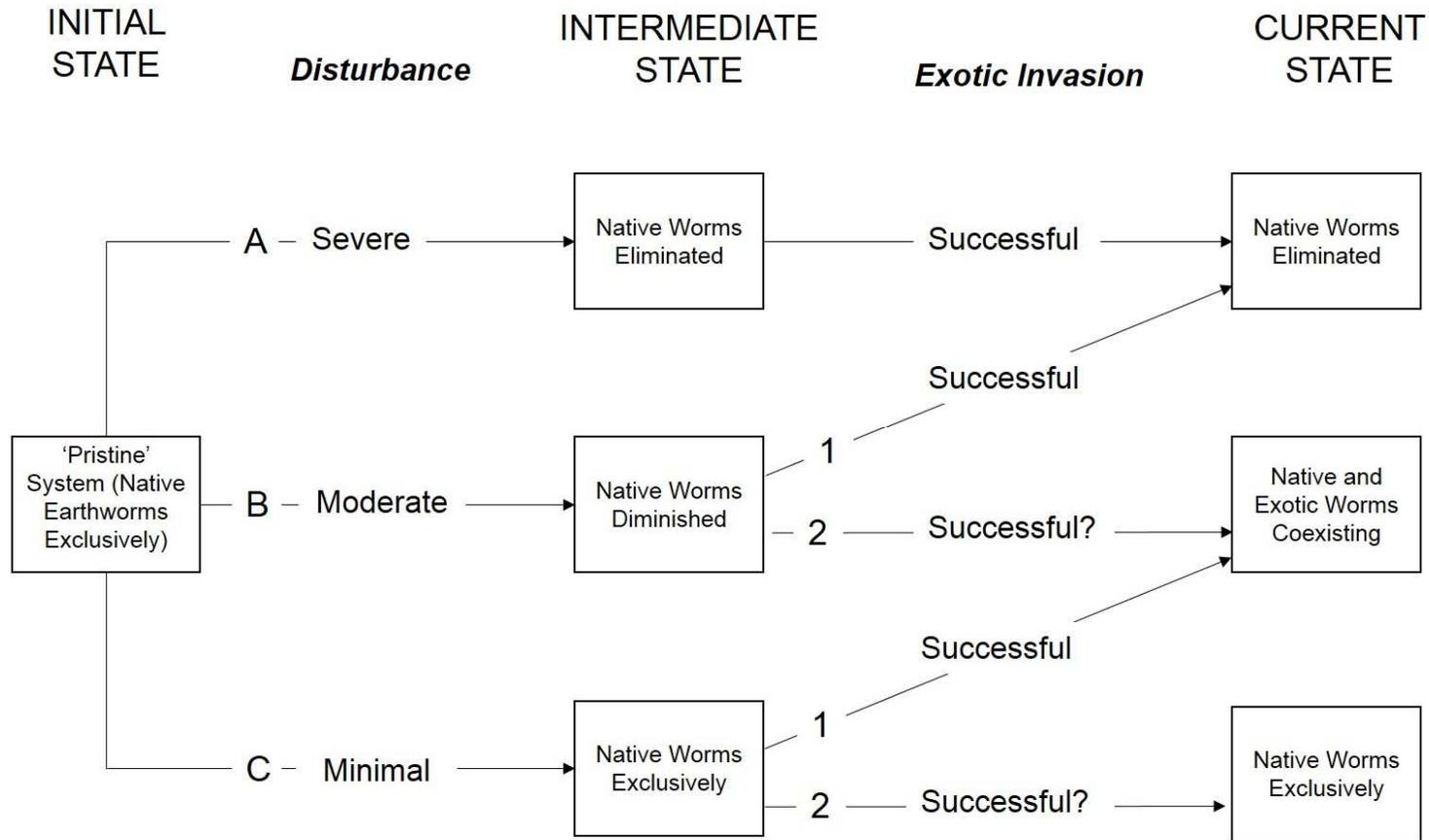
Figure 7. Diagram from Hendrix et al. (2006) showing the hypothesized invasional sequence of non-native earthworms into habitats that experienced different levels of disturbance. Used with permission.

CONCLUSION

Alliaria petiolata is devastating to native forest understory plant assemblages in many regions of the United States and Canada, reducing biodiversity of the native herbaceous layer (Anderson et al. 1996, Godwin 2012) and inhibiting the regeneration of important native tree species (McCarthy 1999, Stinson et al. 2006). The situation is quite bleak, with investigators using words such as "dominant" and "permanent" to describe its residence within North American forests (Nuzzo 2002, Godwin 2012). The purpose of the present study was to document the invasion of *A. petiolata* in the southern Ozark Plateaus, and to characterize the plant's growth and reproductive habits, population densities, seedling mortality, and the habitat types it most commonly invades in this region.

Plants were documented in more than 60 locations in 17 Ozark counties, and through the use of herbarium specimens and other reliable information sources, it appears that the plant has been steadily spreading throughout the region over the past four decades and that the rate of spread is increasing. Habitats in which the plant was found included floodplain forests and mesic upland forests, as well as roadsides with full sun exposure. In some locations infestations were quite dense, but in others, it appeared populations were not yet well-established. At this time, overall population densities are much lower than they are in the Midwest and Northeast, as might be expected for a plant that has only been in a region for about 40 years. In fact, in several floodplain locations only a small, dense cluster of plants was found growing at the base of a tree where flooding had likely deposited seeds. However, based on a few particularly dense populations that have been observed, especially one currently growing at Hobbs State Park, it is very likely that this is just the beginning of a very serious invasion by *A. petiolata*, and that it will be impossible to eradicate if it is allowed to continue to grow unchecked.

In addition, it is clear that winters in the southern Ozarks are sufficiently cold enough to stratify *A. petiolata* seeds, but are also mild enough to allow for a very low rosette mortality rate. Rosettes are able to grow and add biomass in the winter when temperatures are above freezing, which leads to increased seed production in the spring. These factors may contribute to the continued success of *A. petiolata* in the southern Ozarks over time.

Non-native earthworms have been blamed for the localized extinction of a rare fern (Gundale 2002), a decline in the abundance of woodland salamanders and two species of birds (Maerz et al. 2009,

Loss et al. 2012), and have been blamed for facilitating invasions by non-native plant species in northern forests of the United States (Hale et al. 2006, Nuzzo et al. 2009). Analyses of data collected in plots within the southern Ozarks does not indicate that the presence of non-native earthworms is causing a decrease in plant species richness or in leaf litter volume in this region. The data collected also did not support the hypothesis that non-native earthworms would be more abundant in plots inhabited by *A. petiolata*, and therefore may be facilitating invasions by the invasive plant. However, non-native earthworms were documented in nearly every plot sampled, and were found to be far more abundant in those plots than were native earthworms, which were present in small amounts in only three plots. Despite the of lack of information on the abundances and distributions of native earthworms in the region prior to this study, it is reasonable to conclude, based on species richness the region, that they were once far more abundant than they are today, and as a result of ecological disturbances followed by invasion of their habitats by non-native earthworms (Hendrix et al. 2006), their numbers have decreased dramatically over time.

Invasions by non-native species are a major ecological issue because of their ability to reduce biodiversity of native species. *Alliaria petiolata* is one of the most important and destructive invasive plants in forests of the eastern United States, and it is now a member of hardwood forests in the Ozarks. Because *A. petiolata* has demonstrated an ability to invade a wide variety of habitat types, is self-fertile, is capable of altering pH and soil nutrient levels in the soil when plants reach sufficient densities (Rodgers et al. 2008), and shades out native tree seedlings (Stinson et al. 2006), its presence in the southern Ozarks should be taken very seriously when it is observed growing, and populations should be removed before they become too large to manage. It is the sincere hope of the author that the present study will not only contribute to the growing body of work being conducted on *A. petiolata*, but that it will be used locally as a source of information for land managers in both private and public sectors to effectively control the plant in this region.

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APPENDIX A

All vascular plant species identified each plot. A = invaded. B = non-invaded. Site codes: E1 = Lake Eucha 1; E2 = Lake Eucha 2; SP = Spavinaw Creek; GR = Gravette; BF = Baker Ford; DE = Decatur; PI = Pineville; MH = Mt. Hersey; HA = Hasty; TB = Tyler Bend. Note: * = non-native species. ** = invasive species. Nomenclature follows Gentry et al. (2013).

	Site																			
	E1		E2		SP		GR		BF		DE		PI		MH		HA		TB	
Species	A	B	A	B	A	B	A	B	A	B	A	B	A	B	A	B	A	B	A	B
TREES																				
Acer negundo L.			x		x		x	x	x		x				x	x	x	x	x	
Acer rubrum L. var. *rubrum*			x	x	x		x	x	x	x	x				x		x	x	x	x
Acer saccharum Marshall var. *saccharum*			x						x		x		x		x					
Aesculus glabra Willd.	x																			
Asimina triloba (L.) Dunal									x						x		x		x	x
Carya cordiformis (Wangenh.) K. Koch	x		x		x		x	x	x											
Carya tomentosa (Lam.) Nutt.			x																	
Carya sp.	x																			
Celtis laevigata (Willd.)	x				x				x						x					
Cercis canadensis L. var. *canadensis*							x	x	x	x								x		
Cornus florida L.				x					x		x	x								
Diospyros virginiana L.								x												
Fraxinus americana L.			x	x			x				x		x							
Fraxinus pennsylvanica Marshall				x								x								x
Gleditsia triacanthos L.												x			x		x			
Juglans nigra L.	x				x								x							
Juniperus virginiana L. var. *virginiana*																				x
Liquidambar styraciflua L.				x					x	x					x					
Maclura pomifera (Raf.) C.K. Schneid.				x		x			x											

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Species	E1 A	E1 B	E2 A	E2 B	SP A	SP B	GR A	GR B	BF A	BF B	DE A	DE B	PI A	PI B	MH A	MH B	HA A	HA B	TB A	TB B
TREES (Cont.)																				
Morus rubra L.	x																x			
Nyssa sylvatica Marshall				x				x					x				x			
Ostrya virginiana (Mill.) K. Koch				x					x	x							x	x		
Pinus echinata Mill.								x			x									
Platanus occidentalis L.					x										x					
Prunus serotina Ehrh.				x				x			x	x								
Ptelea trifoliata L.									x											
Quercus alba L.							x			x	x	x			x					
Quercus falcata Michx.												x				x		x		
Quercus muehlenbergii Engelm.	x			x			x	x							x	x				
Quercus rubra L.				x				x	x	x										x
Quercus sp. (red oak group)																				x
Quercus sp. (white oak group)																		x		
Rosaceae																		x		
Sassafras albidum (Nutt.) Nees											x	x			x					x
Tilia americana L. var. *americana*			x											x				x		
Ulmus alata Michx.							x	x					x			x		x		
Ulmus americana L.							x	x			x	x	x		x	x				x
Ulmus rubra Muhl.	x																			x
Ulmus sp.			x				x											x		
SHRUBS																				
Arundinaria gigantea (Walter) Muhl.																x				

Species — Site

Species	E1 A	E1 B	E2 A	E2 B	SP A	SP B	GR A	GR B	BF A	BF B	DE A	DE B	PI A	PI B	MH A	MH B	HA A	HA B	TB A	TB B
SHRUBS (Cont.)																				
Castanea ozarkensis Ashe						x							x							
Corylus americana Walter											x									
Frangula caroliniana (Walter) A. Gray											x					x		x		
Lindera benzoin (L.) Blume	x		x		x	x	x	x	x			x			x	x	x	x	x	x
Rhus aromatica Aiton var. *aromatica*										x										
Rosa carolina L.	x										x									
Rubus frondosus Bigelow											x									
Rubus leviculus L.H. Bailey					x															
Rubus sp.																x				
Symphoricarpos orbiculatus Moench	x		x		x			x	x			x	x	x	x		x	x		
Vaccinium stamineum L.											x									
Viburnum prunifolium L.																x				
VINES																				
Aristolochia tomentosa Sims																				
Berchemia scandens (Hill) K. Koch																x				
Campsis radicans (L.) Seem. ex Bureau	x												x							
Cocculus carolinus (L.) DC.																	x			
Dioscorea villosa L.							x													
Euonymus fortunei (Turcz.) Hand.-Mazz.**	x															x				
Lonicera japonica Thunb. ex Murray**			x				x													
Menispermum canadense L.													x							
Parthenocissus quinquefolia (L.) Planch	x		x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x

Species	E1		E2		SP		GR		BF		DE		PI		MH		HA		TB	
	A	B	A	B	A	B	A	B	A	B	A	B	A	B	A	B	A	B	A	B
VINES (Cont.)																				
Smilax glauca Walter						x														
Smilax hispida Raf.	x																			
Smilax sp.									x							x	x	x		
Toxicodendron radicans (L.) Kuntze									x	x					x	x		x	x	x
Vitis sp.			x	x			x	x	x	x						x	x			x
HERBS																				
Agrimonia rostellata Wallr.	x									x						x	x			
Alliaria petiolata (M.Bieb.) Cavara & Grande**	x		x		x		x		x		x		x		x		x		x	
Allium canadense L. var. *canadense*	x																			
Allium sp.			x						x						x					x
Amphicarpaea bracteata (L.) Fernald							x	x	x	x	x									
Aplectrum hyemale (Muhl. ex Willd.) Nutt														x						
Arisaema triphyllum (L.) Schott in Schott & Endl.																x				
Asarum canadense L.					x	x	x	x											x	x
Asplenium platyneuron (L.) Britton, Sterns & Poggenb.							x									x		x		
Asteraceae							x													
Boehmeria cylindrica (L.) Sw.	x						x					x			x					
Botrychium virginianum (L.) Sw.													x							
Bromus pubescens Muhl. ex Willd.	x																			
Cardamine concatenata (Michx.) O. Schwarz				x							x								x	x
Cardamine hirsuta L.*				x	x	x			x								x	x	x	x
Carex blanda Dewey	x				x	x												x		

Species	Site E1		E2		SP		GR		BF		DE		PI		MH		HA		TB	
	A	B	A	B	A	B	A	B	A	B	A	B	A	B	A	B	A	B	A	B
HERBS (Cont.)																				
Carex jamesii Schwein.	x				x	x								x		x		x		
Carex oligocarpa Schkuhr ex Willd.	x																			
Carex sp.			x						x										x	x
Cerastium brachypetalum Pers.*											x									
Cerastium glomeratum Thuill.*					x															
Cerastium sp.									x											
Chaerophyllum procumbens (L.) Crantz			x		x	x			x				x	x		x	x	x	x	x
Chaerophyllum tainturieri Hook.	x	x																		
Chasmanthium latifolium (Michx.) H.O. Yates	x	x			x	x									x	x				
Claytonia virginica L.																x				
Clematis catesbyana Pursh											x									
Commelina sp.			x																	
Corydalis sp.																			x	
Cryptotaenia canadensis (L.) DC.																x			x	x
Cynoglossum virginianum L.																x				
Desmodium cuspidatum (Muhl. ex Willd.) DC ex Loudon							x													
Desmodium glutinosum (Muhl. ex Willd.) A.W. Wood									x	x										
Desmodium nudiflorum (L.) DC.							x	x			x	x	x		x	x				
Desmodium sp.			x	x																
Diarrhena americana P. Beauv.	x				x	x														
Dichanthelium boscii (Poir.) Gould & C.A. Clark									x	x					x	x				
Dichanthelium commutatum (Schult.) Gould																x				

Species	E1		E2		SP		GR		BF		DE		PI		MH		HA		TB	
	A	B	A	B	A	B	A	B	A	B	A	B	A	B	A	B	A	B	A	B
HERBS (Cont.)																				
Elephantopus carolinianus Raeusch.															x					
Elymus virginicus L. var. *virginicus*	x	x			x	x							x	x					x	x
Enemion biternatum Raf.			x	x						x						x				
Erythronium albidum Nutt.																			x	x
Erythronium rostratum W. Wolf			x	x	x															
Galium aparine L.	x	x			x								x	x			x			
Galium circaezans Michx.											x	x								
Galium concinnum Torr. & A. Gray									x	x										
Galium tricornutum Dandy*							x				x	x								
Galium sp.			x				x												x	
Geranium maculatum L.												x								
Geum canadense Jacq.													x			x				
Glechoma hederacea L.*												x								
Hydrastis canadensis L.																			x	
Hydrophyllum appendiculatum Michx.																		x		
Impatiens capensis Meerb.														x						
Lamium purpureum L.*	x	x	x		x	x			x				x	x			x	x	x	x
Laportea canadensis (L.) Wedd.						x			x						x		x		x	x
Lepidium virginicum L.						x														
Microstegium vimineum (Trin.) A. Camus**			x						x								x	x	x	
Myosotis verna Nutt.																			x	
Onoclea sensibilis L.																		x		
Osmorhiza sp.											x									

Species	E1		E2		SP		GR		BF		DE		PI		MH		HA		TB	
Site	A	B	A	B	A	B	A	B	A	B	A	B	A	B	A	B	A	B	A	B
HERBS (Cont.)																				
Oxalis violacea L.															x					
Parietaria pensylvanica Muhl. ex Willd.			x																	
Perilla frutescens (L.) Britton**		x	x			x											x			
Persicaria lapathifolia (L.) Gray															x					
Persicaria virginiana (L.) Gaertn.	x	x			x	x			x						x					
Phacelia hirsuta Nutt.																	x			
Phacelia ranunculacea (Nutt.) Constance															x					
Phegopteris hexagonoptera (Michx.) Fée										x										
Phlox pilosa L. ssp. *pilosa*	x					x														
Pilea pumila (L.) A. Gray															x					
Poa sylvestris A. Gray						x			x						x			x		
Poaceae				x								x								x
Podophyllum peltatum L.							x	x	x				x	x						
Polygonatum biflorum (Walter) Elliott			x		x															
Polymnia canadensis L.															x					
Polystichum acrostichoides (Michx.) Schott							x													
Prenanthes altissima L.										x										
Ranunculus micranthus Nutt. in Torr. & A. Gray	x	x				x							x				x	x		
Ranunculus sp.		x																		
Sanguinaria canadensis L.															x					
Sanicula canadensis L.									x	x					x		x		x	x
Silphium perfoliatum L. var. *perfoliatum*		x																		
Viola striata Aiton					x	x									x	x				x

Species	Site																			
	E1		E2		SP		GR		BF		DE		PI		MH		HA		TB	
	A	B	A	B	A	B	A	B	A	B	A	B	A	B	A	B	A	B	A	B
HERBS (Cont.)																				
Viola villosa Walter	x																			x
Viola sp.							x						x						x	x
Woodsia obtusa (Spreng.) Torr.															x	x				
Zizia aurea (L.) W.D.J. Koch												x								
Stellaria media (L.) Vill.*	x	x	x		x	x			x		x		x	x	x	x	x	x	x	x
Thalictrum thalictroides (L.) A.J. Eames & B. Boivin	x		x						x	x	x					x			x	x
Tradescantia ernestiana E.S. Anderson & Woodson	x																			
Tradescantia ohiensis Raf.					x															
Trillium sp.			x														x			x
Valerianella sp.																			x	x
Verbesina alternifolia (L.) Britton ex Kearney	x	x				x											x	x	x	x
Veronica hederifolia L.*									x										x	
Viola pubescens Aiton	x	x																		
Viola sororia Willd.																x				

APPENDIX B

GPS coordinates of each occurrence of *Alliaria petiolata* known within the southern Ozark Plateaus Ecoregion. Locations marked with an asterisk (*) are those that could not be re-located during the course of the present study. Entries marked in bold were used as study sites. Except where noted in the Source column, occurrences were found during the course of the present study. Note: UARK = University of Arkansas Herbarium; OU = Robert Bebb Herbarium, University of Oklahoma; MO = Missouri Botanical Garden; BUFF = Buffalo National River Vegetation Mapping Project.

Location	Source	County	Elevation (m)	Latitude	Longitude
ARKANSAS					
Decatur		**Benton**	**342**	**36.379510**	**-94.484600**
Decatur, Hwy. 102 and Hwy. 59	UARK; J. Test; #XIII-1-14; 1972	Benton	352	36.371982	-94.469701
Gravette, Hwy 72		Benton	360	36.419830	-94.442740
Gravette		**Benton**	**373**	**36.430730**	**-94.438560**
Hobbs State Park, Sinking Stream Trail		Benton	369	36.296150	-93.959220
Hobbs State Park, Van Winkle Trail		Benton	343	36.298500	-93.957270
Illinois River, Fisher Ford		Benton	287	36.122990	-94.515440
Illinois River, Hwy 59 Bridge		Benton	285	36.107910	-94.532910
Lowell, Hwy 264 and CR 236		Benton	393	36.255502	-94.186480
Maysville Cemetery		Benton	318	36.415590	-94.596990
Maysville, Beaty Creek		Benton	307	36.416848	-94.599810
Mount Pleasant		Benton	397	36.423770	-94.388170
Property of Art Evans	UARK; T. Witsell; #09-173; 2009	Benton	354	36.378250	-94.481820
Sulphur Springs, roadside		Benton	287	36.479800	-94.455490
Harrison, Minnie Harris Park		Boone	322	36.228250	-93.103470

Location	Source	County	Elevation (m)	Latitude	Longitude
ARKANSAS (cont.)					
Eureka Springs, near Hwy 62 and College St.		Carroll	447	36.393810	-93.737810
Mulberry River SW of Cass	UARK; G. Barber; #2394; 1982	Franklin	224	35.662426	-93.844392
Guion*	Brent Baker, pers. comm.	Izard	89	35.931105	-91.951232
White River, Mt. Olive Access*	UARK; S. Nunn; #5430; 2009	Izard	95	35.997347	-92.092614
Draketown		Madison	401	36.020370	-93.870320
BNR, Buffalo Point	UARK; D.R. Bradford; #23; 1987	Marion	148	36.068852	-92.556887
BNR, Buffalo Point	BUFF, 2007	Marion	148	36.067550	-92.575670
BNR, Dillard's Ferry		Marion	160	36.066800	-92.578160
BNR, Rush		Marion	145	36.122404	-92.550270
BNR, Baker Ford		**Newton**	**175**	**35.980750**	**-92.813630**
BNR, Carver		Newton	219	35.982930	-93.041020
BNR, Hasty	BUFF, 2006	Newton	226	36.008450	-93.081890
BNR, Hasty		**Newton**	**233**	**36.007870**	**-93.081350**
BNR, Mt. Hersey	BUFF, 2007	Newton	212	36.011100	-92.952240
BNR, Mt. Hersey		**Newton**	**228**	**36.010350**	**-92.951610**
BNR, Mt. Hersey parking area		Newton	215	36.009730	-92.952730
Mt. Hersey Rd, NE of Mt. Hersey access		Newton	232	36.018890	-92.945160
Mt. Hersey Rd, NW of Mt. Hersey access		**Newton**	231	36.015820	-92.961790
BNR, Arnold Bluff	BUFF, 2007	Searcy	199	35.975830	-92.795250

Location	Source	County	Elevation (m)	Latitude	Longitude
ARKANSAS (cont.)					
BNR, Gilbert		Searcy	174	35.987300	-92.714810
BNR, Grinder's Ferry		Searcy	174	35.984860	-92.744130
BNR, Maumee South		Searcy	161	36.039100	-92.636120
BNR, Richland Creek		Searcy	215	35.927679	-92.892336
BNR, Shine Eye		Searcy	181	35.988080	-92.734590
BNR, Spring Creek		Searcy	188	36.030380	-92.584630
BNR, The Narrows		Searcy	205	35.965725	-92.902528
BNR, Tyler Bend, Rock Wall Trail	**Brent Baker, pers. comm.**	**Searcy**	**181**	**35.981230**	**-92.753200**
BNR, Tyler Bend Campground	UARK; M.A. Beer; #92-135; 1992	Searcy	179	35.989402	-92.762467
BNR, Tyler Bend near launch ramp	BUFF, 2006	Searcy	176	35.990170	-92.764160
BNR, Tyler Bend picnic area	BUFF, 2007	Searcy	180	35.989480	-92.765990
BNR, Tyler Bend, River View Trail		Searcy	216	35.983140	-92.766730
BNR, Woolum		Searcy	203	35.969910	-92.883160
BNR, Woolum	BUFF, 2007	Searcy	199	35.968960	-92.886990
St. Joe, Baker Ford Rd.		Searcy	264	35.989970	-92.820740
Optimus, White River		Stone	96	36.033760	-92.058200
Little Red River, Rumley Rd.*	UARK; M.A. Beer; #92-136; 1992	Van Buren	271	35.787980	-92.510860
Fayetteville, Mud Creek		Washington	361	36.119320	-94.143660
White River, Baptist Ford, S of Greenland	UARK; J. Lanter; #13.2; 1980	Washington	377	35.981570	-94.174640

Location	Source	County	Elevation (m)	Latitude	Longitude
ARKANSAS (cont.)					
White River, Baptist Ford, S of Greenland	UARK; N. Green; #109; 1984	Washington	377	35.981570	-94.174640
White River, SE of Fayetteville	UARK; R. Reese; #11; 2001	Washington	364	36.014416	-94.142478
White River, West Fork, Dye Creek Rd.		Washington	392	35.940710	-94.187100
White River, S of West Fork		Washington	403	35.913910	-94.179890
MISSOURI					
Roaring River State Park	MO; D. Aubrey; #29; 2002	Barry	341	36.589060	-93.843400
Brandsville*	MO; B. Summers; 9643; 2001	Howell	286	36.704223	-91.693884
Noel		McDonald	251	36.518890	-94.483040
Noel city park		McDonald	257	36.544360	-94.488150
Pineville		**McDonald**	**279**	**36.571270**	**-94.409100**
Elk River		McDonald	263	36.588230	-94.391040
Elk River, Cowskin Access		McDonald	234	36.631296	-94.588600
OKLAHOMA					
Lake Eucha	**OU; A.A. Reznicek; 9769; 1994**	**Delaware**	**237**	**36.355040**	**-94.794850**
Lake Eucha		**Delaware**	**269**	**36.346920**	**-94.822810**
Choleta, CR 4530		Delaware	246	36.391664	-94.944125
Spavinaw Creek	**OU; A. Buthod; #AB-8123; 2009**	**Delaware**	**207**	**36.402050**	**-94.964390**
Disney		Mayes	232	36.479657	-95.007591

APPENDIX C

Graphs showing the concentrations of variables measured in soil samples of invaded and non-invaded plots in 2011 and 2012.