

Community Mulch Piles May Contribute to Dispersal of *Brachyponera chinensis* (Hymenoptera: Formicidae)¹

Emilee M. Poole² and Scott Horn

USDA Forest Service, Southern Research Station, Athens, GA 30602

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It is well documented that human-mediated dispersal of nonnative species plays a large role in the success of invasion (Liebhold and Tobin 2008, Annu. Rev. Entomol. 53: 387–408; McClure 1990, Environ. Entomol. 19(1): 36–43; Suarez et al. 2001, Proc. Natl. Acad. Sci. U. S. A. 98(3): 1095–1100). Nonnative invasive insects, particularly ants, are found across many terrestrial ecosystems worldwide (Bradshaw et al. 2016, Nat. Commun. 7: 12986). Aspects of their biology, colony and nest structure, diet, habitat preferences, and the ease of dispersal make invasive ants challenging pests (Holway et al. 2002, Annu. Rev. Ecol. Syst. 33: 181–233), and they often prove difficult to manage once established (Morrison et al. 2004, Biol. Invasions 6: 183–191). As climate conditions continue to change, predictions show that many problematic ant species may be capable of global range expansion into novel habitats (Bertlesmeire et al. 2013, PLoS ONE 8(10): e75438). With the costs and logistical hurdles of early detection, quarantine, and eradication (Martinez et al. 2019, Biol. Invasions 22: 75–100), there are mounting pressures to prevent additional introductions of invasive ants. Some invasive ant species threaten human and pet health with stings and severe reactions, and there are additional concerns over economic losses in agricultural industries (Lard et al. 2002, Southw. Entomol. Suppl. 25: 123–137; Lard et al. 2006, An Economic Impact of Imported Fire Ants in the United States of America, Texas A&M Univ., College Station, TX). Furthermore, invasive ant species may cause ecological damage by negatively affecting native ant species (Holway et al. 2002; Porter and Savignano 1990, Ecology 71(6): 2095–2106), native insect–plant interactions, plant distributions, nutrient cycling (Milligan et al. 2011, J. Ecol. 110(2): 359–373), and arthropod diversity. Studies also have shown that invasive ants can be detrimental to numerous vertebrate and invertebrate species (Hooper-Bùi et al. 2004, Sociobiology 44(3): 401–418; Siddiqui et al. 2021, Environ. Sci. Pollut. Res. Int. 28: 54362–54382).

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²Corresponding author (email: Emilee.poole@usda.gov).

Until recently, the Asian needle ant (*Brachyponera chinensis* Emery) has maintained a generally negligible presence since it was first recorded in the United States in the 1930s (Smith 1934, Ann. Entomol. Soc. Am. 27: 557–564). On the basis of previous work (Guénard and Dunn 2010, PLoS ONE 5: e11614; Guénard et al. 2018, Fla. Entomol. 101(4): 649–656), *B. chinensis* has increased in abundance, and its range continues to expand throughout terrestrial ecosystems in the eastern United States. Efforts have been made to document the current range and potential ecological impacts of *B. chinensis* (Bertelsmeier et al. 2013; PLoS ONE 8(10): e75438; Guénard and Dunn 2010; Merchlinsky et al. 2023, Ecol. Entomol. 48(5): 588–596). Previous studies have focused on the ability of *B. chinensis* to affect native biodiversity. For example, work has shown that *B. chinensis* can displace native ant species, particularly small, seed-dispersing ants (Guénard and Dunn 2010; Spicer-Rice and Silverman 2013, Anim. Behav. 86(3): 497–506) and further the impact by altering the distribution of ant-dispersed native plants (Rodriguez-Cabal et al. 2012, Biol. Invasions 4(3): 557–565; Warren 2015, Biol. Invasions 17: 3153–3162). Along with ecological impacts, public health concerns have been raised over the ability of *B. chinensis* to deliver a potent sting which often causes severe localized reactions that can escalate to anaphylaxis and life-threatening allergic reactions (Bae et al. 1999, Korean J. Prev. Med. 32: 421–426; C. M. Hawley, pers. comm.; Lee et al. 2009, Clin. Exp. Allergy 39: 602–607; Nelder et al. 2006, J. Med. Entomol. 43: 1094–1098). This species does not have obvious indicators of their presence, such as mounds, but rather are found in damp habitats (Brown 1958, Acta Hymenopterol. 1(1): 1–50; Smith 1934), in and around coarse woody debris, landscape, construction, and agricultural materials (E.M.P. and S.H., pers. obs.). These nesting habits, along with their success as small colonies, further the difficulty in determining the presence of *B. chinensis* and implementing any management efforts.

Brachyponera chinensis is prevalent in undisturbed forests to highly disturbed urban habitats (Guénard and Dunn 2010). Because of their preference for generally damp environments (Zungoli et al. 2008, Asian Needle Ant, *Pachycondyla chinensis* (Emery), Clemson Univ. Cooperative Extension Service, Clemson, SC; Pecarovic et al. 2010, PLoS ONE 5(10): e13222), mulch or large volumes of wood chips seem to attract *B. chinensis* (Suehiro et al. 2017, Sci. Rep. 7: 15016). Therefore, our objective was to use a long-standing community mulch pile as a model system to determine if it could be serving as a source for unintentional ant dispersal.

Fieldwork was conducted at Harris Shoals Park in Watkinsville, GA (N 33° 51'46.1442", W 83°24'29.1384"), a small, high-traffic park situated around Calls Creek. The park is 30.2 ha and partially forested with recreational amenities. Most of the forested areas are dominated by hardwoods, such as sweetgum (*Liquidambar styraciflua* L.), yellow poplar (*Liriodendron tulipifera* L.), red maple (*Acer rubrum* L.), and water oak (*Quercus nigra* L.) interspersed with loblolly pines (*Pinus taeda* L.). Mulch samples were collected from a large, well-established community-accessible mulch pile comprised of chipped tree and limb debris that was placed directly on mineral soil at the edge of a forest. Debris and decaying material not collected by the public is not removed from the pile, and new mulch is continuously added on top of existing material. Therefore, upon time of collection, the mulch pile varied in age.

Mulch samples were collected on 17 July and 27 September 2023. At each collection time, ten 4,310-cm³ samples were randomly collected from the upper and lower layers of the mulch pile ($n = 20$). The lower layer was characterized by old, decomposing, moist debris near the soil, whereas the upper layer was characterized by large, dry, whole wood chips. Ants were extracted into ethanol using Berlese funnels (a 25-W heat lamp and 30-cm funnel on top of a 13.25-L bucket), and samples were collected after 72 h. The number of *B. chinensis* were totaled from each sample.

All statistical analyses were performed in R version 4.2.2 with the rstatix package (Kassambara 2023, <https://CRAN.R-project.org/package=rstatix>). We compared *B. chinensis* abundance between mulch layers for both time periods using the nonparametric Wilcoxon rank sum test, since the data set did not meet the assumptions of a *t* test.

In total, 1,982 *B. chinensis* specimens were collected, with *B. chinensis* making up 98.7% of all ants collected. *Brachyponera chinensis* abundance in the upper and lower layers differed significantly in July (Wilcoxon rank sum test, $W = 85.5$, $P = 0.0073$), whereas there was little difference in abundance in September (Wilcoxon rank sum test, $W = 57.5$, $P = 0.59$). *Brachyponera chinensis* collected per sample ranged from 0 to 58 individuals in the upper layer and 0 to 704 in the lower layer in July, whereas collections ranged from 0 to 216 and 0 to 116 in the upper and lower layers, respectively, in September. The median *B. chinensis* abundance in the upper layer and lower layers for July was 0.5 (interquartile range [IQR] = 2.75) and 40 (IQR = 99.2), and median *B. chinensis* abundance in the upper layer and lower layers for September was 9 (IQR = 29.5) and 10 (IQR = 63.2), respectively (Fig. 1).

Brachyponera chinensis can invade a variety of terrestrial habitats, including undisturbed forested environments, putting arthropod communities, ecological processes, and humans at risk (Guénard and Dunn 2010; Guénard et al. 2018; Rodriguez-Cabal et al. 2012; Ulyshen et al. 2023, Curr. Biol. 33: 1346–1350). To our knowledge, this is the first effort to investigate a potential human-mediated dispersal mechanism of *B. chinensis*. Although there were significantly more ants collected in the lower layer of the mulch in July, there is still potential to amass *B. chinensis* when only targeting newer mulch in the upper layer. In fact, 50% of samples collected from the top layer in June contained at least one needle ant, and 90% of samples from all other layers and collection dates contained at least one needle ant. This observed difference between mulch layers is likely attributed to the increased moisture and reduced temperature fluctuations and solar exposure the lower layer provides, particularly in summer (Iqbal et al. 2020, Bull. Natl. Res. Centre 44: 75), since there was no statistical difference in *B. chinensis* abundance between the layers in September. It is likely that the high numbers in both the upper and lower layers, 216 and 704 individuals, respectively, were collected from active nests (MacGown 2009, Mids. Entomol. 2: 88–89).

Brachyponera chinensis nests vary dramatically in size, and it has been estimated that a nest can range from a few individuals to thousands (MacGown 2009; Zungoli et al. 2008). As a successful invader, *B. chinensis* can use both polygynous and polydomous behavior, so colony structure can include multiple interconnected nests and queens (Eyer et al. 2018, Mol. Ecol. 27(23): 4711–4724; Klotz et al. 2008; Urban Ants of North America and Europe Identification, Biology, and

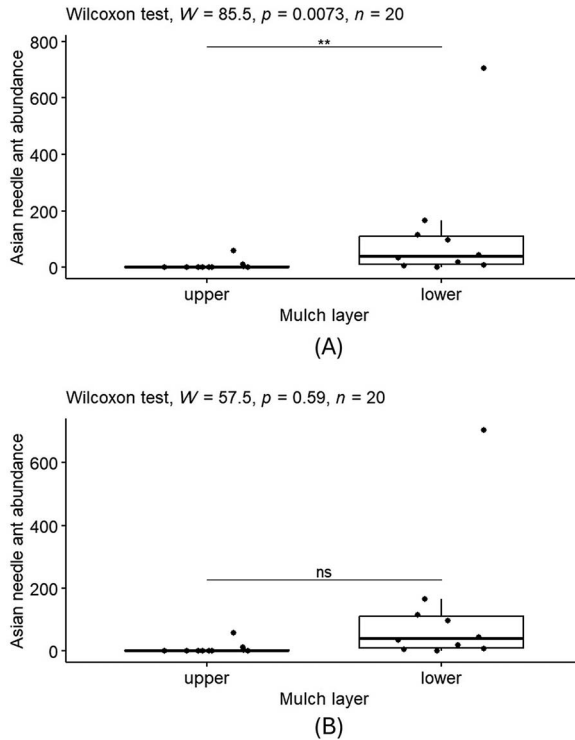


Fig. 1. Number of *Brachyponera chinensis* in upper and lower mulch layers in July (A) and September (B).

Management, Cornell Univ. Press, Ithaca, NY; Robinson 2014, Curr. Opin. Insect Sci. 5: 37–43). When gathering large volumes of mulch, there is potential to collect entire nests or colonies and unknowingly distribute queens, workers, and brood. Alternatively, citizens moving mulch can distribute partial colonies with workers and brood comprised of reproductive males and females, potentially leading to colony founding at a new site. Evidence supports that *B. chinensis* nests can function without a queen (Allen 2017, Biology and Behavior of the Asian Needle Ant, *Brachyponera chinensis* (Emery), Ph.D. Dissertation, Clemson Univ., Clemson, SC; Murata et al. 2017, Entomol. Sci. 20(1): 86–95). Over half the *B. chinensis* nests collected by Gotoh and Ito (2008, Insectes Soc. 55: 98–104) in its native range lacked queens. In lab conditions, workers collected without a queen will establish a new nest site and maintain the remaining brood; the longest surviving individuals in the lab with no brood or queen was 509 d (E.M.P., pers. obs.). Additionally, relocated workers may be able to integrate if added to an existing colony with little to no aggression (D. B. Booher, pers. comm.), thus strengthening an existing population of *B. chinensis*.

The full extent of Asian needle ant impact is unknown; however, accidentally aiding the already expanding range of the species jeopardizes biodiversity of the immediate area, puts adjacent habitats at risk (Bednar and Silverman 2011,

Insectes Soc.58(4): 459–467; Suehiro et al. 2017, Sci. Rep. 17: 15016; Ulyshen et al. 2023), and positions humans in proximity to this significant pest. Additionally, new introductions near streams and waterways have the potential to spread downstream (S.H., pers. obs.). These potential cascades initiated by accidental dispersal can increase the connectivity of existing populations and expedite the expansion of Asian needle ant in both natural and anthropogenic habitats. Studies are currently underway in our lab to determine at-risk arthropod species in invaded ecosystems.

In summary, during both sampling dates, *B. chinensis* nests with brood were present throughout an established community mulch pile with active workers foraging on the surface. We speculate that residents are loading mulch for landscapes, gardens, and other outdoor projects and are unknowingly moving this invasive ant to their homes, which raises concerns over increasing exposure and likelihood of stings and potential allergic reactions to this invasive pest. Additionally, residents are unintentionally adding to existing populations of *B. chinensis* or dispersing it into new habitats. These actions have the potential to form populations that will likely coalesce with its existing range (Liebhold and Tobin 2008). Additional work is needed to determine rate of success for establishment and spread after distribution and the likelihood of dispersing a queen. Our results demonstrate that community mulch piles may serve as a reservoir for Asian needle ant populations and a source for distribution of this detrimental invasive species. There are indications that *B. chinensis* can exhibit unicolonial nest structure, where nests are interconnected and form an expansive “super colony” (Warren et al. 2020, Biol. Invasions 22: 813–825), which could complicate any efforts for management and control. In an effort to reduce needle ant dispersal, we recommend placing mulch piles and other bulk organic materials (i.e., soils, compost, rocks, pine nuggets) on man-made surfaces (i.e., concrete or cement) to prevent soil contact and limit prolonged moisture, a preferred element of Asian needle ant habitat and other successful invasive ant species (Holway 1998, Oecologia 11: 206–12; Mackay and Mackay 2010, The systematics and biology of the New World ants of the genus *Pachycondyla* (Hymenoptera: Formicidae), Edwin Mellen Press, Lewiston, NY; Smith 1934). In addition, limiting the time that the mulch is present and encouraging rapid turnover is also likely to reduce accumulated moisture, decomposition of material, colonization by potential prey (Jordan and Jones 2006, Urban Ecosyst. 10: 87–95; Silverman et al. 2006, J. Econ. Entomol. 99: 1757–1760), and overall suitability for Asian needle ants.