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A new species of *Neostenoptera* Meunier (Diptera: Cecidomyiidae: Winnertziinae) from Hawai'i

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# A new species of *Neostenoptera* Meunier (Diptera: Cecidomyiidae: Winnertziinae) from Hawai'i

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**Abstract.** A new paedogenetic midge (Diptera: Cecidomyiidae: Winnertziinae: Heteropezini) from Oʻahu Island, Hawaiʻi, *Neostenoptera hawaiiensis* Plakidas, Nguyen, and Ferro, **new species**, is described and illustrated. A key to all species in the genus is provided. Specimens were emergent from deadwood gathered at Waimea Arboretum and Botanical Garden. *Neostenoptera appalachiensis* Plakidas and Ferro were collected from the same set of samples in Hawaiʻi, and additional specimens are reported from Georgia and South Carolina, three **new state records**. The discovery of two paedogenetic midges in Hawaiʻi poses a unique set of questions as to their possible mode of arrival on an island ecosystem. We briefly address the possibility that both species are simply "hitchhikers" that went undetected at ports of entry.

Key words. Heteropezini, paedogenetic midges, emergence chamber.

ZooBank registration. urn:lsid:zoobank.org:pub:73D4C822-5C81-4291-9019-2AC0BE96800A

#### Introduction

Following the discovery of the first paedogenetic midge from Denmark, *Miastor metraloas* Meinert, 1864 (Diptera: Cecidomyiidae: Winnertziinae: Heteropezini), there have been eight additional new genera and 18 new species (Gagné and Jaschhof 2021, p. 626), with the newest paedogenetic midges identified as *Neostenoptera appalachiensis* Plakidas and Ferro, 2016, and *Nikandria australis* Jaschhof and Jaschhof, 2017. These are exceptionally rare finds for collectors in that paedogenetic midges are all micro-dipterans, typically 1–2 millimeters long and translucent to light yellow-brown in color making them difficult to locate in samples and even more difficult to slide mount for microscopic study. Adults emerge once a year typically in late spring or early summer and are short lived. Collecting them can prove to be an arduous task.

Neostenoptera Meunier, 1902 were first reported from Hawai'i by Evenhuis et al. (2018) from three locations on O'ahu. Specimens were collected using a yellow sticky board trap and were not conspecific with Neostenoptera appalachiensis. Additional specimens of Neostenoptera were collected as part of the first C-MAIKI project (see below) and are described as a new species.

### Materials and Methods

The first C-MAIKI (Center for Microbiome Analysis through Island Knowledge and Investigation) project was formed by an interdisciplinary consortium of faculty at the University of Hawai'i at Mānoa to assess the diversity and function of microbes across an entire watershed on the island of Oahu, Hawai'i (Hynson et al. 2018). The portion of the project concerning this research is described below.

At each of six sites in Waimea Arboretum and Botanical Garden (N 21.63°, W -158.05°, Oʻahu Island, Honolulu Co., Hawaiʻi, USA) three samples of dead wood were collected and placed in emergence chambers (design based on Ferro and Carlton 2011). The wood was approximately 1–4 cm diameter and decay class two (based on Pyle and Brown 1999). Samples were collected during the spring (March – April) of 2018 (start dates differ by site due to accessibility issues) and sealed into emergence chambers. Specimens were collected approximately once a week for up to 12 weeks and retained for identification.

Specimens for study are deposited in the following collections:

BPBM Bernice P. Bishop Museum, Honolulu Hawai'i, USA

CUAC Clemson University Arthropod Collection, Clemson University, Clemson, South Carolina, USA

JPPC John Plakidas Personal Collection, Pittsburgh, Pennsylvania, USA

UHM University of Hawai'i at Mānoa, Honolulu Hawai'i, USA

**USNM** National Museum of Natural History, Washington, D.C., USA: holotype male and one male and one female paratype

In total, 45 specimens were examined, 40 males and five females. Twenty-six specimens (21 males and five females) were dehydrated in 91% isopropyl alcohol, cured in clove oil, and permanently slide mounted in Canada balsam. The remaining 19 specimens were retained in 91% isopropyl alcohol filled vials for future study. Adult terminologies are mostly from McAlpine et al. (1981) and Jaschhof and Jaschhof (2013). Adults were imaged using a Zeiss Axioskop compound microscope (100X total magnification) and Canon EOS 7D mark ii digital camera body. Images were stacked by hand in 10 um increments using fine focus and assembled with Helicon Focus.

# Systematic Entomology

Diptera: Cecidomyiidae Newman, 1834 Subfamily Winnertziinae Panelius, 1965 Tribe Heteropezini Shiner, 1868

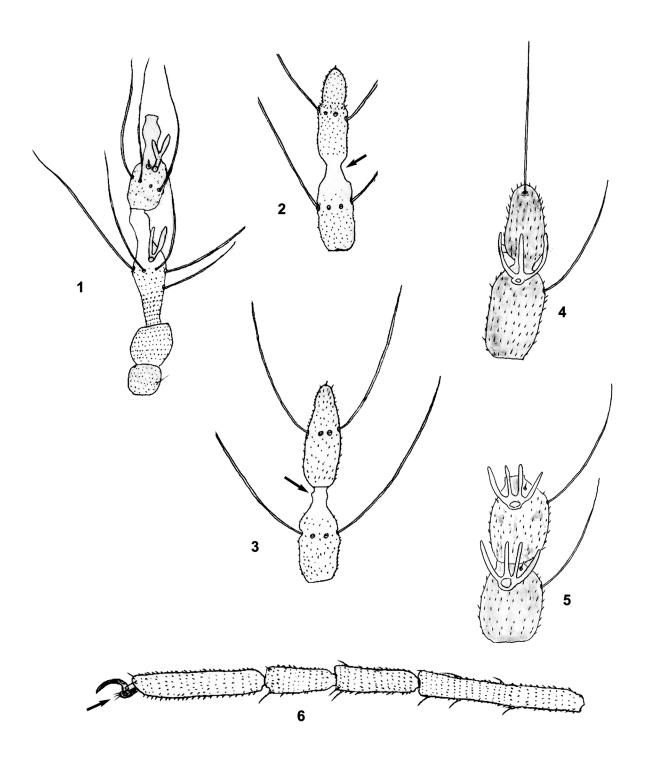
#### Neostenoptera Meunier, 1902

*Neostenoptera* Meunier 1902: 102. Type species *N. kiefferi* (type lost, known only from three illustrations and brief description) (Meunier 1901).

#### Neostenoptera hawaiiensis Plakidas, Nguyen, and Ferro, new species

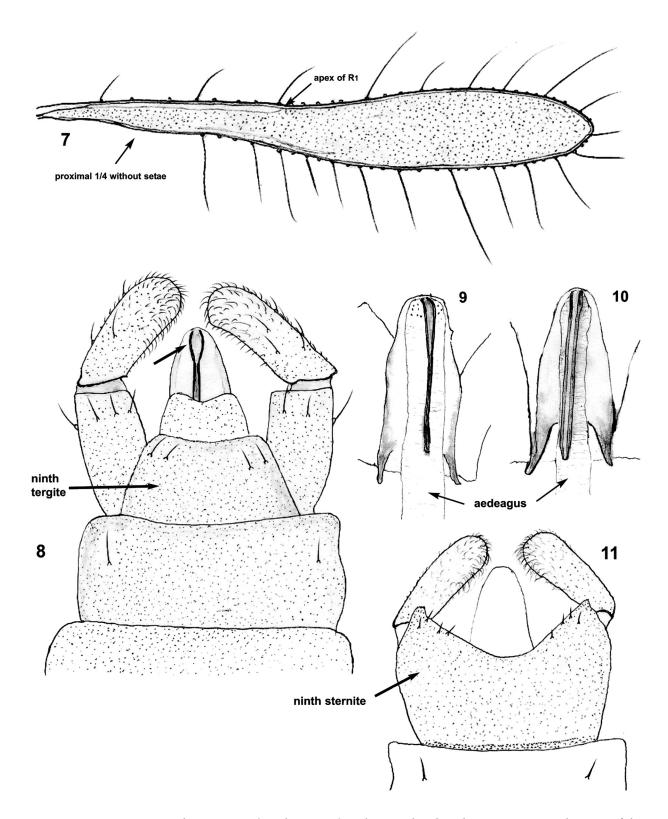
Figures 1-12, 16-17, Map 1

**Diagnosis.** Adult, body length (measured from head to end of genitalia) male 1.0-1.2 mm (n=10); female 1.4-1.6 mm (n=5). Color: pale translucent-yellow, the eyes are reddish-brown, with one smaller brown spot below each eye; wings teneral, with only veins R1 and CuA1 present and a fringe of long brown setae along the wing margin except for the proximal  $\frac{1}{4}$  along the wing base (Fig. 7). *Neostenoptera hawaiiensis* most closely resembles N. appalachiensis but can be distinguished in the following manner. Female antennae with nine flagellomeres, flagellomere 9 shorter and narrower than 8 and with one long dark brown seta distally (Fig. 4), a species defining trait. In contrast, the female flagellum of N. appalachiensis has ten flagellomeres, flagellomere 10 with a short coniform seta distally (Fig. 15). The male of N. hawaiiensis has the ejaculatory apodeme (Fig. 8–10) lightly sclerotized, as long as the aedeagus, with a forked apex, a species defining trait. The tegmen (Fig. 9–10) is lightly



**Figures 1–6.** *Neostenoptera hawaiiensis.* **1)** Male scape to flagellomere 2. **2–3)** Variation in male flagellomeres 10+11. **2)** Arrow points to the variation in the stem of flagellomere 10. **3)** Arrow points to the longer stem of flagellomere 10. **4)** Female flagellomeres 8+9. **5)** Female flagellomeres 3+4. **6)** Male, four-segmented tarsus, midleg, arrow to empodium.

 $\mathbf{4} \cdot \mathbf{June} \ 24,2022$  Plakidas et al.



**Figures** 7–11. *Neostenoptera hawaiiensis*. 7) Male wing. 8) Male genitalia, dorsal, arrow points to the apex of the forked ejaculatory apodeme. 9–10) Male genitalia, variation in shape of tegmen and apex of ejaculatory apodeme. 11) Male genitalia, detailing the shape of the ninth sternite, ventral.

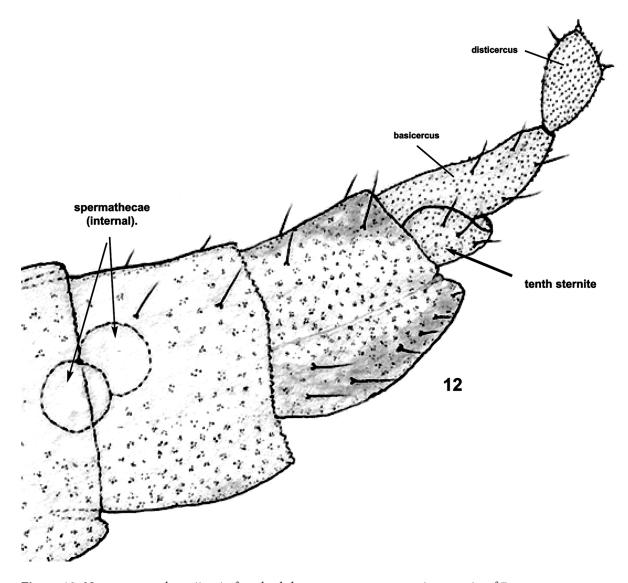
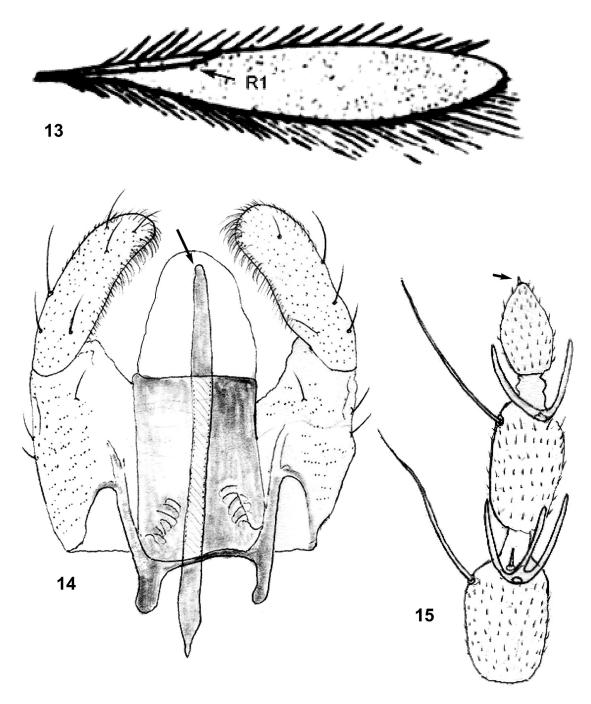


Figure 12. Neostenoptera hawaiiensis, female abdomen segments: posterior margin of 7 to cercus.

sclerotized laterally and extends to the height of the aedeagus. In contrast, the ejaculatory apodeme of *N. appala-chiensis* has a tubular apex and the tegmen does not reach the full height of the aedeagus (Fig. 14).

**Description.** Adult. Body length (measured from head to end of genitalia) male  $1.0-1.2 \,\mathrm{mm}$  (n=10), female  $1.4-1.6 \,\mathrm{mm}$  (n=5). Color, pale translucent-yellow. **Head.** Eyes situated laterally; eye bridge complete, without facets at vertex. Occiput with two setae. Palpus and labellum absent, frons bare. Antennal scape with one seta; pedicel with horizontal rows of microtrichia, no setae. **Female flagellum.** Flagellum with nine flagellomeres each with two digitate sensoria on flagellomeres 1-8 (Fig. 4-5), flagellomere 1 with two setae on the node, flagellomeres 2-8 with one or two setae on the node; flagellomere 9 shorter and narrower than 8 and with one long dark brown seta distally, a species defining trait. **Male flagellum.** Flagellum with eleven flagellomeres, flagellomeres 1 and 2 with single digitate sensoria, flagellomeres 3-11 lack sensoria (Fig. 1-3). **Thorax and chaetotaxy.** Pronotum with four setae, two dorsocentral setae and two dorsolateral setae, one of which is elongated. Lateral sclerites bare except for a fine covering of microtrichia; scutellum non-setose. Wing (Fig. 7): elongate, narrow, with a row of long light brown setae along the entire wing margin except on the proximal  $\frac{1}{4}$ ; membrane entirely microtrichose. Vein R1 closely joined to C, reaching the wing margin before midlength; vein R5 absent; vein CuA1 visible near wing base. Halters translucent yellow, with one seta near the base and one seta on the knob. Legs with fore femur inflated



**Figures 13–15.** *Neostenoptera* spp. **13)** *Neostenoptera kiefferi*, male wing (after Meunier 1901: Fig. 19). **14)** *Neostenoptera appalachiensis*, male genitalia, arrow to tubular apex of ejaculatory apodeme. **15)** *Neostenoptera appalachiensis*, female flagellomeres 8–10, arrow to coniform seta at apex of tenth flagellomere.

distally, mid and hind leg femora slender or only slightly inflated distally; tarsi four-segmented (Fig. 6); tarsal claws falcate, empodium shorter than claws.

Male abdomen and chaetotaxy. Tergites and sternites membranous; tergite 1 with two lateral setae; tergites 2–7 with two setae situated anteriorly and posteriorly, and two closely approximated trichoid sensillae; tergite 8 (Fig. 8) with two setae and two trichoid sensillae. Sternite 1 without setae; sternites 2–7 with four setae and two



**Figures 16–17.** *Neostenoptera hawaiiensis.* **16)** *Neostenoptera hawaiiensis*, habitus, male, paratype. **17)** *Neostenoptera hawaiiensis*, habitus, female, paratype.

closely approximated trichoid sensillae; sternite 8 with two setae and two trichoid sensillae. Sternite 9 extending to the height of the gonocoxites with a few setae along the posterior margin (Fig. 11). Tergite 9 (Fig. 8) with four setae at the posterior lateral margins, no trichoid sensillae; cercus bilobed, longer than tergite 9; hypoproct (sternite 10) not discernable. Gonocoxites fused along the anterior margin, covered with microtrichia and a few setae situated distally. Gonostyli about 2.75× longer than wide, covered with setulae, sparsely covered with setae,

**Table 1.** Paratype collection locality coordinates, emergence dates, sample codes, and institution in which they are deposited. All specimens have label data: USA: Hawai'i, Honolulu Co., Waimea Arboretum and Botanical Garden, C-MĀIKI Institute, dead wood emergence.

Coordinates	Emergence dates	Sample code	Paratypes
N 21.6336°, W -158.05209°	9–17 April 2018	C-1-1	4 M, alcohol, UHM
N 21.6336°, W –158.05209°	17-23 April 2018	B-1-1	1 M, alcohol, CUAC
N 21.63094°, W –158.04771°	17–23 April 2018	A-2-1	4 M, slide, BPBM; 1 F, 2 M, slide, CUAC; 1 M, slide, JPPC; 2 M, slide, UHM; 1 M, slide, USNM
N 21.63094°, W -158.04771°	23-30 April 2018	C-2-1	1 F, 1 M, alcohol, BPBM
N 21.6336°, W -158.05209°	30 April-5 May 2018	C-1-1	5 M, alcohol, CUAC
N 21.63094°, W –158.04771°	30 April–7 May 2018	C-2-1	2 M, slide, CUAC; 1 F, slide, JPPC; 1 F, 1 M, slide, UHM
N 21.63094°, W -158.04771°	7–14 May 2018	C-2-1	2 M, slide, BPBM; 1 M, slide, CUAC; 2 M, slide, JPPC; 2 M, slide, UHM; 1 F, slide, USNM
N 21.6336°, W -158.05209°	14-21 May 2018	A-1-1	1 F, slide, BPBM;
N 21.6336°, W -158.05209°	14-21 May 2018	C-1-1	2 F, alcohol, UHM
N 21.6203°, W -158.01466°	2-9 July 2018	A-4-1	2 M, alcohol, BPBM
N 21.6203°, W –158.01466°	16–30 July 2018	A-4-1	3 M, alcohol, BPBM
N 21.6203°, W -158.01466°	30 July-6 August 2018	A-4-1	1 M, slide, BPBM

the inner margin with longer, stouter hair-like setae forming a light comb of hairs. Ejaculatory apodeme (Fig. 8–10) lightly sclerotized, as long as aedeagus, with forked apex, a species defining trait. Tegmen (Fig. 9–10) lightly sclerotized laterally extending to the height of the aedeagus.

Female abdomen and chaetotaxy. Tergites and sternites 1–7 similar to male. Tergites 8 and 9 with two anterior and two posterior setae; tergite 10 imperceptible; sternite 8 without setae; sternite 9 bilobed each lobe with seven setae; sternite 10 (hypoproct) bilobed with three setae per side. Cercus two-segmented (Fig. 12), disticercus shorter than basicercus, with horizontal rows of microtrichia and two short dorsal setae located at the distal corners. Two circular dark brown sclerotized spermathecae at level of tergites 7 and 8.

Larva and pupa. Unknown.

**Type material.** Holotype male, labeled: USA: Hawaii, Honolulu Co., GPS 21.63094 -158.04771; Waimea Arboretum & BG. 17-23 April 2018; C-MĀIKI Institute, dead wood emergence, A-2-1. Deposited in USNM.

**Designated paratypes** (Table 1). All specimens have label data: USA: Hawaiʻi, Honolulu Co., Waimea Arboretum and Botanical Garden, C-MĀIKI Institute, dead wood emergence. Specific emergence dates and sample code numbers are associated with appropriate specimens.

Species concept. The concept of "species" is a human-induced attempt to generalize and simplify a complex and cluttered natural world. J. B. S. Haldane (1956) put it more eloquently: "The concept of a species is a concession to our linguistic habits and neurological mechanisms". Our concept of species is also a concession to the shortness of a human life in relation to the length of evolutionary time. As such, a "unified" or "universal" species concept may not exist and it's reasonable to expect future workers will bring about new species concepts or refine old ones. The validity of the species described herein will be assessed and reassessed by future workers whether we explicitly state which species concept is being invoked. Despite these drawbacks there may be some utility in stating outright that the Morphological Species Concept (sensu Mayden 1997) was used when defining this species.

**Etymology.** The specific name, *hawaiiensis*, is Latin meaning "from Hawai'i", in reference to the collection site in Waimea Arboretum, Honolulu Co., Hawai'i.

**Bionomics.** Specimens were collected using an emergence chamber from dead wood 1–4 cm in diameter. The dead wood samples were sequestered in emergence chambers during March when presumably only larvae were present. The peak of adult emergence was during April and May, with a few more specimens emerging into August. Specimens of *Neostenoptera* sp. reported by Evenhuis et al. (2018) were collected April, May, August, and September from yellow sticky boards, suggesting that they were also present as adults throughout the summer and flew.

#### Key to the species of Neostenoptera Meunier

In the following key morphological traits, which readily separate both males and females of *N. appalachiensis* and *N. hawaiiensis*, are presented. These two species are easily separated by comparing male genitalia or number of flagellomeres in the females.

1.	Wing uniformly elliptical (Fig. 13); vein R1 prominent and distinctly separate from costa
_	Wing not uniformly elliptical (Fig. 7); vein R1 close to the costa
2.	Vein R5 visible near midlength; scutellum with two setae
_	Vein R5 clearly absent; scutellum without setae
3.	Male genitalia with apex of ejaculatory apodeme forked (Fig. 8); female with nine flagellomeres, the ninth flagellomere with a long, dark brown apical seta (Fig. 4)
_	Male genitalia with apex of ejaculatory apodeme tubular (Fig. 14); female with ten flagellomeres, the
	tenth flagellomere with a short coniform seta (Fig. 15)

#### Neostenoptera appalachiensis Plakidas and Ferro, 2016

Neostenoptera appalachiensis has been sighted or collected three times since it was described (Plakidas and Ferro 2016) (Maps 1–2) each resulting in a new state record. Two photos were placed on bugguide.net by Jonathan Burishkin (bugguide.net/node/view/1271455; bugguide.net/node/view/1271456) of specimens that "came to moth lights at night" in GEORGIA, **new state record**. The photos were taken during 6 August 2016, at USA: Georgia: Bartow Co.: Cartersville (approximately N 34.16°, W –84.79°). The flies are clearly on the light sheet well above the ground and appeared to have flown there.

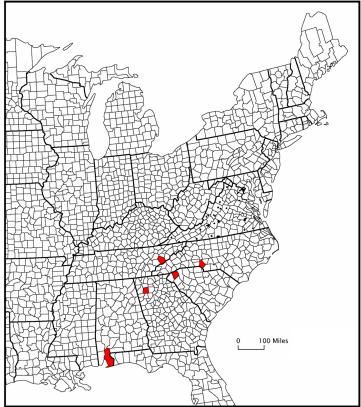
Curt Harden collected *N. appalachiensis* emergent from sweetgum limbs collected from his yard in SOUTH CAROLINA, **new state record**, (USA: South Carolina, Pickens Co., Central, 115 Cedar Creek Circle, N 34.7146°, W –82.7804°). A large sweetgum limb fell from a tree during fall 2020, pieces were placed in an emergence chamber early 2021 and a sample taken 14 May 2021 contained specimens—"there were tons (top of the liquid was thick with them)" (pers. com.)—specimens are deposited in CUAC.

Astonishingly, five female specimens of *N. appalachiensis* were collected in HAWAI'I, **new state record**, from samples containing *Neostenoptera hawaiiensis*. All specimens were from material collected at the same locality but emerged over multiple sampling dates: USA: Hawaii: Honolulu Co.: Waimea Arboretum and Botanical Garden: N 21.6203°, W –158.01466° C-MĀIKI Institute, dead wood emergence, A-4-1: 1 F, 29 May–4 June, slide, USNM; 2 F, 12–18 June, slides, CUAC, BPBM; 1 F, 18–25 June, slide, JPPC; 1 F, 25 June–2 July, alcohol, UHM.

*Neostenoptera appalachiensis* is now known from the following states: Alabama, Georgia, Hawaiʻi, North Carolina, South Carolina, and Tennessee.

**Discussion.** While not impossible, it seems improbable that any *Neostenoptera* are native to Hawaii. For example, if adults were blown from the nearest mainland, California—approximately 3850 km (2390 miles) away, at an average speed of 50 kph (31 mph) they would be airborne for 77 hours and would probably freeze, starve to death, or desiccate in that time. Similarly, if carried on a log or structure floating from any mainland, eggs, larvae, and/ or adults, would have to keep from overheating, drying out, being blown to sea, or poisoned with saltwater for at least several weeks. Given these circumstances it would be unlikely that *Neostenoptera* could arrive at Hawai'i other than by human aid.





Maps 1–2. Neostenoptera collection localities. 1) Neostenoptera spp. collection localities on Oʻahu Island, Hawaiʻi. Neostenoptera hawaiiensis site 1 (sample codes A-1-1, B-1-1, C-1-1), site 2 (sample codes A-2-1, C-2-1), site 3 (sample codes A-4-1). Neostenoptera appalachiensis site 3 (sample code A-4-1). Neostenoptera sp. (Evenhuis et al. 2018) sites 4, 5, and 6. 2) Neostenoptera appalachiensis collection localities (by county) in the continental United States.

Neostenoptera hawaiiensis and N. appalachiensis were both collected within the Waimea Arboretum and Botanical Garden. Sites where woody materials were collected were either within the Botanical Garden itself, or the forest of predominately introduced plants beyond the garden, deeper into the valley. The garden has sections that focus on plants native to Hawai'i, Polynesia, and other island ecosystems, but it also contains many plants from Central America, South America, and Africa.

While the larvae of *Neostenoptera* are unknown, the larvae of three eastern North American paedogenetic species are known to live and reproduce in decaying substrates and mushrooms (Plakidas 2018; Yukawa 2021), and it's likely *Neostenoptera* does the same. Both *Neostenoptera* species, like many other non-native species, probably "hitch-hiked" a ride aboard exotic plants introduced to the island and are unintentional introduced species to Hawai'i from other parts of the world. Micro-dipterans can easily go undetected in plant shipments since their minute larvae are embedded in decaying plant tissue. Neither species have been found to be economically harmful since their larvae are strictly fungivorous.

The above speculation calls into question the ultimate origin of *N. appalachiensis* which itself may have been introduced to Eastern North America, possibly from Central America, South America, or Africa. Recent collections of *Neostenoptera* have come from citizen science, monitoring, and by-catch (residues) from collections made using emergence chambers. As these techniques continue to be used, more observations and specimens should become available and the biogeographic mysteries presented by the genus may be solved.

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