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Two sympatric species of *Antrodiaetus* from southwestern North Carolina (Araneae, Mygalomorphae, Antrodiaetidae)

BRENT E. HENDRIXSON & JASON E. BOND

East Carolina University, Department of Biology, Howell Science Complex N211, Greenville, NC 27858 USA; beh0701@mail.ecu.edu, bondja@mail.ecu.edu

Abstract

Two sympatric species of *Antrodiaetus* (Araneae, Mygalomorphae, Antrodiaetidae) are recorded from southwestern North Carolina: *A. unicolor* (Hentz 1841) and *A. microunicolor* new species. A neotype for *A. unicolor* is designated from DeSoto State Park in Alabama and a description is provided. A new species of *Antrodiaetus* is described from the Coweeta Long Term Ecological Research station in southwestern North Carolina. This new species is sympatric (putatively syntopic) with the closely related *A. unicolor* and can be differentiated from that species on the basis of size, setal characters, coloration, selected morphometric ratios, and non-overlapping breeding seasons. A brief account on the natural history for both species at Coweeta is presented.

Key words: New species, spider taxonomy, temporal isolation, neotype designation

Introduction

The mygalomorph spider genus *Antrodiaetus* Ausserer 1871 currently includes 13 species throughout the United States, western Canada, and Japan. These spiders (Fig. 1) build cryptic, silk-lined subterranean burrows that are concealed by a collapsible turret or collar during daylight hours (Figs. 12–14). After dusk, spiders prop open the collar and wait at the burrow entrance to seize passing insects and other prey items. The taxonomy of this group has not received formal attention since Coyle (1971), although the most recent treatment of the genus by Miller and Coyle (1996) proposed a species-level phylogeny for *Antrodiaetus* and *Atypoides* Pickard-Cambridge 1883. The highest nominal species-level diversity of this genus is in the Pacific Northwest and surrounding areas where at least six species are known (Coyle 1971). In addition, two species are recognized from Japan, another confined to the “mountain islands” of the American Southwest, and the remaining four from the eastern and Midwestern deciduous forests of the United States. Of the latter

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species, *A. unicolor* (Hentz 1841) is the most widely distributed and shows a great deal of morphological variation across its distribution. This species has essentially been used as a "catch-all" name for all *Antrodiaetus* collected south of northern Virginia in the central and southern Appalachian Mountains.

During April 2003, we had the opportunity to study the spider collections at the North Carolina Museum of Natural Sciences and came across several series of putative *Antrodiaetus unicolor* males from the Coweeta LTER site in the southwestern mountains of North Carolina (Fig. 2). These spiders showed remarkable differences in size. These spiders showed remarkable differences in size, and this variation appeared to be correlated with the time of year in which they were collected (i.e., large males were collected earlier than small males, dates were non-overlapping). This spurred an interest to conduct additional fieldwork at Coweeta. In particular, we sought to determine if this was an anomalous observation, whether females showed comparable degrees of morphological and behavioral differentiation, whether habitat segregation could be detected, and ultimately, whether the two "forms" represented different species. Based on the available morphological and behavioral evidence, we propose that the smaller spiders represent a new species and describe it below. We also designate and describe a neotype for *Antrodiaetus unicolor* to fix a type locality for that species.

Methods

Fieldwork.—Forty pitfall traps were installed along Watersheds 2 and 14 at the Coweeta LTER site to collect adult males (additional collecting was carried out at Watersheds 18 and 36). These traps consisted of a 20-ounce plastic cup inserted into a 32-ounce plastic cup and were covered by a 10 X 10 cm piece of roofing tile to reduce flooding. A 50:50 mixture of propylene glycol and 100% ethanol was used as a preservative (cup filled about halfway). The traps were placed in areas with relatively high densities of burrows, near the base of overhanging roots, or random points along ravine slopes. Trap maintenance and inspection was performed at least every other weekend from 13 September–15 November 2003. Specimens collected in the traps were immediately placed into 100% ethanol and transferred to 80% ethanol back at the lab. Adult females and immature specimens were collected directly from their burrows using two techniques. During daylight hours, closed burrows were located by careful inspection of the substrate and excavated. At night, open burrows were located with a headlamp. Spiders were often found at their burrow entrance and a hand trowel was quickly inserted into the substrate just behind the spider to intersect the burrow. All freshly collected material was preserved in 80% ethanol and immediately placed into a freezer at –20°C.

Laboratory methods.—Landmarks for mensuration and terminology essentially follow that of Coyle (1971), except that the maximum diameter of the male pedipalpal tibia (PTT in Coyle 1971) is herein referred to as pedipalpal tibia depth. Abbreviations for measurements and meristic counts are as follows: ALD = anterior lateral eye diameter; ALS = distance between anterior lateral eyes; AMD = anterior median eye "pupil" diameter (Coyle 1971: 326, fig. 73); AMS = distance between "pupils" of anterior median eyes; CL = length of dorsal shield of prosoma; CT = number of cheliceral macroteeth; IFeL = length of femur I; IMeL = length of metatarsus I; ITaL = length of tarsus I; ITiL = length of tibia I; IVFeL = length of femur IV; IVMeL = length of metatarsus IV; IVTaL = length of tarsus IV; IVTiL = length of tibia IV; OQW = ocular quadrangle width; PFeL = pedipalpal femur length; PTiL = pedipalpal tibia length; PTiD = pedipalpal tibia depth; SL = sternum length; SW = sternum width.

Specimen repositories and other abbreviations are as follows: American Museum of Natural History, New York (AMNH); East Carolina University, Greenville, North Carolina (specimens will be deposited in USNM; ECU-USNM); North Carolina Museum of Natural Sciences, Raleigh (NCSM); United States National Museum (Smithsonian Institution), Washington, District of Columbia (USNM). All ECU specimens are accompanied by a "MY" (i.e., mygalomorph) database number. Other abbreviations in the text include: cm = centimeter; ICS = inner conductor sclerite of the male pedipalp bulb; l = left side; LTER = Long Term Ecological Research; mm = millimeter; n = number of specimens; OCS = outer conductor sclerite of the male pedipalp bulb; r = right side.

Morphometric statistics were calculated on the basis of ten adult males for both species at the Coweeta LTER site, five adult females of both species from Coweeta, and four adult males of *Antrodiaetus unicolor* from northeastern Alabama (i.e., new type locality). All measurements, reported in millimeters, were performed on a Leica MZ12.5 stereomicroscope equipped with a calibrated ocular micrometer scale; measurements are accurate ± 0.02–0.1 mm depending on the magnification used.

Female spermathecae were dissected from the ventral opisthosomal wall, optically cleared overnight in approximately 20 µl of clove oil, and immediately observed in ethanol. The illustration was made in Adobe Illustrator vector graphics software (ver. 10, Adobe Systems Inc., San Jose, CA) by tracing a digital photograph taken through a stereomicroscope.

Digital images were taken with a MicrOptics Inc., Digital Imaging System. The map coverages (i.e., Coweeta basin, watershed boundaries, streams) of the Coweeta LTER site were obtained from their webpage via the University of Georgia (see Coweeta LTER 2003 in references) and imported/edited in ArcView GIS software (ESRI 2002). Latitude and longitude data are reported in decimal degrees (DD.DD°).

Taxonomy

Family Antrodiaetidae Gertsch 1940

Brachybothriinae Simon 1892: 193. Type genus, *Brachybothrium* Simon (=*Antrodiaetus* Ausserer).

Brachybothriidae Simon: Pocock 1903: 346.
Acattymidae Kishida 1930: 34. Type genus, *Acattyma* L. Koch (=*Antrodiaetus* Ausserer).
Antrodiaetinae Gertsch 1940: 236. Type genus, *Antrodiaetus* Ausserer.
Antrodiaetidae Gertsch: Coyle 1971: 330–331; Raven 1985: 124; Eskov and Zonshtein 1990: 333.

Genus *Antrodiaetus* Ausserer 1871

Antrodiaetus Ausserer 1871: 136; Coyle, 1971: 331–332. Type species by monotypy, *Mygale unicolor* Hentz.
Acattyma L. Koch 1878: 760 (synonymized by Yaginuma 1960). Type species by monotypy, *Acattyma roretzi* L. Koch.
Brachybothrium Simon 1884: 314 (synonymized with *Antrodiaetus* by Kishida 1930). Type species by subsequent designation (Simon 1892), *Brachybothrium pacificum* Simon.
Nidivalvata Atkinson 1886: 129 (synonymized with *Brachybothrium* by Simon 1890). Type species by subsequent designation (Coyle 1971), *Nidivalvata marxii* Atkinson.
Anthrodiaetus Ausserer: Simon 1890: 312 (unjustified emendation).

Antrodiaetus unicolor (Hentz 1841)

(Figures 1, 3–5, 9–10; Tables 1–2)

Mygale unicolor Hentz 1841: 42; Hentz 1842: 57, pl. 7, fig. 5.
Mygale gracilis Hentz 1841: 42 (synonymized by Coyle 1971: 335); Hentz 1842: 56, pl. 7, fig. 4.
Antrodiaetus unicolor: Ausserer 1871: 136; Roewer 1942: 189; Bonnet 1955: 335; Coyle 1971: 335–344, figs. 113, 120, 130, 138, 145–146, 158, 173–174, 188–194, 234–240, 270–279, 313, 315, 318; Gertsch & Platnick 1979: 4, figs. 5–6; Eskov & Zonstein 1990: 354: figs. 28–29.
Closterochilus gracilis: Ausserer 1871: 142; Roewer 1942: 190.
Eurypelma bicolor: Marx 1883: 24 (incorrect subsequent spelling).
Eurypelma gracilis: Marx 1883: 24.
Brachybothrium accentuatum Simon 1884: 315 (synonymized by Coyle 1971); Roewer 1942: 189; Bonnet 1955: 906.
Nidivalvata marxii Atkinson 1886: 110–111, 113, 116, 130–131, pl. 5, figs. 8–10, 13, 17–18, 23 (synonymized by Coyle 1971).
Nidivalvata angustata Atkinson 1886: 130, 113, 117 (synonymized by Coyle 1971).
Brachybothrium marxi: Simon 1890: 310; Roewer 1942: 190; Bonnet 1955: 906.
Brachybothrium angustatum: Simon 1890: 310; Petrunkevitch 1911: 52; Roewer 1942: 190; Bonnet 1955: 906.
Anthrodiaetus unicolor: Simon 1890: 312.
Brachybothrium unicolor: Comstock 1912: 249.
Brachybothrium pacificum: Barrows 1918: 298 (misidentification); Barrows 1925: 493, pl. 37, figs. 17–22 (misidentification).
Missulena gracilis: Petrunkevitch 1939: 213; Bonnet 1957: 2939.
Antrodiaetus bicolor: Vogel 1962: 246.

Type data.—United States: *Alabama:* Dekalb County: DeSoto State Park near Fort Payne (34.50°N, 85.62°W), July–August 1937 (collector unknown, probably W.B. Jones), male neotype (herein designated) (AMNH).

FIGURE 1. Live habitus of adult female *Antrodiaetus* from Coweeta. A, *A. unicolor* (MY 2300); B, *A. microunicolor* new species (MY 2422). Note the size difference. Scale bar = 5 mm.

Hentz (1841) described *Mygale unicolor* on the basis of a female specimen from Alabama. In the same paper, he described a male, also from Alabama, which he named *M. gracilis*. Unfortunately, both specimens have been destroyed and the exact locality from which these spiders were collected in Alabama is unknown. Coyle (1971) decided to synonymize the latter species under *Antrodiaetus unicolor*, but did not designate a neotype for the species. It is necessary to designate a neotype for *A. unicolor* at this time to establish its identity and to set a type locality for the species. A significantly larger scale study on this species is underway and preliminary data suggests that *A. unicolor* actually represents a complex of “cryptic species”; therefore, a fixed locality is necessary for the anticipated nomenclatural matters ahead.

We selected an adult male from DeSoto State Park in northeastern Alabama as the neotype for the following reasons: (1) male mygalomorph spiders tend to have more meaningful and useful diagnostic morphological characters; (2) to maintain the type locality of *A. unicolor* in Alabama; (3) most populations studied in Alabama appear relatively homogeneous and likely belong to the same “morphological” species (Coyle 1971; Hendrixson pers. obs.); (4) the population is sufficiently large (Coyle 1971; Hendrixson pers. obs.); and (5) this population of *A. unicolor* is likely to persist because its habitat is protected within state park boundaries.

Other specimens examined.—United States: *Alabama:* Dekalb County: DeSoto State Park near Fort Payne (34.50°N, 85.62°W): July–August 1937 (W.B. Jones, 1 male (AMNH); ditto, October 1937 (collector unknown), 1 male (AMNH); ditto, December 1937 (W.B. Jones), 1 male (AMNH); *North Carolina:* Macon County: Coweeta Hydrologic Station (LTER): Coweeta Watershed unknown: 27 September–13 October 1978, in pitfall traps (L. Reynolds), 4 males (NCSM). Coweeta Watershed 2 (35.07°N, 83.44°W): 13 October 1978, in pitfall traps (L. Reynolds), 12 males (NCSM); ditto, 27 September–11 October 2003, in pitfall traps (B.E. Hendrixson & C.J. Dreiling), 2 males (ECU-USNM, MY 2390, 2391). Coweeta Watershed 7 (35.06°N, 83.44°W): 13 October 1978, in pitfall traps (L. Reynolds), 1 male (NCSM). Coweeta Watershed 14 (35.05°N, 83.43°W): 13 September 2003 (B.E. Hendrixson, R.E. Chester, J.L. Roberts & C.L. Spruill), 4 females (ECU-USNM, MY 2300–2303); ditto, 13–27 September 2003, in pitfall traps (B.E. Hendrixson), 3 males (ECU-USNM, MY 2314–2316); ditto, 27 September 2003 (B.E. Hendrixson), 1 male, 1 female (ECU-USNM, MY 2317, 2323).

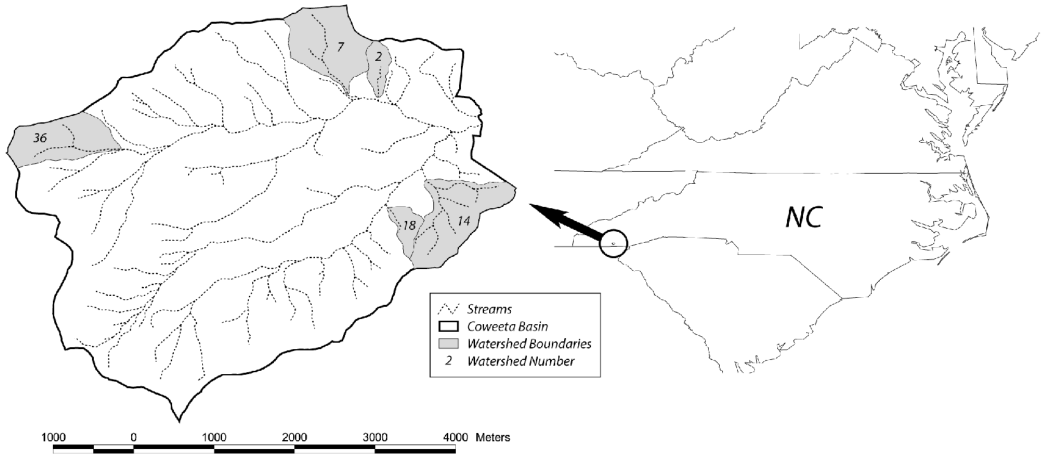
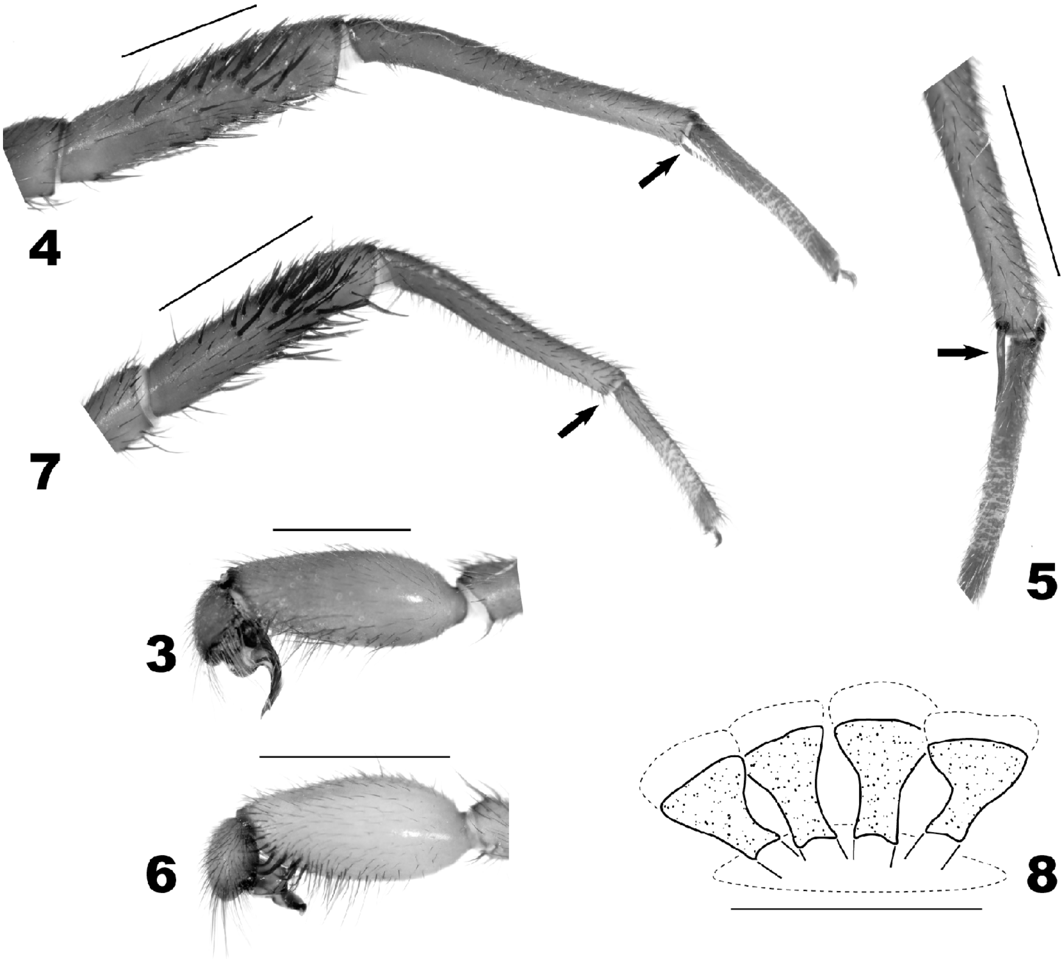


FIGURE 2. Map of the Coweeta LTER site, showing its location in southwestern North Carolina (NC) and the specific watersheds from which specimens in this study were collected.



FIGURES 3–8. Neotype male of *Antrodiaetus unicolor* from Alabama (3–5), holotype male of *A. microunicolor* new species (6, 7), and female (MY 2402) of *A. microunicolor* new species (8): 3, right pedipalp tibia, cymbium and bulb, prolateral aspect; 4, tibia, metatarsus and tarsus I showing mating clasper, prolateral aspect (arrow indicating presence of macroseta A on the ventral aspect of metatarsus); 5, closer view of the ventral aspect of metatarsus I (arrow indicating presence of macroseta A); 6, right pedipalp tibia, cymbium and bulb, prolateral aspect (note relative robustness of tibia); 7, tibia, metatarsus and tarsus I showing mating clasper, prolateral aspect (arrow indicating absence of macrosetae on the ventral aspect of metatarsus); 8, spermathecae (solid lines indicate heavily sclerotized area; dotted lines indicate areas with little sclerotization). Note: apparent differences in pedipalp bulb morphology between the two species are photographic artifacts. To prevent damage to the specimens, the bulbs were not twisted to obtain the same view. No significant differences were observed between the two species. Scale bars for appendages = 2 mm; for spermathecae = 0.5 mm.

Diagnosis.—Of the three species of *Antrodiaetus* currently recognized from the eastern United States (i.e., *A. unicolor*, *A. robustus*, *A. microunicolor* new species), *A. unicolor* can be recognized by the following combination of characters: presence of macroseta A on male metatarsus I (rarely absent, or rarely with macroseta B); at least one-fifth of macrosetae on male prolateral tibia I ensiform; presence of thickened convergent medial setae just posterior to the pedicel on the opisthosoma (on immature and female specimens). For comparisons to *A. microunicolor* new species, please refer to the diagnosis of that species found below.

Description.—*Neotype male: Coloration* (in alcohol)*:* Specimen has been preserved for over 65 years and is more or less dark reddish-brown throughout; this coloration probably is not indicative of the original spider. *Prosoma:* Head region slightly elevated from thoracic region. Setae moderately dense along peripheral edges of dorsal shield of prosoma; setae sparsely distributed on dorsal surface of dorsal shield of prosoma posterior to fovea. Sternum and labium moderately to densely setose. *Opisthosoma:* Three heavily sclerotized, completely continuous tergites on dorsal surface; posterior patch smaller than others but mostly indistinct from second. Entire opisthosomal surface densely covered with setae, interspersed with some slightly more elongated and thickened setae posteriorly; tergites accompanied by a few thickened setae. Ventral surface of opisthosoma with 30 epiandrous gland spigots located just anterior to genital opening. *Chelicerae:* Anterior dorsal prominence weak. Upper ectal (retrolateral) surface devoid of setae. *Pedipalps* (Fig. 3)*:* Tibia moderately slender (PTiL/PTiD = 2.44). ICS tip below level of OCS; ICS tip well-sclerotized, tapered to a narrow point; OCS tip well-sclerotized, blunt, weakly serrated. *Leg I:* Mating clasper (located on prolateral surface of tibia) consisting of 13 ensiform, 2 attenuate macrosetae, centered at approximately 2/3 distance from proximal to distal end of tibia (Fig. 4). Prolateral, ventral, distal aspect of tibia with a macroseta. Retrolateral, ventral aspect of tibia with 7 ensiform macrosetae; distal-most macroseta of group positioned at approximately 4/5 of distance from the proximal to distal end of tibia. Macroseta A (Coyle 1971, fig. 70) present on ventral aspect of metatarsus (Figs. 4–5); a moderately thickened seta is located at position B, but is not considered a macroseta. Metatarsus moderately sinuous in ventral view. *Measurements* (mm)*:* CL = 6.13; SL = 3.35; SW = 2.90; CT (l/r) = 10/9; PFeL = 3.65; PTiL = 3.30; PTiD = 1.35; IFeL = 5.85; ITiL = 4.05; IMeL = 5.00; ITaL = 2.85; ALD = 0.40; AMD = 0.12; ALS = 0.42; AMS = 0.20; OQW = 1.22.

Variation.—*Males from DeSoto:* Three additional adult males of *Antrodiaetus unicolor* from the new type locality at DeSoto State Park were examined during the course of this study. They do not differ significantly from the neotype in any important characters. The number of macrosetae making up the mating clasper is variable. One specimen also possesses macroseta B on the ventral surface of metatarsus I. A summary of measurements can be found in Table 1.

Specimens from Coweeta: A total of 23 adult males and five adult females were studied from Coweeta. Males compare favorably to those at DeSoto State Park. Most of the males possessed macroseta A on the ventral surface of metatarsus I; four males also had macroseta B; and one male also had macrosetae B and F. Two males collected during the 13–27 September 2003 pitfall trap series (MY 2314, 2316) were divergent from the others by the following characters: (1) the absence of macrosetae on the ventral surface of metatarsus I; (2) darker coloration; (3) legs slightly more setose; (4) absence of a macroseta on the prolateral, ventral, distal aspect of tibia I (this macroseta is absent in a few other males as well); and (5) mating clasper macrosetae centered at approximately 1/2 the distance from the proximal to distal end of tibia I. These two specimens are herein referred to as *Antrodiaetus unicolor* because of their large size (CL 7.30 and 6.30 mm, respectively) and breeding season, but they will be studied in greater detail at a future date. The females do not differ from one another in a meaningful way. A summary of measurements can be found in Tables 1 and 2.

TABLE 1. Selected measurements (in mm) for adult male *Antrodiaetus*. The first row for each species shows the range of measurements observed; the second row indicates the mean and standard deviation in the measurements for the given sample size. AL = males of *A. unicolor* from DeSoto State Park in northeastern Alabama; COW = males of *A. unicolor* from the Coweeta LTER site in southwestern North Carolina.

Species	CL	IFeL	ITiL	IMeL	ITaL	PFeL	PTiL	PTiD
A. unicolor (AL)	5.44–6.38	5.15–5.85	3.70–4.05	4.50–5.00	2.50–2.85	3.30–3.65	2.95–3.25	1.15–1.35
n = 4	6.05±0.42	5.68±0.35	3.94±0.16	4.78±0.21	2.69±0.14	3.55±0.17	3.18±0.16	1.29±0.09
A. unicolor (COW)	5.63–7.06	5.05–6.20	3.60–4.35	4.30–5.25	2.35–2.75	3.30–3.90	3.05–3.60	1.35–1.50
n = 10	6.47±0.42	5.81±0.35	4.07±0.25	4.80±0.27	2.57±0.13	3.71±0.19	3.35±0.15	1.43±0.06
A. microunicolor	3.75–4.50	3.35–4.10	2.40–2.85	2.75–3.20	1.60–1.85	2.25–2.70	2.05–2.35	0.95–1.10
new species n = 10	4.15±0.29	3.72±0.25	2.64±0.14	3.02±0.16	1.74±0.09	2.47±0.15	2.23±0.11	1.05±0.06

TABLE 2. Selected measurements (in mm) for adult female *Antrodiaetus*. The first row for each species shows the range of measurements observed; the second row indicates the mean and standard deviation in the measurements for the given sample size. COW = females of *A. unicolor* from the Coweeta LTER site in southwestern North Carolina.

Species	CL	SL	SW	IFeL	ITiL	IMeL	ITaL	IVFeL	IVTiL	IVMeL	IVTaL
A. unicolor (COW)	7.10–8.20	3.70–4.40	3.20–3.80	5.05–5.85	3.00–3.60	3.05–3.60	1.60–1.90	4.95–5.65	3.00–3.40	4.20–4.95	1.70–1.95
n = 5	7.80±0.48	4.10±0.29	3.52±0.29	5.47±0.33	3.35±0.25	3.36±0.23	1.78±0.13	5.30±0.33	3.20±0.19	4.66±0.34	1.83±0.10
A. microunicolor new species	5.25–6.88	2.88–3.63	2.44–3.13	3.70–4.65	2.30–2.80	2.10–2.55	1.25–1.50	3.50–4.50	2.05–2.45	2.85–3.60	1.30–1.50
n = 5	5.82±0.69	3.14±0.34	2.66±0.29	4.04±0.38	2.50±0.20	2.27±0.21	1.34±0.11	3.85±0.41	2.19±0.17	3.12±0.30	1.37±0.08

Distribution.—Widespread throughout the eastern United States, particularly in the southern and central Appalachian Mountains. Please refer to Map 1 in Coyle (1971) for a more thorough picture of the known distribution of this species.

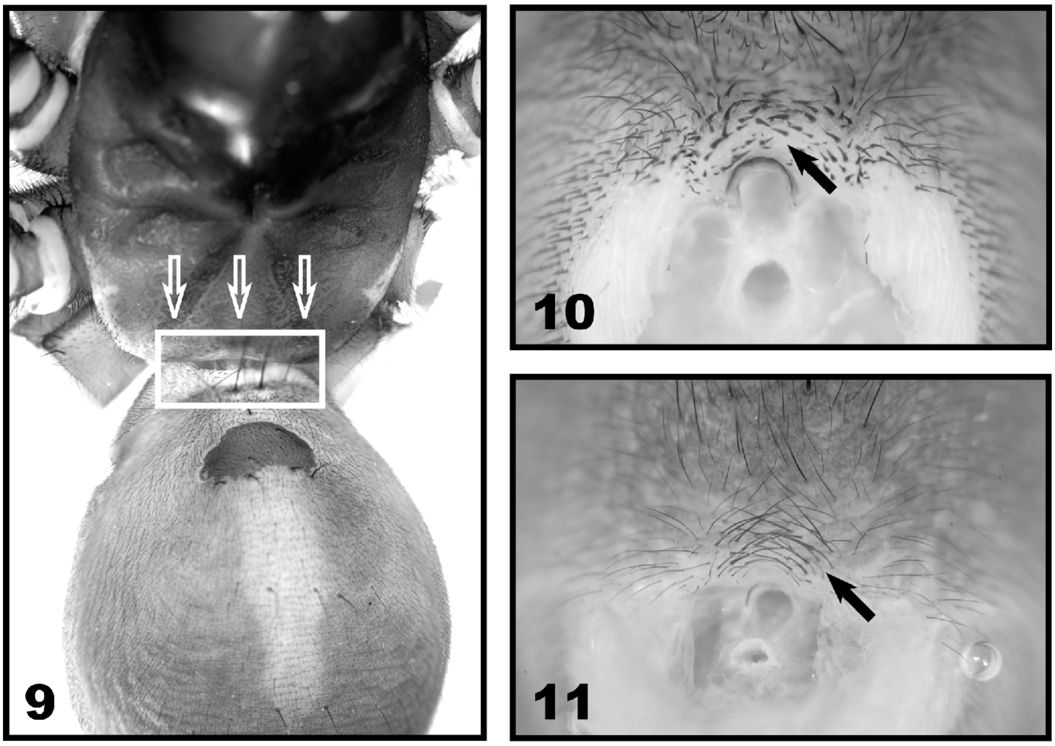
Antrodiaetus microunicolor **new species**

(Figures 1, 6–8, 11–14; Tables 1–2)

Type data.— United States: *North Carolina:* Macon County: Coweeta Hydrologic Station (LTER) Watershed 2 (35.07°N, 83.44°W), 24 November 1978 (L. Reynolds), male holotype (UNSM); ditto, 19 December 1978, in pitfall traps (L. Reynolds), 8 paratype males (NCSM); ditto, 25 October 2003 (B.E. Hendrixson), 2 paratype females (ECU-USNM, MY 2401, 2402); ditto, 8 November 2003 (B.E. Hendrixson), 1 paratype male, 1 paratype female (ECU-USNM, MY 2425, 2422). Coweeta Watershed 7 (35.06°N, 83.44°W): 19 December 1978, in pitfall traps (L. Reynolds), 2 paratype males (NCSM). Coweeta Watershed 14 (35.05°N, 83.43°W): 25 October–8 November 2003, in pitfall traps (B.E. Hendrixson), 2 paratype males (ECU-USNM, MY 2420, 2421). Coweeta Watershed 18 (35.05°N, 83.44°W): 15 November 2003 (B.E. Hendrixson, P.E. Marek & D.A. Beamer), 1 paratype female (ECU-USNM, MY 2448). Coweeta Watershed 36 (35.06°N, 83.47°W): 15 November 2003 (B.E. Hendrixson, P.E. Marek & D.A. Beamer), 1 paratype female (ECU-USNM, MY 2441).

Diagnosis.—The new species appears to be most closely allied to *Antrodiaetus unicolor.* Males of the new species are readily identified by the following characters (Coweeta *A. unicolor* characters in parentheses): exceptionally small size: CL < 4.50 mm (CL > 5.60 mm); the absence of macrosetae on the ventral, distal surface of metatarsus I (at least one macroseta usually present); and breeding season: late October through December (September through mid-October). In addition, males of the new species have a slightly more robust pedipalp tibia, although the ranges between the two species are more or less continuous: PTiL/PTiD = 2.09–2.24 (2.26–2.52). Females of both species are fairly homogeneous in morphology (Figs. 1A–1B) and pose some problems with identification, although the following characters may provide some diagnostic utility (Coweeta *A. unicolor* characters in parentheses): smaller size: CL < 7.00 mm (CL > 7.00 mm); lighter coloration: dorsal shield of prosoma, legs, and tergite usually yellowish-brown (dark chocolate brown); and convergent median dorsal setae just posterior to pedicel thin, sometimes somewhat thickened, and tapered (thorn-like); the latter character appears to work well for identifying immature specimens from Coweeta as well. The following morphometric ratio may provide some value: IVMeL/CL = 0.52–0.57 (0.59–0.61).

Antrodiaetus microunicolor new species can be differentiated from *A. robustus* by some of the same characters (*A. robustus* characters listed in parentheses, Coyle 1971): smaller size (male CL 5.40–6.60 mm; female CL 6.20–9.30 mm); male metatarsus I macrosetae (usually with macroseta A and B present). They also differ by the male prolateral tibia I macrosetae (less than one-fifth ensiform).



FIGURES 9–11. Adult females of *Antrodiaetus unicolor* (9, 10) and *A. microunicolor* new species (11) from Coweeta: 9, dorsal view showing location (indicated by white box) of convergent setae on opisthosoma posterior to pedicel (direction of view needed to observe setae indicated by white arrows); 10, opisthosoma, thorn-like setae, frontal view; 11, opisthosoma, thin and tapered setae, frontal view.

Description.—*Holotype male: Coloration* (in alcohol)*:* Specimen has been preserved for over 25 years and appears to have been bleached; we have decided to describe the coloration of a recently collected specimen instead. Base color of dorsal shield of prosoma, pedipalps, legs II–IV light grayish-tan, distal segments lighter. Eyes underlined with black pigment. Femur I light grayish-tan; patella I grayish-brown; tibia, metatarsus, tarsus I orangish-red. Chelicerae darker than dorsal shield of prosoma. Sternum pale grayish-yellow, labium darker. Opisthosoma purplish-gray; tergites darker than opisthosomal surface; second tergite somewhat darker than dorsal shield. *Prosoma:* Head region slightly elevated from thoracic region. Setae moderately dense along peripheral edges of dorsal shield; setae sparsely distributed on dorsal surface of dorsal shield of prosoma posterior to foveal groove. Sternum and labium moderately to densely setose. *Opisthosoma:* Three heavily sclerotized patches on dorsal surface; posterior patch smaller than others but mostly continuous with the second. Entire opisthosomal surface densely covered with setae, interspersed with some slightly more elongated and thickened setae posteriorly; tergites accompanied by a few thickened setae. Ventral surface of opisthosoma with 25 epiandrous gland spigots located just anterior to genital opening. *Chelicerae:* Anterior dorsal prominence weak. Upper ectal (retrolateral) surface devoid of setae. *Pedipalps* (Fig. 6)*:* Tibia moderately robust (PTiL/PTiD = 2.14). ICS tip below level of OCS; ICS tip well-sclerotized, tapered to a narrow point; OCS tip well-sclerotized, blunt, weakly serrated. *Leg I:* Mating clasper consisting of 16 ensiform, 5 attenuate macrosetae, centered at approximately 2/3 distance from proximal to distal end of tibia (Fig. 7). Prolateral, ventral, distal aspect of tibia with a macroseta. Retrolateral, ventral aspect of tibia with 4 ensiform, 1 attenuate macrosetae; distal-most macroseta of group positioned at approximately 2/3 distance from proximal to distal end of tibia. Prolateral, ventral aspect of tibia with 5 ensiform macrosetae. No macrosetae present on ventral aspect of metatarsus (Fig. 7). Metatarsus slightly sinuous in ventral view. *Measurements* (mm)*:* CL = 4.50; SL = 2.25; SW = 2.00; CT (l/r) = 10/9; PFeL = 2.70; PTiL = 2.35; PTiD = 1.10; IFeL = 4.10; ITiL = 2.85; IMeL = 3.15; ITaL = 1.85; ALD = 0.28; AMD = 0.12; ALS = 0.38; AMS = 0.16; OQW = 0.88.

Paratype female (MY 2402)*: Coloration* (in alcohol)*:* Dorsal shield of prosoma, opisthosomal tergite, pedipalps, and legs yellowish-brown, head region slightly darker. Eyes underlined with black pigment. Chelicerae light brown. Sternum orangish-brown, labium darker. Abdomen yellowish-brown with faint purple pigment posterior of tergite; cordate mark weakly indicated as pale longitudinal band along midline. *Prosoma:* Head region strongly elevated from throacic region. Setae moderately dense along peripheral edges of dorsal shield of prosoma; setae sparsely distributed on dorsal surface of dorsal shield of prosoma posterior to thoracic groove. Sternum and labium moderately to densely setose. Sternum with three pairs of sigilla, anterior-most pair somewhat reduced. *Opisthosoma:* Spermathecae (Fig. 8) consisting of four receptacles; stalk and bowl well-sclerotized; stalk not expanded basally; bulb somewhat flattened. Dorsal background setae sparsely to moderately long; tergite with a few thickened setae. Convergent median dorsal setae just posterior to pedicel thin and tapered, not thorn-like (see Figs. 9–11 for a comparison with *Antrodiaetus unicolor*). *Chelicerae:* Rastellum well-developed. Upper ectal (retrolateral) surface devoid of setae. *Measurements* (mm)*:* CL = 5.25; SL = 2.88; SW = 2.44; CT (l/r) = 12/11; PFeL = 2.70; IFeL = 3.70; ITiL = 2.30; IMeL = 2.10; ITaL = 1.25; IVFeL = 3.50; IVTiL = 2.05; IVMeL = 2.90; IVTaL = 1.30; ALD = 0.34; AMD = 0.14; ALS = 0.54; AMS = 0.16; OQW = 1.20.

Variation.—A total of 14 males and five females were studied from Coweeta. Three males lacked a macroseta on the prolateral, ventral, distal aspect of tibia I (a character also found in some males of *Antrodiaetus unicolor*). A very small male (MY 2421, CL = 3.60 mm) had macroseta A on the ventral aspect of metatarsus I, but we tentatively assign it to *A. microunicolor* new species on the basis of its size and breeding season. One female (MY 2441) had moderately more thickened convergent medial dorsal setae on the opisthosoma just posterior to the pedicel. However, these setae do not appear as well developed and thorn-like as they do in females of *A. unicolor* from Coweeta (Fig. 10), and look to be broken at their apices (i.e., they do not appear tapered). Because of their small size, adult females were identified solely on the basis of whether they contained offspring in their burrows. The lower limit of the length of the dorsal shield of prosoma for adult females is unknown. One female (MY 2401) is fairly large (CL = 6.88 mm), nearly as big as the smallest confirmed adult females of *A. unicolor*, but its dorsal shield color and opisthosomal convergent medial setae compare favorably to the other specimens belonging to the new species. A summary of measurements can be found in Tables 1 and 2.

Remarks.—Populations of *Antrodiaetus unicolor* (as delineated by Coyle 1971) containing unusually small males (i.e., ≈CL 4.00 mm) were also examined to determine the diagnostic utility of the metatarsus I macrosetal character. Small males from Pittsburgh, Pennsylvania (population A of Coyle 1971) and Duke Forest, North Carolina (population N) each possess macroseta A. The diminutive male that Coyle (1971) examined from west of Lake City, Tennessee (population L) was unavailable for study.

At least three other populations of *Antrodiaetus* containing unusually small adult females (as determined by the presence of offspring in their burrows) have been discovered throughout the course of fieldwork in western North Carolina and eastern Tennessee. These spiders compare favorably to females of *A. microunicolor* new species, but adult males are unavailable from these sites and we choose to hold off assigning them to any particular species at this time.

Before establishing *Antrodiaetus microunicolor* new species, other available names (i.e., those synonymized under *A. unicolor*) were considered for resurrection. As discussed above, Hentz (1841) described the species *Mygale gracilis* from Alabama, but the holotype was destroyed, and therefore, its exact identity is unknown; Coyle (1971) synonymized this name with *A. unicolor* and we feel that decision is well justified and should be maintained. Atkinson (1886) described two species, *Nidivalvata marxii* and *N. angustata*, from Chapel Hill, North Carolina. These two “species” are likely conspecific with material examined from Duke Forest in Durham and Orange County, North Carolina; these populations, located within the eastern piedmont, are at least 400 kilometers from Coweeta and are not conspecific with *A. microunicolor* new species. Finally, Simon (1884) described *Brachybothrium accentuatum* on the basis of an immature female from North Carolina. This specimen does not share characters indicative of the new species from Coweeta (e.g., thin and tapered setae). In addition, because immature mygalomorph spiders are nearly impossible to identify due to their lack of diagnostic characters, and because the precise location within North Carolina is unknown, we agree with Coyle’s (1971) decision to synonymize and maintain this name under *A. unicolor.*

Distribution.—The new species is presently known only from the type locality, at the Coweeta LTER site in the southern Appalachian Mountains near Otto, North Carolina (Fig. 2).

Etymology.—The specific epithet refers to the diminutive size of this species and its affinity to *Antrodiaetus unicolor.*

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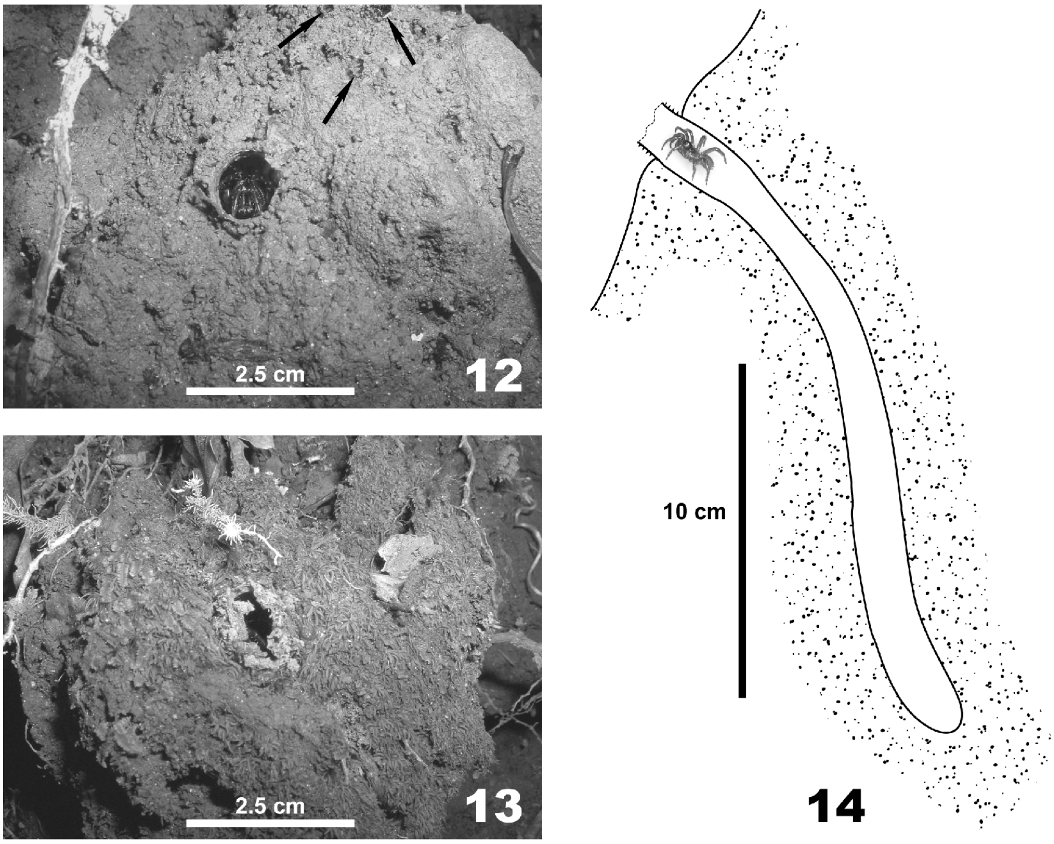
Both species have been collected more or less side-by-side at elevations ranging from 690–1120 m. Optimal habitat conditions compare favorably to that described for *Antrodiaetus unicolor* by Coyle (1971). The macrohabitat is more or less humid and cool with a dense deciduous canopy of mostly *Quercus*, *Acer* and *Betula*. The understory consists of nearly impenetrable patches of *Rhododendron maximum* and *Kalmia latifolia*. Burrow aggregations for both species are most abundant along stream banks and ravine slopes, and are typically located at the base of overhanging roots that provide protection from flooding. The soil is predominantly damp and spongy, although sandy loam-like soils are common directly along streams. Burrows have also been found in moss mats, among rock crevices, and beneath decaying logs. No differences in microhabitat could be detected between the two species. Both species appear to be highly abundant at all localities throughout Coweeta.



FIGURES 12–14. Burrows of adult female *Antrodiaetus microunicolor* new species: 12, waiting at entrance for prey (arrows indicate burrows belonging to immature *Antrodiaetus*); 13, partially closed burrow (this photograph was taken shortly after dusk); 14, diagram illustrating internal structure of burrow.

Burrows of both species do not appear structurally different (no data collected on *Antrodiaetus unicolor* burrows, but see Coyle 1971). Adult female burrows of *A. microunicolor* new species (Figs. 12–14) are lightly to heavily lined with silk, and slightly widened near the entrance and base. The burrow entrance is commonly constructed almost perpendicular to the substrate; however, burrows frequently change direction in response to roots, stones, and other obstacles. Burrows ranged from approximately 8–10 mm in diameter and 15–20 cm in depth.

Breeding season is defined as the time when adult males are actively searching for females. Based on pitfall trap sampling, males of *Antrodiaetus unicolor* probably begin wandering in mid-September and continue until mid-October. Males of *A. microunicolor* new species emerge from their burrows in late October and presumably remain active through December. We have found no evidence for even a slight overlap in breeding seasons (i.e., two independent pitfall trap series failed to recover both species synchronously). Interestingly, no adult males for either species were collected from their burrows prior to their perceived breeding season. This suggests that they may take extra care concealing their burrows prior to emergence. Live males were only observed on two occasions (*A. unicolor* on 27 September and *A. microunicolor* new species on 8 November), both at least an hour after dusk.

Initial observations suggest that females demonstrate an interesting pattern of seasonal activity as well, but more data must be collected to describe these general trends. Adult female burrows of *Antrodiaetus unicolor* were observed early in the collecting season, coinciding roughly with the period of male wandering. No females were observed with eggsacs or offspring in their burrows, and only one female (MY 2323) appeared gravid. Furthermore, no large burrows (indicative of adult female *A. unicolor*) appeared active later in the season. Conversely, adult females (n = 5) of *A. microunicolor* new species were only observed later in the collecting season: 25 October–15 November. Each was found with second instar offspring scattered throughout the burrow. It is unknown whether females brooding offspring are capable of breeding in sequential seasons. Offspring were only collected from two females, these ranging in number from 98 to 140 (the actual number is more than this because not all individuals could be collected).

An additional burrowing mygalomorph spider species was found associated with *Antrodiaetus* at Coweeta. A single male belonging to the trapdoor spider genus *Ummidia* (family Ctenizidae) was collected in the same pitfall trap as two males of *A. unicolor* during the 27 September–11 October pitfall-trapping series at Watershed 2. Careful inspection and light scraping of the surrounding banks did not uncover any burrows of this species.

No data on predators, prey, or parasites were collected. *Antrodiaetus* undoubtedly feed upon the multitude of small arthropods available (see Coyle 1971 for a summary of prey items taken by *A. unicolor*).

Discussion

This manuscript is the first in a series of studies investigating the systematics and evolution of the *Antrodiaetus unicolor* "species complex" in the eastern United States. The discovery of two species at Coweeta is intriguing and provides some insight into the meaningfulness of the morphological variation first observed by Coyle (1971) in this group of difficult spiders. Furthermore, it presents an attractive system for exploring broader scale questions in evolutionary biology (e.g., maintenance of sympatric species, character displacement, delimitation of species boundaries).

This species complex poses an interesting problem for detecting and delimiting species boundaries. As already discussed, *Antrodiaetus unicolor* (*sensu lato*) exhibits a great deal of variation, both within and between populations. Conversely, a number of (at least partially) isolated and allopatric populations show little to no morphological differentiation. Is this indicative of a single widespread species or numerous localized "cryptic species"? Coyle (1971) discussed the difficulty in interpreting this variation and declined to describe any new species from the extensive material he examined. Some populations were divergent in a number of characters, but intermediate forms were common. To compound this complex issue, numerous populations are known only from females or immature specimens. Female and immature mygalomorph spiders pose a particular challenge to taxonomy because of their paucity of useful characters. In most cases, females will be assigned to a species on the basis of known males from the area. Clearly, this is warranted if only one species is known from that specific locality. However, when closely related species are sympatric or if males are unknown, little confidence can be placed in these criteria when morphological divergence is scarce.

Detecting and defining species boundaries are arguably the most significant endeavors in systematic biology, if not simply for their importance in improving our understanding of biodiversity. Unfortunately, the above-mentioned morphology (and male) biased taxonomy incontrovertibly underestimates true evolutionary diversity in this group of spiders. To fully unravel this species complex will require a multifaceted approach. Approaches to delimiting species boundaries using molecular techniques (e.g., DNA sequencing) have great potential for recovering diversity that is difficult to decipher on the basis of morphology alone (e.g., highly conservative or variable populations). In addition, molecules provide an independent source of data that allows for an explicit reassessment of traditional taxonomies based on morphology in a rigorous hypothesis-testing manner. Numerous studies in recent years have convincingly demonstrated "cryptic diversity" in arthropod groups previously defined solely on the basis of morphology (e.g., pseudoscorpions, Wilcox et al. 1997; trapdoor spiders, Bond et al. 2001; araneomorph spiders, Hedin & Wood 2002; millipedes, Bond & Sierwald 2002, Bond et al. 2003) and some have argued that in such cases, a molecular approach to defining species using DNA taxonomy "may be the only way to reveal the true level of biodiversity" (Proudlove & Wood 2003). Furthermore, some authors have used molecular characters for species diagnoses (e.g., Bond & Sierwald

2003) *in conjunction* with studies of morphological variation. Our future studies on the *Antrodiaetus unicolor* species complex will most certainly utilize a “total evidence” approach that will take into account morphological and molecular variation (e.g., Puorto et al. 2001; Wiens & Penkrot 2002; Lee 2004), within the context of a robust phylogenetic framework. Concordance from multiple lines of evidence will likely prove to be the most rigorous approach for delimiting species.

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