Copyright © 2011 · Magnolia Press

Article



Redescription of *Hypogastrura gracilis*, synonymy of *Ceratophysella quinidentis* with *C. duplicispinosa*, and additional information on *C. adexilis* (Collembola: Hypogastruridae)

JIGANG JIANG¹, WENYING YIN¹, JIANXIU CHEN² & ERNEST C. BERNARD^{3,4}

¹Institute of Plant Physiology & Ecology, Shanghai Institutes for Biological Sciences, Chinese Academy of Sciences, Shanghai 200032, China; Department of Life Science, Hunan University of Arts and Science, Changde 415000, China. E-mail: jjgangnanjing@163.com; wyyin@sibs.ac.cn ²Department of Biology, Nanjing University, Nanjing 210093, Jiangsu, China. E-mail: chenjx@nju.edu.cn ³The University of Tennessee, Entomology and Plant Pathology, 2431 Joe Johnson Drive, 205 Plant Sciences, Knoxville, TN, USA 37996-4560. E-mail: ebernard@utk.edu

⁴Corresponding author

Abstract

Hypogastrura gracilis Folsom is redescribed from cotypes and Chinese specimens, and a lectotype and 4 paralectotypes are designated. Specimens referable to *Ceratophysella adexilis* Stach are described. *Ceratophysella quinidentis* (Jia, Shi & Chen) is reevaluated from type specimens and recognized as a new junior synonym of *C. duplicispinosa* (Yosii).

Key words: Ceratophysella cf. adexilis, taxonomy

Introduction

Many of the world's species of Hypogastruridae were described well before the systematic importance of mouthpart morphology, chaetotaxy, and sensilla arrangement was adequately appreciated. Numerous Asian species described 40 or more years ago are difficult to recognize because the original descriptions are very incomplete compared with current standards, which has led to descriptions of synonymous taxa or to the outright inability to confidently identify even common, widespread species. The objective of this paper is to clarify three of these problematic taxa. *Hypogastrura gracilis* Folsom (1899), described from Japan, is redescribed from a type specimen and compared with other Asian reports and with Chinese specimens. Specimens referable to *Ceratophysella adexilis* Stach, 1964, already known from China, are described from Nanjing City. Type material of *Chinogastrura quinidentis* Jia, Shi & Chen, 2005 was examined to determine its similarity to *Ceratophysella duplicispinosa* (Yosii, 1954), which is known from Japan, China, Korea, and the Russian Far East. Assignment of chaetotaxy types (A or B) in *Ceratophysella* spp. follows Gisin (1947) and Fjellberg (1998). Clavate tenent hair designations follow Deharveng (1983). Antennal segments are abbreviated Ant. I–IV. Thoracic and abdominal segments are abbreviated Th. I–III and Abd. I–VI. Fore, middle and hind legs are abbreviated as legs I–III.

Taxonomy

Hypogastrura gracilis (Folsom, 1899)

Figs 1-3

Achorutes gracilis Folsom, 1899: 263

Type material. Female lectotype (on slide) and 4 paralectotypes (in alcohol), Japan, Yanaka, Tokyo, Japan, 14 November 1894, deposited in the Museum of Comparative Zoology, Harvard University, Cambridge, Massachusetts, USA.

Other material. 16 females and 6 males, China, Jiangsu Province, Lianyungang City, Huaguoshan Park, 6 November 2006, in soil under bricks and piles of decayed peanut plants, collected by Jian-xiu Chen *et al.*; collection number C9530, deposited in the Department of Biology, Nanjing University, China.

Redescription. Body length: up to 1.9 mm; color in ethanol violet to violet-black dorsally, pale to yellow ventrally and on tibiotarsi, furca and ventral tube. Tegumentary granulation fine and uniform, dorsum of Abd. V with 9-19 (usually 13–15) granules between setae p_1 .

Body setae similar in length. Dorsal cephalic setae weakly differentiated (Figs 1A, B). Seta v_2 longer than v_1 ; setae p_1 , p_3 and p_5 slightly blunt, and longer than p_2 , p_4 and p_6 (Fig. 1A). Head ventrally with 6 proximal, 4 basomedial, 5 basolateral and 3 postlabial setae (Fig. 2D). Th. I with 3+3 setae. Th. II with 3 rows of setae; m_3 present (lectotype) or absent (Chinese specimens), m_2 , m_5 or m_6 rarely absent, m_7 and p_4 sensilliform. Th. III with 3 rows of setae; m_2 , m_5 , p_3 . present, m_3 absent in lectotype; m3 present, m2, m5 absent in Chinese specimens; m_7 and p_4 sensilliform, ratio of sensillum p_4 to common seta $p_5 2.1-3.0:1$. Setae absent on thoracic sterna. Abd. I–III each with 3 rows of setae, but setae and their positions on Abd. I in lectotype partially obscured: 5+5 in anterior row, a_3 and a_5 absent; 3+3 in middle row, as m_3 , m_4 and m_6 or m_7 ; 7+7 in posterior row, as p_{1-7} , p_5 sensilliform (Fig. 1A). Abd. IV with 3 rows of setae: 5+5 in anterior row; 3+3 in middle row, m_2 absent; 6+6 in posterior row, p_5 sensilliform, ratio of p_5 to common seta $p_4 2.4-2.5:1$ in Chinese specimens, slightly less in lectotype (Figs 1A, B). Abd. V with 2 rows of setae, 5+5 in anterior row; 6+6 in posterior row, $p_1:p_2$ ratio $1.5:1; p_3$ sensilliform (Fig. 1).

Head without tubercles and spines. Labrum (Figs 2A, B) with granulation similar to that of body; 6 apical papillae, outer papillae rounded to conical, setal formula 4/5, 5, 4. Maxilla with lamella 1 longer and other lamellae shorter than maxillary teeth; short marginal filaments present on lamellae 2 and 3; dense and fine denticles present on lamella 1 and inner side of lamellae 4–6; lamella 5 with large process (conspicuous "shoulder") (Fig. 2C). Maxillary outer lobe with 2 sublobal hairs (Fig. 2F). Labial palpus with 5 papillae (A–E), lateral process, guard setae a_1 , b_{1-4} , d_{2-4} , e_{1-6} present, d_1 and e_7 absent (Fig. 2E).

Antennae (Figs 2G–I) 0.60–0.80 and 0.12–0.15 times length of cephalic diagonal and body length, respectively. Ant. IV with simple or weakly lobed subapical bulb; subapical vesicle (os) and microsensillum (ms) present; 3 dorsoexternal and 1 dorsointernal sensilla, curved and somewhat blunt, ventrally with 2 shorter sensilla near apex. Antenna III organ with 2 short rods in separate foveae and 2 guard sensilla. Ventral microsensillum present on Ant. III. Ant. II with 13 setae. Ant. I with 7 setae, without seta *p*. Eyes 8+8; eye patch with 3 setae, Oc_2 longer than Oc_1 and Oc_3 (Fig. 2J).

Postantennal organ subequal to nearest eye in diameter, composed of 4 lobes; anterior 2 lobes larger than posterior lobes, accessory tubercle absent (Fig. 2J).

Unguis well developed, with 1 inner tooth at 2/3 distance of its inner edge from base and 2 small lateral teeth near the apex (Fig. 3A). Unguiculus with broad basal lamella, tip of apical filament reaching 2/3–4/5 distance of inner edge of unguis. Legs I–III respectively with 1, 2, 3 setae on subcoxa 1; 0, 3, 3 on subcoxa 2; 3, 8, 7 on coxa; 7, 7, 7 on trochanter; 12, 13, 12 on femur; and 19, 19, 18 on tibiotarsus including 2, 3, 3 clavate tenent hairs (Figs 3A–D): on tibiotarsus I, setae A1 and A2 clavate; on tibiotarsi II and III, A1, A2, and A7 clavate; seta m present.

Ventral tube with 4+4 setae (Fig. 3E). Tenaculum with 3 teeth on each ramus, no seta on corpus. Manubrium with about 10+10 posterior setae. Dens posteriorly with fine granules and 7 setae (Figs 3F, G), length ratio of outer basal seta to inner basal seta 1.6–2.5:1 (average 2:1). Mucro with narrow outer lamella, without inner lamella; length ratio of dens to mucro 3.0–3.5:1 (average 3:1). Two anal spines on Abd. VI slightly curved, conical, slightly shorter than basal papillae. Adult female and male genital plates with 21–27 and 30–45 setae, respectively (Figs 3H, I).

Remarks. Five cotypes of this species were borrowed from the Museum of Comparative Zoology, Harvard University, Cambridge, Massachusetts, and one specimen was mounted for microscopic observation.

Folsom (1899) described *Hypogastrura gracilis* from Tokyo, but the description is incomplete by current standards. Although the species was redescribed by Yosii (1960), knowledge of its characters still was considered insufficient to place it in a key to Palaearctic Hypogastruridae (Thibaud *et al.* 2004). Korean specimens were described by Park & Park (2006), but without a detailed analysis of the chaetotaxy. Neither of these redescriptions was based on type specimens.



FIGURE 1. *Hypogastrura gracilis*: A, Dorsal chaetotaxy of specimen from Jiangsu province, China; B, Partial dorsal chaetotaxy of lectotype. S = sensilliform seta.



FIGURE 2. *Hypogastrura gracilis*: A, Labrum of lectotype; B, Labrum of Chinese specimen; C, Maxilla of Chinese specimen: a, inner view, b, ventral view; D, Ventral cephalic chaetotaxy, Chinese specimen; E, Labial palpus, Chinese specimen; F, outer maxillary lobe, Chinese specimen; G–H, Dorsal view of antenna and ventral view of Ant. IV, respectively, Chinese specimen; I, Dorsal view of antenna, lectotype; J, Postantennal organ and eye patch, Chinese specimen.

Examination of a type specimen of *H. gracilis* established that the unguis possesses distal lateral teeth, a character not reported by Folsom (1899). The examination also confirmed the characters reported by earlier authors, although some of the setae were obscured or not found due to intestinal contents and crystalline formations in the body, especially in the region of Abd. I–III. Nevertheless, it is clear that the Chinese specimens should be assigned to this species. *Hypogastrura gracilis* is a member of the *H. viatica* species group (Yosii 1960) on the basis of



FIGURE 3. *Hypogastrura gracilis*: A, Distal part of tibiotarsi, lectotype: a, tibiotarsus I, b, tibiotarsus II, c, tibiotarsus III; B–D, Tibiotarsus I, tibiotarsus II, hind leg, respectively, Chinese specimen; E, Ventral tube; F, Dens and mucro, lectotype; G, Furca, Chinese specimen: a, dorsal view of left side, b, lateral view of mucro; H–I, Female and male genital plates, respectively, Chinese specimens.

multiple clubbed tenent hairs (usually 2,3,3–4) on the tibiotarsi. In the key of Thibaud *et al.* (2004) this species will trace to *H. viatica* (Tullberg). However, the macrochaetae of Th. I–Abd. IV of *H. gracilis* are all short and of

approximately equal length; whereas, many of the p-row setae of *H. viatica* are much longer than the a-row setae (Jordana *et al.* 1997, Yosii 1960). Also, *H. gracilis* possesses 7 dental setae (6 on *H. viatica*). Dental seta number separates *H. gracilis* from all other *H. viatica*-group species except *H. barguzini* Babenko in Babenko *et al.* (1994) and *H. purpurescens* (Lubbock). *Hypogastrura barguzini* differs from *H. gracilis* in having m-row setae on Abd. V (lacking in *H. gracilis*) and having body sensilla shorter than the common setae (longer in *H. gracilis*). In *H. purpurescens*, the metatibiotarsus has 2 clavate tenent hairs (A1, B2), whereas *H. gracilis* has 3 (A1, A2, A7).

Characters of *H. gracilis* from China conform well with the lectotype in the possession of 6 anterior papillae on the labrum (more pronounced and rounded on lectotype, conical on Chinese specimens), dens with seven setae, and mucro with small outer lamella. However, several apparent chaetotaxic differences, based on the redescription of Yosii (1960), led us to conclude initially that the Chinese specimens were a separate species closely related to *H. gracilis*. These differences include the apparent lack of m-setae on Abd. I–III (present on Chinese specimens) and absence of seta m_4 on Th. II (present on Chinese specimens). In the lectotype and Chinese specimens, the m-setae are advanced so far anteriorly that they could be interpreted as a-setae. The basic chaetotaxic plan as illustrated by Yosii (1960, p. 259, "key figure") indicated a complete m-row for Abd. I–II. In most species of *Hypogastrura* s. 1. as illustrated by Yosii, the m-row in these segments is usually reduced or absent. Most recent authors have accepted the presence of a reduced m-row consisting of m_3 and m_4 (Babenko *et al.* 1994, Jordana *et al.* 1997), but Christiansen & Bellinger (1998) illustrated Abd. III as having only a- and p-setal rows. For convention the Babenko-Jordana interpretation is accepted here, even though m_3 and m_4 are located in the precise positions they would be if they were setae a_3 and a_5 slightly displaced posteriorly.

Identification of Korean specimens as *H. gracilis* (Park & Park 2006) is not entirely certain. The lectotype and Chinese specimens of *H. gracilis* have 1+1 distal lateral teeth on the unguis, setae a_3 and m_3 on Th. III, and sensilliform seta p_5 on Abd. IV. The Korean specimens were not described as having lateral ungual teeth; lacked setae a_3 and m_3 on Th. III; and possessed seta p_4 on Abd. IV as a sensillum. However, the other characters described for Korean *H. gracilis* fit the species well. The lateral ungual teeth may be minute and very difficult to detect initially unless the unguis is seen in dorsal or ventral view (see Fig. 2A). Furthermore, hypogastrurid body setae can vary from specimen to specimen and even between left and right sides. Occasional extra or missing setae are frequent, which may account for the p_4 position of the Abd. IV sensillum and the variation in Th. III chaetotaxy in the Korean specimens, and for chaetotaxic differences with the specimens redescribed by Yosii (1960).

Ceratophysella cf. *adexilis* Stach, 1964 Figs 4, 5

Material examined. Six females and 2 males on slides, many in alcohol, Nanjing City: Qingliangshan Park, in soil under a pile of tiles. Collection number C9462, deposited in Nanjing University

Remarks on Chinese specimens. Body length up to 1.6 mm, color violet-black. Dorsal body granules moderately fine, coarser on most of Abd. V; dorsum of Abd. V with 13–15 granules between p₁ setae.

Body setae well differentiated (Figs 4A, B), Type A. Th. I with 3+3 setae. Th. II with three rows of setae, as a_{1-2} , a_{4-6} , m_1 , a_{5-7} and p_{1-6} . Th. III with three rows of setae, as a_{1-6} , m_1 , a_{5-7} and p_{1-6} ; m_7 and p_4 sensilliform (Fig. 4A). Abd. I–III each with 3 rows of setae, as a_{1-3} , m_{3-4} , a_6 and p_{1-6} ; a_2 ' seta present. Abd. IV with 3 rows of setae, a_1 absent. Abd. V with 2 rows of setae, p_3 sensilliform.

Antenna shorter than diameter of head. Ant. III sense organ composed of two short rods located in small grooves and two blunt sensory hairs (Fig. 5E). Eversible sac strongly developed. Ant. IV ventrally with weakly developed sensory field of ten pointed file setae (Fig. 5F). Ant. IV dorsally with seven sensilla (Fig. 5E). Seta *i*, subapical vesicle (os) and microsensillum (ms) present. Apical bulb bilobed.

Postantennal organ typical for the genus (Fig. 5A), occasionally with irregular lobes (Fig. 5B), 2–2.5× diameter of neighbouring ocellus. Accessory tubercle finely granulated and rounded (Figs 5A, B). Labrum with granules on corpus, without rounded apical papillae, setae formula 4/5, 5, 4. Maxilla (Fig. 5C) with 6 lamellae, lamella 1 longer than maxillary teeth, with coarse denticles and apical third with thickened marginal filaments; lamellae 2 and 3 with long marginal filaments; lamella 4 with denticles, ventrally with shorter filaments or longer denticles; lamella 5 with denticles and dorsally with short filaments). Maxillary outer lobe with one sublobal hair (Fig. 5D).

Unguis with distinct inner mid-ventral tooth; lateral teeth minute or not visible (Fig. 5G). Unguiculus almost half length inner side of unguis, with broad, rounded basal lamella and short apical filament. Tibiotarsal tenent hair as long as unguis, acuminate. Tibiotarsi I–III respectively with 19, 19, 18 setae.

Ventral tube with 4+4 setae, tenaculum with 4+4 teeth. Furca well developed. Ratio of manubrium:dens:mucro 1.7–2.2:1.8–2.1:1. Dens dorsally finely granulated, with 7 setae, 2 inner apical setae slightly thicker than other setae (Fig. 4C). Mucro boat-like, rounded at tip, with well-developed outer lobe (Fig. 4C). Anal spines long, slightly curved, pale yellowish, located on high papillae touching at their bases. Anal spines + papillae longer than inner side of unguis.

Discussion. We tentatively apply the name *adexilis* to these specimens, but the status of the species itself is uncertain as it cannot reliably be distinguished from several congeners (see Jia *et al.* 2010). *Ceratophysella adexilis* was described from the vicinities of Beijing and Nanjing. It has type A chaetotaxy and belongs to the *C. denticulata* group. Information on body length and pigmentation is lacking in the original description. According to the original description, the submedian and lateral macrosetae of all terga are smooth and very long (Stach 1964). However, Babenko *et al.* (1994) pointed out that some type specimens of *C. adexilis* had serrated macrosetae. Therefore, specimens collected from Qinglianshan Park in Nanjing City are consistent with characters of the type description. *Ceratophysella adexilis* is very similar to *C. communis* (Folsom), but their relationship to each other cannot be clarified until type specimens of the many similar species are comparatively examined (Jia *et al.* 2010).

Distribution. Nanjing, Beijing (Stach 1964).



FIGURE 4. Ceratophysella cf. adexilis: A, Dorsal thoracic chaetotaxy; B, Dorsal abdominal chaetotaxy; C, Dens and mucro.



FIGURE 5. *Ceratophysella* cf. *adexilis*: A, Postantennal organ and eyes; B, Atypical postantennal organ; C, Maxilla; D, Outer maxillary lobe; E, Third and fourth antennal segments, dorsal view; F, Third and fourth antennal segments, ventral view; G, Tibiotarsus.



FIGURE 6. *Ceratophysella quinidentis* type specimens (= *C. duplicispinosa*): A, Third and fourth antennal segments, dorsal view; B, Third and fourth antennal segments, ventral view; C, Labrum; D, Apex of tibiotarsus; E, Furca, dorsal view of right side.

Ceratophysella duplicispinosa (Yosii, 1954)

Fig. 6

Hypogastrura duplicispinosa Yosii, 1954: 781 Chinogastrura punctata Rusek, 1967: 186 Chinogastrura quinidentis Jia, Shi & Chen, 2005: 241 **new synonym** Ceratophysella quinidentis Jia, Skarżyński & Li 2010: 61

Material examined. *Ceratophysella quinidentis* male holotype, 2 female and 2 immature male paratypes, 55 additional juveniles, Nanjing University, Nanjing City, Jiangsu Province, China, 26 February 1990, K. Christiansen, coll., deposited in Nanjing University as #8118.

Ceratophysella duplicispinosa. Two females, Zhejiang Province, core area of Western Tianmu Mountain, 14 Apr. 2005, coll. Chen *et al.*, No. C9248; 3 males, 1 female from Hunan Province, Changde City, Huayanxi Forest Park, 27 Oct. 2005, Coll. Jigang Jiang, No. 9294; 3 males and 2 juveniles, Guangdong Province, Heshan Hilly Land Interdisciplinary Experimental Station, Chinese Academy of Science, 15 Sept. 2006, Coll. Chen *et al.*

Remarks. The type specimens of *C. quinidentis* agree well with the description of *C. duplicispinosa* (Yosii 1954), as well as that of Russian specimens (Babenko *et al.* 1994). Reexamination of the *C. quinidentis* type material verified the presence of a trilobed apical bulb on Ant. IV and ventral file of 20–25 truncate setae (Fig. 6B), and the presence of a typical *Ceratophysella* labrum (Fig. 6C). However, some characters of *C. quinidentis* were inaccurately described. The fourth antennal segment was found to have 5 rather than 3 sensilla (Fig. 6A). The unguis possesses proximal and distal lateral teeth (Fig. 6D). With regard to tenacular teeth, one specimen has 5+5 teeth, another has 4+5 teeth, and the rest have 4+4 teeth. The dens is strongly granulate dorsally and has 7 setae, of which 2 are thickened (Fig. 6E).

The comparison of *C. quinidentis* with *C. duplicispinosa* (Jia *et al.* 2005, Table 1) contained several errors. For instance, *C. duplicispinosa* was cited as having 4 dorsal manubrial setae and 6 dental setae, and lacking ventral file setae on Abd. IV. However, *Ceratophysella duplicispinosa* has 20–35 setae present in the antennal file, 6–7 setae on the dens (Babenko *et al.* 1994, Rusek 1967, Yosii 1954), and at least 8 setae on each side of the manubrium (Yosii 1954). Mature specimens from Zhejiang, Hunan and Guangdong have 20–30 sensilla present in the ventral file of Ant. IV, while immature specimens usually have fewer than 15 setae in the file. Based on the analysis of the type specimens, there are no characters left that separate these two taxa. The best diagnostic character for this species is the transformation of the p_1 setae of Abd. V to thick spines. Therefore, we consider *C. quinidentis* a junior synonym of *C. duplicispinosa*.

Acknowledgements

The present study was supported by the Ministry of Science and Technology of the People's Republic of China (2006FY120100), a China Postdoctoral Science Foundation funded project (No. 20100470743), and also by the Scientific Research Fund of the Hunan Provincial Education Department (09C698). The authors also gratefully acknowledge the support of the K.C. Wang Education Foundation, Hong Kong. Finally, we express our appreciation to Dariusz Skarżyński and Anatoly Babenko for their careful reviews and insightful comments.

References

Babenko, A.B., Chernova, N.M., Potapov, M.B. & Stebaeva, S.K. (1994) *Collembola of Russia and adjacent countries: Family Hypogastruridae*. Nauka, Moscow, 336 pp. [In Russian with English summaries].

Christiansen, K.A. & Bellinger, P. (1998) The Collembola of North America North of the Rio Grande. Part I: Introduction, General, Families Poduridae and Hypogastruridae. Grinnell College, Grinnell, Iowa, 425 pp.

Deharveng, L. (1983) Morphologie évolutive des Collemboles Neanurinae en particulier de la lignée néanurienne. *Travaux de Laboratoire d'Ecobiologie des Arthropodes Edaphiques, Toulouse*, 4, 1–63.

Fjellberg, A. (1998) The Collembola of Fennoscandia and Denmark. Brill, Leiden, 184 pp.

Folsom, J.W. (1899) Japanese Collembola, Part II. Proceedings of the American Academy of Arts and Sciences, 34 (9), 261–274.

- Gisin, H. (1947) Notes taxonomiques sur quelques especes suisses des genres *Hypogastrura* et *Xenylla*. *Mitteilungen der Schweizerischen Entomologischen Gesellschaft*, 20, 341–344.
- Jia, J.-L., Shi, S.-D. & Chen, J.-X. (2005) A new species of the genus *Chinogastrura* (Collembola: Hypogastruridae) from China. *Entomotaxonomia*, 27 (4), 241–245.
- Jia, J.-L., Skarżyński, D. &. Li, Y. (2010) *Ceratophysella taiguensis* sp. nov. (Collembola, Hypogastruridae) from China, with an annotated checklist of Chinese *Ceratophysella* Börner, 1932. *Zootaxa*, 2644, 57–63.
- Jordana, R., Arbea J.I., Simón, C. & Luciáñez, M.J. (1997) Fauna Iberica, Vol. 8. Collembola Poduromorpha. *Museo Nacional de Ciencias Naturales, Madrid*, 807 pp.
- Park, K.H. & Park, N.Y. (2006) Two new species of *Ceratophysella* (Collembola: Hypogastruridae) from Korea. *Florida Ento-mologist*, 89 (4), 489–496.

Rusek, J. (1967) Beitrag zur Kenntnis der Collembola (Apterygota) Chinas. Acta Entomologica Bohemoslovaca, 64, 184–194.

Stach, J. (1964) Materials to the knowledge of Chinese collembolan fauna. Acta Zoologica Cracoviensia, 9, 21–26.

Thibaud, J.-M., Schulz, H.-J. & Gama Assalino, M.M. da (2004) Hypogastruridae. *In*: Dunger, W. (Ed.) Synopses on Palearctic Collembola. Vol. 4, *Abhandlungen und Berichte des Naturkundemuseums Görlitz*, 75 (2), 1–287.

Yosii, R. (1954) Springschwänze des Ozé-Naturschutzgebietes. Scientific Researches of the Ozegahara Moor, 777-830.

Yosii, R. (1960) Studies on the Collembolan genus Hypogastrura. The American Midland Naturalist, 64 (2), 257–281.