

Gregarines Infecting *Ischnura* spp. in Texas, U.S.A., Including Description of *Septemlaterospora rasberryi* n. gen. n. sp. (Apicomplexa: Actinocephalidae: Acanthosporinae) and Revision of *Steganorhynchus dunwoodyi* (Apicomplexa: Actinocephalidae: Menosporinae)

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GREGARINES INFECTING *ISCHNURA* SPP. IN TEXAS, U.S.A., INCLUDING DESCRIPTION OF *SEPTEMLATEROSPORA RASBERRYI* N. GEN. N. SP. (APICOMPLEXA: ACTINOCEPHALIDAE: ACANTHOSPORINAE) AND REVISION OF *STEGANORHYNCHUS DUNWOODYI* (APICOMPLEXA: ACTINOCEPHALIDAE: MENOSPORINAE)

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ABSTRACT: *Septemlaterospora rasberryi* n. gen. n. sp. (Apicomplexa: Eugregarinida: Actinocephalidae: Acanthosporinae) is described from adults of *Ischnura ramburii* (Odonata: Zygoptera: Coenagrionidae). *Septemlaterospora* n. gen. is distinguished by the form of the oocysts: terminally truncated heptagonal bipyramids bearing 7 equatorial spines, 1 at each equatorial vertex, 7 terminal spines obliquely inserted at each pole, 1 at each vertex created by polar truncation; 21 spines total. The holdfast is compound, comprising a terminal epimerite and intercalating diamerite; epimerite is a thick disk or linearly crateriform sucker; diamerite is short (less than half of the total holdfast length) and very broadly obdeltoid. Association occurs immediately before syzygy and is cephalolateral and biassociative. Gametocysts are spherical with a conspicuous hyaline coat. Lacking conspicuous sporoducts, they dehiscence by simple rupture. *Steganorhynchus dunwoodyi* is redescribed utilizing a new complete taxonomic data set, consisting of a larger set of metric characters and based on uniformly prepared, permanent specimens. New host and geographic records are reported for *Calyxocephalus karyopera*, *Domadracunculus janovyi*, *Nubenocephalus secundus*, and *Steganorhynchus dunwoodyi*, and the type host of *D. janovyi* is amended.

All known gregarines of Nearctic odonates (both Anisoptera and Zygoptera) are members of the family Actinocephalidae. Seven species, assigned to 6 genera across 3 subfamilies, have been described from Zygoptera in North America, north of Mexico: *Actinocephalus carrilynnae* Richardson and Janovy, 1990 (Actinocephalinae); *Nubenocephalus nebraskensis* Clopton, Percival, and Janovy, 1993 (Acanthosporinae); *Domadracunculus janovyi* Clopton, 1995 (Menosporinae); *Hoplorhynchus acanthatholius* Percival, Clopton, and Janovy, 1995 (Menosporinae); *Steganorhynchus dunwoodyi* Percival, Clopton, and Janovy, 1995 (Menosporinae); *Calyxocephalus karyopera* Clopton, 2004 (Acanthosporinae); and *Nubenocephalus secundus* Hays, Cook, and Clopton, 2007 (Acanthosporinae). This is considerably less than known diversity of other biogeographical regions that have been surveyed more intensively, e.g. the Indian subcontinent (e.g., Kori and Amoji, 1985; Sarkar, 1995, 1997) and Oriental faunal regions (e.g., Obata, 1953; Hoshide, 1959, 1977), but this is more likely the result of disparity in survey effort than true Nearctic faunistic poverty (Clopton, 1995; Hays et al., 2007). As part of an ongoing survey effort to expand the geographic survey area and the taxonomic breadth of odonate hosts examined for eugregarine parasites, we surveyed the eugregarine fauna of adults and naiads of three species of *Ischnura* (Zygoptera: Coenagrionidae) from several localities in Texas.

During this survey, we collected a heretofore unknown gregarine species from adults of *Ischnura ramburii* and *Ischnura posita* (Odonata: Zygoptera: Coenagrionidae). The gregarines recovered are referable to the family Actinocephalidae and subfamily Acanthosporinae, but they are taxonomically distinct from its constituent genera. Herein we describe the new taxon, and, additionally, we provide the first report of *Steganorhynchus dunwoodyi* since its original description, including new host and locality records, report a new complete morphological data set,

and delineate an extended gregarine morphometric set for *Steganorhynchus*. We further report new host and locality records for an additional 4 gregarine species.

MATERIALS AND METHODS

Populations of *Ischnura* spp. were collected from the following locations in Texas: Collins Pond, Polk County (30.410°N, 94.128°W); Beaver Slide Pond, Polk County (30.385°N, 96.176°W); city of Livingston Pond, Polk County (30.712°N, 94.951°W); Center for Biological Field Studies Pond (CBFS), Sam Houston State University, Walker County (30.746°N, 95.474°W); Parker Pond, Walker County (30.699°N, 95.527°W); Raspberry Pond, Walker County (30.692°N, 95.435°W); Lone Star Pond, Walker County (30.715°N, 95.517°W); Steinhagen Lake, Tyler County (30.853°N, 94.174°W); Double Lake, San Jacinto County (30.550°N, 95.133°W); Oil Field Pond, Madison County (31.641°N, 96.328°W); Wildlife Viewing Area Pond, Willacy County (26.482°N, 97.748°W); Lake Texana, Jackson County (29.019°N, 96.563°W); and Herron Pond, Taylor County (32.134°N, 99.708°W).

Damselflies were isolated, abdomen-down, in 5-ml test tubes with ~1 ml of water. Shed gametocysts were isolated from collected feces or collected directly from the rectum on postmortem examination, triple rinsed in spring water, and placed in individual silicon-stoppered 4 × 12-mm glass microvials (BioQuip Products, Gardena, California) with ~50 µl of spring water and observed daily for maturation and dehiscence. Fresh preparations of oocysts were examined as wet mounts.

Parasite preparations followed the methods of Clopton (2004a). Adult damselflies were eviscerated, and their alimentary canals were dissected in insect muscle saline (Belton and Grundfest, 1962). Permanent parasite preparations were made using wet smears of gregarines and host gut tissues. Smears were fixed by inverted flotation in hot AFA (ethanol, formalin, and acetic acid). Fixed smears were stained in Harris' hematoxylin and eosin-xytol, dehydrated in an ethanol series, cleared in xylene, and mounted in Damar balsam.

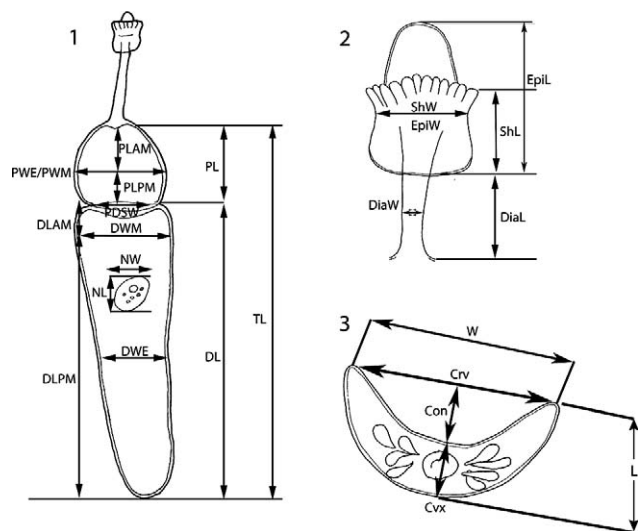
Observations of all ontogenetic stages were made using an Olympus B-Max 52 compound microscope (Olympus America, Inc., Melville, New York) with ×10, ×40, ×60, and ×100 universal and planapochromatic objectives with either phase contrast condensers or differential interference contrast prisms. Digital photographs were taken with an Olympus DP-72 digital camera through the aforementioned microscope. Morphometrics of all ontogenetic stages were taken from the digitized images using cellSens Standard (Olympus Corporation).

Terminology for developmental stages generally follows that of Levine (1971) with modifications suggested by Clopton et al. (1993) and Hays et al. (2007). Terminology for shapes of planes and solids follows Clopton (2004b) and Hays et al. (2007). The metric characters used herein are

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FIGURES 1–3. Morphometric character set for trophozoites and oocysts of *Steganorhynchus* species mapped onto *Steganorhynchus dunwoodyi*. (1) Trophozoite characters mapped onto a mature trophozoite. (2) Epimerite characters. (3) Oocyst characters. (Con, oocyst concavity; Crv, oocyst curvature; Cvx, oocyst convexity; DiaL, diameter length; DiaW, diameter width; DL, deutomerite length; DWE, deutomerite width equatorial; DWM, deutomerite width maximum; DLAM, deutomerite length anterior maximum; DLPM, deutomerite length posterior maximum; EpiL, epimerite length; EpiW, epimerite width; L, oocyst length; NDS, nuclear/anterior length to septum; NL, nuclear length; NW, nuclear width; PDSW, width of protomerite-deutomerite septum; PL, protomerite length; PWE, protomerite width equatorial; PWM, protomerite width maximum; PLAM, protomerite length anterior maximum; PLPM, protomerite length posterior maximum; ShL, width of sheath surrounding epimerite; ShW, width of sheath surrounding epimerite; TL, total length; W, oocyst width).

consistent with those outlined and illustrated for the Actinocephalidae (Clopton, 2004a; Clopton et al., 2007; Cook et al., 2011), and are as follows: diameter length (DiaL), diameter width (DiaW), deutomerite length (DL), deutomerite width equatorial (DWE), deutomerite width maximum (DWM), deutomerite width maximum anterior length (DLAM), deutomerite width maximum posterior length (DLPM), epimerite length (EpiL), epimerite width (EpiW), nuclear/anterior length to septum (NDS), nuclear length (NL), nuclear width (NW), oocyst length (OL), width of oocyst at equator (OW), width of oocyst at polar truncation (OWP), oocyst residuum diameter (OrD), oocyst residuum eccentricity or distance from oocyst residuum to oocyst pole (OrPD), width of protomerite-deutomerite septum (PDSW), protomerite length (PL), protomerite width equatorial (PWE), protomerite width maximum (PWM), protomerite with maximum anterior length (PLAM), protomerite width maximum posterior length (PLPM), total length (TL). Metric characters used herein for the oocysts of *Calyxoccephalus* are consistent with those outlined for the Acanthosporinae (Clopton, 2004a) and are as follows: length of dorsal or polar oocyst spine (DSL), length of oocyst equatorial spine (ESL), distance from oocyst equator to oocyst pole (OLA), oocyst length (OL), width of oocyst at equator (OW), width of oocyst at polar truncation (OWP), oocyst residuum diameter (OrD), oocyst residuum eccentricity or distance from oocyst residuum to oocyst pole (OrPD). Additional metric characters used herein for the epimerite and oocysts of *Steganorhynchus*, delineated in Figures 1–3, are as follows: length of sheath surrounding epimerite (ShL), width of sheath surrounding epimerite (ShW), oocyst length measured as distance along greatest absolute vertical axis (L), oocyst width measured as distance along greatest absolute horizontal axis (W), oocyst curvature measured as distance from “tip-to-tip” along basal horizontal axis (Crv), oocyst convexity measured as distance from interior concave margin to basal horizontal axis along the vertical axis of symmetry (Cvx), and oocyst

concavity measured as distance from interior concave margin to basal horizontal axis along the vertical axis of symmetry (Con). Measurements are presented in μm as mean values followed parenthetically by range values, standard deviations, and sample sizes.

RESULTS

Three species of *Ischnura* were collected; *Ischnura ramburii*, *Ischnura posita*, and *Ischnura hastata*, collectively hosting 8 species of actinocephalid gregarines (Table I). *Steganorhynchus dunwoodyi*, originally described from *Ischnura verticalis*, was collected from several locations in Texas from all 3 species of *Ischnura*, and a new, complete morphological data set is reported from *I. hastata* from Raspberry Pond, Walker County, in the redescription that follows. A new genus of Acanthosporinae was collected from 3 populations of *I. ramburii* and 1 population of *I. hastata*. Metric data and relative morphometrics and ratios are presented in the description that follows. All collections of *Calyxoccephalus karyopera*, *Domadracunculus janovyi*, *Nubenocephalus secundus*, and *Steganorhynchus dunwoodyi* represent new host and locality records for these species. We were not able to obtain oocysts of *Hoplorhynchus* sp. collected from *I. ramburii* at 3 locations, but it is clearly not *Hoplorhynchus acanthatholius*, Percival, Clopton, and Janovy, 1995, the only other species of *Hoplorhynchus* known from North American odonates. Specimens collected in the present study have an epimerite bearing 16 equidistant retroarcuate hooks, while *H. acanthatholius* possesses an epimerite bearing 8 equidistant retroarcuate hooks.

We examined the host voucher specimen from the original species description of *D. janovyi* deposited in the Entomology Museum, Department of Entomology, Texas A&M University, College Station, Texas (accession number TAMU 611), and determined that it is in fact *I. ramburii*. Thus, the type host for *D. janovyi* is amended to *Ischnura ramburii*. Voucher specimens of all host species are deposited in the Sam Houston State University Entomology Collection (SHSUE), Department of Biological Sciences, Sam Houston State University, Huntsville, Texas, as follows: *I. hastata* (SHSUE 001,173, authors ID AJS12292), *I. posita* (SHSUE 001,309, authors ID TJC12535), and *I. ramburii* (SHSUE 000,668).

DESCRIPTION

Septemlaterospora n. gen.

Diagnosis: Eugregarinorida Leger, 1892, sensu Clopton (2000); Septatorina Lankester, 1885, sensu Clopton (2000); Stenophoricae Levine, 1984, sensu Clopton (2000); Acanthosporinae Leger, 1892, sensu Clopton (2000); with the characters of the genus *Septemlaterospora* n. gen.: holdfast compound, comprising a terminal epimerite and intercalating diamerite, epimerite a thick disk or linearly crateriform sucker, diamerite short (less than half of the total holdfast length) and very broadly obdeltoid; association late, cephalolateral, biassociative; gametocysts spherical, hyaline coat present, conspicuous sporoducts absent, dehiscing by simple rupture to release individual oocysts en masse; oocysts terminally truncated heptagonal bipyramids bearing 7 equatorial spines, 1 at each equatorial vertex, 7 terminal spines obliquely inserted at each pole, 1 at each vertex created by polar truncation; 21 spines total.

Taxonomic summary

Type species: *Septemlaterospora rasberryi* n. sp.

Etymology: The generic name *Septemlaterospora* is taken from the Latin roots septem meaning seven and laterum meaning side. The name is given to reflect the heptagonal structure of the oocysts, which is unique among acanthosporine genera.

TABLE I. Collecting sites of *Ischnura hastata*, *Ischnura posita*, and *Ischnura ramburii* indicating species of gregarines collected from each host species.

Location/coordinates	Host species	Host stadium	Gregarine species
Polk County, Collins Pond, 30.410°N, 94.128°W	<i>I. hastata</i>	Adult	<i>Steganorhynchus dunwoodyi</i>
		Naiad	<i>St. dunwoodyi</i>
	<i>I. posita</i>	Naiad	<i>St. dunwoodyi</i>
Polk County, Beaver Slide Pond, 30.385°N, 96.176°W	<i>I. ramburii</i>	Adult	<i>Hoplorhynchus</i> sp.
	<i>I. hastata</i>	Naiad	<i>St. dunwoodyi</i>
	<i>I. posita</i>	Naiad	<i>Domadracunculus janovyi</i>
	<i>I. ramburii</i>	Naiad	<i>St. dunwoodyi</i>
Polk County, city of Livingston Pond, 30.712°N, 94.951°W	<i>I. posita</i>	Adult	<i>St. dunwoodyi</i>
	<i>I. ramburii</i>	Adult	<i>Calyxocephalus karyopera</i>
	<i>I. hastata</i>	Naiad	<i>St. dunwoodyi</i>
Walker County, Center for Biological Field Studies, 30.746°N, 95.474°W		Adult	<i>St. dunwoodyi</i>
	<i>I. posita</i>	Naiad	<i>St. dunwoodyi</i>
		Adult	<i>St. dunwoodyi</i>
	<i>I. ramburii</i>	Naiad	<i>St. dunwoodyi</i>
			<i>Hoplorhynchus</i> sp.
Walker County, Parker Pond, 30.699°N, 95.527°W	<i>I. posita</i>	Adult	<i>St. dunwoodyi</i>
Walker County, Raspberry Pond, 30.692°N, 95.435°W	<i>I. hastata</i>	Adult	<i>Nubenocephalus</i> sp.
			<i>St. dunwoodyi</i>
	<i>I. posita</i>	Adult	<i>St. dunwoodyi</i>
	<i>I. ramburii</i>	Adult	<i>C. karyopera</i>
			<i>Septemlaterospora raspberryi</i> n gen. n. sp.
Walker County, Lone Star Pond, 30.715°N, 95.517°W	<i>I. hastata</i>	Adult	<i>Nubenocephalus secundus</i>
	<i>I. ramburii</i>	Adult	<i>St. dunwoodyi</i>
	<i>I. posita</i>	Adult	<i>Se. raspberryi</i>
Tyler County, Steinhagen Lake, 30.853°N, 94.174°W			<i>St. dunwoodyi</i>
	<i>I. ramburii</i>	Adult	<i>Nubenocephalus</i> sp.
			<i>Se. raspberryi</i>
San Jacinto County, Double Lake, 30.550°N, 95.133°W	<i>I. hastata</i>	Adult	<i>Se. raspberryi</i>
			<i>St. dunwoodyi</i>
	<i>I. ramburii</i>	Adult	<i>C. karyopera</i>
			<i>Se. raspberryi</i>
Madison County, Oil Field Pond, 31.641°N, 96.328°W	<i>I. hastata</i>	Adult	<i>N. secundus</i>
Willacy County, Wildlife Viewing Area, 26.482°N, 97.748°W	<i>I. ramburii</i>	Adult	<i>Hoplorhynchus</i> sp.
Jackson County, Lake Texana, 29.019°N, 96.563°W	<i>I. ramburii</i>	Adult	<i>N. secundus</i>
Taylor County, Herron Pond, 32.134°N, 99.708°W	<i>I. posita</i>	Adult	<i>S. raspberryi</i>
			<i>D. janovyi</i>
			<i>Nubenocephalus</i> sp.

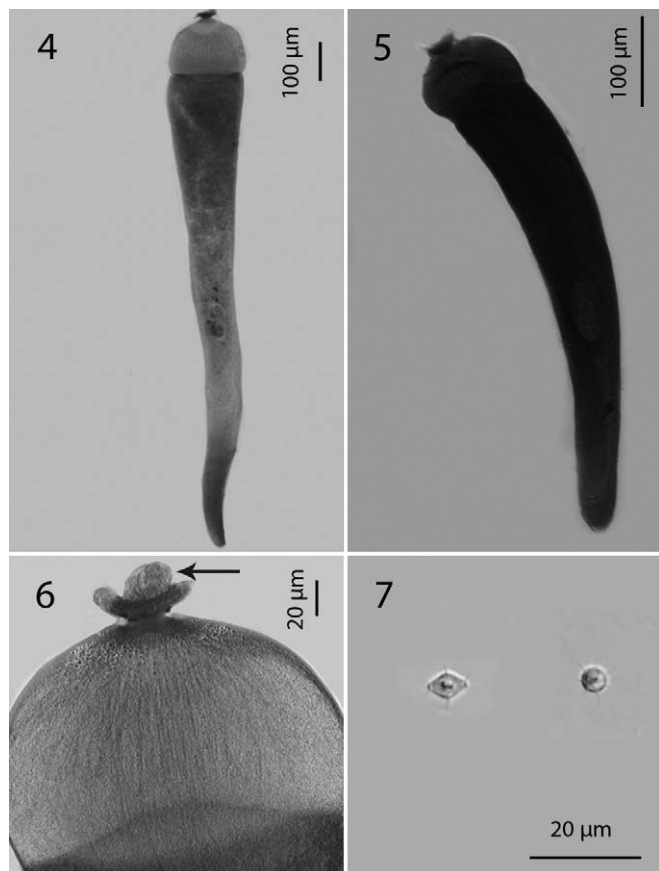
Remarks

Septemlaterospora n. gen. is clearly referable to the Actinocephalidae based on gamontic association, the timing of syzygy, and the mode of gametocyst dehiscence and Acanthosporinae based on oocyst characters, but it is clearly distinguished from known acanthosporine genera by oocyst shape and number of spines, and by epimeritic structure. Of the 10 acanthosporine genera previously described infecting odonates (Léger, 1892; Ellis, 1914; Obata, 1953; Sarkar, 1981; Sarkar and Haldar, 1981; Kori and Amoji, 1985; Clopton et al., 1993; Sarkar, 1995; Clopton, 2004a), 4 have polyhedral oocysts in the form of a bipyramid. The intercalation of rectangular faces between the opposing bipyramid poles of *Septemlaterospora* distinguishes the new genus from *Prismatospora* Ellis, 1914. The heptagonal bipyramid shape of the oocysts of *Septemlaterospora* distinguishes it from *Munkundaella* Sarkar, 1981, *Nubenocephalus* Clopton, Percival, and Janovy, 1993, and *Calyxocephalus* Clopton, 2004, all of which have oocysts that are hexagonal bipyramids. Although the new genus is distinguished upon oocyst morphology alone, the epimerite structure of the new genus is unknown among the 4 described acanthosporine genera possessing polyhedral oocysts.

Septemlaterospora raspberryi n. sp.

(Figs. 4–7)

Mature trophozoite (Figs. 4–6): Solitary, attached to host intestinal epithelium with linearly crateriform epimerite; epimerite (Fig. 6) a linearly crateriform sucker; diamerite short, less than half total holdfast length; total epimerite length (TEL) 21.6 (11.3–39.9 ± 6.2, 56); EpiL 14.7 (7.1–31.0 ± 4.5, 56); EpiW 35.0 (20.8–62.5 ± 8.7, 56); DiaL 7.1 (2.5–13.1 ± 3.1, 56); DiaW 16.6 (8.0–39.0 ± 6.0, 56). Protomerite very shallowly oblong, PL 83.1 (41.3–147.7 ± 25.5, 56); PWE 142.9 (37.1–266.2 ± 53.1, 56); PWM 144.3 (45.0–266.2 ± 52.7, 56); PLAM 47.1 (19.9–92.6 ± 16.0, 56); PLPM 35.5 (0.1–74.6 ± 17.3, 56); PDSW 128.6 (44.8–248.1 ± 49.2, 56); PL/PWE 0.6 (0.3–1.8 ± 0.3, 56); PL/PWM 0.6 (0.4–1.5 ± 0.2, 56); PL/PDSW 0.7 (0.4–1.5 ± 0.2, 56); PLAM/PL 0.6 (0.4–1.0 ± 0.1, 56); PLAM/PLPM 1.7 (0.7–10.0 ± 1.6, 56); PWM/PWE 1.0 (1.0–1.2 ± 0.04, 56); PWM/PDSW 1.1 (1.0–1.4 ± 0.1, 56). Deutomerite deeply obtrullate, DL 741.1 (367.5–1213.5 ± 223.3, 56); DWE 77.3 (31.7–220.2 ± 35.6, 56); DWM 141.9 (55.5–308.8 ± 57.8, 56); DLAM 30.5 (5.6–137.5 ± 23.1, 56); DLPM 713.0 (302.6–1161.3 ± 212.7, 56); DL/DWE 10.7 (2.7–21.9 ± 3.9, 56); DL/DWM 5.7 (1.9–10.6 ± 1.7, 56); DL/PDSW 6.3 (2.4–13.5 ± 2.1, 56); DWM/DWE 1.9 (1.1–3.1 ± 0.4, 56); DWM/PDSW 1.1 (0.9–1.6 ± 0.1, 56). Total length (TL) 823.7 (408.8–1,342.8 ± 240.5, 56); TL/PL 10.2



FIGURES 4–7. Mature trophozoites and oocysts of *Septemlaterospora rasberryi* n. gen., n. sp., differential interference microscopy. (4–5) Mature trophozoites. (6) Epimerite/diamerite complex; arrow indicates host tissue still attached to epimerite. (7) Oocysts.

(6.8–19.3 $\pm$ 2.5, 56); DL/PL 9.2(5.8–18.3 $\pm$ 2.5, 56); DWM/PWM 1.0 (0.6–1.3 $\pm$ 0.1, 56); TL/DL 1.1 (1.1–1.2 $\pm$ 0.03, 56). Nucleus ellipsoid, placement slightly variable but usually located in the anterior third of deutomerite; nuclear karyosomes present: NW 31.3 (15.7–51.3 $\pm$ 7.6, 56); NL 55.4 (30.8–94.4 $\pm$ 15.1, 56); NDS 213.6 (4.0–711.5 $\pm$ 201.4, 56); NL/NW 1.8 (1.0–3.6 $\pm$ 0.5, 56); DL/NL 13.8 (7.0–28.3 $\pm$ 4.2, 56); TL/NL 15.3 (8.0–31.2 $\pm$ 4.5, 56); DL/NDS 14.4 (1.2–197.0 $\pm$ 28.8, 56). Relative morphometric ratios: EpiL > DiaL, EpiW > DiaW, EpiL < PL, DiaL < PL, PWM > DWM, PL < PWM.

Gamonts and association: Solitary gamonts rare, isogamontic, similar in morphology and size to mature trophozoites, but lacking holdfast structures; gamonts enter cephalolateral association immediately before syzygy.

Gametocysts: White in color, roughly orbicular, persistent gelatinous epicyst. Gametocysts stored in spring water dehisced by simple rupture in 3–4 days; oocysts released singly.

Oocysts (Fig. 7): Axially symmetric with polar truncations, very uniform in size and shape, heptagonal bipyramidal with slight polar truncations, biconical in lateral view, polyhedral in polar view. OL 7.6 (6.9–7.8 $\pm$ 0.3, 23); OW 5.2 (4.8–5.5 $\pm$ 0.2, 23); OWP 1.5 (1.4–1.6 $\pm$ 0.07, 23). Distance from oocyst equator to oocyst pole (OLA) 3.4 (3.1–3.6 $\pm$ 0.15), bearing 7 equatorial spines, 1 at each equatorial vertex, length of equatorial spine (ESL) 2.4 (2.1–2.8 $\pm$ 0.19); 7 terminal spines obliquely inserted at each pole, 1 at each vertex created by polar truncation, DSL 1.6 (1.5–1.7 $\pm$ 0.07, 23); oocyst residuum present and smooth, OrD 1.8 (1.6–2.1 $\pm$ 0.1, 23); OrPD 2.1 (1.7–2.6 $\pm$ 0.3, 23). Oocysts released from gametocyst by simple rupture as individual oocysts en masse. Indices: OL/OW 1.4 (1.4–1.4 $\pm$ 0.0, 23); OL/OWP 5.4 (5.4–5.4 $\pm$ 0.0, 23); OL/OrPD 3.6 (3.6–3.6 $\pm$ 0.0, 23); OL/DSL 4.6 (4.6–4.6 $\pm$ 0.0, 23); OW/OWP 3.7

(3.7–3.7 $\pm$ 0.0, 23); OL/ESL 4.5 (3.8–4.8); OW/ESL 3.0 (2.8–3.4); OW/DSL 3.2 (3.4–4.8); OL/OrD 4.2 (3.4–4.8); OW/OrD 2.8 (2.4–3.1).

Taxonomic summary

Type host: *Ischnura ramburii* Selys (Odonata: Zygoptera: Coenagrionidae).

Type locality: Steinhagen Lake, Tyler County, Texas (30.853°N, 94.174°W).

Host vouchers: One symbiotype specimen is deposited in the Sam Houston State University Entomology Museum, Sam Houston State University, Huntsville, Texas (accession number SHSUE 000,668).

Other hosts: *Ischnura hastata*.

Other localities: Raspberry Pond, Walker County (30.692°N, 95.435°W); Lone Star Pond, Walker County (30.715°N, 95.517°W); Steinhagen Lake, Tyler County (30.853°N, 94.174°W); and Double Lake, San Jacinto County (30.550°N, 95.133°W).

Site of infection: Trophozoites and gamonts were collected from the entire length of the mesenteron. Gametocysts were collected in host hindgut, rectum, and feces.

Specimens deposited: The type series is deposited in the Harold W. Manter Laboratory for Parasitology (HWML), Division of Parasitology, University of Nebraska State Museum, Lincoln, Nebraska. The holotype is a hapantotype slide HWML 100,055 (authors' slide AJS12803A). The paratype series consists of 2 additional slides containing trophozoites and gamonts deposited in 2 lots as follows: HWML 100,056 (authors' slide AJS12803B) and HWML 100,057 (authors' slide AJS12804). Seven additional voucher specimens containing trophozoites and gamonts are deposited in 2 lots as follows: HWML 100,058–HWML 100,059 (authors' slides AJS11283 and AJS11231b) and HWML 100,060–HWML 100,064 (authors' slides AJS12166, TJC11106d, TJC11085d, TJC11106b, and TJC11076b). Voucher specimens retained by the authors are deposited in the Sam Houston State University Parasite Museum (SHSUP), Sam Houston State University, Huntsville, Texas. The voucher series consists of 35 slides containing trophozoites and gamonts deposited in 1 lot (SHSUP 000,001–000,035) as follows: AJS11325, AJS11328b, AJS11258, AJS11194, AJS11299, AJS11291, AJS11255, AJS11250, AJS11350a, AJS11350b, AJS11355a, AJS11355b, AJS11376, AJS112647, AJS11229, AJS11230a, AJS11231a, AJS112172, TJC11107, TJC11074a, TJC111073, TJC11082b, TJC11084b, TJC11085a, TJC11085b, TJC11085c, TJC11085d, TJC11089b, TJC11093b, TJC11093c, TJC11095, AJS112973, AJS112223, AJS11214, AJS11259.

Etymology: The specific epithet is an honorific and a sign of our gratitude to Tom and Linda Rasberry who graciously gave the authors permission to collect on their property and who have been continuously supportive of our research efforts, particularly those involving student training.

Remarks

This work establishes *Septemlaterospora* within the Acanthosporinae with the description of *Septemlaterospora rasberryi*.

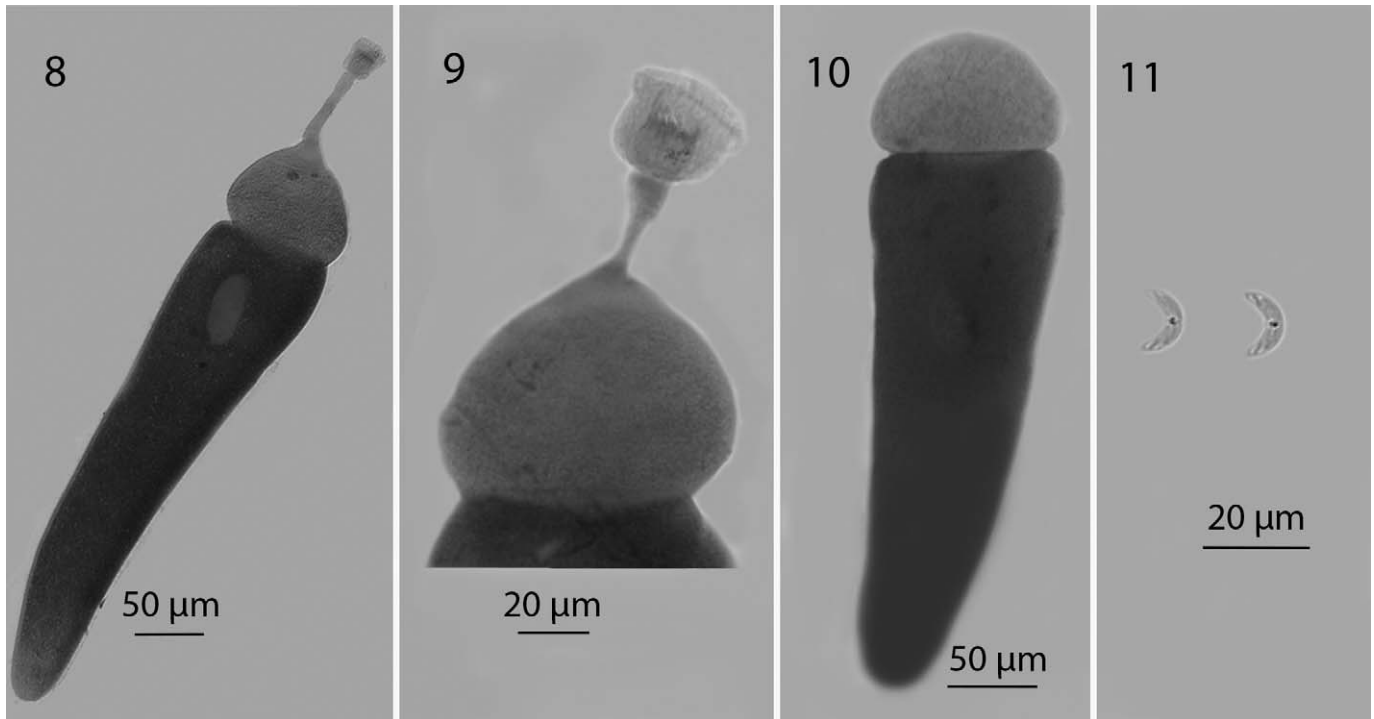
REDESCRIPTION

Steganorhynchus Percival, Clopton, and Janovy, 1995 Revised

Revised diagnosis: Eugregarinorida Leger, 1892, sensu Clopton (2002); Septatorina Lankester, 1885, sensu Clopton (2002); Actinocephalidae Leger, 1892, sensu Clopton (2002); Menosporinae Leger, 1892 sensu strictu; with characters of the genus *Steganorhynchus* Percival, Clopton, and Janovy, 1995, revised as follows: holdfast compound, comprising a terminal epimerite and intercalating diamerite, epimerite an ovoidal papilla enclosed in a very broadly panduriform retractable sheath, diamerite long (greater than half of the total holdfast length); oocysts axially asymmetric, shallowly luniform to luniform, without spines or thickenings at their poles, compressed in the plane perpendicular to the major axis; development solitary; association late, cephalolateral.

Steganorhynchus dunwoodyi Percival, Clopton, and Janovy 1995 (Figs. 4–7)

Mature trophozoite (Figs. 8–11): Solitary, attached to host intestinal epithelium with ovoidal epimerite enclosed in very broadly panduriform retractable sheath, anterior margin of sheath pleated; diamerite long,



FIGURES 8–11. Ontogenetic stages of *Steganorhynchus dunwoodyi*, differential interference microscopy. (8) Mature trophozoites. (9) Epimerite/diamerite complex. (10) Gamont. (11) Oocysts.

greater than half the total holdfast length; total epimerite length (TEL) variable (probably due to mechanical stretching during permanent slide preparation) 223.1 (92.5–384.4 $\pm$ 96.0, 11), EpiL 21.5 (18.5–33.4 $\pm$ 4.1, 11); EpiW 15.8 (12.0–20.3 $\pm$ 2.6, 11); DiaL 201.69 (70.5–365.9 $\pm$ 97.7, 11); DiaW 27.3 (14.8–37.5 $\pm$ 6.7, 11). Protomerite broadly to very broadly ovoid, PL 74.8 (39.1–93.6 $\pm$ 15.6, 27); PWE 64.3 (28.8–143.2 $\pm$ 24.4, 27); PWM 75.7 (31.3–145.8 $\pm$ 25.2, 27); PLAM 53.9 (36.9–77.9 $\pm$ 12.1, 27); PLPM 20.9 (10.4–52.0 $\pm$ 9.4, 27); PDSW 59.9 (22.1–144.7 $\pm$ 23.5, 27); PL/PWE 1.2 (0.6–2.2 $\pm$ 0.3, 27); PL/PWM 1.0 (0.6–1.5 $\pm$ 0.2, 27); PL/PDSW 1.3 (0.6–1.9 $\pm$ 0.3, 27); PLAM/PL 0.7 (0.5–0.8 $\pm$ 0.07, 27); PLAM/PLPM 2.8 (1.2–5.1 $\pm$ 1.0, 27); PWM/PWE 1.1 (1.0–1.4 $\pm$ 0.1, 27); PWM/PDWS 1.2 (1.0–1.8 $\pm$ 0.1, 27). Deutomerite narrowly obdeltoid, DL 324.7 (113.4–497.0 $\pm$ 98.3, 27); DWE 67.5 (29.4–106.3 $\pm$ 21.5, 27); DWM 84.1 (34.2–158.3 $\pm$ 30.2, 27); DLAM 31.0 (10.9–87.7 $\pm$ 20.1, 27); DLPM 292.4 (102.7–461.7 $\pm$ 90.1, 27); DL/DWE 4.9 (3.1–7.4 $\pm$ 1.2, 27); DL/DWM 3.9 (2.8–6.6 $\pm$ 0.9, 27); DL/PDSW 5.6 (3.1–8.5 $\pm$ 1.2, 27); DWM/DWE 1.2 (1.0–2.5 $\pm$ 0.2, 27); DWM/PDSW 1.4 (1.0–1.8 $\pm$ 0.1, 27). Total length (TL) 398.5 (152.6–618.2 $\pm$ 111.0, 27); TL/PL5.2 (3.8–7.5 $\pm$ 0.09, 27); PL/DL0.2 (0.1–0.3 $\pm$ 0.05, 27); DWM/PWM 1.1 (0.7–1.3 $\pm$ 0.1, 27); TL/DL 1.2 (1.1–1.3 $\pm$ 0.05, 27). Nucleus elliptoid, placement slightly variable but usually located near the center of the deutomerite; nuclear karyosomes apparent: NW 26.6 (19.3–35.6 $\pm$ 4.8, 27); NL 42.1 (22.5–65.3 $\pm$ 11.1, 27); NDS 93.4 (6.1–256.0 $\pm$ 65.4, 27); NL/NW 1.5 (1.0–2.4 $\pm$ 0.3, 27); TL/NL 9.5 (6.5–16.2 $\pm$ 2.0, 27); DL/NDS 7.9 (1.6–72.8 $\pm$ 14.1, 27). Relative morphometric ratios: EpiL < DiaL, EpiW > DiaW, EpiL > PL, DiaL < PL, PWM > DWM, PL > PWM.

Gamonts and association (Fig. 10): Solitary gamonts abundant, isogamontic, similar in morphology and size to mature trophozoites, but lacking holdfast structures; gamonts enter cephalolateral association immediately before syzygy.

Gametocysts: White in color, roughly orbicular, persistent gelatinous epicyst. Gametocysts stored in spring water dehisced by simple rupture in 3–4 days; oocysts released singly.

Oocysts (Fig. 11): Axially asymmetric, shallowly luniform to luniform, compressed in the plane perpendicular to the major axis, smooth (without spines), uniform in size and shape, only slightly variable in size; L 7.6 (7.3–8.1 $\pm$ 0.2, 14); Cvx 3.9 (3.6–4.1 $\pm$ 0.2, 14); W 11.0 (10.1–11.8 $\pm$ 0.5, 14); Crv 9.9 (9.2–10.3 $\pm$ 0.3, 14); Con 3.6 (3.4–3.9 $\pm$ 0.2, 14).

Taxonomic summary

Type host: *Ischnura verticalis* (Say, 1839).

Type locality: Oak Lake, Lancaster County, Nebraska, U.S.A.

Other hosts: *Ischnura hastata*, *Ischnura posita*, and *Ischnura ramburii*.

Other localities: Collins Pond, Polk County (30.410°N, 94.128°W); Beaver Slide Pond, Polk County (30.385°N, 96.176°W); city of Livingston Pond, Polk County (30.712°N, 94.951°W); Center for Biological Field Studies, Sam Houston State University, Walker County (30.746°N, 95.474°W); Parker Pond, Walker County (30.699°N, 95.527°W); Raspberry Pond, Walker County (30.692°N, 95.435°W); Lone Star Pond, Walker County (30.715°N, 95.517°W); Steinhagen Lake, Tyler County (30.853°N, 94.174°W); Double Lake, San Jacinto County (30.550°N, 95.133°W).

Site of infection: Trophozoites and gamonts were collected from the entire length of the mesenteron. Gametocysts were collected in host hindgut, rectum, and feces.

Specimens deposited: The holotype (one slide USNM No. 82821) is deposited in the United States National Museum (USNM) Helminthological Collections, Beltsville, Maryland. A voucher series is deposited in the Harold W. Manter Laboratory for Parasitology (HWML), Division of Parasitology, University of Nebraska State Museum, Lincoln, Nebraska. The voucher series comprises 10 slides containing trophozoites and gamonts deposited in 2 lots as follows: HWML 100,050–HWML 100,054 (authors' slides, AJS11199, AJS12661, AJS12662, TJC11008, and TJC11047) and HWML 100,065–HWML 100,069 (authors' slides, AJS11134, TJC11005, AJS11203, TJC11036, and TJC11043B). Additional voucher specimens retained by the authors are deposited in the Sam Houston State University Parasite Museum (SHSUP), Sam Houston State University, Huntsville, Texas. The voucher series consists of 78 slides containing trophozoites and gamonts deposited in 1 lot (SHSUP 000,036–114) as follows: AJSH12613, TJC12265, TJC12277a, TJC12277b, TJC12279a, AJSH12621, TJC12299b, TJC12283b, TJC12283a, TJC12280, AJSH12622, AJS12616, AJSH12612, AJS12668, TJC12321a, TJC11024, AJSH12678, AJSH12800, AJSH12737, AJSH12796, AJSH12780, AJSH12777, AJSH12772, AJSH12736, AJSH12533, AJSH12526, AJSH12582, AJSH12413, AJSH12746, AJSH12751, AJSH12755, AJS10315, AJS10328, AJS10329, AJS10351, AJS10355, AJS10357, AJS10411, AJS10433, AJS10441, AJS10452, AJS10470, AJS10640, AJS10646, AJS10647, AJS10652, AJS10653, SIS10170,

TABLE II. Gregarine species known from zygopteran hosts in North America, north of Mexico.

Gregarine species	Type host	Other known hosts	References
<i>Actinocephalus carrilynnae</i>	<i>Enallagma civile</i>	<i>Ischnura verticalis</i>	Richardson and Janovy (1990), Percival-Cook et al. (2001)
<i>Calyxocephalus karyopera</i>	<i>Calopteryx maculata</i>	<i>Ischnura ramburii</i>	Clopton (2004a), this study
<i>Septemlaterospora rasberryi</i>	<i>Ischnura ramburii</i>	<i>Ischnura hastata</i>	This study
<i>Domadracunculus janovyi</i>	<i>Ischnura ramburii</i> *	<i>Ischnura posita</i>	Clopton (1995), this study
<i>Hoplorhynchus acanthatholius</i>	<i>Enallagma civile</i>	None	Percival et al. (1995), Percival-Cook et al. (2001)
<i>Nubenocephalus nebraskensis</i>	<i>Argia bipunctulata</i>	<i>Enallagma civile</i> <i>Ischnura verticalis</i> <i>Argia translata</i> <i>Argia tibialis</i>	Clopton et al. (1993), Percival-Cook et al. (2001), Cielocha et al. (2011)
<i>Nubenocephalus secundus</i>	<i>Argia sedula</i>	<i>Argia immunda</i> <i>Argia moesta</i> <i>Argia apicalis</i> <i>Argia vivida</i> <i>Argia translata</i> <i>Argia tibialis</i> <i>Argia fumipennis</i> <i>Ischnura hastata</i> <i>Ischnura ramburii</i>	Hays et al. (2007), Cielocha et al. (2011), this study
<i>Steganorhynchus dunwoodyi</i>	<i>Ischnura verticalis</i>	<i>Enallagma civile</i> <i>Ischnura posita</i> <i>Ischnura ramburii</i> <i>Ischnura hastata</i>	Percival et al. (1995), Percival-Cook et al. (2001), this study

* Originally reported as *Ischnura verticalis* (Clopton, 1995), now confirmed as *I. ramburii*.

SIS10182, AJS10609, AJS10350, AJS10608, AJS10340, AJS10628, SLS10187, SLS10188, SLS10192, SLS10198, SLS10199, SLS10202, SLS10215, AJS11364, AJS11365a, AJS11365b, TJC12248a, TJC12248b, TJC12252, JJC12254, AJS11186, AJS11191, AJS11193, AJS11312, AJS11334, AJS12388, AJS12387, AJS11210, AJS11143, AJS11158.

Remarks

This is the first report of *S. dunwoodyi* since its original description, and *I. hastata*, *I. posita*, and *I. ramburii* are new host records for this gregarine species. All Texas collection sites are new locality records for this species. The original description was based on live trophozoites observed and measured as fresh preparations in water. Although no more than 10 individual gregarines were measured from each host, data from live specimens are highly variable due to heat and osmotic stress and myoneme constriction (Clopton, 2012). This redescription, based on uniformly prepared, permanent specimens, stabilizes the taxon by providing a complete new taxonomic data set that utilizes a larger set of metric characters consistent with those previously outlined for the Actinocephalidae (Clopton, 2004a; Clopton et al., 2007; Cook et al., 2011).

DISCUSSION

This study expands the known host and geographic ranges of *C. karyopera*, *D. janovyi*, *N. secundus*, and *St. dunwoodyi*, increases the taxonomic breadth of damselfly hosts examined for gregarine parasites, and contributes to our long-term goal of comprehensive discovery and systematic revision of the Nearctic Eugregarinida with the description of *Septemlaterospora rasberryi* n. gen. n. sp.

Our results are consistent with the notion that gregarines of North American Zygoptera utilize hosts based on stable transmission dynamics and host physiological homogeneity rather than as a function of phylogenetic assemblage (Cielocha

et al., 2011). *Nubenocephalus secundus* parasitizes at least 9 species in 2 genera of coenagrionid damselflies, *N. nebraskensis* parasitizes at least 5 species in 3 genera of coenagrionids, *St. dunwoodyi* parasitizes at least 5 species in 2 genera of coenagrionids, *Se. rasberryi* parasitizes at least 2 species in 1 genus of coenagrionids, *D. janovyi* parasitizes at least 2 species in 1 genus of coenagrionids, *A. carrilynnae* parasitizes at least 2 species in 2 genera of coenagrionids, and *C. karyopera* parasitizes at least 2 species in 2 families of Zygoptera (Table II). Furthermore, 4 of these gregarine species, *N. secundus*, *A. carrilynnae*, *St. dunwoodyi*, and *D. janovyi*, are known to parasitize both adult and larval damselflies. This suggests that actinocephalid gregarines parasitizing damselflies are tracking a resource that is widespread and phylogenetically conserved across host life-cycle stages, genera (*Argia*, *Enallagma*, and *Ischnura* are in the family Coenagrionidae), and, in some cases, across host families (*Calopteryx* is in the family Calypterigidae). Given that all gregarine/odonate studies to date have examined only a small number of the 134 species in 22 genera across 5 families of Zygoptera in the United States, this observed pattern can only be supported or refuted by more data from a larger number of potential host species across a broader geographic range.

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this material are those of the authors(s) and do not necessarily reflect the views of the Big Thicket Foundation.

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