DESCRIPTIONS OF FOUR NEARCTIC *PROCLADIUS* SKUSE PUPAL EXUVIAE

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Abstract

Associated *Procladius* Skuse (*Holotanypus* Roback) material from the Sublette collection was found to have specimens with undescribed immature stages. Pupal exuviae of two species are described for the first time: *P. barbatulus* Sublette and *P. clavus* Roback. Adult and larval features are included for *P. barbatulus*. In addition, specimens of Nearctic *P. denticulatus* Sublette are compared to Palearctic *P. signatus* (Zetterstedt) to determine if exuviae features can separate these species or suggest that *P. signatus* is a senior synonym. Finally, associated pupal and larval traits of *P. prolongatus* Roback are compared to the original description, corroborating the theory that features are unique enough to warrant subgeneric status.

Introduction

Within Procladius Skuse, Holotanypus Roback is a diverse subgenus in the Nearctic region with 18 described species (Ashe and O'Connor 2009), of which six have not previously been described in the pupal stage. While comparing recently collected material to the Sublette collection at the University of Minnesota, a number of associated specimens were noted within Procladius, including two species (P. barbatulus Sublette and P. clavus Roback) with undescribed pupal exuviae. Along with these, we also compare an associated specimen of the Nearctic P. denticulatus Sublette to the morphologically similar Palearctic P. signatus (Zetterstedt) to determine if the pupal stage can be used to better distinguish these species. Finally, we used eight associated rearings from Alaska to reevaluate if P. prolongatus Roback is unique enough to have a separate subgenus designation (Roback 1980, 1982).

Materials and Methods

All specimens in the University of Minnesota, St. Paul (UMSP) collection were originally identified by J. E. Sublette and adult features were reconfirmed for this study using Roback (1971). *P*.

prolongatus reared associations were provided by Malcom Butler and will be added to the UMSP collection. Measurements and terminology follow Sæther (1980). Additional specimens of *P. denticulatus* were identified by W. P. Coffman and D. Oliver, while additional specimens of *P. signatus* were identified by P. H. Langton.

Results

Procladius (Holotanypus) barbatulus Sublette

Material examined. USA: California, 1.7 miles east of Benton Hot Springs, 21-VIII-67, col. Jon Martin.

Adult male (n = 1). Total length approximately 3.7 mm.

Head. AR 1.80. Temporal setae 23-24. Clypeus with 15 setae. Palpomere lengths (μ m): 50, 70, 105, 148, 238.

Thorax. Antepronotals 8-9. Dorsocentrals 19. Prealars 16-17. Acrostichals 40. Prescutellars 7. Supraalar 1. Scutellars 22.

Wing. VR 1.46. Brachiolum with 3 setae; R with 14-17 setae, R_1 with 13 setae, R_{4+5} with 19-23 setae. Squama with 32-36 setae. Extended part of costa 100 µm long. Wing length 2.1 mm. Total length/wing length 1.76. Dark spot over r-m extending to part of m-cu.

Legs. Spur of front tibia 55 μ m. Spurs of middle tibia 38-40 μ m and 48-50 μ m. Spurs of hind tibia 38 μ m and 73 μ m. Width at apex of front tibia 58-60 μ m; middle tibia 53-58 μ m; hind tibia 70-73 μ m. Comb with 11 setae; shortest seta 30 μ m long, longest seta 58-63 μ m long. One pseudospur present on ta₁ of all legs and one pseudospur on ta₂ of mid and hind legs. Pseudospurs 30-43 μ m long. Additional measurements and proportions are in Table 1.

Hypopygium. Tergite IX with 21 setae. Transverse sternapodeme 195 μ m. Phallapodeme 75 μ m. Strut ratio 2.84. Gonocoxite 213 μ m long, gonostylus

	fe	ti	ta ₁	ta ₂	ta ₃	ta ₄	ta ₅
p ₁	770, 780	940, 970	690	340	250	170	120
p_2	850	910, 940	580, 590	300	210, 230	140, 150	100
p ₃	770, 780	1050	700, 740	350, 370	250, 260	160, 170	120,130
	LR	BV	SV	BR			
p ₁	0.61	2.73	2.42	2.92	-		
p_2	0.70, 0.71	3.08, 3.12	2.92, 2.95	2.69			
p ₃	0.65, 0.67	2.86, 2.88	2.51, 2.56	2.71, 2.85			

Table 1. Procladius barbatulus, male. Lengths (in µm) and proportions of legs.

100 μm long, gonostylus ratio 10. HR 2.13; HV 3.70.

Pupa (n = 1). Total length 4.80 mm.

Cephalothorax. Faint brown, mostly translucent. Thoracic horn (Fig. 1) brown; 380 μ m long; with triangular teeth, sometimes rounded apically; no reticulation; plastron plate length 75 μ m; horn width, plastron plate width and apical constriction of horn chamber not measurable.

Abdomen. Color pattern 1, entirely translucent. Shagreen (Fig. 2) of T IV medially with small points (1-2 μ m) often arranged in short rows and similar in size to medial points on T VIII. LS taeni-

ae of VII at 0.45, 0.53, 0.80, 0.93 segment length. LS taeniae of VIII at 0.30, 0.48, 0.63, 0.80, 0.93 segment length. Anal lobe length 500 μ m; 35-36 spines on outer margin, often bifid or trifid; largest spines 25-27 μ m long with spine size similar for much of the margin; smaller spines apically with inner 2-3 spines on a protuberance (Fig. 3).

Larva (n = 1)

Head. Head capsule length about 650 μ m. Antenna in Fig. 4. Antennal segment 1-4 lengths (μ m): 155-157, 20, 5-6, 2. AR 3.95-4.05. Basal antennal segment 30-33 μ m wide; 4.8-5.2 times as long as wide; ring organ 0.73 from base; blade 27.5 μ m long. Mandible 135-140 μ m long. Maxilla features



Figures 1-3. 1, *Procladius barbatulus* thoracic horn (Scale = 0.1 mm); 2, tergite IV medial shagreen (Scale = 0.025 mm); 3, tergite VIII and anal lobe (Scale = 0.1 mm).



Figures 4-5. 4, Procladius barbatulus antenna, mandible, and dorsomental teeth; 5, ligula (Scale = 0.025 mm).

not distinguishable. Dorsomental teeth in Fig. 4 and ligula in Fig. 5.

Abdomen. Features not distinguishable.

Remarks. As only the second specimen reported, after the holotype, adult and larval traits were included for comparison to Sublette (1964) and Roback (1971). In Roback (1980) this specimen keys to couplet 18 for *P. freemani*, and in Sæther (2010) to couplet 9 and *P. freemani*. With only a single specimen, it is not certain that any feature can be used to distinguish *P. barbatulus* from *P. freemani* in the pupal stage. Features span all three of Roback's variants (with var. 2 the best fit) and are also similar to specimens described by Sæther. The larva keys to couplet 14 in Roback and appears indistinguishable from other species in this couplet.

Habitat may be the same as most *P. freemani*, which is typically lakes, and the type specimen is from Hume Lake, a reservoir in Sequoia National Forest. Based on slide information, the Benton Hot Springs specimen presented here may be from spring-fed desert streams or related pools.

Known range for *P. barbatulus* is California, USA, in Fresno and Mono Counties.

Procladius (Holotanypus) clavus Roback

Material examined. Canada: British Colombia, Box 4 Lk., Riske Cr. B.C., 1-VI-1970, col. R. Cannings.

Adult males (n = 3).

Hypopygium. Tergite IX with 44-62, 53 setae (n = 3). Transverse sternapodeme 280-307, 296 μ m long (n = 3). Phallapodeme 100-113, 106 μ m long (n = 6). Strut ratio 2.10-2.50, 2.26 (n = 6). Gono-coxite 220-250, 239 μ m long (n = 6). Gonostylus 118-138, 130 μ m long (n = 6). No lobes present

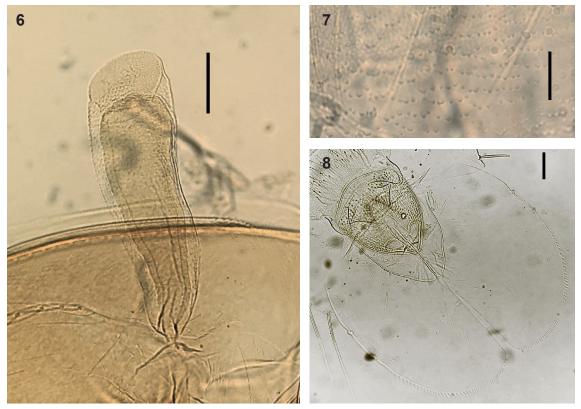
for measuring the gonostylus ratio. HR 1.72-1.92, 1.84 (n = 6).

Pupa (n = 3). Total length 6.37 mm, range 6.1-6.5 mm

Cephalothorax. Faint brown, mostly translucent. Thoracic horn (Fig. 6) 450-520, 483 μ m long (n = 4); 140-150, 145 μ m wide (n = 2); with teeth; no reticulation; plastron plate 70-98, 84 μ m long (n = 4); 106-120, 113 μ m wide (n = 2); apical constriction of horn chamber 70-85, 78 μ m wide (n = 2); length/width of horn 3.27-3.85, 3.56 (n = 2); width of plastron plate/width of horn 0.79-0.80, 0.80 (n = 2); width of plastron plate/width of constriction 1.41-1.51, 1.46 (n = 2).

Abdomen. Color pattern 1, entirely translucent. Shagreen (Fig. 7) of T IV medially with small points (2 μ m) arranged in loosely connected, elongate rows and smaller than median points of T VIII (3-5 μ m). LS taeniae of VII at 0.50, 0.69, 0.86, 0.96 (n = 3) segment length. LS taeniae of VIII at 0.30, 0.45, 0.61, 0.77, 0.93 (n = 3) segment length. Anal lobe length 700-820, 787 μ m; 43-54, 49 (n = 5) spines on outer margin; largest spines (22-30 μ m) posterolateral, usually with gaps narrower than spine base; basal spines broadly triangular; apical spines 10-15 μ m, directed posteriorly, and usually on a small protuberance or clustered together with a shared base (Fig. 8).

Remarks. All specimens were pharate, making adult features difficult to observe so only hypopygium measurements were included. In Roback (1980) these specimens key to couplet 13, in Sæther (2010) to couplet 6, and are similar to *P. culiciformis*. Sæther's measurements of *P. culiciformis* were generally similar to Sublette's Riske Creek specimens, but it is possible that *P. clavus* can be distinguished by the wider apical constriction.



Figures 6-8. 6, *Procladius clavus* thoracic horn (Scale = 0.1 mm); 7, tergite IV median shagreen (Scale = 0.025 mm); 8, segment VIII and anal lobe (Scale = 0.1 mm).

tion of the horn chamber, which also leads to a smaller ratio for width of plastron plate/width of constriction. Sæther describes *P. culiciformis* anal lobe inner margins as having small spines, which are not present in *clavus* specimens. Finally, figures of the thoracic horn in both Sæther and Roback show the *P. culiciformis* apical constriction as having a rather long neck, while in *clavus* the neck is short enough that it was hard to determine the width, and therefore the horn may resemble *P. paragretis* or *P. ruris*.

Known range is British Colombia and Nunavut, Canada.

Procladius (Holotanypus) prolongatus Roback

Material examined. USA: Alaska, Loon Pond, a tundra pond on the west edge of Prudhoe Bay, 3-VII-1980, col. Malcom Butler.

Pupa (n = 8), 4 males, 4 females. Total length 7.65-8.47, 8.02 mm (n = 8)

Cephalothorax. Light brown. Thoracic horn (Fig. 9) 630-760, 682 μ m long (n = 16); 240-300, 265 μ m wide (n = 13); covered evenly with short teeth; no reticulation; plastron plate 140-200, 170 μ m long (n = 16); 252-330, 279 μ m wide (n = 13); apical constriction of horn chamber 110-148, 130 μ m wide (n = 5) with a short neck often obscured

by the large, dark plastron plate; an oval aperture between the neck and atrium is often elongate and distinct; length/width of horn 2.3-2.78, 2.57; width of plastron plate/width of constriction 1.89-2.35, 2.06 (n = 5). Respiratory atrium, neck and plastron plate dark brown. Plastron plate width usually equal to or wider than horn width.

Abdomen. Color pattern 3, but usually a solid light brown with no pale patches. Shagreen (Fig. 10) of T IV medially with elongate (4-7 μ m) single sharp points. Shagreen on T VIII similar in size, shape, and pattern, although less robust than on other tergites. LS taeniae of VII at 0.38, 0.62, 0.81, 0.96 (n = 6) segment length. LS taeniae of VIII at 0.16, 0.35, 0.57, 0.78, 0.95 (n = 6) segment length. Anal lobe (Fig. 11) length 920-1000, 964 µm; 30-48, 40 (n = 16) spines on outer margin; largest spines 40-70 µm posterolateral, with distance between spines equal or wider than width of spine bases; smallest apical and lateral spines 10-20 µm long; posterior edge indented, apical spines clustered on a projection (except one specimen with no projection and no indentation along posterior edge).

Remarks. Roback (1980) described features that suggested subgeneric status for *P. prolongatus*, and he formally implemented this distinction among *Holotanypus* groups in Roback (1982). From associated Prudhoe Bay material, we can

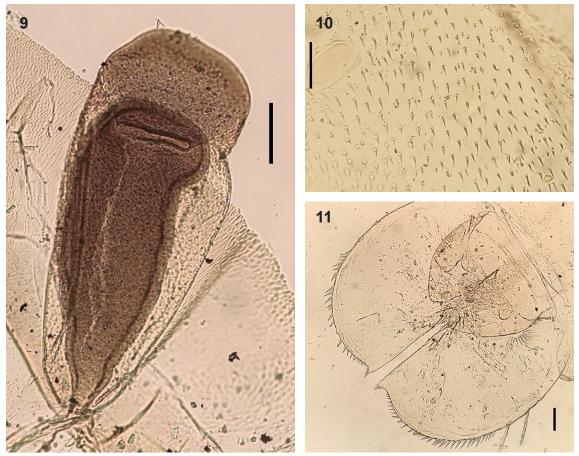


Figure 9-11. 9, *Procladius prolongatus* thoracic horn (Scale = 0.1 mm); 10, tergite IV medial shagreen (Scale = 0.025 mm); 11, anal lobe (Scale = 0.1 mm).

provide insight into the immature traits used to support the subgenus.

The anal lobes from Roback's Barrow, Alaska, specimens were described as "unusually shaped" with a "projecting outer curve". The Prudhoe Bay specimens usually do have a rounded lateral shape, but none are as expansive as in Roback's description and some also have lateral edges that are straight. Based on descriptions in Roback (1980) and material from Isle Royale, Michigan, P. dentus also has a large anal lobe that can be rounded laterally. P. dentus and prolongatus can both have a fold near the respiratory organ neck, but in both species this feature is not always observed. Therefore, these two features are not diagnostic and without a series of specimens may create confusion when using the key in Roback (1980). These species may be reliably separated by tergite IV and VIII shagreen, which is elongate $(4-7 \ \mu m)$ in P. prolongatus and short (1-4 µm) in P. dentus. In addition, the number of spines on the anal lobe will distinguish these species, with 30-48 in P. prolongatus and 56-63 (from Isle Royale) and 68 (from the single specimen reported by Roback) in *P. dentus.*

Larvae have one proleg that is apparently diagnostic in *Procladius*, with a wide, triangular base and a tight apical hook (see figure 244, Roback 1980). Prudhoe Bay tundra pond specimens have this feature. Larvae of *P. dentus*, though undescribed, should presumably have simple proleg claws, similar to all other known larvae in *Holotanypus* (Roback 1982).

Known range is Nunavut, Canada, and Alaska, USA.

Procladius (Holotanypus) denticulatus Sublette

Material examined. USA: New Hampshire, Mirror Lake, 14-24-VI-74, reared male, col. Rhoda Walter.

Pupa (n = 1). Total length 5.45 mm.

Cephalothorax. Thoracic horn 420-430 μ m long; 100-110 μ m wide; with teeth; atrial wall with weak reticulation; plastron plate length 50 μ m, width 90 μ m; apical constriction of horn chamber 62 μ m wide; length/width of horn 3.9-4.2; width of plastron plate/width of horn 0.82; width of plastron plate/width of constriction 1.45 (Fig. 12).

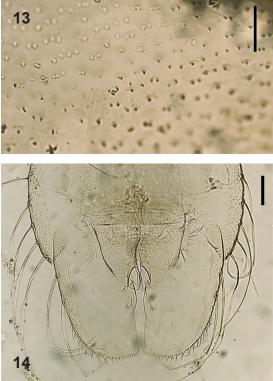
Abdomen. Tergites with color pattern 3, minimal



Figures 12-14. *Procladius denticulatus*. 12, thoracic horn (Scale = 0.1 mm); 13, tergite IV medial shagreen (Scale = 0.025 mm); 14, segment VIII and anal lobe (Scale = 0.1 mm).

contrast. Shagreen of T IV medially with small points (1-2 μ m, Fig. 13) often arranged in short rows and smaller than median points on T VIII (3-4 μ m). LS taeniae of VII at 0.62, 0.81, 0.93, 0.98 segment length. LS taeniae of VIII at 0.30, 0.50, 0.67, 0.83, 0.93 segment length. Anal lobe length 530 μ m; 29 spines on outer margin; largest spines (33-37 μ m) posterolateral with gaps wider than the base of the spines; anterior and apical spines smaller (as small as 5 μ m), more closely set, with one apical spine on a small protuberance apically (Fig. 14).

Remarks. Separating this species from P. signatus, which has been suggested as a possible senior synonym (Roback 1971), is difficult since the two species are exceedingly alike. Including an additional three specimens (collected by W. P. Coffman and D. Oliver) to the one described here, we observe that on the whole *P. denticulatus* is smaller (ThL 400-430 µm), whereas P. signatus is larger (ThL 480-624 µm, mean = 546, n = 13). Roback (1980) gives the range for P. denticulatus ThL as $370-540 \ \mu m \ (mean = 468) \ and \ Sather \ (2010) \ a$ ThL range of 393-577 μ m (mean = 469), so there is overlap. However, the means are well separated so ThL should be a useful feature unless a specimen falls into the intermediate range and geographic provenance is unknown. Size may be useful for total exuviae length as well, with a range of 5.7-8.5 mm (mean = 6.6) for *P. signatus* (Langton 1991).



In addition, points on tergite IV are smaller on *P. denticulatus* (1-2 μ m), which are smaller than on tergite VIII (3-4 μ m), whereas points on the two tergites for *P. signatus* are of similar size (4 μ m, Langton 1991). This feature is independent of exuviae size. A final structural difference that may hold for *P. signatus* is that the strong points of the thoracic horn wall continue over the 'shoulder', where the horn constricts apically, and onto the 'hood', the apical region containing the plastron plate (Fig. 15). In contrast, *P. denticulatus* points on the hood are reduced in size and density.

Procladius denticulatus is widespread in the northern Nearctic.

Discussion

For the two newly described pupal exuviae, *P. barbatulus* does not appear to be distinguishable morphologically from *P. freemani*, but *P. clavus* does have a distinct set of features and appears to be most similar to *P. culiciformis*. In *P. prolongatus* there are distinct features in immature life stages that will separate it from similar species.

Whether *P. denticulatus* and *P. signatus* are at two ends of a cline from the Eastern Palearctic to the Nearctic, or if they are two distinct species that are only slightly diverged from a common ancestor, cannot be fully answered here. But our results do suggest that most specimens can be separated using the thoracic horn length and tergite IV sha-

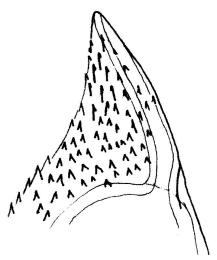


Figure 15. Procladius signatus, apex of thoracic horn.

green. As Roback (1980) points out, some intermediate specimens will complicate identification, particularly when a comparing a "larger series from a broad geographical range."

Two exuvial features may be worth consideration for future *Procladius* revisions. First, variability in the pattern of tergite shagreen shows this is not necessarily a stable feature in some *Procladius* species, yet spinule size and comparison between T IV and T VIII appear useful. Finally, variation in size and distribution of points on the 'shoulder' and 'hood' of the thoracic horn should be investigated in this genus.

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