

## New records of Chironomidae (Diptera) of Tuvalu

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**Abstract** – Here we report the chironomid fauna of Tuvalu for the first time. Four species belonging to two subfamilies were collected on Fongafale Island. At least one species inhabited a freshwater environment. We describe male imagines of *Comptosmittia longipalpis* sp. n., *Pseudosmittia tuvaluensis* sp. n. and *Pseudosmittia amplexivirga* sp. n. We also describe the morphology of *Chironomus* sp. larvae inhabiting a wet taro swamp.

**Key words:** Chironomidae / Diptera / Funafuti Atoll / new species / Tuvalu

### Introduction

To date the chironomid faunas of the Pacific islands have been studied in detail in the Samoan Islands (Edwards, 1928), Marquesas Islands (Edwards, 1933), Hawaiian Islands (Hardy, 1960) and in a broad sense in Micronesia (Tokunaga, 1964), but not yet in the state of Tuvalu. Laird (1956) reported the absence of Chironomidae from aquatic habitats on Funafuti Atoll, Tuvalu. Low-profile Pacific islands such as those of Tuvalu could suffer from inundation caused by rising sea levels. Further, Tuvalu is currently suffering from the effects of inappropriate land development (Yamano *et al.*, 2007). There is therefore an urgent need to study the florae and faunas of Pacific low-profile islands. In 2009, the second and third authors of this study visited Fongafale Island in Tuvalu and collected chironomid specimens. Here we report the morphology and cytochrome *c* oxidase I (*COI*) DNA sequences of the Tuvaluan chironomids for the first time.

### Methods and terminology

The general terminology and abbreviations follows Sæther (1980). The definition of VR2 (venarum ratio 2) of *Pseudosmittia* follows Sæther (2004). Length ratio of palpomeres 5/3 follows Lin *et al.* (2013). All materials were collected by the second author. Adults were collected with a sweep net. Larvae were collected with a D-frame net. Collections were attempted at four sites on Fongafale, the most densely populated island in Tuvalu (Fig. 1). Site 1 is located in a wetland near a transmission tower. Site 2 is a stand of bushes on the west coast, the side facing a lagoon

of the atoll. Site 3 is a mangrove forest on a ridge between a small brackish lake and the ocean. Site 4 is located in a taro (*Colocasia esculenta*) swamp under cultivation near an airstrip. The adults and larvae were preserved in 80% or 99.5% ethanol, respectively. Microscope slides were prepared with Canada balsam using the usual methods (Pinder, 1989). Measurements are given as ranges followed by a mean (when 3 or more measurements were taken), followed by the number measured (*n*) in parenthesis. All specimens studied here, including the types of the new species, have been deposited in the National Institute for Environmental Studies, Tsukuba, Japan.

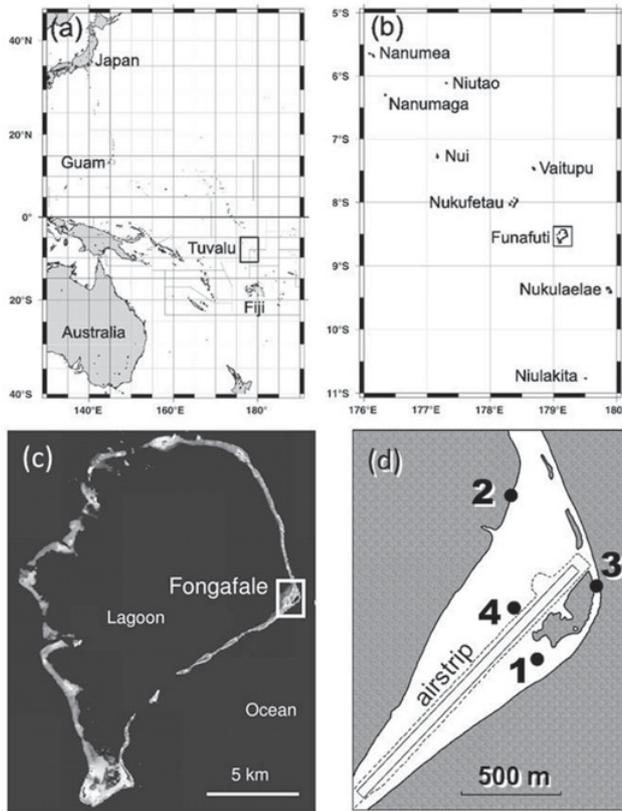
The specimens of *Comptosmittia longipalpis*, *Pseudosmittia tuvaluensis* and *Chironomus* sp. were subjected to DNA extraction for direct sequencing of a mitochondrial gene encoding *COI*, described by Kondo *et al.* (submitted), although PCR products were not recovered from *C. longipalpis* samples. The only sample of *Pseudosmittia amplexivirga* had been prepared for a slide, before the DNA analysis was planned. The length of the obtained sequence was 658 bp, excluding the primer region. Nucleotide sequence data are available in the DDBJ/EMBL/GenBank databases under the accession numbers AB769378–AB769382. After DNA extraction, the recovered translucent specimens were preserved in 99.5% ethanol and used for additional morphological observations.

### Subfamily Orthoclaadiinae

#### *Comptosmittia longipalpis* sp. n. (Fig. 2)

Type material: Holotype male, TUVALU: Funafuti Atoll, Fongafale Island, a wetland near the transmission

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**Fig. 1.** Map of sampling sites. (a) Tuvalu and adjacent countries in the Pacific; (b) Tuvalu; (c) aerial photograph of Funafuti Atoll; (d) locations of sampling sites on Fongafale Island.

tower (Site 1), 13.iii.2009, sweep net, K. Satake (NIES No. 1305, slide mounted with Canada balsam). Paratypes: 2 males, as holotype (NIES No. 1306–1307).

Diagnostic characteristics: The imago is distinguished by having no virga, complicated inferior volsellae on gonocoxites, weakly dentated megasetae, moderately developed anal point with concave margins, bare wing membranes, weakly developed anal lobe, strongly sinuous  $Cu_1$ , setae-bearing supraalar area and relatively long terminal palpomere.

### MALE ( $n=3$ , except when otherwise stated)

Total length (mm): 1.80–2.02, 1.93. Wing length (mm): 0.84–0.94, 0.89. Total length/wing length:

	fe	ti	ta <sub>1</sub>	ta <sub>2</sub>	ta <sub>3</sub>	ta <sub>4</sub>	ta <sub>5</sub>
p <sub>1</sub>	395–436, 410	436–467, 447	298–316, 304	158–182, 167	117–129, 122	72–75, 73	55–63, 59
p <sub>2</sub>	392–474, 427	396–422, 405	212–236, 224	89–104, 96	69–72, 71	45–47, 46	46–52, 50
p <sub>3</sub>	435–499, 462	465–502, 478	293–315, 305	135–138, 136	128–133, 130	41–68, 64	59–61, 60
	LR	BV	SV	BR			
p <sub>1</sub>	0.68–0.69, 0.68	2.73–2.81, 2.76	2.80–2.86, 2.82	1.8–2.7, 2.4			
p <sub>2</sub>	0.53–0.56, 0.55	3.95–4.15, 4.03	3.53–3.83, 3.72	1.6–2.3, 1.9			
p <sub>3</sub>	0.61–0.67, 0.64	3.07–3.39, 3.19	2.91–3.26, 3.08	2.5–3.8, 3.3			

2.05–2.33, 2.18. Wing length/length of profemur: 2.10–2.23, 2.16. Coloration entirely dark brown, scutellum slightly paler.

### Head

Antenna brown, 13 flagellomeres; flagellar whorl brown, normally developed; groove beginning about flagellomere 3 or 4; no apical strong seta. AR 0.51–0.56, 0.53. Eye bare, without dorsomedial elongation. Inner verticals 3 (2); outer verticals 5; no postorbitals. Clypeus with 16–18, 17 setae. Tentorium 101–117, 108  $\mu$ m long; 15–17, 16  $\mu$ m wide. Stipes 112–116, 114  $\mu$ m long. Palpomere lengths ( $\mu$ m): 20–22, 21 (2); 28–31, 30 (2); 46–49, 48 (2); 55 (1); 102–107, 105 (2); palpomeres 2 and 3 seem to be fused. Length ratio of palpomeres 5/3 2.08–2.33, 2.21 (2). A few sensilla clavata on palpomere 3, straight, like short sensilla chaetica.

### Thorax

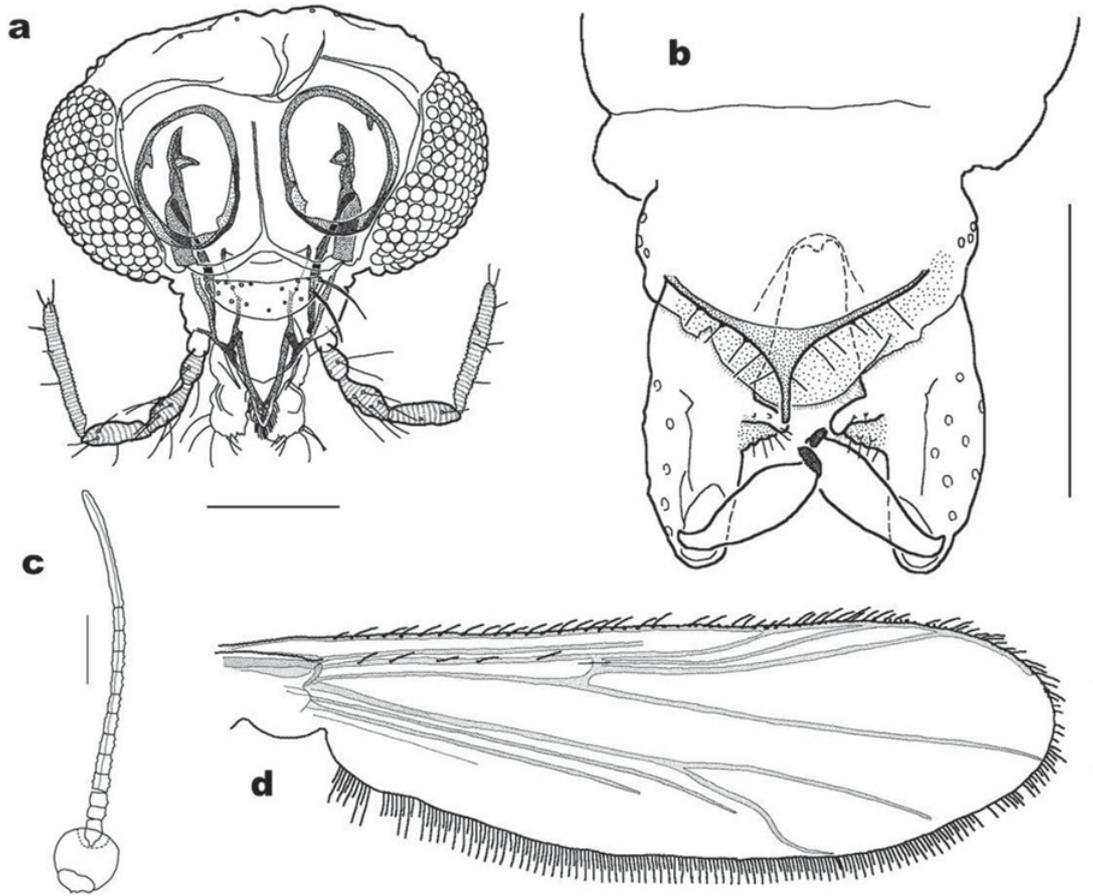
Anteprenotum without setae. Dorsocentrals normal, uniserial, arising from small pale pits, 10–11, 10; acrostichals start some distance from anteprenotum, short and scalpellate, 8–9, 8; prealars 3–4, 3; supraalars 1–2, 1. Scutellum with 5–6, 6 (2) setae in single transverse row.

### Wing

VR 1.44–1.47, 1.46. Wing membrane quite bare. Anal lobe moderately developed but not protruding. Costal extension 105–112, 108  $\mu$ m long. R with 3–6, 4 setae, other veins bare, except for marginal setae of costa.  $Cu_1$  strongly sinuous. Squama bare.

### Legs

Small pulvilli present. Spur of front tibia 30–33, 32  $\mu$ m (2) long; spurs of middle tibia 17  $\mu$ m (2) and 13–15, 14  $\mu$ m (2) long; spurs of hind tibia 33–36, 35  $\mu$ m (2) and 12–15, 14  $\mu$ m (2) long. Comb of 8 setae, 17–33  $\mu$ m long. Pseudospurs lacking. Lengths ( $\mu$ m) and proportions of legs:



**Fig. 2.** *Comptosmittia longipalpis* sp. n. Male. (a) Head; (b) hypopygium; (c) antenna; (d) wing. Scale bar = 0.1 mm.

### Hypopygium

Anal point 24.2–31.0, 26.9  $\mu\text{m}$  long, 28.2–40.8, 34.5  $\mu\text{m}$  (2) wide at base; with 6–7 strong lateral setae along concave lateral margins, void of microtrichiae at tip; laterosternite IX with 3–5, 4 setae. Phallapodeme 40–44, 42  $\mu\text{m}$  (2) long; transverse sternapodeme nearly straight, with rounded oral projections, 41–49, 45  $\mu\text{m}$  (2) long. Virga absent. Gonocoxite 112–124, 120  $\mu\text{m}$  long, with partially broad inferior volsella with dorsal rounded projection free of microtrichia and ventral lobe covered with microtrichiae. Gonostylus 53–58, 55  $\mu\text{m}$  (2) long, with semicircular, 6- $\mu\text{m}$ -long, vestigially dentate megaseta. HR 1.93–2.37, 2.15 (2); HV 3.37–3.85, 3.61 (2).

### Remarks

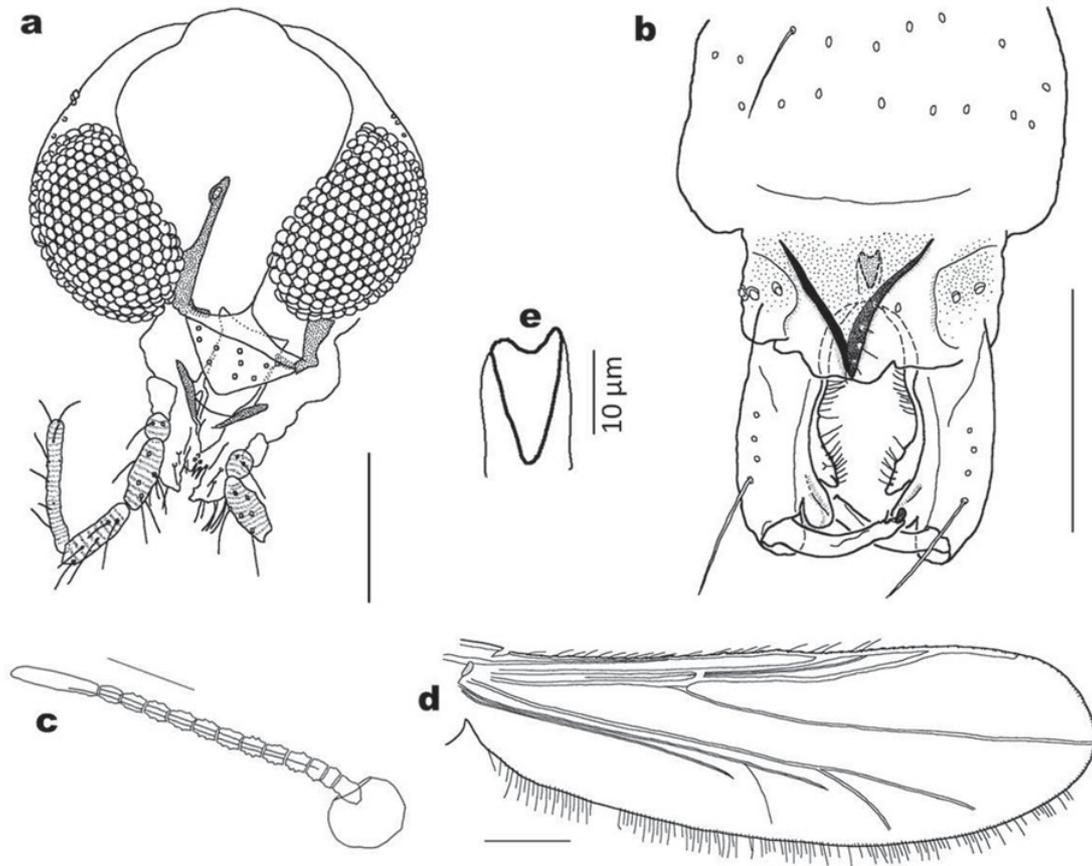
*C. longipalpis* apparently keys to *Comptosmittia dentispina* Sæther, 1981 in the review of the genus *Comptosmittia* Sæther, 1981 by Mendes *et al.* (2004) and Lin *et al.* (2013), because it has no protruding of the anal lobe of wing, 13-segmented flagellomeres of antenna, no virga, the complicated inferior volsella and the dentate

megaseta. However, *C. longipalpis* can be separated from *C. dentispina* by the darker thorax, quite bare wing, weak but conspicuous anal lobes, vestigial dentation of megaseta and the larger length ratio of palpomere 5/3 (2.21). The male of *C. dentispina* has pale brown thorax, the wing with 2–4 setae on membrane, the reduced anal lobe and the strong dentation of megaseta (Sæther, 1981). With the palpomere lengths presented by Sæther (1981), a calculation of the length ratio of palpomere 5/3 of *C. dentispina* is 1.12.

### *Pseudosmittia tuvaluensis* sp. n. (Fig. 3)

Type material: Holotype male, TUVU: Funafuti Atoll, Fongafale Island, swamp in a mangrove forest (Site 3), 11.iii.2009, sweep net, K. Satake (NIES No. 1301, slide mounted with Canada balsam). Paratypes: seven males, as holotype (NIES No. 1302–1303, D0466–D0469). Direct sequencing identified three haplotypes of *COI* gene from four specimens. The three haplotypes differed from each other by two nucleotides at three sites. The accession numbers were AB769378 to AB769380.

Diagnostic characteristics: The male imago is distinguished from other congeners with completely bare eyes, forked postcubitus veins, a broadly triangular



**Fig. 3.** *Pseudosmittia tuvaluensis* sp. n. Male. (a) Head; (b) hypopygium; (c) antenna; (d) wing; (e) virga. Scale bar = 0.1 mm, except for virga.

anal point with straight side margins and sharply pointed apex, a broad and short virga, inferior volsellae on gonocoxites with accessory lobes situated posterior to inferior volsella, minute square cristae dorsalis, ultimate flagellomere without apical seta, and a very low AR.

### MALE ( $n=3$ , except when otherwise stated)

Total length (mm) 1.48–1.79, 1.65. Wing length (mm) 0.71–0.80, 0.74. Total length/wing length 2.08–2.52, 2.24. Wing length/length of profemur 2.32–2.35, 2.33 (2). Coloration blackish brown.

#### Head

Antenna brown, 13 flagellomeres; flagellar whorl brown, normally developed; groove beginning about flagellomere 3 or 4; no apical strong seta. AR 0.31 (3). Eye bare, without dorsomedian elongation. Inner verticals 4–5, 5; outer verticals 4–5, 4; no postorbitals. Clypeus with 10–11, 10 setae. Tentorium 97–103, 100  $\mu\text{m}$  long; 16–18, 16  $\mu\text{m}$  wide. Palpomere lengths ( $\mu\text{m}$ ): 22; 20–27, 24; 50–57, 52; 61–64, 63; and 92–97, 94. Palpomere 3 with a

few indistinct sensilla clavata; palpomere 4 without sensilla clavata.

#### Thorax

Anteprepronotum with 3–4, 4 setae. Dorsocentrals normal, uni- to biserial, arising from small pale pits, 17–20, 19; acrostichals arising at paler area in the center of scutum, 2; prealars 4–5 4, no supraalars. Scutellum with 6 setae in single transverse row.

#### Wing

VR 1.73–1.93, 1.80. VR2 1.50 (1). Costal extension 18–76, 38  $\mu\text{m}$  long. All veins bare, except for marginal setae of costa; costal extension with sparse marginal setae. Squama bare. Postcubitus vein forked.

#### Legs

Pulvilli absent. Spur of front tibia 32  $\mu\text{m}$  long, spurs of middle tibia 17–20, 19  $\mu\text{m}$  long, spurs of hind tibia 41–44, 43  $\mu\text{m}$  and 15  $\mu\text{m}$  long. Comb of 13 setae 23–41  $\mu\text{m}$  long. Lengths ( $\mu\text{m}$ ) and proportions of legs:

	fe	ti	ta <sub>1</sub>	ta <sub>2</sub>	ta <sub>3</sub>	ta <sub>4</sub>	ta <sub>5</sub>
p <sub>1</sub>	302–345, 320	339–353, 346	167–178, 174	71–79, 76	61–62, 61	41–44, 43	46–51, 48
p <sub>2</sub>	341–350, 346	342–368, 357	172–178, 176	79–86, 84	60–69, 65	37–48, 44	40–48, 46
p <sub>3</sub>	314–340, 331	346–371, 361	190–218, 208	96–104, 101	103–108, 105	43–50, 47	44–47, 46
	LR	BV	SV	BR			
p <sub>1</sub>	0.49–0.52, 0.50	3.69–3.72, 3.68	3.70–3.93, 3.82	2.0–2.4, 2.2			
p <sub>2</sub>	0.48–0.50, 0.49	3.52–3.96, 3.71	3.97–4.04, 3.99	2.0–2.4, 2.2			
p <sub>3</sub>	0.55–0.59, 0.58	2.97–3.03, 3.01	3.26–3.47, 3.33	1.9–2.8, 2.3			

## Hypopygium

Anal point 54–60, 58 µm long. Laterosternite IX with 3 setae. Phallapodeme 48–55, 53 µm long; transverse sternapodeme 59–75, 64 µm long, straight, without oral projections. Virga broad, triangular, 15–19, 17 µm long. Gonocoxite 111–136, 120 µm long, with large bifurcate inferior volsella. Gonostylus 57–60, 58 µm long; with minute preapical crista dorsalis; with 6–8, 7 µm long megaseta. HR 1.88–2.35, 2.06; HV 2.59–3.01, 2.84.

## Remarks

Combination of no strong seta on antennal apex, the broadly triangular anal point, the forked postcubitus, the low AR (about 0.3), the simple virga and the duplicate inferior volsella separate *P. tuvaluensis* from other congeners except for an Oceanian species *Pseudosmittia dupla* (Tokunaga, 1964). However, *P. tuvaluensis* can be separated from *P. dupla* by the posteriorly situated accessory lobe of inferior volsella and the small but apparent crista dorsalis on the gonostylus. *P. dupla* has an accessory lobe just covered by an inferior volsella and no crista dorsalis on a gonostylus (Tokunaga, 1964; Ferrington and Sæther, 2011).

## *Pseudosmittia amplexivirga* sp. n. (Fig. 4)

Material examined: 1 male, TUVAlU: Funafuti Atoll, Fongafale Island, a swamp in a mangrove forest (Site 3), 11.iii.2009, sweep net, K. Satake (NIES No. 1304, slide mounted with Canada balsam).

Diagnostic characteristics: The male imago is distinguished from other congeners with completely bare eyes, forked postcubitus veins, a broadly triangular anal point with straight side margins and a sharply pointed apex, a slender virga with lateral lamellae, a large triangular inferior volsella on a gonocoxite and a preapical crista dorsalis on a gonostylus.

## MALE IMAGO (n=1)

Total length 1.93 mm. Wing length 0.86 mm. Total length/wing length 2.26. Coloration entirely dark brown.

## Head

Antenna brown, 13 flagellomeres; flagellar whorl brown, normally developed; groove beginning at flagellomere 3; no apical strong seta. AR 0.70. Eye bare, without dorsomedian elongation. Inner verticals 3; outer verticals 2; no postorbitals. Clypeus with 13 setae. Tentorium 95 µm long, 20 µm wide. Stipes 80 µm long. Apex of palpomere 4, with a somewhat thicker apical spine, extend ventrally. Palpomere lengths (µm): 23, 23, 49, 104, 101; palpomeres 2 and 3 seem to be fused. Palpomere 3 with a few indistinct sensilla clavata; palpomere 4 without sensilla clavata.

## Thorax

Anteprenotum without setae. Dorsocentrals normal, uniserial, arising from small pale pits, 15; acrostichals 2, arising at the center of scutum; prealars 3; no supraalars. Scutellum with 3 setae in single transverse row. Postcubitus veins forked.

## Wing

VR 1.65, VR2 1.43. Wing membrane quite bare. Anal lobe moderately developed. Costa not produced. All veins bare, except for marginal setae of costa. Squama bare.

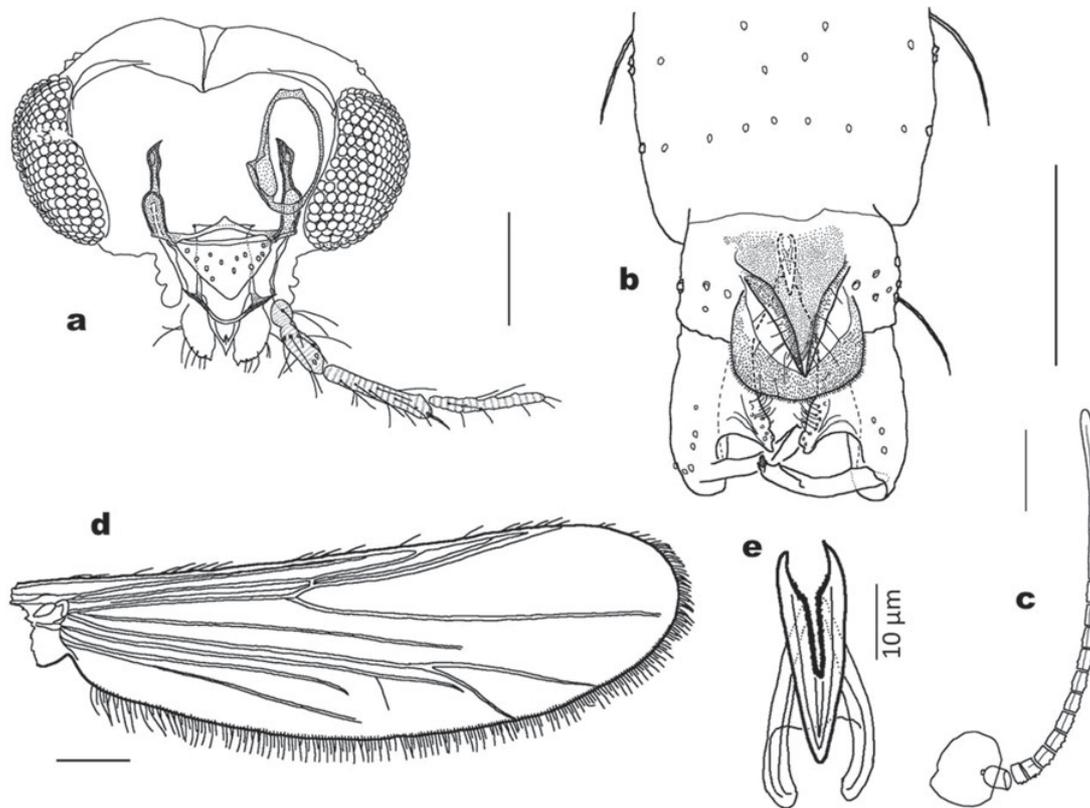
## Legs

Pulvilli absent. Spur of front tibia 32 µm long, of middle tibia 27 µm long, spurs of hind tibia 38 µm and 25 µm long. Comb of 11 setae, 21–30 µm long. Lengths (µm) and proportions of legs:

	fe	ti	ta <sub>1</sub>	ta <sub>2</sub>	ta <sub>3</sub>	ta <sub>4</sub>	ta <sub>5</sub>
p <sub>1</sub>	380	400	237	122	96	61	50
p <sub>2</sub>	424	429	234	119	83	56	55
p <sub>3</sub>	393	425	249	147	141	66	53
	LR	BV	SV	BR			
p <sub>1</sub>	0.59	3.09	3.28	3.4			
p <sub>2</sub>	0.54	3.48	3.65	3.3			
p <sub>3</sub>	0.58	2.62	3.29	4.4			

## Hypopygium

Anal point 59 µm long. Phallapodeme 56 µm long; transverse sternapodeme 61 µm long, without oral



**Fig. 4.** *Pseudosmittia amplexivirga* sp. n. Male. (a) Head; (b) hypopygium; (c) antenna; (d) wing; (e) virga. Scale bar = 0.1 mm, except for virga.

projections, arched. Virga 28  $\mu\text{m}$  long, with a pair of lateral lamellae. Gonocoxite 112  $\mu\text{m}$  long, with large triangular inferior volsella. Gonostylus 81  $\mu\text{m}$  long; with minute square preapical crista dorsalis; with 7  $\mu\text{m}$  long megaseta. HR 1.84; HV 2.36.

## Remarks

Combination of no strong seta on antennal apex, the broadly triangular anal point, the lamellate virga, the protruding triangular inferior volsella and the forked postcubitus may separate this species from other congeners. Apart from structure of virga, above mentioned characters are shared by *Pseudosmittia kraussi* (Tokunaga, 1964), *Pseudosmittia palauensis* (Tokunaga, 1964) and *Pseudosmittia triangula* (Tokunaga, 1964), which have stout and simple virga and have distribution within Oceanian region (Tokunaga, 1964; Ferrington and Sæther, 2011). According to Ferrington and Sæther (2011), lamellar structures flanking virga are known among three species belonging to the *brevifurcata*-group, namely, *Pseudosmittia lamellata* Andersen *et al.*, 2010, *Pseudosmittia propetropis* Ferrington and Sæther, 2011 and *Pseudosmittia tropis* Andersen *et al.*, 2010 recorded from Afrotropical and Neotropical regions. However, all of them quite differ from *P. amplexivirga* by having the unbranched postcubitus, the narrow and blunt anal point and the weak inferior volsella appressed to the gonocoxite.

## *Chironomus* sp. Tuvalu (Fig. 5)

Material examined: 4 larvae, TUVALU: Funafuti Atoll, Fongafale Island, a puddle in a taro swamp, 11.iii.2009, D-frame net sweeping, K. Satake (NIES No. 1312-1314, D0481). Twenty-seven 4th instar larvae and two 3rd instar larvae of probably the same species as slide specimens, stored in 99.5% ethyl alcohol. The partial *COI* gene sequence from one specimen was obtained (accession number AB769382).

## Larva, 4th instar ( $n=3$ )

Head capsule yellow, with dark mentum. Antenna segment lengths ( $\mu\text{m}$ ): 54–60; 16–17; 3–4; 4–5; 3; blade 68–78; subsidiary 4–5  $\mu\text{m}$ ; style 7–8. AR 1.9–2.2. Lauterborn organs 4–5  $\mu\text{m}$ . Ring organ basal. Very strong (but pale) seta on 1st antennal segment, 18–21  $\mu\text{m}$  long, arising from mid-segment and extending to near apex of segment 1. Mentum width 110–113  $\mu\text{m}$ , dorsal surface smooth. Mandible 120–132  $\mu\text{m}$ . Premandible 200  $\mu\text{m}$ , apical tooth blunt, brush normally developed.

Labrum narrowed and tapering anteriorly. SI plumose, SII simple. Labral lamellae normally developed. Pecten epipharyngis composed of a combed plate with about 15 teeth. Premandible with two apical teeth. Mentum with a trifold median tooth, and six lateral teeth in each side, second lateral tooth appressed to first lateral tooth making

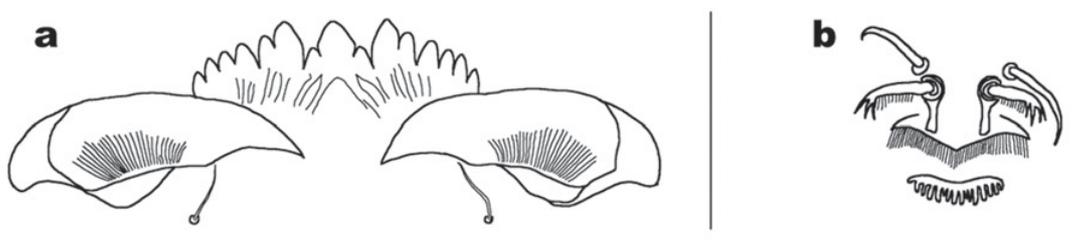


Fig. 5. *Chironomus* sp. Tuvalu. Larva. (a) Mentum; (b) labium. Scale bar = 0.1 mm.

the notch between them shallow, second and third teeth divided by a deep notch, third to six lateral teeth gradually shorten, fourth lateral tooth slightly shorter than the trend.

Abdomen ( $n = 1$ ). Anterior parapods crowned with simple dark yellow claws. Procercus not pigmented, 6 apical setae 325  $\mu\text{m}$  long. Posterior parapod claws large, curved, simple, dark yellow. Seventh abdominal segment with a pair of caudolateral processes. Eighth abdominal segment with two pairs of long blood gills.

## Remarks

This larva belongs to medium-sized species among the genus *Chironomus* Meigen, 1803. Morphology of this larva is typical of the genus, with a pair of lateral tubules and two pairs of ventral tubules. The partial *COI* sequences will be helpful for confirmation to associate with adult specimens in future. The habitat of this species is a shallow freshwater puddle in a taro swamp.

## General characteristics of the Tuvaluan chironomid fauna

In this study, four species belonging to three genera of chironomids were found in Tuvalu. The previously known habitats of *Comptosmittia* have been phytotelmata (Cranston and Kitching, 1995); therefore, axils of *Pandanus*, for example, seemed to be a candidate habitat in Tuvalu, although such habitats have not yet been examined. The habitats of *Pseudosmittia* are uncertain in many cases. At least two *Pseudosmittia* species are aquatic, but many species are said to be terrestrial or semi-terrestrial (Cranston *et al.*, 1989). As larvae of *Chironomus* sp. Tuvalu were found only in a taro field, the habitat of this species is apparently fresh water. Thus, at least *Chironomus* sp. Tuvalu and probably *C. longipalpis* depend on inland freshwater environments in Tuvalu.

Each genus found in Tuvalu has widespread distribution among oceanic islands. *Comptosmittia* have been recorded from Micronesia (Tokunaga, 1964) and Lesser Antilles (Sæther, 1981). Each species of this genus is endemic to a certain island group or an archipelago. *Pseudosmittia* have been recorded from the same distribution as *Comptosmittia* (Tokunaga, 1964; Sæther, 1981), plus in Samoa (Edwards, 1928), Greater Antilles (Sublette

and Wirth, 1972), the Seychelles (Sæther, 2004) and Macaronesia (Murray *et al.*, 2004). Several species of this genus are endemic to certain island groups, whereas several species are widespread (for example *Pseudosmittia brachydicerana*, recorded from Micronesia, Hawaii, Jamaica and several other tropical islands; Andersen *et al.*, 2010; Ferrington and Sæther, 2011). *Chironomus* have been recorded from the same distribution as *Pseudosmittia* (Edwards, 1928; Tokunaga, 1964; Spies and Reiss, 1996; Murray *et al.*, 2004; Sæther, 2004), plus in the Marquesas (Edwards, 1933). Thus, at the generic level, the chironomids found in Tuvalu are very common among oceanic islands, whereas at the specific level they are proper to Tuvalu. The Tanypodinae, *Cricotopus* and *Tanytarsus*, common members of island chironomid faunae, have not yet been collected in Tuvalu.

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