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FIRST LARVAL DESCRIPTION OF THE COASTAL GENUS AND SPECIES
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ABSTRACT

The late-instar larva of *Phucobius simulator* Sharp is described for the first time, being also the first larval description of the genus *Phucobius* Sharp. Nine unknown larvae were collected in association with adults of *P. simulator* from seashores of Korea and Japan. The unknown larvae were identified as *P. simulator* by DNA sequencing. Diagnostic characters of the species are provided, with illustrations.

Key words: *Phucobius simulator*, larval description, seashore, Staphylinidae

RESUMEN

Se describe por primera vez el último estadio larval de *Phucobius simulator* Sharp, siendo también la primera descripción de una larva del género *Phucobius* Sharp. Se recolectaron nueve larvas desconocidas en asociación con los adultos de *P. simulator* de la costa de Corea y de Japón. Se identificaron las larvas desconocidas como *P. simulator* por medio de la secuenciación de ADN. Se provee caracteres diagnósticos e ilustraciones de la especie.

Palabras Clave: *Phucobius simulator*, descripción larval, costa, Staphylinidae

The genus *Phucobius* Sharp contains 8 species (Herman 2001) and is confined to the seashores of the Oriental and eastern Palearctic Regions, and East Africa. Adults of the genus *Phucobius* are similar to those of the genus *Cafius* Stephens but lack spines on the anterior tibiae (Moore & Legner 1976; Smetana 1995). Adults and larvae are found under accumulated decaying seaweeds and logs. To date no late-instar larvae of the genus *Phucobius* have ever been described.

In this paper, we describe late-instar larvae of *P. simulator* for the first time through the association of larvae-adults with DNA sequences, and provide diagnostic characters with illustrations.

LATE-INSTAR LARVA OF
PHUCOBIUS SIMULATOR SHARP
(FIG. 1)

Diagnostic Combination

Among coastal staphylinid genera, late-instar larvae of *Phucobius simulator* are recognized by the following combination of characters: neck present; antenna with article 2 longer than 3, ar-

ticle 3 with 2 solenidia and 1 campaniform sensillum (Fig. 2); stipes a little shorter than maxillary palpus, mala with 1 apical seta (Fig. 3); mandible slender, with tiny internal tooth (Fig. 4); nasale middle 3 teeth distinctly separated from lateral teeth (Fig. 5); ligula short, 0.32 times as long as labial palpomere 1 (Fig. 6); tarsungulus with 3 spines (Fig. 7); tergite X shorter than 1st article of urogomphi.

Description

Length 11.0-13.0 mm. General body shape elongate, flattened, parallel-sided (Fig. 1). Body brown, but head light brown, abdomen dark brown.

Head

Sub-quadrate, almost equally wide from apical to basal margin. About 0.8 times as long as wide. Four stemmata present. Ecdysial sutures distinct and complete from near antennal insertion. Antenna (Fig. 2), 4-articled. Length of articles 1st = 4th < 3rd < 2nd; article 1 wider than

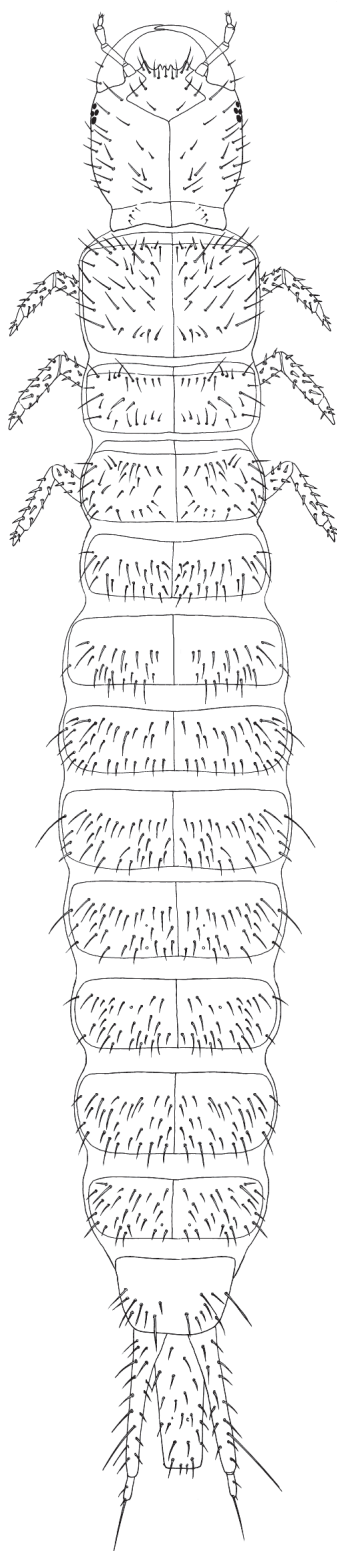


Fig. 1. *Phucobius simulator*. Habitus of larva, length 12.0 mm.

long, transverse; 1 campaniform sensillum present on 1/3 of article 2; article 3 with 2 solenidia (IIS1 and IIS2) and 1 campaniform sensillum, 1 corn-type sensory appendage present; article 4 with 4 solenidia (IVS1–IVS4); article 3 and 4 each with 3 setae. Mentum with 3 pairs of setae. Gular sutures convergent in middle of head, divergent to apex.

Mouthparts

Maxilla (Fig. 3). Stipes a little shorter than maxillary palpus; mala with 1 seta at apex, small, elongate; maxillary palpus with 4 articles, a separate sclerotization forming a short ring at base in form of an extra article present, 1 seta and 1 campaniform sensillum present; length ratio of palpomeres 1st : 2nd : 3rd : 4th = 1:1.43:1.14:0.36; width of palpomeres 1st = 2nd > 3rd > 4th. Mandible (Fig. 4) slender, 2 macrosetae present along outer surface, falciform, undivided at acute apex, tiny internal tooth present on apical region, almost symmetrical. Nasale (Fig. 5). Anterior margin of nasale with 9 teeth divided into 3 distinct clusters (1 middle and 2 lateral), each cluster with 3 teeth; middle 3 teeth pointed (LT4 and LT5), central tooth (LT5) smallest, the last and penultimate teeth (LT 1 and LT2) very weak. Ten setae present on each side of midline. Labium (Fig. 6). Labial palpus with 3 articles; length ratio of palpomeres 1st : 2nd : 3rd = 1.0:0.47:0.16, conical ligula short, 0.32 times as long as palpomere 1, pubescent at base.

Thorax

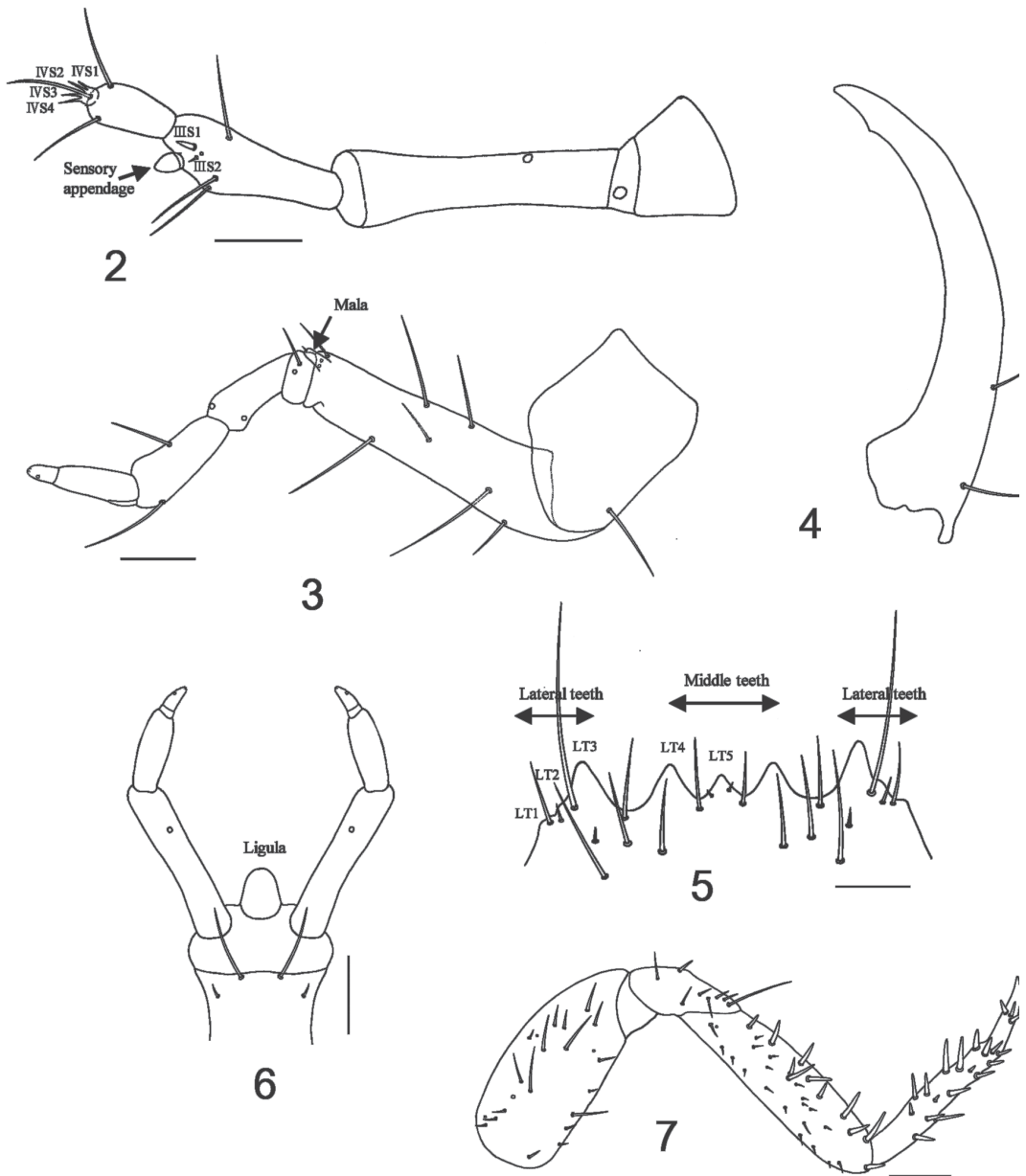
Pronotum subquadrate, sclerotized, setae scattered at sides and on disc. Pronotum about 2 times longer than mesonotum. Mesonotum length subequal to metanotum, both as long as posterior margin of pronotum.

Legs (Fig. 7)

Coxa, trochanter, femur, tibia, and tarsungulus distinguishable, tarsungulus with 3 articulated spines.

Abdomen

Abdominal tergites I–VIII transverse, parallel-sided, slightly narrowed to apex. Tergites and sternites divided by mid-longitudinal line; tergite X about 5.0 times longer than wide. Urogomphi two-articled, longer than tergite X; article 1 slender, longer than tergite X; article 2 with 2 small setae and 1 large seta arising from apex.



Figs. 2-7. *Phucobius simulator*. 2, Antenna, dorsal aspect; 3, maxilla, dorsal aspect; 4, mandible, dorsal aspect; 5, nasale, dorsal aspect; 6, labium, ventral aspect; 7, anterior leg, dorsal aspect. Scale bars = 0.1 mm.

Specimens Examined

KOREA: Gyeongnam Prov., Geoje Isl., Hwangpo beach, 28-VI-2004, J.-S. Park and K.-J. Ahn, ex under seaweeds (CNUIC, 1); Gangwon Prov., Gangneung-si, Geumjin-ri, Okgye beach, N 37° 37' 37.2" E 129° 03' 04.8" 7 m, 4-IX-2013, K.-J. Ahn, I. S. Yoo, J. H. Song, under log near estu-

ary (CNUIC, 2); Japan: Hokkaido, Nemuro, Shunkunitai, 24-VIII-1999, K.-J. Ahn, ex under stones on salt marsh (CNUIC, 6).

Remarks

Most specimens were collected under logs or stones in an estuary (Fig. 8). The late-instar larva of *Phucobius simulator* resembles that of *Cafius*



Fig. 8. Collection site of *Phucobius simulator* larvae in an estuary at Okgye Beach, Korea. Inset is a beetle larva collected in the microhabitat (under log or stone) indicated by an arrow.

vestitus (Sharp) in shape and structures of mouth-parts including the teeth on the nasale. They are also similar in the shape of antenna and urogomphi. But they differ from each other in the ratios of following characters presented in Table 1.

ADDITIONAL MOLECULAR EVIDENCE FOR THE IDENTIFICATION OF LATE-INSTAR LARVAE OF *P. SIMULATOR*

Unknown larvae were collected with adults of *Phucobius simulator* from seashores of Korea and Japan, and we attempted to associate them by gene sequencing of individuals. The partial

cytochrome oxidase II gene was sequenced from the unknown larvae and several identified adult specimens of *Phucobius* (see Jeon & Ahn 2007 for the method). The sequences are deposited in GenBank under accession numbers (Table 2).

In previous studies of the genus *Cafius* known to be closely related to *Phucobius* (Jeon & Ahn 2005, 2007; Jeon et al. 2012), the intraspecific p-distance of *cytochrome oxidase II* gene varied in the range 0-2.93% and the minimum interspecific p-distance was 11.79%. The pairwise distances of *P. simulator* between adult and larva were 1.23-1.72%, placed within the range of intraspecific p-distances compared to the former results.

We added the present *Phucobius* data (Table 2) to the previous *Cafius* data (Jeon & Ahn 2007) and performed a parsimony analysis with CO II genes in TNT 1.1 (Goloboff et al. 2007) using the implicit enumeration option. The length of the COII sequences included in the analysis was 344 bp. The branch support values were estimated by bootstrapping with 100 replications. The analysis showed that specimens of a single species formed cohesive assemblages (Fig. 9). The unknown larva grouped unambiguously with the adult specimens of *Phucobius simulator*. Therefore, we identified the unknown larvae as probable late-instar larvae of *P. simulator*.

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TABLE 1. DIFFERENCES BETWEEN LATE-INSTAR LARVAE OF *PHUCOBIUS SIMULATOR* AND *CAFIUS VESTITUS*.

	<i>P. simulator</i>	<i>C. vestitus</i>
LT1 and LT2 on nasale	not distinct	distinct
length ratio of LT5 to LT4	about 0.75	about 0.87
length ratio of ligula to labial palpomere 1	about 0.32	about 0.67
length ratio of labial palpomere 1 to 2	about 2.13	about 1.36

TABLE 2. SPECIES, COLLECTION INFORMATION, AND GENBANK ACCESSION NUMBERS FOR CYTOCHROME OXIDASE II SEQUENCES INVESTIGATED IN THIS STUDY TO IDENTIFY LARVAE COLLECTED IN ASSOCIATION WITH ADULTS OF *PHUCOBIUS SIMULATOR* FROM SEASHORES OF KOREA AND JAPAN.

Species	Collection information	GenBank accession
<i>Phucobius simulator</i> (larva)	Korea: Geoje, Hwangpo Beach,	EF450220
<i>P. simulator</i> (adult)	Korea: Jeju, Seongsan, Ilchulbong,	EF450221
<i>P. simulator</i> (adult)	Korea: Jeju, Seongsan, Ilchulbong,	EF530919
<i>Phucobius</i> sp. (adult)	Australia: Queensland, Daintree N.P., Beach of Wonga	EF530920
<i>Phucobius</i> sp. (adult)	Philippines: Panglao Isl., Libaong beach	EF530921

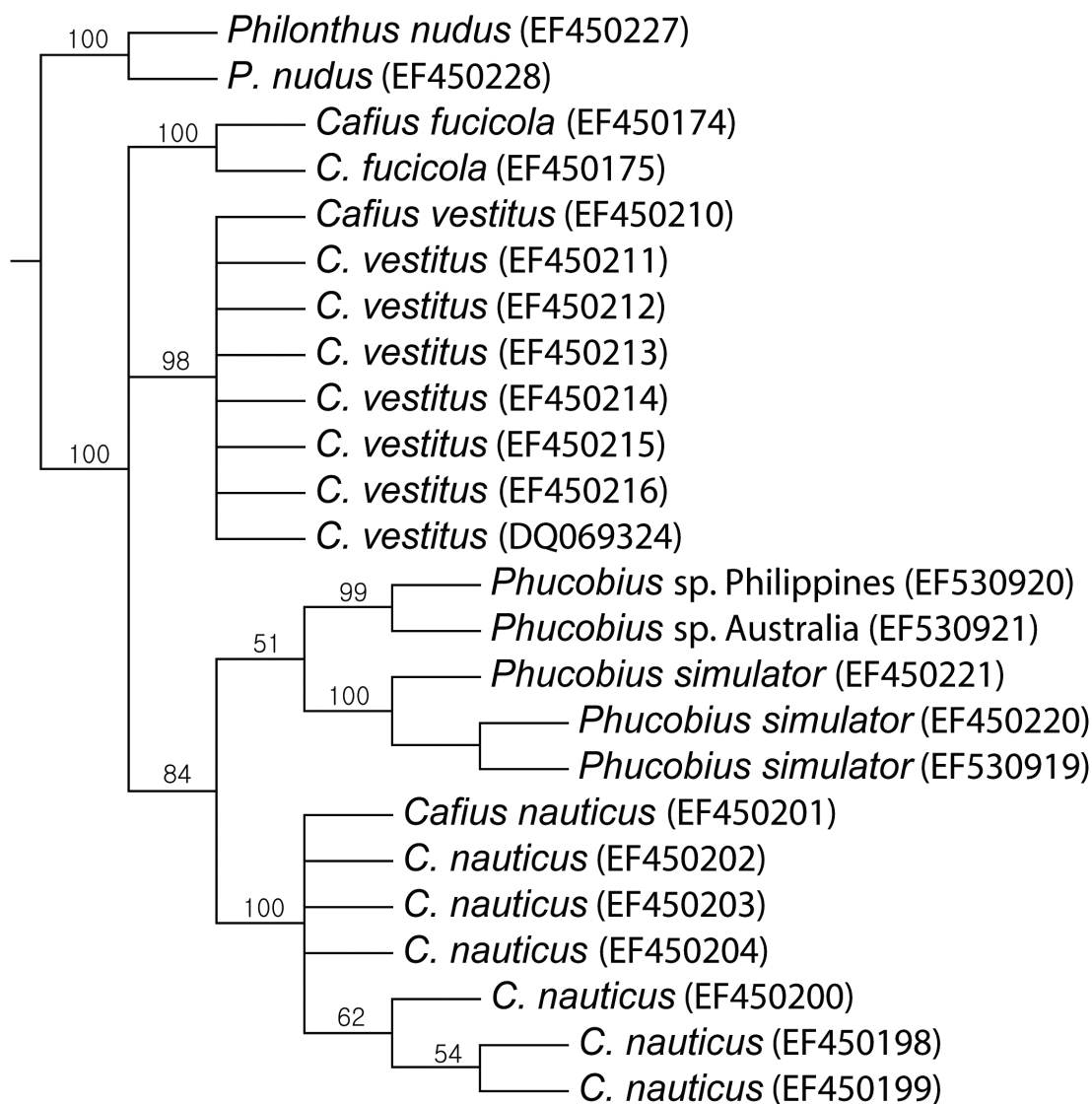


Fig. 9. Bootstrapping results in TNT based on 100 replications with branch support values.

A parsimony analysis in TNT 1.1 was performed on the CO II *Phucobius* and *Cafius* 344 bp data, and the branch support values were estimated by bootstrapping with 100 replications.

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