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Description of a New Species of *Neocollyris* W. Horn, 1901 (Coleoptera: Cicindelidae) with a Detailed Description of the Larval Development Observed *In Situ* on a Native Non-endemic *Premna odorata* Blanco (Lamiaceae) in the Philippines

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This paper presents *Neocollyris* (*Neocollyris*) *anichtchenkoi* sp. nov., a new species of *Neocollyris* W. Horn, 1901 (Coleoptera: Cicindelidae) from Luzon Island, Philippines. Larval development was documented *in situ* in *Premna odorata* Blanco (Lamiaceae). Scanning electron micrograph images of the larvae are also provided. This is the first record of a new species with notes on the larval development of a Philippine endemic *Neocollyris*.

Keywords: Collyridini, larvae, Luzon, taxonomy

INTRODUCTION

As an archipelagic country, the Philippines has a high rate of island endemism. Unfortunately, its unique biodiversity is threatened by anthropogenic pressures, making it one of the biodiversity hotspots (Myers *et al.* 2000). With about a quarter of pre-colonial forests remaining (Pulhin *et al.* 2021), many species are threatened with extinction but remain poorly studied. The Philippine Coleoptera is one such taxa that is still understudied. It is represented by about 8,000 species, including more than 150 species of tiger beetles from the family Cicindelidae, most of which are endemic (Cabras *et al.* 2016). Recent efforts have added new beetle species (Anichtchenko and Medina 2019, 2020; Medina *et al.* 2019; Medina *et al.* 2020a, b, 2021b, 2023). An interesting group of tiger beetles is arboreal tiger beetles that include the genera *Therates* Latreille 1816, *Tricondyla* Latreille 1822, and *Neocollyris* Horn 1901, which have larvae that reside in living or rotting wood unlike epigeic species (Medina *et al.* 2021a; Marohomsalic *et al.* 2021; Trautner and Schawaller 1996; Toki *et al.* 2017; Yamamoto *et al.* 2018). Like other genera of Collyridini, the genus *Neocollyris* has an elongated body and prominent eyes (Horn 1931). Recent phylogenies place *Neocollyris* as a sister taxon with *Tricondyla* (Duran and Gough 2020), with the former being differentiated by its elongated and parallel elytra (Horn 1931). Until now, the Philippine *Neocollyris* is represented by 30 species and subspecies, with Luzon having seven (Cabras *et al.* 2016; Anichtchenko and Medina 2019). Its larvae have been recorded to inhabit *Coffea arabica* L. (Rubiaceae), *C. liberica* Hiern (Rubiaceae), *Machilus thunbergii* Siebold & Zucc. (Lauraceae), *Mallotus japonicus* (L.f.) Mü ll. Arg. (Euphorbiaceae), *Bischofia javanica* Blume (Phyllanthaceae), *Miscanthus sinensis* Andersson (Poaceae), and *Phragmites vallatoria* (L.) Veldkamp (Poaceae) (Toki *et al.* 2017).

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Recently, a dead female tiger beetle was seen hanging from a young branch of *Premna odorata* Blanco (Lamiaceae), a widespread species native to the Philippines (de Kok 2013). Further examination revealed that this was a new species of *Neocollyris* that oviposited in the plant. This is also the first record of *P. odorata* as a larval host for the genus. Interestingly, this discovery was made in a residential area where *P. odorata* was unintentionally planted, highlighting the importance of urban biodiversity and the unexpected discoveries that can be made with understudied taxa in disturbed ecosystems (Freitag 2013).

Herein, *N. anichtchenkoi* sp. nov. is described based on specimens from Luzon, Philippines along with its larval development in the young branches of *P. odorata.*

MATERIALS AND METHODS

Taxonomic Treatment

Morphological characters were observed under Leica MZ 12.5 stereomicroscope. Habitus images were taken using a Canon EOS 6D digital camera equipped with an MP-E 65-mm macro lens mounted in StackShot macro rail automated with Helicon Remote version 4.3.0.w. All images were stacked using Helicon Focus version 8.1.1 and processed using a licensed Photoshop CS6 Portable software version. Measurements of various body parts are as follows: total body length (BL) from anterior margin of clypeus to apex of elytra (without labrum), length of compound eyes (LCE), width of compound eyes (WCE), length of labrum (LL) from anterior margin of clypeus to anterior margin of labrum (without apical tooth), width of labrum (WL), length of mandible (LM), length of pronotum (LP) along midline, width of pronotum at widest point (WP); length of elytra (LE) from humerus to apex of sutural angle (without sutural tooth), width of elytra (WE) at widest point, length of aedeagus (LA), and width of aedeagus (WA) in broadest sense; / separates different lines on a label, and // separates different labels. All measurements are given in millimeters (mm).

Comparative material and specimens used in this study are deposited in the following collections:

AAc	Alexander Anichtchenko Collection, Daugavpils, Latvia
DMC	Decier Oliver Mapile Collection, Calasiao, Philippines
DOrSU	Davao Oriental State University Beetle Collections, Mati City, Philippines
JWc	Jürgen Wiesner Collection, Wolfsburg, Germany
MMCP	Milton Medina Collections, Tagum City Philippines
PNM	Philippine National Museum, Ermita, Manila, Philippines
SNSD	Senckenberg Naturhistorische Sammlungen Dresden, Germany

Observation of Larval Development

On the morning of 15 Jun 2023, a dead female was spotted on a young branch of *Premna odorata* Blanco with its ovipositor still attached to the internode of the branch. The dead specimen was still pliable, indicating that it died recently prior to discovery. The branch together with the specimen were collected and examined. An egg was found inside the excavated cavity, confirming that *P. odorata* is used by *Neocollyris* as a larval host. Subsequent examination of young branches of two *P. odorata* plants within a residential lot in Calasiao, Pangasinan, Luzon, Philippines yielded multiple cavities containing eggs and larvae. No other Cicindelidae was observed apart from the *Neocollyris* described herein; thus, all immature stages collected are confidently identified as this species.

Immature specimens were preserved in 70% ethanol, and some were first fixed in hot water, and all are currently deposited in DMC. Larval instars were determined by counting the number of setae at the margins of the galea and comparing the relative sizes of the head capsule, following Gwiazdowski *et al.* (2020). Images were either taken with a Redmi Note 10 Pro smartphone with a built-in macro lens camera or a 48 MP FHD microscope camera attached to an unbranded trinocular microscope. Images were stacked using Helicon Focus version 8.2.2 and processed using a licensed Photoshop CS6 Portable software version. Measurements are given in millimeters using an INGCO® digital caliper. Scanning electron microscope (SEM) TM4000Plus was used to provide images of the larval instars.

TAXONOMY

***Neocollyris anichtchenkoi* Medina & Mapile, sp. nov.**
Zoobank: urn:lsid:zoobank.org:pub:5F6DFD13-79B4-448C-889E-F9CC6322BBDB
Figure 1A.

Type material. Holotype: male: PHILIPPINES: Luzon, Pangasinan, Calasiao, Brgy. Nalsian Centro, 15.vi.2023, D. Mapile leg. (MMCP), printed on a red card, to be deposited at PNM. **Paratypes: 1 female,** same label as holotype, printed on a red card; **1 male,** PHILIPPINES: Luzon, Pangasinan, Calasiao, Brgy.

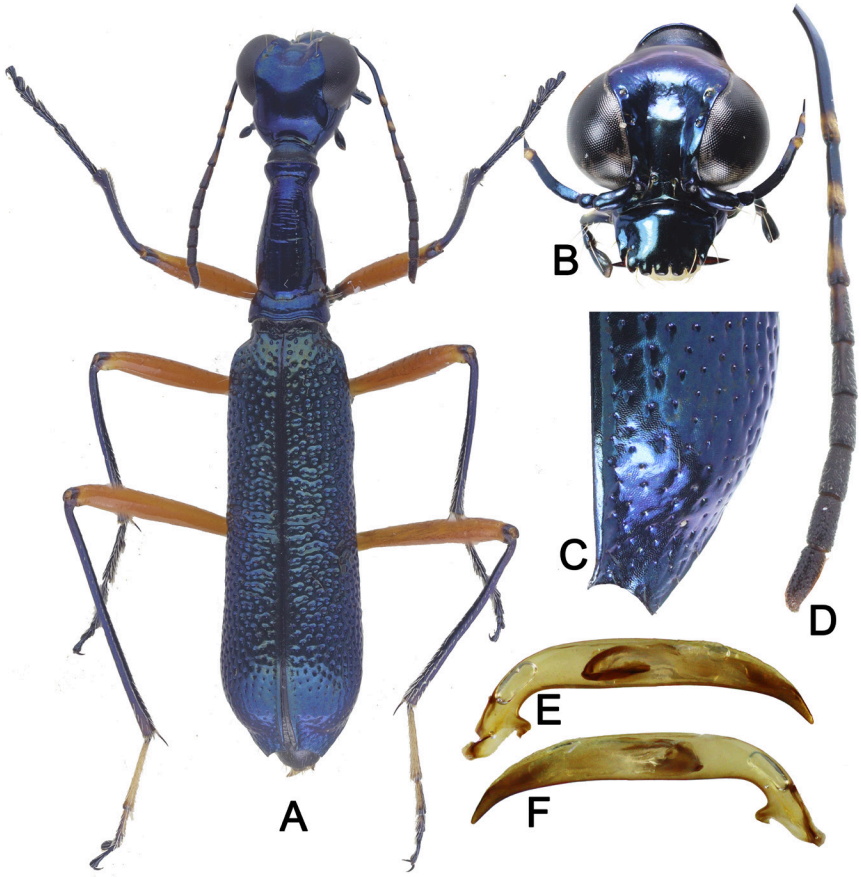

Figure 1. [A] Habitus of *Neocollyris anichtchenkoi* sp. nov. sp; [B] frons; [C] apical third of elytra showing the apex; [D] enlarge view of the antennae; [E–F] aedeagus on both lateral sides.

Nalsian Centro, 29.v.2022, D. Mapile leg. (DMC); **1 female,** PHILIPPINES: Luzon, Pangasinan, Calasiao, Brgy. Nalsian Centro, 15.vii.2023, D. Mapile leg. (DMC). Each specimen also bears a red label under the locality label that states: "Holotype [or Paratype] / *Neocollyris (Neocollyris) anichtchenkoi* / Medina & Mapile, sp. nov." Eggs and larvae desribed herein are in DMC. All materials will be deposited at PNM.

Description

Measurements. Dimensions, holotype male: [BL] 11.0 mm, [LCE] 1.0 mm, [WCE] 0.5 mm, [LL] 1.0 mm, [WL] 1.0 mm, [LM] 1.0 mm, [LP] 2.5 mm, [WP] 1.2 mm, [LE] 7.0 mm, [WE] 2.0 mm, [LA] 2.0 mm, and [WA] 0.3 mm.

Male (integument). Head, prothorax, elytra, and abdomen metallic blue, with prothorax more lustrous and darker; femora light brown; tibia plus front and middle tarsi metallic blue, with hind tarsi first and second tarsomeres whitish and the rest of the segments metallic blue.

Head. Base narrow, strongly sinuate; vertex glabrous, slightly rugose with dense very fine punctuation; dorsally, a steep carina between vertex and near basal margin; with two supraorbital setae, one near margin between vertex and frons, another at the side of frons; clypeus metallic blue to metallic green, with two setae near apical margin; eyes twice longer than wide, matte black lined with white irregular lines, broader at front; mentum yellowish, parallel elongate; genae as long as wide; labrum raised dorsally, as long as wide, with two sharp teeth at the sides, five blunt teeth at the middle, with seven to eight long yellowish setae arranged in between teeth; mandibles lustrous, darkly colored, slightly recurved, and a little longer than labrum.

Antenna. Almost reaching basal margin of pronotum (~ 4 mm); scape enlarged with long yellowish setae at apex, covered with very fine micro-sculpture, almost invisible; pedicel almost rounded, glabrous, short; antennomere III flat, recurved near the middle, individually longer than antennomeres IV and V, lined with few short setae at the sides, apex with short yellowish maculation; antennomeres IV and V semi-flattened to semi-rounded, almost same length, covered with few short erect yellowish setae, with yellowish maculation from the

middle towards apex; antennomeres VI to X dark brown, more thickened, rounded, and elongate, densely covered with short semi-erect whitish setae; antennomere XI shorter than antennomere X, densely covered with whitish semi-erect setae, lanceolate towards the apex.

Prothorax. Bottle-shaped, narrower at apex, widest at base; highly sinuate near apical margin; covered with long erect whitish setae concentrated at propleuron and prosternum; pronotum with few erect setae at the sides.

Elytra. More than thrice longer than wide, with dense deep punctures from the base, finer near elytral declivity towards apex; narrower at the base, widest near apex; with a tinge of metallic red at humeri and very faint transverse metallic red line at the middle from lateral margin almost reaching suture; apex caudate with a short sutural spine; with few long erect whitish to yellowish setae arranged randomly.

Mesosternum lustrous metallic green with a tinge of metallic blue, densely covered with long whitish setae. Mesepimeron parallel, elongate, with deep puncture near the humeral angle, covered with fine erect whitish setae. Metepisternum parallel widest at the base, with few semi-erect whitish setae lining the apical margin, with elongated depression near the elytral margin. Metasternum slightly raised at midline, lustrous bluish green, with semi-erect whitish setae along the apical margin. Procoxa raised, elongate, lustrous metallic green, with few erect whitish setae at base; mesocoxa and metacoxa not raised. Trochanter of front legs with one erect whitish seta, absent in both middle and hind legs. Femora of front and middle legs with few erect whitish setae, absent in femora of hind legs. Tibia with two tibial spurs, sparsely covered with semi-erect whitish setae, concentrated at the apex; hind tibia apex with whitish maculation. Tarsi of front and middle legs lustrous metallic blue, densely covered with short whitish semi-erect setae; first and second tarsomeres of hind legs whitish, the rest dark metallic blue, densely covered with recumbent whitish setae. Claws are simple, lustrous black.

Abdominal ventrites sparsely covered with thin long erect whitish setae, majorly at the middle; ventrites I-IV metallic blue with a tinge of metallic green at the midline; ventrite V metallic blue. Aedeagus six to seven times longer than wide, slightly sinuate at the base, parallel-sided, elongated, widest near the middle, slowly tapering towards the apex.

Female. There are no significant morphological differences between the male and female beetles except the females are a little larger, have shorter antennae, and have a slight hump near the apex of the pronotum.

Differential diagnosis. This new species belongs to the sub-genus *Neocollyris* s. str. and is similar to *Neocollyris (Neocollyris) albitarsi* (Erichson, 1834) by having whitish tarsomeres but differs in having thicker antennomeres and prominent sutural spine. The new species is also similar to *N. emarginata* (Dejean 1825) in terms of body form but can be easily distinguished by the unique shape of the basal part of the head. The aedeagus is distinct from both close congeners (Figures 1E and F).

Etymology. This magnificent new species of *Neocollyris* is named after our good friend and colleague, Dr. Alexander Anichtchenko of Daugavpils University, a world-leading carabidologist who made a significant contribution to the advancement of the taxonomy of tiger beetles and ground beetles in the Philippines and the world.

Distribution. Philippines (Luzon: Pangasinan).

LARVAL DESCRIPTION

Oviposition (Figures 2A1–2). The female *Neocollyris* (*N.*) *anichtchenkoi* sp. nov. carefully selects the softer branch for oviposition, usually about 4–6 mm in diameter. Most of the oviposition is done about 5–36 mm from the nodes of *Premna odorata* Blanco. Using the ovipositor, the female starts to make a circular hole usually between 1.0–2.0 mm in diameter. Based on 23 observed egg and larval cavities, females oviposit singly in excavated cavities.

Ovum (Figure 2B). Mean measurements (n = 8): length 1.9 mm, maximum width 1.0 mm. The egg is yellowish with a transparent chorion sometimes visible, and elliptic to ovate in shape. As the larva develops inside, the egg turns entirely yellow with four dark spots that are its eyes. The exact duration was not determined, but a collected egg hatched after a week.

First instar larva (Figure 2C1-2). Mean measurements (n = 5): body length 5.1 mm, length of head 0.9 mm, maximum width of head 0.9 mm, length of pronotum 0.6 mm, maximum width of pronotum 1.0 mm. The first instar larva is entirely yellow when freshly hatched except for its dark eyes. The head begins to sclerotize starting from the mandibles. It uses its mandibles to create a conspicuous opening where it can fit its head and pronotum for ambush hunting and to further excavate a tunnel in the soft tissue in the direction of plant growth. The head and pronotum are large, sclerotized structures covered in goldish erect setae, with the pronotum more setaceous. Mandibles are relatively shorter and wide. The rest of the body is composed of soft tissues with a ring of golden erect setae at each abdominal segment. Tergite V bears three short denticles on both sides surrounded by golden erect setae. Tergite X with minute denticles.

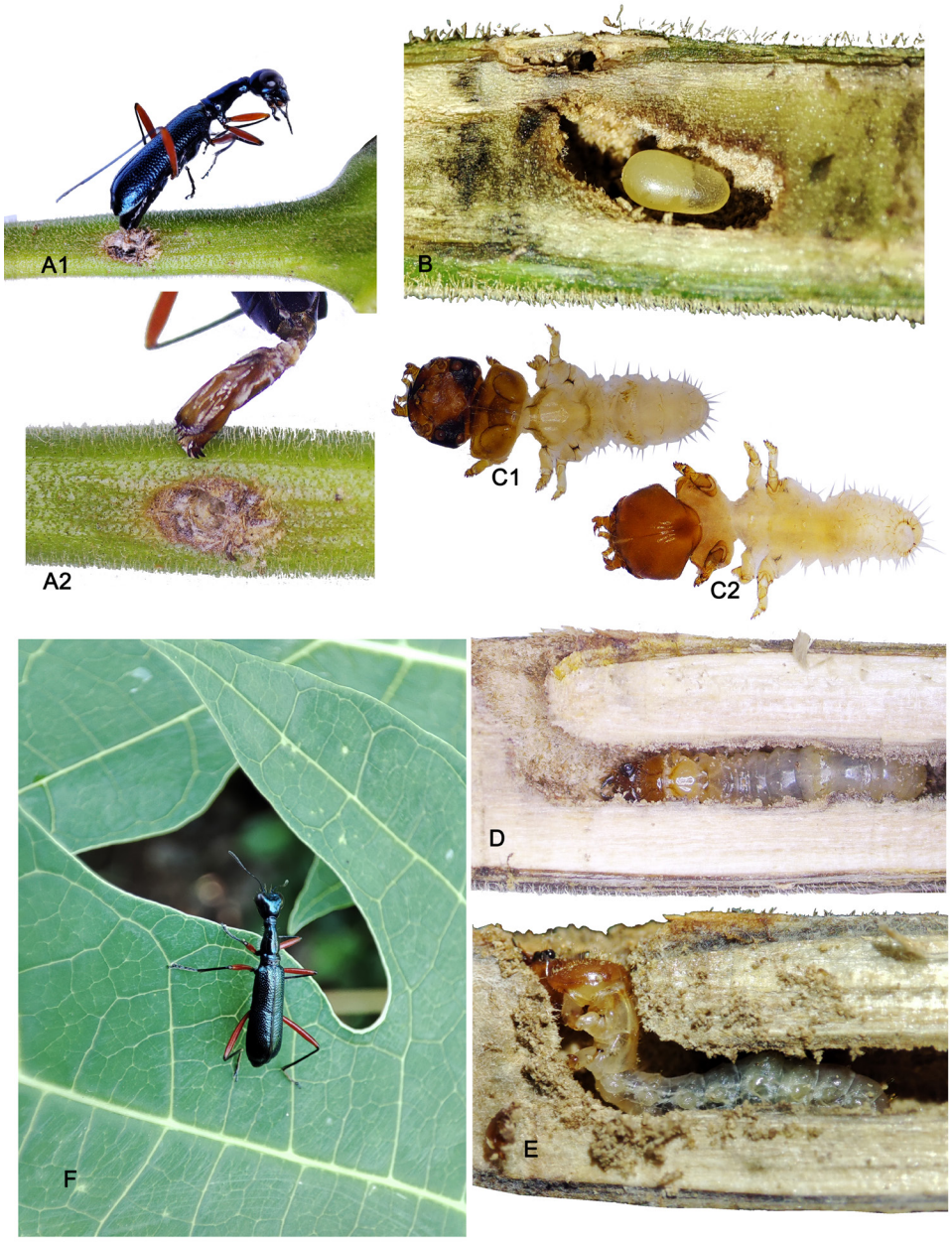

Figure 2. [A1–A2] Deposition of female *Neocollyris anichtchenkoi* sp. nov. in *Premna odorata* Blanco; [B] position of the egg inside the tunnel in *P. odorata* Blanco; [C1–C2] first instar, dorsal, and ventral aspects respectively; [D] between 2nd and third larval instar resting inside the tunnel in *P. odorata* Blanco; [E] larvae in ambush position; [F] fully grown adult *in situ*.

Second instar larva (Figure 2D). Mean measurements (n = 4): body length 8.9 mm, length of head 1.4 mm, maximum width of head 1.3 mm, length of pronotum 0.9 mm, and maximum width of pronotum 1.5 mm. The second instar larva is similar to the preceding instar but larger in size and has two setae on the inner margin of the galea.

Third instar larva (Figure 2E). Measurements (n = 1): body length 9.0 mm, length of head 1.8 mm, maximum width of head 2.0 mm, length of pronotum 1.2 mm, and maximum width of pronotum 2.0 mm. The third instar larva is similar to the preceding instar but larger in size and has three setae on the inner margin of the galea. The setae covering the head and pronotum are more evident and longer.

Pupae

No observations were made. Adult (Figure 2F). Like its congeners, N. (N.) anichtchenkoi sp. nov is an active predatory species. Most adult sightings were on P.

odorata and nearby foliage where they most likely hunt due to the abundance of insects inhabiting or feeding on P. odorata. Adults are mostly active in the morning and have only been sighted from May–June after three years of observation, coinciding with the start of the wet season. Both male and female beetles can be seen skittishly walking on a leaf before flying to the next one, and seldom seen running on the stems, and have never been observed on the ground. They are easily spotted due to their conspicuous metallic appearance, but they are difficult to document and capture due to their good eyesight and skittish tendencies, often resulting in swift flight to higher foliage when approached.

Habitat. *In situ* observations came from a residential lot in Calasiao, Pangasinan in Luzon, Philippines. There are two mature P. *odorata* that are used by *N.* (*N.*) *anichtchenkoi* sp. nov. as larval hosts. The surrounding vegetation, comprising of intentionally planted fruit trees and vegetables [*e.g. Annona squamosa* L. (Annonaceae)*, Citrus* × *microcarpa* Bunge (Rutaceae)*, Persea americana* Mill. (Lauraceae)*, Annona muricata* L. (Annonaceae)*, Solanum melongena* L. (Solanaceae)*, Capsicum* sp. (Solanaceae), *etc.*] and unintentionally planted weeds, shrubs, and trees [*e.g.* grasses (Poaceae), *Ficus* sp. (Moraceae), *P. odorata*], provides habitat and attracts various insects, which have supported the *N.* (*N.*) *anichtchenkoi* sp. nov. population. Unlike forest-dependent species, *N.* (*N.*) *anichtchenkoi* sp. nov. appears to be tolerant of human disturbance, considering that most

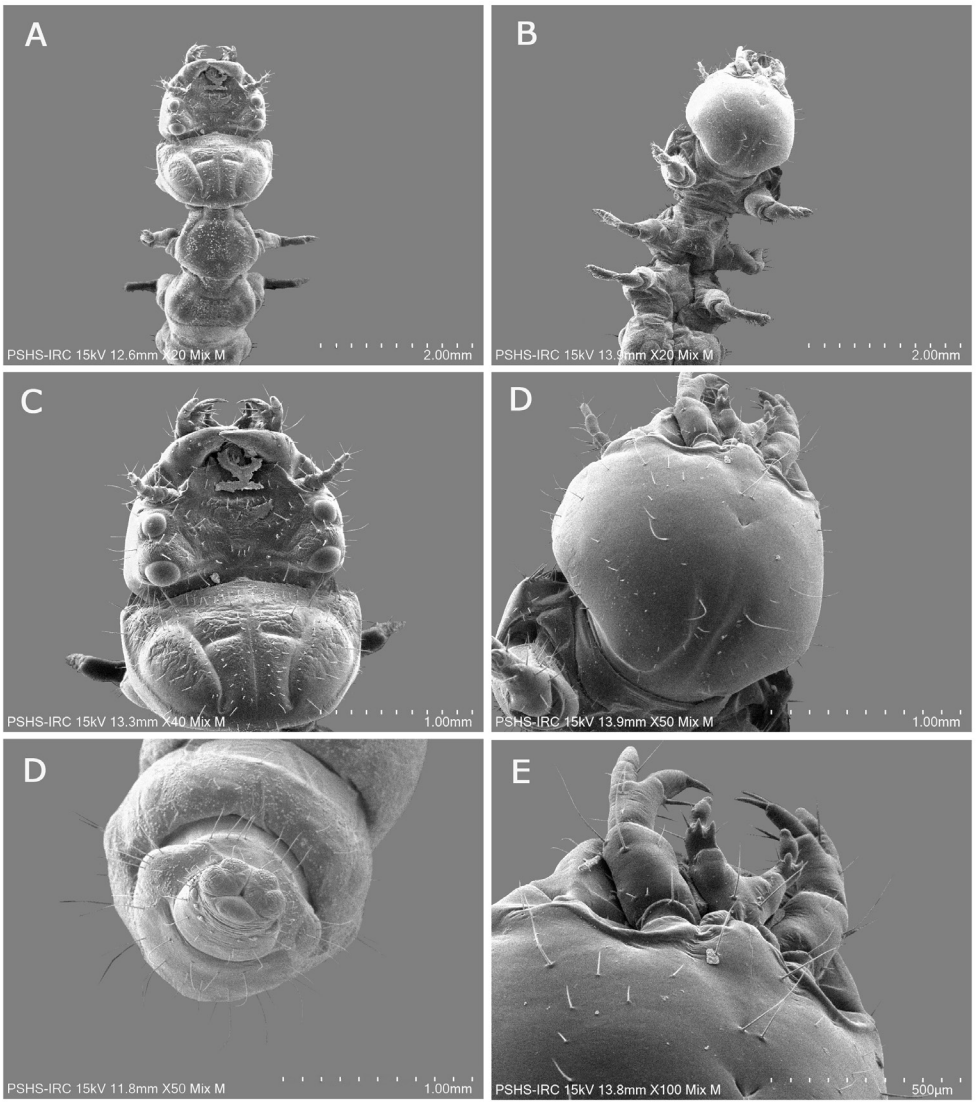

Figure 3. SEM images of the larvae: [A] dorsal aspect of the second instar; [B] ventral aspect of the second instar; [C] head, dorsal; [D] head, ventral; [D] pre-developed pygidium and anal appendages; [E] mouthparts, ventral aspect.

of Pangasinan is highly disturbed and cleared of forests.

Remarks. Unlike the ground-burrowing larvae of most species, the larval stages have relatively short legs and inhabit thin branches. These significantly restrict their locomotion, developing entirely in the cavity where they were oviposited. Thus, the placement of the eggs by the female beetle is crucial for the survival of the larvae.

Larvae feeding behavior (S1–S2). Larval feeding behavior is based on *in situ* and rearing observations. The larvae are ambush predators where they protrude their heads flush with the tunnel opening and widely open their mandibles. *Premna odorata* attracts various insects that can be seen nectaring on its flowers or running across its branches (pers. obs.). This makes it a suitable hunting ground for both larval and adult tiger beetles. Larvae have been observed to naturally prey upon flies (Diptera) and ants (*Crematogaster* sp.) (Hymenoptera: Formicidae). Once it has caught prey, the larva retreats to its tunnel, where it chews its prey using its jaws and maxilla. It spews a brown liquid to further macerate the prey into a bolus. It drinks the juices from the bolus while it further masticates and macerates the prey with the brown liquid. After that, it holds still while drinking the juices from the bolus. The larva then expels the leftover material out of the opening.

ACKNOWLEDGMENT

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STATEMENT ON CONFLICT OF INTEREST

The authors declare that there is no conflict of interest during the preparation of this manuscript.

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APPENDIX

Video S1. *Neocollyris anichtchenkoi* sp. nov. feeding behavior on offered fruit fly. An immobilized fruit fly was fed to a third instar larva which it actively pursued and consumed. Youtube link: https://www.youtube.com/watch?v=T-9PAB7pcbU

Video S2. *Neocollyris anichtchenkoi* sp. nov. feeding behavior on naturally caught ant (*Crematogaster* sp.). A collected larval tunnel was longitudinally cut, which exposed a beetle larva in the middle of consuming an ant (*Crematogaster* sp.). Youtube link: https://www.youtube.com/watch?v=C2GfXpcOPNU