Description of a New Cruranthura Species from the Miyako Islands, Japan, with Its DNA Barcode (Isopoda: Anthuroidea: Paranthuridae)

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We describe *Cruranthura viridis* sp. nov. based on four specimens collected from the subtidal zone near Irabu Island, Miyako Islands, Okinawa, Japan. This species differs from four congeneric species in having (1) small body size (2.53– 3.38 mm), (2) moss green body color, (3) eyes, (4) pleonite 1 longer than pleonite 2, (5) telson length 2.06–2.33 times telson width, (6) propodus of pereopod 1 with 6–10 inner spiniform setae, and (7) the endopods of pleopods 2–5 uniarticulate, with plumose setae. We also determined the partial sequence of the cytochrome *c* oxidase subunit I (COI) gene from the holotype specimen. We discuss differences in piercing mouthparts among paranthurid genera.

Key Words: Califanthura, Colanthura, coral rubble, Crustacea, Cymothoida, mandible, mouthparts, SEM images.

Introduction

The paranthurid genus Cruranthura Thomson, 1946 differs from the other six confamilial genera by the combination of following characters: (1) percopod 7 absent in nonmanca individuals, (2) pleonite 6 delineated dorsally from an oval telson, (3) the head longer than wide, and (4) pleonites 1 free and 2-5 fused dorsally (Poore 2001; Shiraki et al. 2022). The genus currently contains four valid species: C. caeca (Mezhov, 1976), known from the Kurile Islands, Russia (Mezhov 1976); C. furneauxi (Poore, 1981), occurring in Tasmania and New South Wales, Australia (Poore 1981, 1984); C. peroni (Poore, 1981), occurring in Brisbane, Queensland, and New South Wales, Australia (Poore 1981, 1984); and the type species C. simplicia Thomson, 1946, occurring in Western Australia (Thomson 1946). The first two species occupy intertidal or subtidal marine habitats, whereas the latter two are estuarine.

Here we describe a new *Cruranthura* species collected subtidally around Irabu Island, Miyako Islands, Okinawa, Japan and sequenced part of its cytochrome c oxidase subunit I (COI) gene. We also discuss differences among paranthurids in the specialized mouthparts used for piercing.

Materials and Methods

Four individuals were collected from coral rubble at $10-20 \,\text{m}$ depth on the north reef of Irabu Island

(24°51'57.7"N 125°09'41.9"E), Miyako Islands, Okinawa, southwestern Japan, on 25 April 2022. They were photographed and fixed in 70% ethanol on 25 April 2022, and transferred to 99% ethanol on 12 May 2022. The methods used for dissection, preparation of slides, light microscopy, and drawing were as described in Shiraki et al. (2021). One ovigerous female was bisected into anterior and posterior parts in ethanol, treated with hexamethyldisilazane (HMDS; Nation 1983), and observed at 15kV accelerating voltage with an S-3000N scanning electron microscope (SEM; Hitachi, Tokyo, Japan). Body length was measured from the tip of the anterolateral lobe of the head to the tip of the telson, and body width at the widest portion of pereonite 6. Measurements were made axially from digital images by using Adobe Illustrator CC and ImageJ (http://rsb.info.nih.gov/ ij). The specimens examined have been deposited in the Invertebrate Collection of the Hokkaido University Museum (ICHUM), Sapporo, Japan.

DNA was extracted from right pereopod 1 of the holotype by using a NucleoSpin Tissue XS Kit (Macherey-Nagel, Germany). The COI sequence was amplified by PCR with primers LCO1490 and HCO2198 (Folmer et al. 1994) under the following conditions with KOD One (Toyobo, Japan): 45 cycles of 98°C for 10 seconds, 56°C for 5 seconds, and 68°C for 1 second. The nucleotide sequence was determined by direct sequencing with a BigDye Terminator Kit ver. 3.1 and an ABI 3730 Genetic Analyzer (Life Technologies, USA). The COI sequence was deposited in the International Nucleotide Sequence Database (INSD) through the DNA Data Bank of Japan.

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Taxonomy

Superfamily **Anthuroidea** Leach, 1814 Family **Paranthuridae** Menzies and Glynn, 1968 Genus **Cruranthura** Thomson, 1946

Type species. *Cruranthura simplicia* Thomson, 1946, fixed by monotypy.

Diagnosis. See Poore (2001).

Cruranthura viridis sp. nov.

[New Japanese name: Kappa-ashitarazu-uminanafushi] (Figs 1–5)

Diagnosis. Body small (length 2.53–3.38 mm) and moss green in color. Eyes present. Pleonite 1 longer than pleonite 2. Telson length 2.06–2.33 times telson width. Propodus of pereopod 1 with 6–10 spiniform setae. Endopods of pleopods 2–5 uniarticulate, with plumose setae.

Etymology. The specific name is from the Latin adjective *viridis* (green), referring to the moss green body color.

Material examined. Holotype: ovigerous female (ICHUM8565, 8 slides and 1 vial), body length 3.08 mm; coral rubble, 10–20 m depth, Irabu Island (24°51′57.7″N 125°09′41.9″E), Miyako Islands, Okinawa, southwestern Japan, 25 April 2022, collected by Shoki Shiraki. Paratypes: two ovigerous females, ICHUM8566 (body length 3.38 mm; 12 slides and 1 vial) and ICHUM8567 (body length 2.80 mm; 1 slide and 1 vial), and one female lacking oostegites, ICHUM8568 (body length 2.53 mm; 2 slides and 1 vial); collection data for paratypes as for holotype.

Sequence. Partial COI sequence (658 bp, encoding 219 amino acids) from holotype (INSD accession number LC775393).

Description of female based primarily on holotype. Body (Figs 1A-D, 2A, B) color moss green, length 11.54 times body width, slender. Head (Fig. 2C) length 1.35 times head width; rostrum not extending past anterolateral lobes; eyes dorsolateral, small, black; eye length 0.13 times head length. Pereonites 1–7 (Fig. 2A) with length ratios 1. 18:1.07:1.19:1.25:1.22:0.78:0.15 relative to head length; pereonite 7 (Figs 2A, B, 5C, D) reduced, not hidden laterally but rarely visible, lacking pereopod 7 in adults. Pleonite 1 free; pleonites 2-5 articulated laterally but fused dorsally and indicated by folds (Figs 2A, B, 5C, D); pleonite 1 1.72 times as long as pleonite 2. Pleonite 6 delineated dorsally, 0.53 times as long as combined length of pleonites 1-5, delineated dorsally from telson. Telson (Fig. 4H) oval, length from basal narrowest point to the tip 2.12 times width, with six dorsal simple setae and twelve distal simple setae.

Antennula (Fig. 2D) with three peduncular articles and one flagellar article. Peduncular article 1 longest, with two outer plumose sensory setae; article 2 with two distal plumose sensory setae and distal simple seta; article 3 with distal plumose sensory seta and three distal simple setae. Flagellar article with four distal aesthetascs and three distal simple setae. Antenna (Fig. 2E) with five peduncular articles and one flagellar article. Peduncular article 1 naked; article 2 longest, with two distal simple setae and distal seta (tip broken); article 3 with two distal simple setae; article 4 with two distal plumose sensory setae and two simple setae; article 5 with outer plumose sensory seta, seven simple setae, and distal seta (tip broken). Flagellar article with numerous distal simple setae.

Mandible (Fig. 5A, B) not articulated with head, without palp.

Maxilla (Fig. 2F) slender, with 14 teeth and narrow lamel-la.

Maxilliped (from paratype ICHUM8566; Figs 2G, 5A, B) biarticulate; article 1 (presumably corresponding to basis and palp, except for terminal palp article) with seven simple setae; article 2 minute, with four simple setae. Epipod (Fig. 5A, B) oval.

Pereopod 1 (Fig. 3A) subchelate. Basis with dorsal plumose sensory seta and one dorsal and one ventrodistal simple setae. Ischium with ventrodistal simple seta. Merus with two dorsal and two ventrodistal simple setae. Carpus triangular, with five ventral simple setae. Propodus broad, with 10 inner spiniform setae and one outer and two dorsodistal simple setae. Palm with proximal trapezoidal projection and 10 simple setae. Dactylus with three ventral and four inner simple setae. Unguis naked, length about 0.4 times dactylus length.

Pereopod 2 (Fig. 3B) subchelate, narrower than pereopod 1. Basis with two dorsal plumose sensory setae, four simple setae, and dorsal seta (tip broken). Ischium with two ventral simple setae. Merus with two dorsal and three ventral simple setae. Carpus triangular, with three ventrodistal simple setae. Propodus oval, with dorsodistal plumose sensory seta and two dorsodistal simple setae. Palm with five spiniform setae with sensory bristle sensu Negoescu (1994) and three simple setae. Dactylus with two ventral, three ventrodistal, and four inner simple setae. Unguis naked, length about one-third dactylus length.

Pereopod 3 (Fig. 3C) similar to pereopod 2 except in number of simple setae.

Pereopod 4 (Fig. 3D) narrow. Basis with three dorsal plumose sensory setae and three simple setae. Ischium with two ventral simple setae. Merus with two dorsal and two ventrodistal simple setae. Carpus trapezoidal, with two ventral spiniform setae with sensory bristle, dorsal plumose sensory seta, and one dorsal and two ventrodistal simple setae. Propodus with two ventral spiniform setae with sensory bristle, dorsodistal plumose sensory seta, and two dorsodistal and three ventral simple setae. Dactylus with one ventral, three ventrodistal, and three outer simple setae. Unguis naked, length about one-third dactylus length.

Pereopod 5 (Fig. 3E) similar to pereopod 4 except in number of plumose sensory setae and simple setae on basis.

Pereopod 6 (Fig. 3F) similar to pereopod 4 except in number of plumose sensory setae on basis and simple setae on merus and propodus. Propodus with two dorsodistal trifurcate spiniform setae.

Pleopod 1 (Fig. 4A) protopod with three inner hooks.



Fig. 1. *Cruranthura viridis* sp. nov., holotype, ovigerous female. A, B, Dorsal and right views of living individual; C, D, dorsal and left views of fixed specimen. Scale bars: 1 mm.

Exopod operculiform, with sinuate outer distal margin and 13 plumose setae (possibly 14; one pore observed); three simple setae on surface. Endopod with six plumose setae.

Pleopod 2 (Fig. 4B) protopod with two inner hooks. Exopod with five plumose setae and outer simple seta. Endopod uniarticulate, with three plumose setae.

Pleopods 3–5 (Fig. 4C–E) similar. Protopods with one or two simple setae. Exopods with four plumose setae and outer simple seta. Endopods uniarticulate, with three plumose setae.

Uropod (Fig. 4F, G) with triangular-prism-shaped protopod bearing dorsodistal simple seta, and three inner ventral and four outer ventral plumose setae. Exopod (Fig. 4F) broad, oval, with 24 plumose setae (possibly 25; one pore observed), seven simple setae, and two setae (tip broken). Endopod (Fig. 4G) with eight plumose sensory setae, three plumose setae, 11 simple setae, and five setae (tip broken).

Variation. Table 1 indicates differences in length measurements and ratios among the four specimens examined. All specimens shared the following characters: (1) the body color was moss green; (2) eyes were present; and (3) pleonite 1 was longer than pleonite 2. The ratio of telson length to telson width ranged from 2.06 to 2.33 among three specimens (it could not be measured for ICHUM8567,

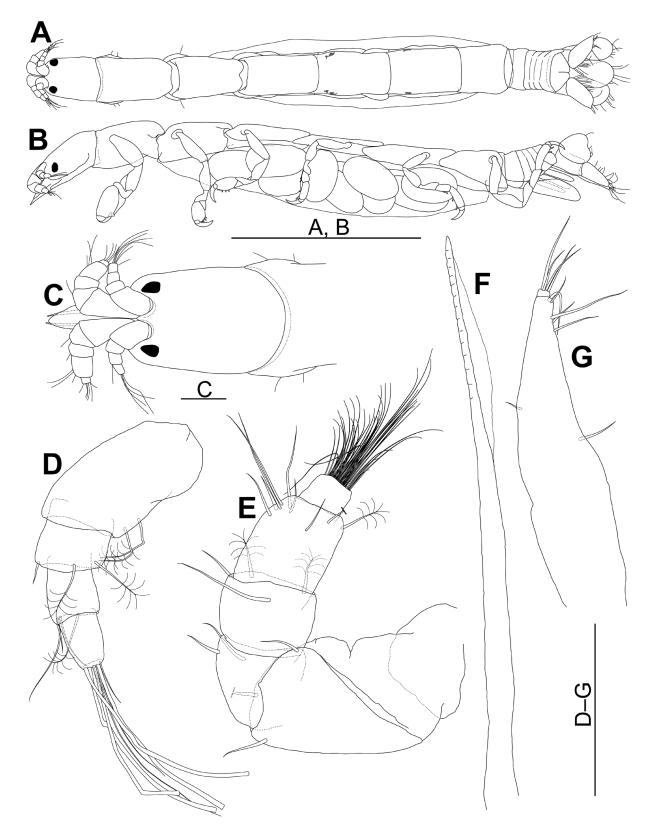


Fig. 2. *Cruranthura viridis* sp. nov., ovigerous females, holotype (A–F), and paratype (ICHUM8566: G). A, B, Body, dorsal and lateral views; C, dorsal view of head; D, right antennula; E, right antenna; F, left maxilla; G, maxilliped. Scale bars: A, B, 1 mm; C–G, 100 μm.

which was used for SEM observation). The number of spiniform setae on the propodus of pereopod 1 ranged from 6 to 10 among seven pereopods of four specimens (it was not counted for right pereopod 1 of the holotype, which was used for DNA extraction). **Genetic information.** In BLAST searches (Altschul et al. 1990), the COI sequence most similar to the holotype COI sequence was from the insect "Cecidomyiidae sp." (INSD accession number OM561009; identity score 80.66%, query cover 99%; Bukowski et al. 2022) rather than from a

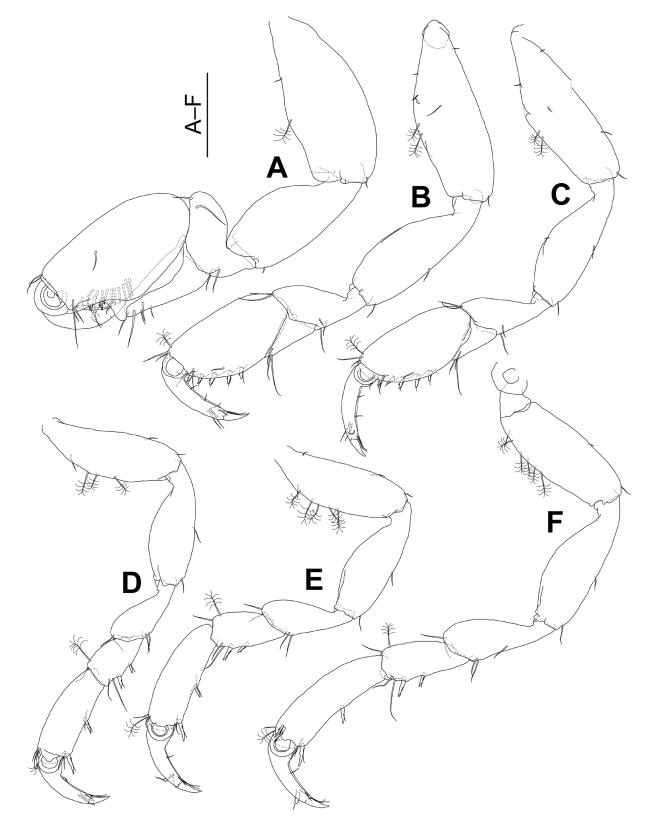


Fig. 3. Cruranthura viridis sp. nov., holotype, ovigerous female. A-F, Left pereopods 1-6. Scale bar: 100 µm.

paranthurid, though there are several paranthurid COI sequences in the database. In a BLAST search with the "Organism" option set to "Anthuridea", the sequence most similar to ours was from *Colanthura pigmentata* Kensley, 1980 (KR095339; identity score 81.03%, query cover 88%; Song and Min 2015). **Remarks.** *Cruranthura viridis* sp. nov. differs from congeners in the suite of characters listed in Table 2. This paper is the first report of *Cruranthura* from Japan.

All paranthurid isopods have piercing-type mouthparts specialized for sucking (Poore 2001), but the degree of specialization differs among genera. We can roughly divide

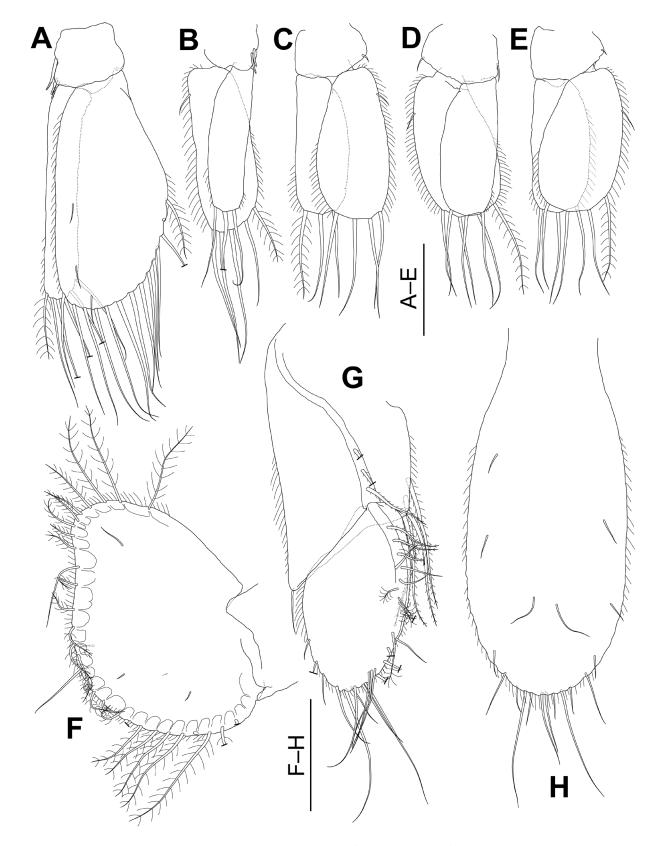


Fig. 4. *Cruranthura viridis* sp. nov., holotype, ovigerous female. A–E, Left pleopods 1–5; F, left uropodal exopod; G, right uropodal endopod; H, telson. Scale bars: 100 μm.

them into two groups: a moderately specialized group (*Del-tanthura* Shiraki, Shimomura, and Kakui, 2022, *Paranthura* Bate and Westwood, 1866, and *Pseudanthura* Richardson, 1911); and a highly specialized group (*Califanthura* Schultz,

1977, *Colanthura* Richardson, 1902, *Cruranthura*, and *Cruregens* Chilton, 1882). The mouthparts in the former group retain the mandibular palp, the articulation between the mandible and head, and the articulation between the maxil-

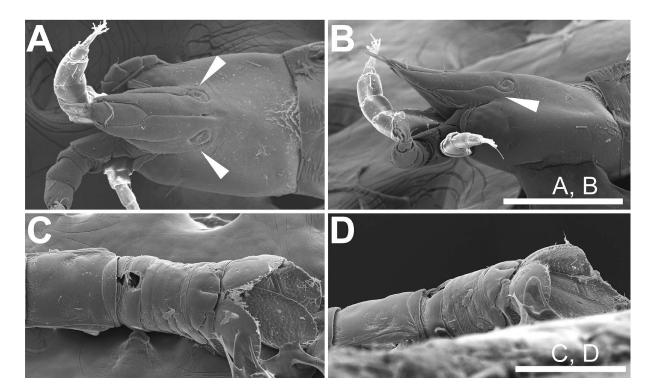


Fig. 5. SEM images of *Cruranthura viridis* sp. nov., paratype (ICHUM8567), ovigerous female, pereopods detached. A, B, Ventral and right views of head; C, D, dorsal and left views of pleon (the hole on pleonite 1 was made by an accidental needle prick during dissection). Arrowheads, the fusion between the mandible and head. Scale bars: A, B, 200 µm; C, D, 300 µm.

Table 1. Length measurements and ratios for the type specimens of *Cruranthura viridis* sp. nov. BL, body length; BW, body width; HL, head length; HW, head width; EL, eye length; P1L–P7L, lengths of pereonites 1–7; p1L–p6L, lengths of pleonites 1–6.

Collection No.	BL (mm)	BW (mm)	BL/BW	HL/HW	EL/HL	HL:P1L:P2L:P3L:P4L:P5L:P6L:P7L:p1-5L ^a):p6L	p1L/p2L
ICHUM8565	3.08	0.27	11.54	1.35	0.13	1.00:1.18:1.07:1.19:1.25:1.22:0.78:0.15:0.62:0.33	1.72
ICHUM8566	3.38	0.27	12.52	1.38	0.16	1.00:1.19:1.08:1.18:1.29:1.30:0.96:0.13:0.66:0.31	1.89
ICHUM8567	2.80	0.24	11.64	1.37	0.14	1.00:1.12:1.02:1.12:1.14:1.11:0.67:0.15:0.65:0.29	1.75
ICHUM8568	2.53	0.22	11.54	1.35	0.14	$1.00\!:\!1.08\!:\!1.02\!:\!1.11\!:\!1.16\!:\!1.05\!:\!0.74\!:\!0.15\!:\!0.65\!:\!0.31$	2.02

^{*a*)} Aggregated length of pleonites 1–5.

Table 2. Comparison of selected characters among *Cruranthura* species. BL, body length; p1L, p2L, lengths of pleonites 1 and 2; TL, telson length; TW, telson width; —, no data.

Character	C. caeca	C. furneauxi	C. peroni	C. simplicia	C. viridis sp. nov.	
BL (mm)	7.5-10.8	5.2-9.1	4.5-9.5	5.25-6.00	2.53-3.38	
Color	_	Red brown	Red brown	a)	Moss green	
Eye	Absent	Present	Present	Present	Present	
Relative lengths of p1L and p2L	p1 = p2	p1 <p2< td=""><td>p1>p2</td><td>_</td><td>p1>p2</td></p2<>	p1>p2	_	p1>p2	
TL/TW	2.5	2.5	2.2	2.0	2.1-2.3	
Number of spiniform setae on propodus of pereopod 1	Ca. 35 ^{b)}	Ca. 35	Up to 25	9–12	6-10	
Endopods of pleopods 2-5	Uniarticulate	Uniarticulate	Biarticulate	Biarticulate	Uniarticulate	
Setae on endopods of pleopods 2-5	Present	Present	Absent	Absent	Present	
Habitat	Marine	Marine	Estuarine	Estuarine	Marine	
References	Mezhov (1976), Poore (1984)	Poore (1981, 1984)	Poore (1981, 1984)	Thomson (1946), Poore (1984)	This study	

^{a)} Scattered, darkly pigmented patches present on head, but information on body color lacking (Thomson 1946).

^{b)} Counted from Mezhov (1976: fig. 4).

lipedal basis and palp (cf. Shiraki and Kakui 2022: fig. 1B). The mouthparts in the latter group lack these three structures (cf. Fig. 5A, B; Annisaqois and Wägele 2021: fig. 31). A piercing apparatus that is fixed on the head and lacks joints and unnecessary structures (such as the mandibular palp) will have higher stability and strength, which may enable species in the latter group to pierce more strongly and may have increased the range of possible prey in the group. Poore (2001) suggested that genera in the moderately specialized group (information on the phylogenetic position of *Deltanthura* in Paranthuridae is unavailable) are more basal within Paranthuridae than genera in the highly specialized group. If this is true, the specialization of piercing mouthparts may have steadily increased in the evolution of Paranthuridae. Molecular phylogeny can test this hypothesis.

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Authors Contributions

Shoki Shiraki: Conceptualization; Resources; Investigation; Visualization; Writing – original draft; Funding acquisition. Keiichi Kakui: Supervision; Writing – original draft; Writing – review & editing.

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Declarations

Competing interests. The authors declare no conflicts of interest.

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