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A new species of vent associated *Munidopsis* (Crustacea: Decapoda: Anomura: Galatheidae) from the Western Pacific, with notes on its genetic identification

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Abstract

Munidopsis myojinensis **n**. **sp.**, herein described and illustrated from Myojin Knoll and Northwest Eifuku Seamount in the Western Pacific, is the tenth species of *Munidopsis* known to be associated with active hydrothermal vents. It resembles *M. starmer* Baba & de Saint Laurent, 1992 in general ornamentation and spination of the body but differs in the erectness of the eyespine and form of the telson plates. Information on genetic identification using mitochondrial sequences (COI) is provided. Distributions of the vent associated *Munidopsis* in the Western Pacific and the habitat of the new species are briefly described.

Key words: Anomura, Galatheidae, Munidopsis, Hydrothermal vent, Myojin Knoll, Eifuku Seamount

Introduction

Species of *Munidopsis* (Crustacea: Galatheidae) are found in a variety of shallow to deepwater habitats throughout the Atlantic, Indian and Pacific Oceans (Ambler 1980, Baba 2005, Chevaldonnè & Olu 1996, and Macpherson, in press). Since the first discovery of hydrothermal vents on the Galapagos Rift in 1977 (Corliss *et al.* 1979), numerous biological communities associated with vents have been reported (Desbruyères *et al.* 1982, 1994; Hessler & Lonsdale 1991; Tunnicliffe 1991; Hashimoto *et al.* 1989), among which are species of *Munidopsis* (see Williams 1988). Species of *Munidopsis* are also known from other reducing environments such as cold seeps and whale carcasses (Chevaldonnè & Olu 1996; Martin & Haney 2005; Macpherson, in press).

The genus *Munidopsis* is represented by more than 150 species in the Indo-Pacific and around 70 species in Atlantic Ocean with nine from active hydrothermal vent systems in the world oceans (Williams 1988; Williams & Baba 1989; Baba & de Saint Laurent 1992; Baba 1995, 2005; Schnabel & Bruce 2005; Macpherson & Segonzac 2005; Osawa *et al.*, 2006a, b; Macpherson, in press). To date, four species of *Munidopsis* have been described from Western Pacific hydrothermal vents: *M. marianica* Williams & Baba, 1989 from Mariana Back Arc Basin, *M. lauensis* Baba & de Saint Laurent, 1992 from Lau and North Fiji Basins, *M. starmer* Baba & de Saint Laurent, 1995, from North Fiji Basin respectively. Three species are

reported from Eastern Pacific vent fields: *M. alvisca* Williams, 1988, and *M. lentigo* Williams & Van Dover, 1983 (see Martin & Haney 2005; Desbruyères *et al.* 2006), and *Munidopsis* sp. near *M. recta* Baba, 2005 (the material reported by Van Dover *et al.* (1985) under *M. subsquamosa* Henderson, 1885; see Baba (2005); Macpherson & Baba in Desbruyères *et al.* 2006)). Two species, *M. acutispina* Benedict, 1902 and *M. exuta* Macpherson & Segonzac, 2005 are reported from Atlantic hydrothermal vents.

Molecular phylogenetic data for the Galatheidae are scarce and the only published studies are as follows: study on *Munidopsis* species based on protein electrophoresis (Creasy *et al.* 2000), analysis of mitochondrial cytochrome c oxidase subunit I (COI) for cryptic species of *Raymunida* Macpherson & Machordom, 2000 (Machordom & Macpherson 2001), COI & 16S rRNA for rapid radiation and cryptic speciation of *Munida* Leach, 1820 (Machordom & Macpherson 2004), and analysis of nuclear 18S rRNA for establishment of a new family Kiwaidae (Macpherson *et al.* 2005). Further molecular studies on anomuran crabs have been mainly restricted to the families Porcellanidae (Stillman & Reeb 2001), Aeglidae (Perez-Losada *et al.* 2002), and other taxa (Cunningham *et al.* 1992; Schubart *et al.* 2002; Ahyong & O'Meally, 2004).

Here we describe and illustrate morphological characters of a new species of *Munidopsis* collected from Myojin Knoll, Izu-Ogasawara Arc, South of Japan and Northwest Eifuku Seamount, Mariana Volcanic Arc, Western Pacific (Fig. 1). The present study is also the first to attempt genetic identification of Western Pacific species of vent associated *Munidopsis* using a molecular marker. We also provide some environmental information obtained during in-situ observations.



FIGURE 1. Locality map, indicating collection sites.

Material and methods

The material reported here was collected by the Japan Agency for Marine-Earth Science & Technology (JAM-STEC). Measurements of specimens, given in millimeters (mm), indicate carapace length including the rostrum. Holotype and paratypes are deposited in the National Science Museum, Tokyo (NSMT), The Los Angeles County Museum of Natural History (LACM), The Smithsonian Instituition, Washington DC (USNM) and in JAMSTEC. The other, non-type specimens are deposited in JAMSTEC and at the University of Mexico campus in Mazatlán, Sinaloa (EMU).

Twenty-three specimens (five from Myojin Knoll and 18 from Northwest Eifuku Seamount) were selected for DNA analysis. Genomic DNA was isolated from the muscle preserved in ethanol (12–20 mg) treated with the Qiagen Dneasy, according to manufacturer's instructions (Qiagen Inc., USA). The COI sequences were amplified by Polymerase chain reaction (PCR) using the universal primers HCO 2198 and LCO 1490 (Folmer *et al.* 1994). In a final volume of 50 μ l, the PCR mix contained 3 μ l of DNA template, 5 μ l of 10X buffer, 4 μ l of dNTP (supplied by manufacturer), 1 μ l each of forward and reverse primer, 0.25 μ l of Taq polymerase (Promega Inc., WI) and 35.75 μ l of sterile water. PCR conditions for amplification of the gene regions were as follows: 95°C (1 min.), 35 cycles of 95°C (1 min.), 40° C (1min), 72°C for (1.5 min.) and a final extension at 72°C (7 min). The amplified fragments of around 660 bp were purified using a Wizard SV gel and PCR clean up kit prior to sequencing both strands using Big Dye Terminator sequencing reactions (Applied Biosystems).

The forward and the reverse DNA sequences obtained for each specimen were aligned and checked using the Mac Vector program after removing the primer regions. Additionally, all the alignments were checked visually. Dendrograms were constructed using Neighbor Joining in Mac Vector. *Munida taenia* was used as an outgroup (GenBank accession number AY 351007).

The GenBank accession numbers for the sequences acquired during this study are as follows: *M. myojin-ensis* n. sp. (Myojin Knoll) (EF 143603); *M. myojinensis* (Eifuku Seamount) (EF 157849); *M. lauensis* (Desmos Site) (EF 157850); *M. lauensis* (Mariner site) (EF 157851); *M. lauensis* (Hine Hina) (EF 157852); *M. lauensis* (Brothers Seamount) (EF 157853).

Systematics

Order Decapoda

Family Galatheidae Samouelle, 1819

Genus Munidopsis Whiteaves, 1874

Munidopsis myojinensis, n. sp. (Figs. 2–5)

Material examined. Type material. Holotype: ovigerous female, 26.11 mm (NSMT-Cr16877), taken by manned submersible, *Shinkai 2000*, dive # 2K 1009 and the support ship *Natsushima* from Myojin Knoll 32°06.29'N, 139° 52.19' E, 7 May 1998, 1288 m. Allotype: 1 male, 22.05 mm (NSMT-Cr16879), NW Eifuku Seamount, 21°29.24'N, 144°02.48'E, 27 October 2005, 1625 m, dive HD# 492. Other paratypes: 1 male, 20.54 mm (LACM CR2004-019.1), NW Eifuku Seamount, 21°29.26' N, 144°02.50'E, 11 April 2004, 1573 m, R793-33-01; 1 female 24.26 mm (USNM), NW Eifuku Seamount, 21°29.26'N, 144°02.51'E, 10 April 2004, 1576 m, R792-08-01; 1 female, 22.63 mm (NSMT-Cr16877), Myojin Knoll, 32°06.30'N, 139°52.04'E, 29 June 1999, 1340 m, *Shinkai 2000*, dive # 2K 1112.



FIGURE 2. *Munidopsis myojinensis* holotype female (NSMT-Cr16877), a, carapace, lateral; b, right P1, dorsal; c, right P2, lateral; d, dactylus of P2, lateral e, thoracic sternum, f, endopod of right third maxilliped; g, posterior part of abdomen and telson; h, left antennal peduncle, ventral view; i, left antennular basal article, ventral. Scales: a, b, c = 3 mm; e, g = 3mm; d, f, h, i = 1 mm.

Additional material. Myojin Knoll: 1 female 24.48 mm (JAMSTEC 018307), 32°06.25'N, 139°52.17'E, 4 July 1999, *Shinkai 2000*, dive # 2K 1116; 3 females, 27.4 mm, 23.19 mm, 18.75 mm, 1 male, 22.05mm (JAMSTEC 057776–057779), 32°06.21'N, 139°52.17'E, 25 June 2004, 1219 m, *Hyper-Dolphin*, dive # 312; 2 females, 21.66 mm, 22.54 mm (JAMSTEC 058045–058046), 32°06.21'N, 139°52.17'E, 25 June 2004, 1219 m, *Hyper-Dolphin*, dive # 312.

Northwest Eifuku Seamount: 2 females, 22.63 mm, 21.22 mm, 2 males, 20.40 mm, 21.79 mm (JAM-STEC 061128–061132), 21°29.252'N, 144°02.489'E, 27 October 2005, 1625 m, *Hyper-Dolphin*, dive # 492; 2 males, 17.95 mm 16.23 mm, 1 female, 17.06 mm (JAMSTEC 061325–061327), 21°29.24'N, 144°02.49'E, 27 October 2005, 1625m, *Hyper- Dolphin*, dive # 492; 1 female, 19.30 mm (EMU-7229), 21° 29.26'N, 144°2.50'E, 11 April 2004, 1573 m, R793-33-01; 1 female, 20.25 mm, (EMU-7230), 21°29.26'N, 144°2.52'E, 10 April 2004, 1576 m, R792-08-01; 9 females, 26.20 mm, 22.29 mm, 27.61 mm, 21.71 mm, 17.27 mm, 19.07 mm, 28.77 mm, 21.22 mm, 25.75 mm (JAMSTEC 061241–061258), 21°29.252'N, 144°02.52'E, 29 October 2005, 1582 m, *Hyper-Dolphin*, dive # 494.

Diagnosis. Carapace covered with scale like short ridges; epigastric spine strong and well developed. Frontal margin oblique; antennal spine present, stronger than anterolateral spine. Lateral margin spinous, spine at end of anterior cervical groove long and strong. Rostrum long, triangular, serrated laterally, slightly upturned. Abdominal tergites unarmed; telson composed of 10 plates. Eyestalk immovable, eyespine long, cornea rounded. P1 bearing numerous elevated scaliform ridges and rugosites; fixed finger without denticulate ridge on distolateral surface. P2–4 relatively long and spinous; P2 reaching tip of P1. Epipods absent from pereopods.

Description. Carapace, exclusive of rostrum, distinctly longer than broad moderately convex transversely, anterior and posterior bifurcation of cervical groove distinct. Rostrum 0.21–0.23 times length of remaining carapace, almost horizontal, occasionally slightly upturned distally, moderately triangular with fine serrations on lateral margin, dorsal surface with sparse coarse setae and distinct median longitudinal carina merging into median tubercles on anterior gastric region. Strongly oblique frontal margin sweeping to antennal spine followed by a distinct anterolateral spine, additional 1 or 2 small spine between antennal and anterolateral spine. Gastric region moderately inflated with prominent pair of epigastric spines and adjacent 1 or 2 small spines (sometimes obsolete), followed by scale like setae but tuberculate, elevated rugae bearing sparse short setae. Cardiac region with moderate, transverse elevation preceded and flanked by distinct grooves. Anterior branchial region with strong lateral spine, rugosites with tendency to being transversely continuous across central part of cardiac region. Pterygostomian flap with obliquely interrupted rugae, distinct posteriorly, obsolescent anteriorly, anterior margin angular.

Abdomen unarmed, transverse ridge on segments 2 or 3 with sparse short stiff setae, segments 4–6 almost smooth, segment 6 with posteromedian margin almost transverse, distal margin of lateral lobes convex. Telson composed of 10 plates, length width ratio 0.78–0.92, median lateral plate fringed with long stiff setae. Eyes moderate in size, well exposed, cornea cupped within broad based immovable ocular peduncle, strong mesiodorsal eyespine directed anterolaterally and reaching mid length of rostrum.

Basal article of antennular peduncle, exclusive of spines, somewhat longer than broad, dorsolateral inflation bearing tubular process often developed into spines, distolateral spine well developed, distoventral mesial margin scalloped, contiguous with small mesiodorsal spine.

Antennal peduncle having article 1 with flat ventral process ending in acute spine and much smaller distolateral spine, article 2 with short distolateral spine, article 3 unarmed, article 4 with a crenulated scalloped ending.

Third maxilliped having ischium shorter than merus when measured in midlateral line, bearing mesial crest armed with finely uniform evenly spaced corneous tipped spines and distoventral spine. Basis with 2 or 3 corneous spines in line with crest on ischium. Merus with 4 irregular small spines on flexor margin and small spine at disto dorsal corner. Dactylus reaching proximal end of ischium when folded.

Thoracic sternum broadened posteriorly. Sternite 3 forming apposed lobe at either side of mid line, irregularly serrate on margin.

First percopod (P1) 1.70–1.81 times as long as carapace excluding rostrum, bearing numerous tuberculate, elevated scaliform ridges and rugosites, long plumose setae more dense ventrally along distomesial margin of merus and carpus. Ischium with mesial row of several spines on distal portion and 1 spine on ventral margin, merus with a row of 4 strong spines terminally, preceded by a row of spines dorsally, 2 or 3 spines ventromesially, often irregular tubercles all along merus. Carpus with well-developed acute spine mesially somewhat proximal to junction with palm, followed by a few spines slightly dorsal, 2 distodorsal spines. Propodus spiny, often with 4 distoventral and 2 distolateral spines; ventral surface tuberculate. Fingers longer than palm (up to 1.25 times in females and 1.5 times in males), slightly curved, opposable margins forming spooned shaped cavity; cutting edges closely fitted, with very fine teeth and tuffs of setae; tips close fitted, spooned; fixed finger without denticulate carina on distolateral surface.



FIGURE 3. *Munidopsis myojinensis* holotype female (NSMT-Cr16877), a, dorsal view, b, lateral view, c, in-situ photograph of Myojin Knoll sampling site; d, in–situ photograph of Northwest Eifuku Seamount sampling site.

Percopods 2–4 (P2–4) relatively long, P2 almost reaching tip of P1, corresponding segments of respective percopods nearly equal in length. Merus with dorsal crest bearing row of spines, ventrolateral margin with strong terminal spine followed by small spines or tubercles, mesially and laterally with row of tubercles or scales. Carpus with tuberculate lateral ridge in parallel with row of dorsal marginal spines. Propodus 1.8 times

as long as dactylus, more setose on merus and carpus, especially on mesial face, with two longitudinal tuberculate dorsal ridges, dorsomesial ridge often bearing few spines, dorsolateral in line with preceeding segments. Dactylus stout, setose, ending in short, curved, corneous claw preceded by 11–13 successively diminishing teeth on flexor margin, each tooth bearing short stiff setae arising from its base.

Epipods absent from pereopods.

Eggs few, measuring $2.1 \ge 2.3$ mm in diameter.

Etymology. Named after the type locality Myojin Knoll, a noun in the genitive.

Variation. The material we examined indicates that females are larger than males. In females, P2 is slightly longer than P1 (cheliped) and reaches or slightly overreaches the tip of P1 when extended. In males, P2 is about the same length as P1, falling just short of the tip of P1 when extended. The telson is broader in females than in males. Both of the aforementioned variations are sexual dimorphic. The number of gastric spines shows considerable variation. Most specimens (including the holotype), bear a prominent pair of epi-gastric spines, but in few paratypes, it is accompanied by 1 or 2 obsolete lateral spines. The eyespine is directed anterolaterally in holotype and a few paratypes, and directed forwards in the remainder.

Remarks. Munidopsis myojinensis closely resembles *M. starmer* in general ornamentation and spination of the body. The eyespine of *M. myojinensis* almost reaches half of the rostrum and is directed anterolaterally, whereas *M. starmer* features a much smaller eyespine compared to rostral length, abdominal segment 6 with a rounded, protruding posterolateral flap distinctly overreaching the posteromedian margin and overhanging anterior junction between the telson and uropodal protopod. *Munidopsis myojinensis* is characterized by transverse abdominal segment 6 without a protruding posterolateral flap. This character links the new species to *M. lauensis*, but these two species have clearly different spination of the carapace and cheliped. *Munidopsis myojinensis* is also allied to *M. subsquamosa* in the carapace spination, but distinguished by the more spinous and tuberculate P1, absence of epipods, and the more strongly curved corneous claws on the dactyli of P2–4.

Distribution. Deep-sea hydrothermal vents viz, Myojin Knoll, 1219–1224 m, in South of Japan and Northwest Eifuku Seamount, 1582–1625 m, in North Mariana Arc.

Molecular analysis. A clear genetic differentiation between *M. myojinensis* and *M. lauensis* from different vent sites (i.e., Desmos, Hine Hina, Mariner Sites and Brothers Seamount in the Western Pacific)was obtained (Fig. 6). This is the first report on the genetic identification of vent associated *Munidopsis* from Western Pacific using a molecular marker, and it shows that both *M. myojinensis* and *M. lauensis* form reciprocally monophyletic clusters on the NJ tree. Monophyly of the both species is supported by a high bootstrap proportion (100%) and few or no differences between specimens of the same species obtained from different localities.

Habitat and ecology. Myojin Knoll consists of many small chimneys, 1–3 m height, and a larger chimney of about 20 m height emitting vigorous transparent hydrothermal fluids. The vent site is characterized by a predominance of deep-sea mussels and barnacles. Dense beds of mussel, *Bathymodiolus septemdierum* Hashimoto & Okutani, 1994 and deep-sea barnacles, *Ashinkailepas seepiophila* Yamaguchi, Newman & Hashimoto, 2004 and *Neoverruca* sp. were observed. Alvinocaridid shrimp, *Alvinocaris brevitelsonis* Kikuchi & Hashimoto, 2000, bythograeid crab, *Austinograea yunohana* Takeda, Hashimoto & Ohta, 2000 and few aggregations of sea anemones were also observed in the video footage. There were thick bacterial mats in the substratum. *Munidopsis* species were seen crawling on the vicinity of the chimneys in the video image (Fig. 3c), most among mussels.

The fauna of Northwest Eifuku Seamount was different from that of Myojin Knoll. Substrate was characterized with volcanic rocks with thick bacterial mass. Barnacles abundant in Myojin Knoll were totally absent. Two species of alvinocaridid shrimps, *Alvinocaris brevitelsonis* and *Opaepele loihi* Williams & Dobbs, 1995 and a few bythograeid crabs, *Austinograea yunohana* were observed. *Munidopsis* were seen either among the deep-sea mussels, mostly seen idle or swiftly swimming back beating their abdomen when some disturbances occur. The density of *Munidopsis* was higher than that of Myojin Knoll (Fig. 3d).



FIGURE 4. *Munidopsis myojinensis* paratype male (LACM CR 2004-019.1); a, carapace, frontal region, dorsal; b, right anterior part of carapace, lateral; c, right P1, dorsal; d, left P1, dorsal, with details of merus, ventral; e, thoracic sternum; f, right P2, lateral; g, right P5, lateral. Scales = 3 mm.



FIGURE 5. *Munidopsis myojinensis* paratype male (LACM CR 2004-019.1); a, right antennal peduncle, dorsal (right) and ventral (left) views; b, left third maxilliped, ventral, and dorsal view of merus; c, left antennular basal article, ventral; d, telson, dorsal. Scales: a-c = 1 mm; d = 3 mm.



FIGURE 6. Genetic relationships between *Munidopsis myojinensis* and *M. lauensis* based on DNA sequences. *Munida taenia* is the outgroup.

Discussion

Species of *Munidopsis* occur in most of the deep-sea habitats worldwide and they comprise an important element of macro fauna of hydrothermal vents, cold seeps and other reducing habitats such as whale carcasses or decomposing wood in deep-sea (Williams & Van Dover 1983, Van Dover *et al.* 1985, Williams 1988, Williams & Baba 1989, Baba 2005, Schnabel & Bruce 2005, Osawa *et al.* 2006a, b, and Macpherson in press). *Munidopsis myojinensis*, herein described from Myojin Knoll and Northwest Eifuku Seamount, is the tenth species of *Munidopsis* associated with active hydrothermal vents and the first to be described from these sites. Four additional undescribed species have been also identified from Brothers Seamount, Kermadec Arc, Hatoma Knoll, East China Sea and Sagami Bay in West Pacific, respectively (Cubelio *et al.* in press a, b).

Considering our present knowledge, the distributional patterns of vent associated *Munidopsis* in West Pacific is rather variable. *M. lauensis* inhabits Western Pacific vent sites such as Manus, Lau and North Fiji Back Arc Basins (Tunnicliffe 1991; Baba & de Saint Laurent 1992) and Brothers Seamount (Cubelio *et al.* in press), which are located about 4500 km apart, suggesting a very wide range for this species. Myojin Knoll in the Izu-Ogasawara Arc, South of Japan, and the Northwest Eifuku Seamount in the North Mariana Volcanic Arc, where *Munidopsis myojinensis* was found, are about 1000 km apart, thus indicating a medium distributional range. Other species such as *M. starmer* and *M. sonne* reported only in the North Fiji Basin (Baba & de Saint Laurent 1992; Baba 1995) and *M. marianica*, found only in the North Mariana Back Arc Basin (Williams & Baba 1989), feature a narrow distribution. The dispersal potential of larvae of *Munidopsis* could be one of the factors limiting distributional pattern in Western Pacific vent fields, but no larval data are presently available.

The results derived from the molecular data are congruent with our morphological study in finding *M. myojinensis* from Myojin Knoll and Northwest Eifuku Seamount to be conspecific. They also support the conspecificity of *M. lauensis* from different vent fields in Western Pacific such as Desmos, Mariner, Hine Hina and Brothers Seamount.

Extensive work on phylogeny on the genus *Munida* by Machordom & Macpherson (2004) suggests a recent speciation in the Western Pacific. Ongoing work on phylogeny of the vent *Munidopsis* also indicates similar patterns (unpublished data). More data from the analysis of organisms with faster evolving genes and the results obtained from these new environments should greatly improve our knowledge of biogeography of these vent organisms.

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