

Rediscovery of rarely encountered sea star, *Dipsacaster sagaminus* Hayashi, 1973 (Paxillosida: Astropectinidae)

Itaru Kobayashi^{1*}, Hiroto Watabe², Kohei Oguchi¹, Hisanori Kohtsuka¹

¹Misaki Marine Biological Station, Graduate School of Science, The University of Tokyo, 1024 Koajiro, Misaki, Miura, Kanagawa 238-0225, Japan. ²Faculty of Fisheries, Kagoshima University, 4-50-20 Shimoarata, Kagoshima, Kagoshima 890-0056, Japan.

*Corresponding author, e-mail: itarukobayashi@mmb.s.u-tokyo.ac.jp, Tel: +81-46-881-4107.

Abstract

A rare deep-sea species, *Dipsacaster sagaminus*, was rediscovered in Japanese waters, with extending its distributional range to the southwestern part of Japan. This is the first-time finding of this species 50 years after the original description. Here, we redescribe the morphology and feeding behavior for compensating the basic features of this species. A brief molecular phylogenetic analysis supported the relativeness of *Dipsacaster* and *Leptychaster*, yet the phylogenetic relationships among species were unresolved.

Key words: sea star; starfish; taxonomy; deep sea; Japan

Introduction

Dipsacaster Alcock, 1893 is a deep-sea genus of the family Astropectinidae, having a stellate and flat body, madreporite hidden by paxillae, unequal-sized supero- and inferomarginal plates, and serially arranged gonads along the arms (Fisher 1906; 1911; 1919). This genus currently encompasses 16 species mainly distributed to Indo-west Pacific to North Pacific oceans (AM Clark 1989; Lane and Vimono 2020). From Japanese waters, four species of *Dipsacaster* have been reported: *D. borealis* Fisher, 1910, *D. grandissimus* Goto, 1914, *D. pretiosus* (Döderlein, 1902), and *D. sagaminus* Hayashi, 1973 (Kogure 2018). Among these species, *D. sagaminus* is unique in sharing some diagnostic characters with *Leptychaster* which is a sister genus of *Dipsacaster* (Mah and Foltz 2011). Nevertheless, no more specimens have been collected after five decades since its original description (Hayashi 1973a) and the redescription of holotype (Hayashi 1973b) collected from Sagami Bay, Japan.

During the survey of deep-sea benthic fauna in Japanese waters, we collected four specimens of *D. sagaminus* (Fig. 1). In this paper, we redescribed the morphology, live coloration, and ecology of this species for compensating the original description. Moreover, we investigated the phylogenetic position of this species by operating a preliminary molecular analysis.

Materials and Methods

Sample collection and observation

Three specimens (NSMT E-14493, 14494, 14495) of *Dipsacaster sagaminus* were collected with a 50 cm biological dredge by the R/V *Rinkai-Maru* of the Misaki Marine Biological Station of the University of Tokyo, and one specimen (NSMT E-14496) was collected with a bottom trawl net by the R/V *Kagoshima-Maru* of the Kagoshima University (Fig. 1). One of these (NSMT E-14495) was kept in a 3L acrylic aquarium that adopted the semi-closed recirculating system at 16–17 degrees. After

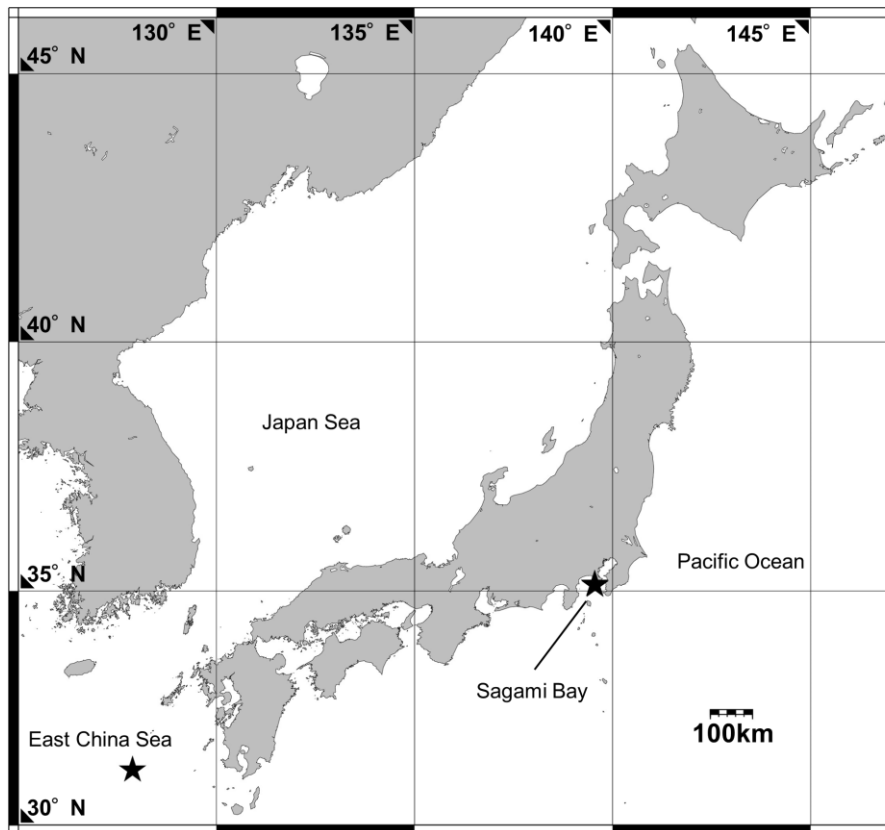


Fig. 1. Sampling sites (solid stars) of *Dipsacaster sagaminus*.

observing the partial decay of captive NSMT E-14495, we fixed it with 99.5% ethanol as with NSMT E-14493 and NSMT E-14494. The specimen collected by the R/V *Kagoshima-Maru* (NSMT E-14496) was fixed with 70% ethanol.

The fixed four specimens were measured the arm length (R) and the interradius length (r) and observed under a dissecting microscope SZX16 (Olympus, Tokyo, Japan). Epidermal tissues and spines entirely covering the body wall were removed from the part of the disk and the arms by applying sodium hypochlorite solution (ca. 5%) to observe the underlying plate's arrangement. Simultaneously, abactinal, superomarginal, inferomarginal, actinal, and adambulacral spines were collected for observing the spine morphology and measuring the spine length.

All four examined specimens were deposited

in the National Museum of Nature and Science, Tsukuba (NSMT).

Molecular experiment

The tube feet of *Dipsacaster sagaminus* were excised for DNA extraction from two individuals. Total DNA was extracted using a Wizard Genomic DNA Purification Kit (Promega Biotec, Madison, WI, USA). A partial region of the cytochrome c oxidase subunit I (COI) gene was amplified by PCR with the following primer set: 5'- ACT GCC CAC GCC CTA GTA ATG ATA TTT TTT ATG GTN ATG CC -3' (COIceF; Hoareau and Boissin 2010) and 5'- TCG TGT GTC TAC GTC CAT TCC TAC TGT RAA CAT RTG -3' (COIceR; Hoareau and Boissin 2010). The obtained PCR products were purified using ExoSAP-IT (Applied Biosystems, Foster City,

CA, USA). The DNA sequences were confirmed using a BigDye Terminator v3.1 Cycle Sequencing Kit (Applied Biosystems) and an ABI 3130 Genetic Analyzer (Applied Biosystems). Two obtained sequences were deposited in the INSDC (International Nucleotide Sequence Database Collaboration) via DNA Data Bank of Japan (DDBJ) under the accession numbers LC771451 (NSMT E-14495) and LC771452 (NSMT E-14494).

Obtained sequences were assembled using GeneStudio Professional Edition ver. 2.2.0.0 (GeneStudio, Inc., Suwanee, GA, USA), providing identical lengths of COI sequences (486 bp) for the two specimens. These sequences were aligned together with five registered sequences in the GenBank by MAFFT ver. 7.222 (Kato and Standley 2013). These sequences were chosen from the clade named “node K” in Mah and Foltz’s (2011) phylogenetic tree since *Dipsacaster* was included in this clade. We manually removed un-overlapped regions of GenBank sequences and the stop codons using MEGA ver. 7.0.26 (Kumar et al. 2016). Reconstructing the phylogenetic tree, GTR+G was selected as a best-fit substitution model by Kakusan4 (Tanabe 2011). The dataset was partitioned by codon position. The maximum likelihood (ML) tree was inferred using RAxML ver. 8.2.9 (Stamatakis 2014). Bootstrap values (BS) were calculated from 1000 replicates.

Results

Taxonomic account

Family Astropectinidae Gray, 1840

Genus *Dipsacaster* Alcock, 1893

Dipsacaster sagaminus Hayashi, 1973

Dipsacaster sagaminus Hayashi, 1973a: 1–4; Hayashi, 1973b: 21–23, pl3, figs 4, 5.

Patagiaster sphaerioplax Kogure and Fujita, 2012: 253–254; Fujita et. al., 2015: 24, 83

Materials examined

NSMT E-14493, west off Jogashima Island, Sagami Bay, Japan, 35°07.055’N, 139°33.409’E, 449–509 m, on February 15, 2017, R=30.3 mm, r=12.4 mm. NSMT E-14494, west off Jogashima Island, Sagami Bay, Japan, 35°08.220’N, 139°32.881’E, 106–119 m, May 14, 2018, R=30.1 mm, r=11.7 mm. NSMT E-14495, west off Jogashima Island, Sagami Bay, Japan, 35°08.298’N, 139°32.784’E, 128–235 m, June 9, 2022, R=14.5 mm, r=6.0 mm. NSMT E-14496, East China Sea, Japan, 31°12.033’N, 127°52.833’E, 140–142 m, May 13, 2023, R=36.5 mm, r=14.8 mm, collected from unknown substrates.

Description

Arms are five, flat, broad proximally, and gradually tapering to the arm tip (Fig. 2A–D). The R/r ratio is 2.4–2.6. The abactinal surface is densely covered by numerous abactinal paxillae composed of abactinal plates and spines (Fig. 3A). The abactinal plates are pawn-shaped, regularly arranged in longitudinal and oblique transverse series in the interradial and the arms, irregularly arranged in the disk’s center (Fig. 2A, B), bearing 13–58 (NSMT E-14495), 14–66 (NSMT E-14493), and 15–89 (NSMT E-14494, 14496) abactinal spines. A midradial abactinal series reaches the terminal plates and is wedged between two superomarginal series in the distal one-third of the arms (Fig. 3B). Abactinal spines are granulated to clavate (Fig. 4A), 0.1–0.2 mm (NSMT E-14495) and 0.2–0.4 mm (NSMT E-14493, 14494, 14496) in length. Papulae are distributed to the interradial and the arm (Fig. 3C). A madreporite is located at the extremity of the

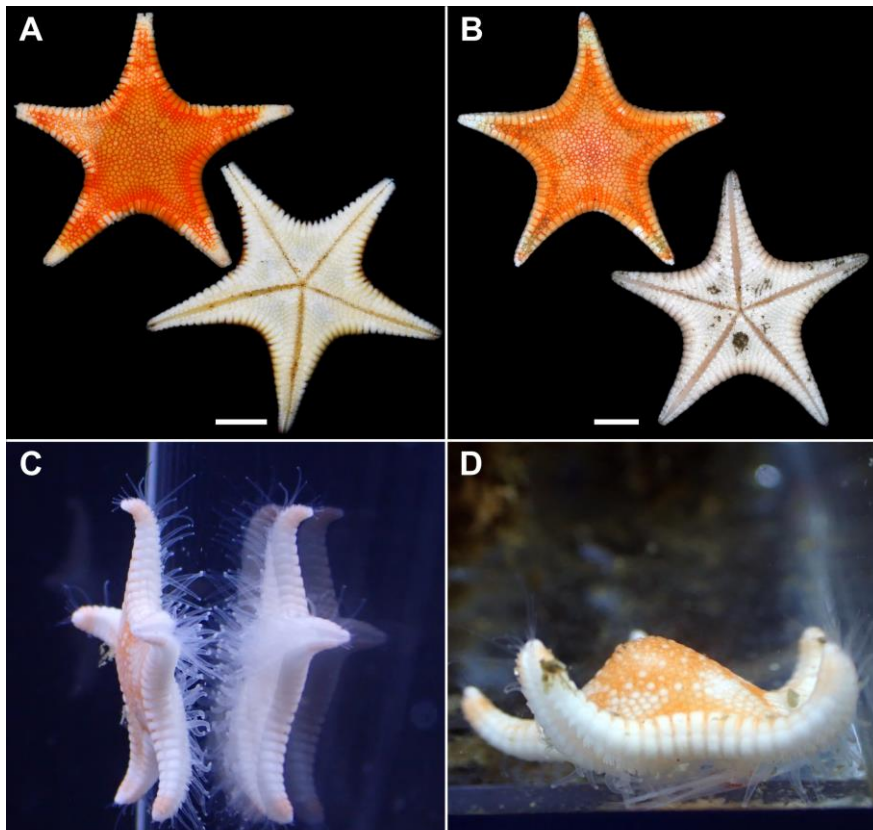


Fig. 2. Live specimens of *Dipsacaster sagamius*. A, NSMT E-14494; B, NSMT E-14493; C–D, NSMT E-14495 in the tank, showing abactinal and actinal sides in A and B extended tube feet in C, and the feeding posture in D.

disk, overhung by neighboring paxillae.

Supero- and inferomarginal plates are transversely elongated rectangular, and arranged in a longitudinal series, respectively (Fig. 3B–F). Superomarginal plates are 13 (NSMT E-14495), 15 (NSMT E-14493), 19 (NSMT E-14494), and 17 (NSMT E-14496) in each series, corresponding to 13, 16, 19, and 17 inferomarginal plates. Inferomarginal plates are slightly wider than the adjacent superomarginal plates and fringed the body outline (Fig. 3B). Between two consecutive supero- and inferomarginal plates, there are the U-shaped fasciolar grooves having almost equal depth and longitudinal width (Fig. 3D). These plates bear numerous granulated to clavate spines (Fig. 4B, C), 0.1–0.3 mm (NSMT E-14495), and 0.2–0.4 mm (NSMT E-14493, 14494, 14496).

Actinal plates are ovoid, convexed, and arranged in five (NSMT E-14495) or six (NSMT E-14493, 14494, 14496) longitudinal series (Fig. 3F). The 1st actinal series is the longest, reaching half of the arm length, and the 2nd to 6th are confined within the interradial area. The proximal-most actinal plates of the 2nd to 4th series are unpaired in NSMT E-14495, and those of the 2nd to 6th series are unpaired in NSMT E-14493, 14494, and 14496. Each actinal plate bears 7–17 (NSMT E-14495), 8–23 (NSMT E-14493, 14494), and 10–38 (NSMT E-14496) actinal spines (Fig. 3G). Actinal spines are conical to clavate (Fig. 4D), 0.2–0.3 mm (NSMT E-14495), and 0.3–0.6 mm (NSMT E-14493, 14494, 14496) in length.

Adambulacral plates are longitudinally elongated, slightly projected toward the ambulac-

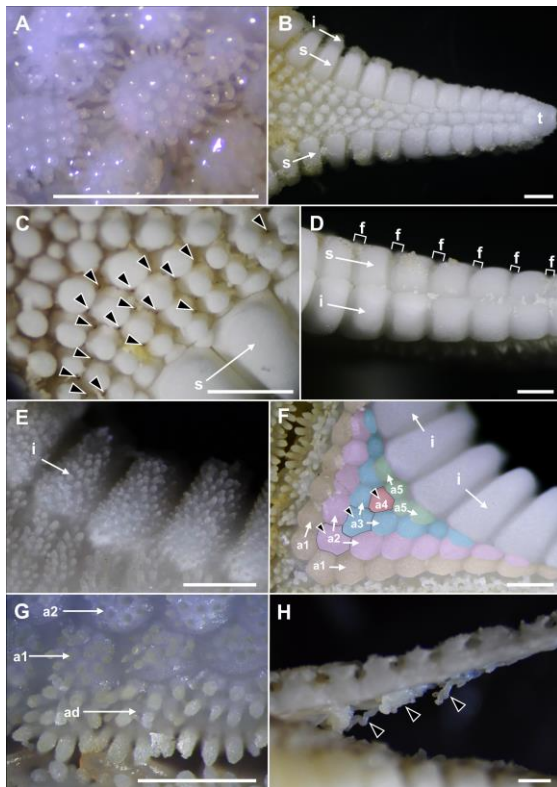


Fig. 3. *Dipsacaster sagaminus*, NSMT E-14495 in A–G and NSMT E-14494 in H. A, abactinal surface at disk; B, denuded abactinal surface at arm; C, denuded abactinal surface at the interradius, showing papular pores (arrowheads); D, denuded lateral surface at the proximal part of arm; E, actinal surface at the interradius; F, denuded actinal surface at the interradius, showing unpaired actinal plates (arrowheads); G, actinal surface at the proximal part of arm; H, lateral view of dissected abactinal body wall, showing gonads (arrowheads). Abbreviations: a, actinal series; ad, adambulacral series; f, fasciolar grooves; i, inferomarginal series; s, superomarginal series. Arabic numerals 1 to 5 indicate the 1st to 5th actinal series, respectively. All scale bars indicate 1 mm.

ral furrow, and arranged in a longitudinal series (Fig. 3G). Adambulacral plates bear 13–17 (NSMT E-14495), 13–23 (NSMT E-14493, 14494), and 18–25 (NSMT E-14496) adambulacral spines arranged in three or more longitudinal rows. These spines are conical, digitated, or clavate (Fig. 4E), 0.3–0.5 mm (NSMT E-14495), 0.5–1.2 mm (NSMT E-14493, 14496), and 0.7–1.0 mm (NSMT E-14494) in length.

Pedicellariae are absent.

Tube feet end with knobby tips, arranged in two longitudinal series (Fig. 2C).

Gonads are longitudinally arranged along the arms in NSMT E-14494 but not observed in unmaturing NSMT E-14495 (Fig. 3H).

The color in the living specimen is orange with a whitish disk and arm tips on the abactinal side and uniform white on the actinal side (Fig. 2A–D).

Distribution

Dipsacaster sagaminus is known from off Jogashima Island, Sagami Bay, Japan (type locality: Hayashi, 1973a), and East China Sea, Japan (this study), 90–509 m in depths.

Feeding

Observations in our aquarium showed the captive *D. sagaminus* (NSMT E-14495) fed on the defrosted krill by swallowing (Fig. 2D). By feeding once a week, we kept this specimen for 11 months.

Phylogenetic analysis

Dipsacaster sagaminus formed a clade with *Leptychaster* and *Dipsacaster* species (BS 88%) (Fig. 5). In this clade, *L. pacificus* was branched at first, and *L. anomalus* was at second from *D. sagaminus* and *D. borealis*. However, these phylogenetic relationships were supported with low supported values (BS 49% and 51%).

Discussion

Dipsacaster sagaminus is recorded from the East China Sea for the first time, extending the distributional range to the southwestern part of Japan. This record is the first time in 50 years since the holotype was described by Hayashi (1973a, b). Describing these specimens resulted in providing body coloration and variations in mor-

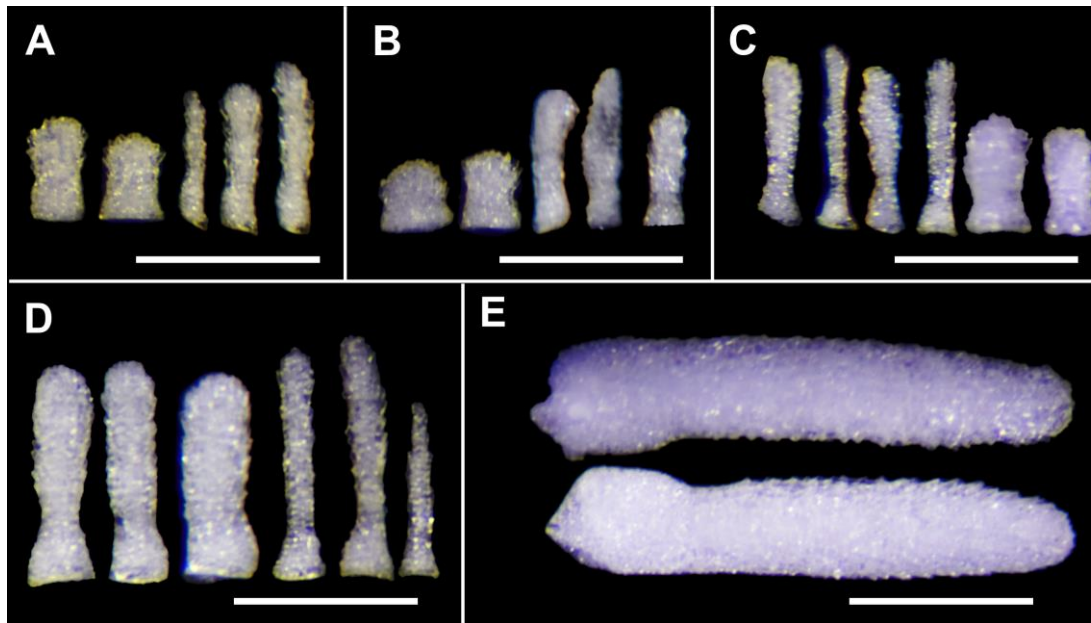


Fig. 4. Spines of *Dipsacaster sagaminus*, NSMT E-14493. A, abactinal spines; B, superomarginal spines; C, inferomarginal spines; D, actinal spines; E, adambulacral spines. All scale bars indicate 0.5 mm.

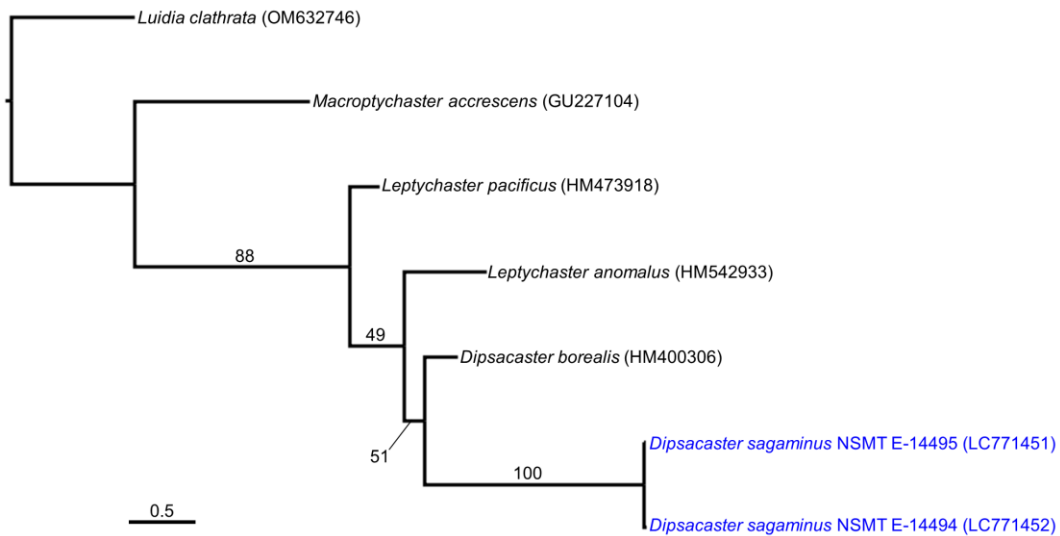


Fig. 5. Molecular phylogenetic tree of two related genera *Dipsacaster* and *Leptychaster* based on COI gene (486 bp). The values on branches indicate bootstrap support. The scale bar indicates substitutions per site in branch length. GenBank accession numbers of COI are shown in parentheses. Blue OTUs indicate the sequences obtained in this study.

phological characters of *D. sagaminus* for the first time. Four specimens we observed agree well with the original description in having only one abactinal series at about the distal one-third of the arms and no enlarged spines on marginal plates (Fig. 3B, E), which is a unique morphological

combination in *D. sagaminus* among 16 congeners (Fisher 1911; Goto 1914; HL Clark 1916; Hayashi 1973a, b; Lane and Vimono 2020). A specimen of *Patagiaster sphaerioplax* Fisher, 1913 from Kumejima Island, Okinawa, Japan (RUMF-ZE-00032) reported by Kogure and

Fujita (2012) and Fujita et al. (2015) clearly belongs to *D. sagaminus* in having only one abactinal series reaching arm tips and lacking enlarged spines on inferomarginal plates, unlike *P. sphaerioplax* that has three abactinal series reaching arm tips and enlarged lanceolate spines on inferomarginal plates (Fisher 1919). Hayashi (1973a, b) presumed that *D. sagaminus* was an intermediate species between two related genera, *Dipsacaster* and *Leptychaster*. Our tree indeed showed a close relationship between these two genera, however, the phylogenetic relationships among species were unresolved since the supporting values were low (Fig. 5). Analysing more species and gene regions would enable to review the classification of these genera.

We observed the captive *D. sagaminus* fed on the krill by swallowing (Fig. 2D). Such a feeding habit without external digestion is consistent with the characteristic of the order Paxillosida (Blake 1990). Carey (1972) investigated the stomach contents of *D. anoplus* and found that the species fed on mollusks, crustaceans, ophiuroids, polychaetes, and sediments, suggesting the species is an omnivore. Although we did not assess the stomach contents in this study, *D. sagaminus* is also likely to be an omnivore. This is the first paper observing the feeding behavior of *Dipsacaster* species.

Acknowledgments

We would like to express our sincere gratitude to Captain Akimasa Habano of R/V *Kagoshima-Maru* at Kagoshima University for providing the specimen from the South China Sea. And we are grateful to Mr. Mamoru Sekihuji and Ms. Michiyo Kawabata at the Misaki Marine Biological Station, the University of Tokyo for operating R/V *Rinkai-Maru*, resulting in collecting the specimens from Sagami Bay. We also thank the crew on board for

the collection of the specimen. The authors would like to thank Mr. Gregorius Altius Pratama (The University of Tokyo) for the English language review. We appreciate the suggestive and helpful comments on the present study by three anonymous reviewers.

References

- Blake, D. B. (1990). Adaptive zones of the class Asterozoa (Echinodermata). *Bull. Mar. Sci.* 46: 701–718.
- Carey, A. G. (1972). Food sources of sublittoral, bathyal, and abyssal asteroids in the northeast Pacific Ocean. *Ophelia*. 10: 35–47.
- Clark, A. M. (1989). An index of names of recent Asterozoa. Part 1: Paxillosida and Notomyotida. *Echinoderm Stud.* 3: 225–347.
- Clark, H. L. (1916). Report on the sea-lilies, starfishes, brittle-stars and sea-urchins obtained by the F.I.S. “Endeavour” on the coasts of Queensland, New South Wales, Tasmania, Victoria, South Australia, and Western Australia. *Biological Results of the Fishing experiments carried on by the F.I.S. Endeavour 1909–1914*, Sydney, pp. 30–32.
- Fisher, W. K. (1906). Starfishes of the Hawaiian Islands. *Bull. U. S. Fish Commission* 23: 987–1130.
- Fisher, W. K. (1911). Asterozoa of the North Pacific and adjacent waters. Part I. Phanerozoa and Spinulosa. *Bull. US. Natl. Mus.* 76: 1–420.
- Fisher, W. K. (1919). Starfishes of the Philippine seas and adjacent waters. *Bull. US. Nat. Mus.* 100: 1–547.
- Fujita, Y., Irimura, S., Kogure, Y., Okanishi, M., Michonneau, F., and Naruse, T. (2015). Catalogue of Materials Deposited in The University Museum (Fujukan), University of the Ryukyus No. 10. Catalogue of Echinodermata Specimens Deposited in The University Museum (Fujukan), University of the Ryukyus. The University Museum (Fujukan), University of the Ryukyus, Okinawa, 106 pp. (In Japanese).
- Goto, S. (1914). A descriptive monograph of Japanese Asterozoa. 1. *Journ. Coll. Sci. Imp. Univ. Tokyo.* 29: 1–808.
- Hayashi, R. (1973a). Seven new species of asteroids from Sagami Bay. *Journal of the College of Liberal Arts, Toyama University, Natural Science* 5: 1–13.
- Hayashi, R. (1973b). The sea-stars of Sagami Bay.

- Biological Laboratory, Imperial Household, Tokyo, pp. 18–23.
- Hoareau, T. B., Boissin, E. (2010). Design of phylum-specific hybrid primers for DNA barcoding: addressing the need for efficient COI amplification in the Echinodermata. *Mol. Ecol. Resour.* 10: 960–967.
- Katoh, K., Standley, D. M. (2013). MAFFT multiple sequence alignment software version 7: improvements in performance and usability. *Mol. Biol. Evol.* 30: 772–780.
- Kogure, Y., Fujita, Y. (2012). A new species of *Neoferdina* and three new records of sea stars (Echinodermata: Asteroidea) collected from Kumejima Islad, southwestern Japan. *Zootaxa* 3367: 252–260.
- Kogure, Y. (2018). A checklist of sea stars (Echinodermata, Asteroidea) from Japanese waters. *Bull. Biogeogr. Soc. Japan.* 73: 70–86. (In Japanese with English abstract).
- Kumar, S., Stecher, G., Tamura, K. (2016). MEGA7: molecular evolutionary genetics analysis version 7.0 for bigger datasets. *Mol. Biol. Evol.* 33: 1870–1874.
- Lane, D. J. W., Vimono, I. B. (2020). Two new species of sea star (Asteroidea, Echinodermata) from mesopelagic depths in the Sunda Strait, Indonesia. *Raffles Bull. Zool.* 68: 662–669.
- Mah, C., Foltz, D. (2011) Molecular phylogeny of the Valvatacea (Asteroidea: Echinodermata). *Zool. J. Linn. Soc.* 161: 769–788.
- Stamatakis, A. (2014). RAxML version 8: a tool for phylogenetic analysis and post-analysis of large phylogenies. *Bioinformatics* 30: 1312–1313.
- Tanabe, A. S. (2011). Kakusan4 and Aminosan: two programs for comparing nonpartitioned, proportional, and separate models for combined molecular phylogenetic analyses of multilocus sequence data. *Mol. Ecol. Resour.* 11: 914–921.

Received: 2 July 2023 | Accepted: 28 July 2023 | Published: 3 August 2023