
БИОРАЗНООБРАЗИЕ ЭКОСИСТЕМ
И ЕГО СОХРАНЕНИЕ

UDC 593.176.1-155(265.72)

DOI: [10.21072/eco.2025.10.3.01](https://doi.org/10.21072/eco.2025.10.3.01)

**FIRST REPORT OF *ACINETA TUBEROSA* (CILIOPHORA) AS AN EPIBIONT
ON A SEA SPIDER (PYCNOGONIDA) FROM NORTHWEST BORNEO (BRUNEI),
WITH NOTES ON CILIATE EPIBIONTS ON PYCNOGONIDS ***

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Abstract: The suctorian *Acineta tuberosa* Ehrenberg, 1834 is reported from the subtidal zone at Bubut, along the South China Sea coastline of Brunei Darussalam, as an epibiont on the sea spider (Pycnogonida) *Hemichela* sp. The found ciliate has a stalked, loricate, flattened triangular cell body, rounded macronucleus and capitate tentacles arranged in two fascicles on two wide and short actinophores. The pycnogonid is also reported here as a host of *A. tuberosa* for the first time. This is also first report of *A. tuberosa* from the Brunei coastal area. In addition, notes on ciliate epibionts of pycnogonids are given.

Keywords: Ciliate, epibiosis, pycnogonid, basibiont, Brunei Darussalam

Introduction

Sea spiders are marine arthropods of the class Pycnogonida, characterized by having long legs and slender bodies. Pycnogonids are considered a suitable basibiont due to their hard cuticle and slow-moving lifestyle, and because they have limited known strategies against fouling, apart from moulting [Arnaud and Bamber, 1987; Wambreuse, Hamel, Mercier, 2021]. Pycnogonids are found to be host to a variety of organisms, like protozoans, poriferans, bryozoans, hydrozoans, polychaetes, nematodes, tunicates, diatoms and multicellular algae. In a few studies, ciliate epibionts have been reported on pycnogonids [Gassovsky, 1916; Pipe, 1982; Kouris et al., 2007; George, Siddiqui, 2020; Wambreuse, Hamel, Mercier, 2021].

Suctorian ciliates are commonly epibionts on marine and freshwater invertebrates such as copepods, cladocerans, nematodes, ostracods, halacarid and hydrachnid mites [e. g. Precht, 1935; Jankowski, 1981; Jankowski, 2007; Dovgal, 1996; Dovgal, 2002; Dovgal, 2013; Fernandez-Leborans, Tato-Porto, 2000; Dovgal, Chatterjee, Ingole, 2008; Dovgal et al., First report ... , 2008; Dovgal, Chatterjee, Ingole, 2009; Dovgal et al., New records of *Praethecacineta* ... , 2009; Ingole et al., 2010; Chatterjee, Fernandez-Leborans, Chan, 2012; Chatterjee et al., New records ... , 2013; Chatterjee, Fernandez-Leborans, Senna, 2013; Chatterjee, Kotov, Fernandez-Leborans, 2013; Chatterjee, Fernandez-Leborans, Marshall, 2014;

*The work of the 2nd author (ID) was carried out within the research topic No 124022400148-4 of A.O. Kovalevsky Institute of Biology of the Southern Seas of RAS. The work of 3rd author (DJM) is supported by Universiti Brunei Darussalam grants UBD/RSCH/1.4/FICBF(b)/2021/033.

Chatterjee, Fernandez-Leborans, Schizas, 2014; Chatterjee et al., 2018; Chatterjee, Dovgal, Fernandez-Leborans, 2019; Chatterjee, Nanajkar, Dovgal, 2019; Chatterjee et al., New records of epibiont ... , 2019; Chatterjee, Dovgal, Nanajkar, 2020; Chatterjee, Dovgal, Sautya, 2022; Chatterjee et al., Report of ... , 2024; Fernandez-Leborans, Chatterjee, Grego, 2012].

The present article documents a suctorian ciliate *Acineta tuberosa* Ehrenberg, 1834 as an epibiont on a pycnogonid from the subtidal zone of the South China Sea, Brunei Darussalam. Furthermore, notes of epibiont ciliates associated with pycnogonids are included in this paper.

Material and Methods

Samples were collected and sorted by the 3rd author (DJM) near Bubut (Brunei Darussalam) ($4^{\circ}46'11''\text{N}$, $114^{\circ}28'11''\text{E}$) in May 2011 (Fig. 1). Benthic sediments from a depth of 15–20 m. The samples were passed through a 300 μm sieve, and the specimens were temporally fixed in 3 % formalin stained with Rose Bengal. After one day the specimens were transferred to and stored in 70 % ethanol. Pycnogonids have been rarely collected from the Brunei Shelf, with only one specimen retrieved from the several Bubut samples. The entire pycnogonid with the ciliates was temporarily mounted on a glass slide in glycerol (stained as above), and is currently stored in the invertebrate collection of DJM. The specimen was examined using a stereomicroscope (Olympus SX10) and an Olympus BX51 compound microscope with differential interference contrast (DIC). Photographs were taken using a DP 28 Olympus digital camera, and Olympus cellSens (Standard) software. Ciliate measurements were conducted utilizing the Top View 3.7 software for the processing of digital images. The systematic position of suctorian ciliates follows Dovgal (2002, 2013).

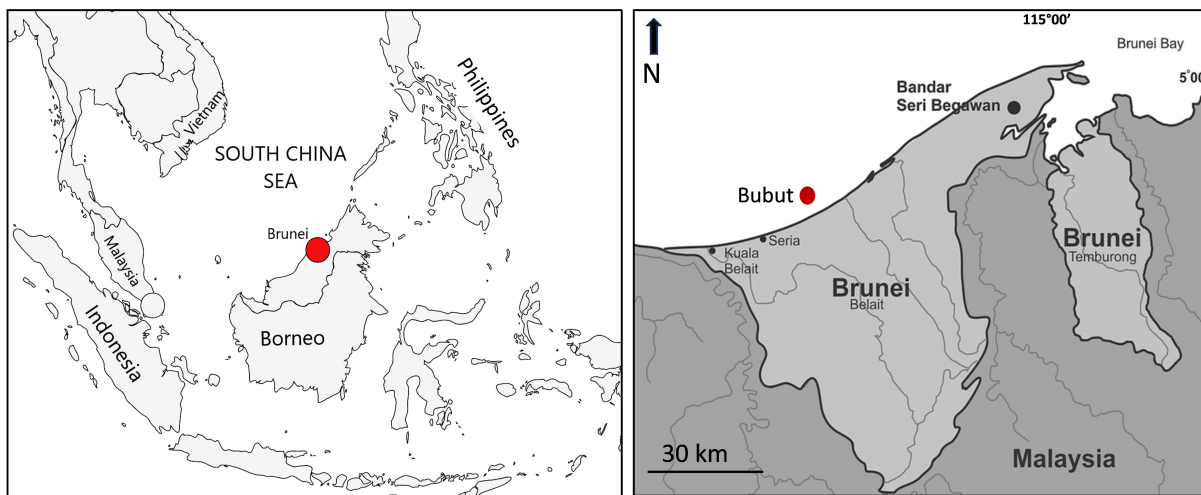


Fig. 1. Map of the collecting site

Рис. 1. Станция отбора проб

Results and Discussion

Basibiont

Class Pycnogonida Latreille, 1810
Order Pantopoda Gerstäcker, 1863
Suborder Eupantopodida Fry, 1978
Superfamily Ascorhynchoidea Pocock, 1904
Family Ammotheidae Dohrn, 1881
Genus *Hemichela* Stock, 1954

Hemichela sp.

One male pycnogonid found at 15–20 m of Brunei Bay as basibiont of ciliates. The basibiont is identified as *Hemichela* sp. The genus *Hemichela* is characterised by the presence of a single dactylus on the chela, which distinguishes it from the closely-related genus *Paranymphe*. The present basibiont specimen is an ovigerous male carrying an egg mass (Fig. 2A and B).

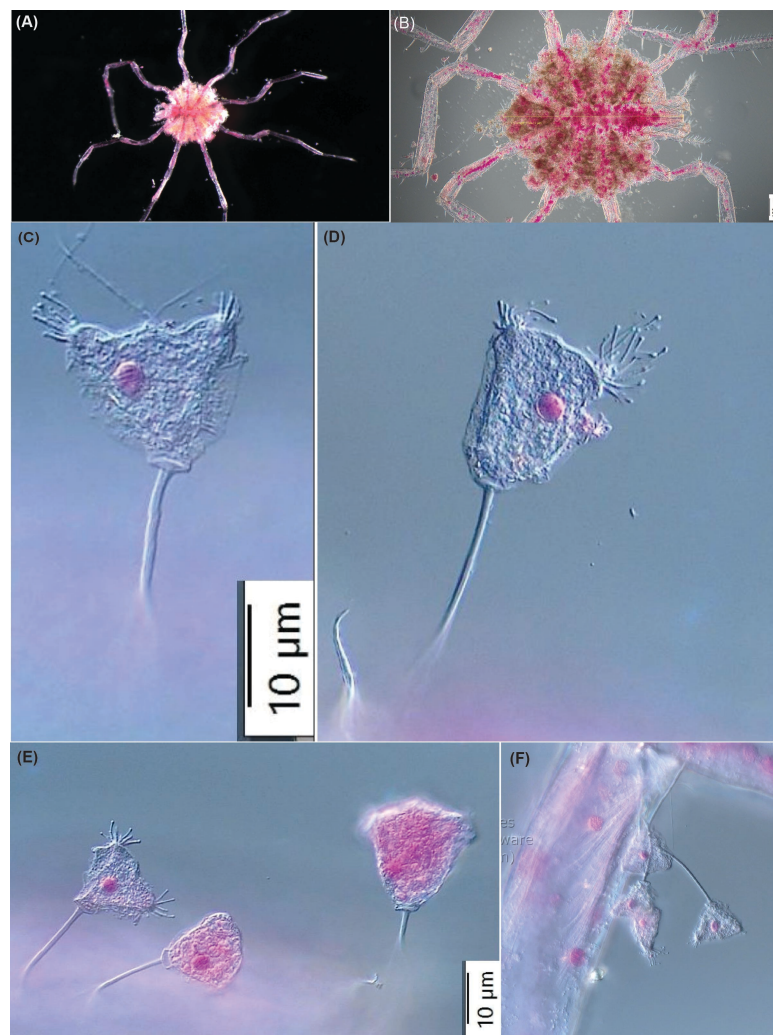


Fig. 2. **A.** Pycnogonid *Hemichela* sp., male (dorsal view), infested with ciliates; **B.** Pycnogonid *Hemichela* sp., male (ventral view), magnified view of egg mass; **C–F.** Magnified views of ciliate *Acineta tuberosa*

Рис. 2. **A.** Самец морского паука *Hemichela* sp. (вид сверху), колонизированный инфузориями; **B.** Самец морского паука *Hemichela* sp. (вид снизу), видно скопление яиц; **C–F.** Увеличенные изображения инфузории *Acineta tuberosa*

Epibiont

Phylum Ciliophora Doflein, 1901

Subphylum Intramacronucleata Lynn, 1996

Class Suctorea Claparède & Lachmann, 1859

Subclass Endogenia Collin, 1912

Order Acinetida Raabe, 1964

Family Acinetidae Ehrenberg, 1838

Genus *Acineta* Ehrenberg, 1834***Acineta tuberosa* Ehrenberg, 1834 (Fig. 2C–F)**

Material examined: Numerous ciliates were found attached on legs of the pycnogonid *Hemichela* sp.

Brief Description: Small (body length up to 19 μm), stalked, loricate suctorian ciliate with flattened triangular cell body and rounded macronucleus, which centrally or some eccentrically located. The developed basal disc is presented in joint between the stalk and the lorica. The actinophores and tentacles were indrawn in some individuals.

Measurements (in μm , based on 7 individuals): Body length 12–19, width 12–16; actinophore length 2–4, width 2–5; macronucleus diameter length 2–4; stalk length 14–31 diameter 1; basal disc width 3–5; tentacle length 2–5. Detail measurements of each of the 7 specimens are given in table 1.

Remarks: The present species is similar to *Acineta tuberosa* Ehrenberg, 1834, but differs from this by its smaller size (12–19 μm in length versus 25–120 μm after [Curds, 1985]) and the presence of a basal disc. However, *A. tuberosa* is a widely distributed and ubiquitous species, with high morphological variability. Thus, the observed differences between the present species and other *A. tuberosa* are insufficient to ascribe new species status with any confidence.

This is a ubiquitous marine and brackish species, observed on inanimate substrates and algae, hydroids, bryozoans, crustaceans, among other in the North, White, Caribbean, Yellow, Caspian, Mediterranean, Black, Azov Seas, the Atlantic, Pacific and Indian Oceans [Kahl, 1934; Wailes, 1943; Chen, Song, Hu, 2005; Fernandez-Leborans, Chatterjee, Grego, 2012; Dovgal, 2013; Chatterjee et al., New records ... , 2013; Chatterjee, Fernandez-Leborans, Senna, 2013; Chatterjee, Kotov, Fernandez-Leborans, 2013; Chatterjee, Dovgal, Nanajkar, 2020; Chatterjee et al., Report of ... , 2024]. The present study represents the first report of *A. tuberosa* on a pycnogonid host. This is also first report of *A. tuberosa* from the Brunei coastal area.

Notes on ciliate epibiont on pycnogonids: There are several groups of organisms identified as epibiotic on pycnogonids. These include organisms like multicellular algae, diatoms, foraminiferans, ciliates, entoprocts, bryozoans, and hydrozoans [King, 1973; Key, Barnes, 1999; Pipe, 1982; Lane et al., 2016; Lane et al., 2018; Kakui, Sekiguchi, 2024]. Table 2 gives a list of ciliates known to be epibionts of pycnogonids. Ciliate epibionts on pycnogonids are represented by peritrichs, suctorians and folliculinid ciliates. Three of these ciliate species have been identified up to species level, and are found also on other basibiont organisms.

Whereas ciliate epibionts are commonly found on organisms of other aquatic taxonomic groups, like isopods, cladocerans, cnidarians, bryozoans, rotifers, nematodes, ostracods, mites [Chatterjee et al., New records ... , 2013; Chatterjee, Fernandez-Leborans, Senna, 2013; Chatterjee, Kotov, Fernandez-Leborans, 2013; Chatterjee et al., 2018; Chatterjee, Nanajkar, Dovgal, 2019; Chatterjee et al., 2023; Chatterjee, Dovgal, Nanajkar, 2020], they are rare on pycnogonids. The reason for this may relate to relatively less investigation of pycnogonids, which themselves are typically rare in benthic samples. Wambreuse, Hamel, Mercier (2021) found that in a single investigation, 26 % of deep sea pycnogonids

were in fact infested by ciliate epibionts. Their samples contained two species belonging to the genus *Ephelota*, and one folliculinid with individuals mainly located on the legs, especially on coxae and femurs.

Lane [Lane et al., 2016] showed that in temperate environments, epibionts are not energetically costly to pycnogonids and, in some circumstances, they may be beneficial. On the other hand, there are observations of epibionts, such as poriferans and bryozoans which cover articulations between leg segments and restrict movement of pycnogonids [Wambreuse, Hamel, Mercier, 2021]. More investigations are needed to better understand the relationship between epibionts and the host pycnogonids. Understanding the role of pycnogonids as basibiont for ciliate and other epibionts may improve our knowledge on the ecosystem functioning of the aquatic biota, with its multifaceted relationships within and between biotic and abiotic factors.

Acknowledgements. First author (TC) thanks Dr. Cengiz Koçak, Department of Hydrobiology, Ege University, Türkiye for his help to identify the pycnogonid (basibiont).

Table 1. Measurements of *Acineta tuberosa* found attached to the pycnogonid *Hemichela* sp. (in µm, based on 7 individuals)

Таблица 1. Морфометрические характеристики *Acineta tuberosa* с морского паука *Hemichela* sp. (в мкм, промерены 7 особей)

No of individual	Body length	Body width	Actinophore length	Actinophore width	Macro-nucleus diameter	Stalk length	Stalk diameter	Basal disc width	Tentacle length
1	13	12	4; 4	3; 5	3	16	1	4	3; 3; 4; 3; 2
2	12	15	2; 4	3; 3	2	17	1	5	4; 4; 4; 3; 3; 5; 3
3	15	12	-	-	4	14	1	4	-
4	19	16	-	-	4	17	1	3	-
5	16	12	3; 3	5; 4	3	23	1	3	3; 2
6	13	15	2; 3	3; 4	4	31	1	3	2; 2; 2; 3
7	13	13	3; 2	3; 2	3	27	1	4	3; 2

Table 2. Finds of ciliates as epibionts on pycnogonid basibionts

Таблица 2. Известные находки инфузорий — эпибионтов морских пауков

Ciliate (epibiont)	Pycnogonid host (basibiont)	Locality	Depth	References	Remarks (if any)
<i>Rhabdophrya nymphonis</i> (Gassovsky, 1916) = <i>Dendrosomides nymphonis</i> Gassovsky, 1916	<i>Nymphon</i> sp.	Kola Bay of the Barents Sea		Gassovsky, 1916; Kahl, 1934	Also found on hydroids, algae and bryozoans from the Barents Sea near Murmansk [Jankowski, 2007; Chatterjee, Dovgal, 2020; Chatterjee, Dovgal, Sautya, 2022]
Unidentified peritrich	<i>Phoxichilidium tubulariae</i>	North Sea	19.5 m depth on steel gas production platform	Pipe, 1982	

Continued on the next page...

Ciliate (epibiont)	Pycnogonid host (basibiont)	Locality	Depth	References	Remarks (if any)
Unidentified suctoria	<i>Phoxichilidium tubulariae</i>	North Sea	19.5 m depth on steel gas production platform	Pipe, 1982	
<i>Ephelota</i> spp (2 species)	<i>Nymphon hirtipes</i>	North Atlantic	315–620 m depth	Wambreuse, Hamel, Mercier, 2021	
Folliculinid	<i>Nymphon hirtipes</i>	North Atlantic	315–620 m depth	Wambreuse, Hamel, Mercier, 2021	
Folliculinid	Unidentified pycnogonid	Juan de Fuca Ridge	Axial Volcano vents, Deep sea	Kouris et al., 2007	
<i>Thecacinetia oregonensis</i> (Murphy, 1965)	<i>Ammothella appendiculata</i> (Dohrn, 1881)	Sandspit, Karachi coast, Pakistan, Northern Arabian Sea	Intertidal	George, Siddiqui, 2020	Also found on nematode <i>Desmodora</i> sp. [Murphy, 1965; Chatterjee et al., 2023]
<i>Acinetia tuberosa</i> Ehrenberg, 1834	<i>Hemichela</i> sp.	Off Brunei Darussalam, Brunei Bay, South China Sea	Subtidal	Present record	

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**ПЕРВОЕ НАБЛЮДЕНИЕ ACINETA TUBEROSA (CILIOPHORA)
КАК ЭПИБИОНТА МОРСКОГО ПАУКА (PYCNOGONIDA)
С СЕВЕРО-ЗАПАДНОГО ПРИБРЕЖЬЯ БОРНЕО (БРУНЕЙ) И КОММЕНТАРИИ
ОБ ИНФУЗОРИЯХ — ЭПИБИОНТАХ ПИКНОГОНИД**

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Аннотация: Суктория *Acineta tuberosa* Ehrenberg, 1834 была обнаружена как эпibiонт морского паука (Pycnogonida) *Hemichela* sp. в сублитеральной зоне побережья Южно-Китайского моря (Бубут, Бруней-Даруссалам). Инфузория с уплощённым треугольным клеточным телом, снабжённым стебельком и раковиной, с округлым макронуклеусом и булавовидными щупальцами, собранными в два пучка на двух широких и коротких актинофорах. Это первая находка *A. tuberosa* на морском пауке, а также первая находка данного вида в побережье Брунея. Приводятся описание инфузории, а также обсуждаются известные случаи эпibiоза цилиат на пикногонидях.

Ключевые слова: инфузория, эпibiоз, пикногонид, базибионт, Бруней-Даруссалам

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Поступила в редакцию 03.04.2025

Принята к публикации 10.07.2025