

Eco-Taxonomic Observations on *Epithemia adnata* (Kützing) Brébisson (Bacillariophyta) from Murguma Reservoir, Purulia, West Bengal; India.

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Abstract

The present paper deals with the eco-taxonomic observations on *Epithemia adnata* (Kützing) Brébisson collected from Murguma reservoir of Purulia district in West Bengal, India. For this purpose Differential interference contrast (DIC) and phase contrast images were taken covering whole size range of the valves and FE SEM also done. Ecological parameters were also considered. Authors tried to compare these taxa with the other six taxa of “*adnata*” group available from scientific literature.

Key Words: *Epithemia adnata*, Murguma, Purulia, West Bengal, India.

Introduction

The genus *Epithemia* Kützing belongs to the family Rhopalodiaceae (Karsten) Topachevs’ky & Oksiyuk under the order Rhopalodiales Mann (Round *et al.*, 1990). Morphologically *Epithemia* is strongly dorsiventral having canal like V shaped raphe and areolae. These are the basic characteristics of Rhopalodiales. Ruck and Theriot, 2011 are of opinion that canal like raphe system originated more than once in course of evolution and forwarded the conception that Rhopalodiales should be placed within Surirellales. They rejected the monophyly of Bacillariales, Rhopalodiales and Surirellales.

Epithemia Kützing a moderately sized genus and according to Guiry and Guiry, 2015 there are 59 species currently accepted taxonomically under this genus. In addition to *Epithemia adnata* (Kützing) Brébisson other six taxa are also placed in this group (“*adnata*” group) according to Vishnyakov *et al.*, 2014. These taxa are differentiated by the characters like cell shape and size, apex shape, extent of raphe curvature and striae/costae density etc. Comparisons of characters of these taxa are given in Table 1. The above mentioned taxa *Epithemia adnata* (Kützing) Brébisson was collected from a reservoir namely Murguma (Plate I, Figs. 1-14). The latitude and longitude of that place are respectively N 23°18'57" and E 86°03'14".

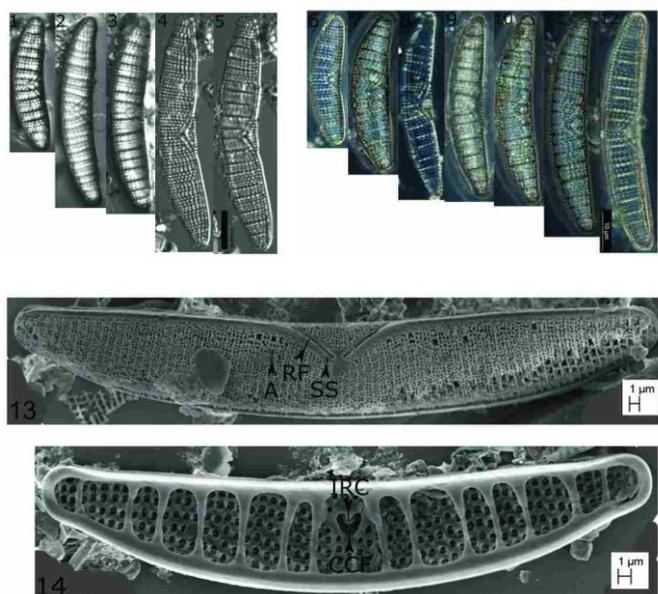


PLATE I. Figs. 1-14: *Epithemia adnata* (Kützing) Brébisson. Figs. 1-5: Differential Interference Contrast (DIC) images. Figs. 6-12: Phase Contrast (PC) images. Fig. 13: External valve view showing raphe fissures (RF), thin silica strip (SS) and areola (A) (SEM).

Fig. 14: Internal valve view showing internal raphe canal (IRC) and central continuous fissure (CCF) (SEM).

Scale bars: DIC bars = 10µm (Figs. 1-5); PC bars = 10µm (Figs. 6-12); SEM bars see in each figure.

Diatom study was initiated in India from 18th century by the diatomologists like Ehrenberg, 1845; Gandhi, 1998 and Sarode and Kamat, 1984. India is extremely rich in diatom diversity. There are greater chances of endemism in several taxa since nobody has taken any initiative except Karthick *et al.*, 2011 & 2013 to study diatom morphotaxonomy in light of modern trends of microscopy considering the current taxonomic tools and concepts establishing the endemism or speciation etc are hard to reconstruct. Therefore the authors decided to explore the taxa in light of modern trends considering the morphology and ecology of the species relating with the collection spots.

Materials and Methods

Study Area

This study area covered the Murguma reservoir. Murguma is a small tribal village in Purulia district of West Bengal, India (Plate II, Figs. 15, 16). The tribal word 'Murguma' means the home for the peacocks. This reservoir or Dam is located on the Saharjor river and is surrounded by hillocks and forests situated near Ajodhya Hills (extended part of Eastern Ghats range).

Sample Collection

Diatom materials were collected from this dam on 21st February, 2015. All together 16 sites (Plate II, Figs. 17-19) were sampled. Epiphytic samples were collected by crushing the submerged roots and stems of aquatic plant materials and resulting suspension was transferred into a glass vials. Epilithic (Plate II, Fig. 19) samples were collected by vigorously scrubbing submerged and semi-submerged stones with a tooth brush and the liquid containing diatoms transferred into another glass vials. Episammic samples were also collected by using a dropper. All samples were preserved in 70% ethanol. pH, temperature, TDS, electrical conductivity and salinity of the spot were measured using PCS multiparameter tester 35 series device.



PLATE II. Figs. 15-19: Murguma Reservoir of Purulia district, West Bengal; India. Fig. 15: Google map image. Fig. 16: Google satellite image. Figs. 17-18: Natural view. Fig. 19: Showing epilithic habitat.

Cleaning Techniques

Sub-samples were cleaned using sodium hypochlorite solution (4% w/v available chlorine) or 30% hydrogen peroxide solution. These procedures are modified from the techniques of Krammer and Lange-Bertalot, 2000; Taylor *et al.*, 2005 and Karthick *et al.*, 2010. The organic coating removed and clean samples were then repeatedly centrifuged at 3000-3500 rpm and alternatively rinsed with distilled water for 4-5 times.

Slide preparation & Microscopy

Small drop of cleaned sample were mounted onto glass slides using MeltMountTM (R.I. 1.704) mounting medium and subsequently observed with an Olympus IX 81 Confocal LS microscope equipped with 100X DIC (oil) optics and photographs were taken with IPP software. Phase contrast images were taken with Leica DM 1000 LED compound light microscope with the help of LAS software.

Scanning electron microscopy was performed using aliquots of the cleaned material air dried on cover slip and mounted on aluminium stub using double sided carbon tape. Stub was sputter coated with gold by Quorum Q 150R model and observed under Zeiss SUPRA 55 FE SEM using accelerating voltage 5 kv and a working distance of 2.4 mm.

The collected preserved materials along with the permanent slides were stored in the Herbarium of Phycology laboratory in the department of Botany, The University of Burdwan for future study and reference purpose.

Results

All 16 sites had alkaline waters (pH 9.23-9.52) and in all site the observed taxa i.e. *Epithemia adnata* (Kützing) Brébisson was found in epilithic and epiphytic conditions. Other ecological parameters of those spots were measured and are noted below:-

Water temperature: 22.0-22.4°C; Electrical Conductivity (EC): 132-134 μ S/cm; Total Dissolved Solids (TDS): 95.2-95.8 ppm; Salinity: 65.8-66.3 ppt.

According to the classification proposed by Round *et al.*, 1990 and Medlin and Kaczmarska, 2004 *Epithemia adnata* (Kützing) Brébisson belongs to:

Division: Bacillariophyta

Sub-division: Bacillariophytina Medlin & Kaczmarska 2004

Class: Bacillariophyceae Haeckel 1878, emend. Medlin & Kaczmarska 2004

Sub class: Bacillariophycidae (Haeckel 1878) Mann 1990

Order: Rhopalodiales Mann 1990

Family: Rhopalodiaceae (Karsten) Topachevs'kyj & Oksiyuk 1960

***Epithemia adnata* (Kützing) Brébisson 1838**

(Pl. I, Figs. 1-14)

(Hustedt 1930, p.385, fig. 729; Krammer & Lange-Bertalot 1997, p. 152, pl. 107, figs. 1-11, pl. 108, figs. 1-3.)

Synonym: *Frustulia adnata* Kützing 1833.

LM morphology: Valves more or less dorsiventral, dorsal margin somewhat convex, ventral margin slightly concave, apices broadly rounded or slightly protracted, observed valve length 35-62.14 μ m, breadth 7.19-10 μ m. Raphe is biarcuate in nature i.e. branches curve from the poles inwardly towards the dorsal side but never reach on that margin, striae are 12-14 in 10 μ m, costae are 3-4 in 10 μ m with 3-6 striae between adjacent costae, costae are more or less parallel to each other or slightly radiate in nature.

SEM morphology: The external raphe fissures (RF) are surrounded by a thin silica strip (SS) on each side, raphe fissures created a V-shaped structure at the centre of ventral margin. In the external valve view we can see a regular and uniform arrangement of domed caps that is linked apically and transapically. These domed caps are usually of four to eight in number and which forms one areola (A). These areolae are very complicated in structure and which makes the identities and boundaries more complex. The raphae fissures are more or less situated in same distance from both margins at the apical region. To the dorsal side of fissure a hyaline band is present and extends along with the valve length.

Internal valve structure shows regularly arranged transapical costae which creates more or less rectangular shapes and interstitial bars. In central part of the valve shows internal raphae canal (IRC) and central continuous fissure (CCF).

Table 1. Comparison of characters of members of “*adnata*” group of *Epithemia* to *Epithemia adnata* (Kützing) Brébisson

Taxon	Valve shape	Length (in 10 µm)	Width (in 10 µm)	Striae (in 10 µm)	Costae (in 10 µm)	Reference
<i>Epithemia adnata</i> (Kütz.) Bréb. 1838	Convex dorsal and somewhat concave ventral margin, apices broadly rounded or slightly protracted.	32 – 62.14	7.19 – 10	12 – 14	3 – 4	Current Study
<i>E. adnata</i> var. <i>minor</i> (Perag. & Hérib.) Patr. 1975	Strongly convex dorsal and somewhat concave ventral margin, apices strongly protracted.	36	9.5	11-13	3-4	Foged , 1984
<i>E. adnata</i> var. <i>porcellus</i> (Kütz.) Patr. 1975	Concave ventral and convex dorsal margin, apices more or less truncate to capitate and slightly reflexed.	40 – 95	8 – 13	10 – 15	3 – 5	You <i>et al.</i> , 2009
<i>E. adnata</i> var. <i>proboscidea</i> (Kütz.) Hende 1954	With almost straight or slightly concave ventral margin and convex dorsal margin, apices rostrate or slight capitate.	40 – 70	8 – 10	10 – 13	2 – 4	You <i>et al.</i> , 2009
<i>E. adnata</i> var. <i>saxonica</i> (Kütz.) Patr. 1975	Valves are shorter and more compact, convex dorsal margin and slightly concave ventral margin, gradually narrow at the apices.	30 – 70	8 – 11	11 – 14	3 – 5	You <i>et al.</i> , 2009
<i>E. selengaensis</i> Vishnyakov, Kulikovskiy & Genkal 2014	Approximately parallel, slightly concave ventral and convex dorsal margin, apices strongly attenuated, abruptly narrowed and rounded.	45 – 64	7 – 9	3 – 8	2 – 4	Vishnyakov <i>et al.</i> , 2014
<i>E. frickei</i> Krammer 1987	Weakly concave ventral and convex dorsal margin, dorsal margin abruptly narrow towards the end of the valve to form depressions, widely rounded apices.	32 – 64	9.5 – 14	10 – 11	2 – 4	Vishnyakov <i>et al.</i> , 2014

Discussion

Epithemia adnata (Kützing) Brébisson was found to grow in more or less standing water having moderately high electrolyte content and high alkalinity. The taxa appears to tolerate elevated water temperatures and since collection spots showed higher TDS than the salinity that means the water is not clean i.e. organic solutes were present in addition to the salt ions. High TDS generally indicates a high alkalinity or hardness of water.

Species of “*adnata*” group of *Epithemia* are characterised by shape of valve, striae in 10 µm and costae in 10 µm and costae in 10 µm. For these features the members of this “*adnata*” group fall into one polytypic species *Epithemia adnata* (Kützing) Brébisson.

According to Vishnyakov *et al.*, 2014 *E. adnata* var. *porcellus* (Kütz.) Patr. and *E. adnata* var. *proboscidea* (Kütz.) Hende should be considered as separate species respectively as *E. porcellus* Kützing and *E. proboscidea* Kützing because of typical morphological features such as attenuate rostrate ends made them to place in separate species rather than treating them under variety of the species *adnata*. But many researchers including the algaeBase consider these taxa under species *adnata*. *E. selengaensis* Vishnyakov, Kulikovskiy & Genkal was misidentified and treated as *E. adnata* var. *porcellus* (Kütz.) Patr. or *E. adnata* var. *proboscidea* (Kütz.) Hende by many scientists.

Conclusion

Epithemia adnata (Kützing) Brébisson shows high diversity in the water bodies of Purulia. Alongside widespread “*adnata*” group of taxa some new taxa may also be discovered from the place by critical investigation as local-endemism is noticed specifically among diatoms.

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