



## **RESEARCH ARTICLE**

## Seven new additions to euglenoid flora of India from petroleum hydrocarbon contaminated water bodies of Naga- Schuppen thrust belt

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#### Abstract

Comprehensive explorations were conducted along the Naga- Schuppen thrust belt of Northeast India, a hydrocarbon province of Southeast Asia, during the years 2021-2023 which revealed altogether 7 new distributional records of freshwater Euglenophycean species for India. Out of the 7 identified taxa, *Phacus corculum, Menoidium tremulum, Trachelomonas crispa, Trachelomonas corniformis, Trachelomonas nadsonii* and *Trachelomonas rugulosa* var. *obliqua* are found to be newly reported algal species not only from India but from the entire Southeast Asia province. All the reported Euglenophyte flora belongs to 2 classes, 2 orders and 3 families. In addition, the systematic accounts of all these species are discussed in detail.

## **Keywords**

New record; India; euglenophytes; diversity; hydrocarbon

#### Introduction

Photoautotrophic algae Euglenophytes are a group of flagellates that are cosmopolitan in nature and are commonly found free-swimmers. These are also often named as Euglenozoa or Euglenoids (1). Almost all the species in this incredibly varied group are unicellular and are capable of living in both fresh and saline water, which is abundant in organic material (2). Generally, higher water temperatures and long days of light during the summer season are found to be congenial for their luxuriant growth in both lotic and lentic water bodies (3). Further, a few members of Euglenophytes have been recently recognised as a tool for water quality monitoring (4).

Studies on algal research in India have documented around 7310 algal species belonging to 835 genera, organised under 11 classes, 95 orders and 255 families (5). Amongst them, the class Euglenophyceae, a remarkable microalgal group, is known to have 516 diverse taxa (384 species, 117 varieties and 15 forms) which include 509 Euglenoids reported from freshwater habitat (6).

Different groups of researchers worldwide, including those from India, have worked significantly on euglenophytes from different habitats (4, 7 -25). In contrast, the primary source of knowledge regarding euglenoids reported from oil contaminated sites is scanty (26). Therefore, in this taxonomic investigation, an attempt has been made to explore freshwater Euglenophycean micro-algal diversity in some crude oil contaminated water bodies located in oil exploration sites, which are organically polluted due to crude oil exploration since 1857 and subsequent leakage and seepage during transportation, refining etc.

## **Materials and Methods**

## Study area

Around 31 crude oil contaminated lentic water bodies of varied sizes located in and around Digboi, Jorajan and Makum oil fields under the Naga-Schuppen thrust belt (Fig. 1), a part of one of the significant hydrocarbon basins in India which houses many running and abandoned oil fields (27) were explored for euglenophytes from May 2021 to April 2023 for 2 years. The TOC (Total Oil Content) was beyond 36.56 g/L in those water bodies on an average throughout the studied year. The region enjoys a subtropical monsoon climate with a cold winter, wet spring and humid summer. The average annual rainfall is  $\pm$  1950 mm.

dehyde and Lugol's iodine solution when necessary. Identification was done based on their morphological features following standard taxonomic monographs, online databases and research publications. Taxa were updated using AlgaeBase (29) and then arranged following a previous report (30).

#### **Results and Discussion**

Altogether, seven Euglenoid species were revealed to occur in different hydrocarbon contaminated water bodies, which include 5 taxa under the genus *Trachelomonas* Ehrenberg, 1834 and one taxa each under *Phacus* Dujardin, 1841 and *Menoidium* Perty, 1852 respectively, during the present taxonomic survey. It is also to be noted that all 7 species were not identified and reported so far from India.

All the species of euglenoid taxa documented

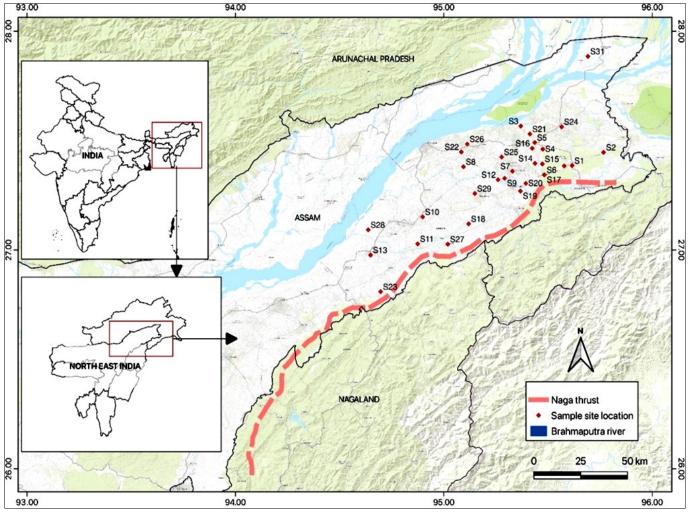


Fig. 1. Map of the study area with 31 locations of crude oil contaminated water bodies along Naga thrust.

#### Sample collection

To enumerate the Euglenophycean diversity in the study area, samples were collected by filtering water through a plankton net with a mesh size of 45  $\mu$ m (28). Concentrated filtrates were then brought to the laboratory with proper labelling. The algal samples were preferably observed in fresh conditions under a Euromex Delphi X Observer series microscope with Image View software and photographs were clicked using a Euromex Digital camera (20 MP). For further observation, preservation was made in 4 % formalduring this investigation are given with detailed morphological descriptions below in tabular form (Tables 1-3). Of these euglenoids, *Phacus corculum* (Fig. 2A), *Menoidium tremulum* (Fig. 2B), *Trachelomonas crispa* (Fig. 2C), *Trachelomonas corniformis* (Fig. 2D), *Trachelomonas nadsonii* (Fig. 2E) and *Trachelomonas rugulosa* var. *obliqua* (Fig. 2F) are found to be new additions to the algal flora not only of India but also to the entire Southeast Asia province. Table 1. Morphological characteristics of species new to India under genus Trachelomonas with their comparison to already reported taxa

Species	Features	Source	Information available on the taxa with source Characters	Present study	
	Shape		Cylindrical, to ellipsoidal	Ellipsoidal, wall covered with minute sharp spines	
	Length		36-40 μm	37.1-40.6 μm	
	Width		16-18 μm	15-18 μm	
Trachelomo- nas crispa Balech 1944: 273, figs 90, 91, 201 (Fig. 2C)	Colour	(31)	Brown	Brown to Dark yellow	
	Collar		Low collar having a circle of short spines around the mouth	Low collar having a circle of short spines around the mouth	
	Flagellum		No mention	Not seen	
	Habitat		Moat, Ditch, Lake	Ditch, Wetland	
	Habit		No mention	Free Floating	
	Shape		Broad ovoid to ellipsoidal, densely punctate	Ovoid, wall densely punctate all over	
	Length	(31)	33-38 μm	33.4-36.2 μm	
Trachelomo- nas corniformis Y.V.Roll 1925: 138, 147, pl. V [5]: fig. 3 Fig. 2D)	Width		24-28 μm	23-28.3 μm	
	Colour		Dark brown	Dark brown	
	Collar		Collar absent but encircled with short spines around the mouth	No collar and encircled with short spines around the mouth	
	Flagellum		Cylindrical, to ellipsoidalF36-40 µm316-18 µm1BrownKLow collar having a circle of short spines around the mouthSNo mentionNMoat, Ditch, LakeCNo mentionFBroad ovoid to ellipsoidal, densely punctateC33-38 µm324-28 µmCCollar absent but encircled with short spines around the mouthNNo mentionNCollar absent but encircled with short spines around the mouthNNo mentionPCollar absent but encircled with short spines around the mouthNNo mentionPFusiform to spindle shaped with wavy margins and densely covered spinesF65-69 µmG19-20.1 µmCBrownAAttenuated into a long neck like process with terminal spikeFPlanktonPPlanktonPPlanktonDApeical pore with a short collar directly inside loricaTPonds, PuddlesPNo mentionPPonds, PuddlesPNo mentionPSpherical, distinct spirally arranged ribsSBrownBApical pore with a short collar directly inside loricaTPonds, PuddlesPNo mentionPPords, PuddlesPNo mentionFWithout collarYWithout collarY	Not seen	
	Habitat		Lake	Ponds, Ditch	
	Habit		No mention	Plankton	
	Shape	(32)		Fusiform, Spherical with densely cov- ered spines	
	Length		65-69 μm	65-69.2 μm	
	Width		19-20.1 µm	20-20.3 μm	
<i>Trachelomonas nadsonii</i> Skv ortzov 1925: 78	Colour		Brown	Brown	
(Fig. 2E)	Collar		Attenuated into a long neck like process with terminal spike	Attenuated into a long neck like pro- cess with terminal spike	
	Flagellum		No mention	Not seen	
	Habitat		Ponds, Lakes, Ditches	Ponds, Paddy field	
	Habit		DW Collar naving a circle or short spines around the mouth spines around the mouth spines around the mouth spines around the mouth spines around the spine around the spines around the spines around	Plankton	
	Shape		Spherical, distinct spirally arranged ribs	Spindle shaped, densely covered with minute spines	
	Length		14-21 μm	14.6-20 μm	
Trachelomo- nas rugulosa var. obligua Bo	Width		20-22 μm	20-22.2 μm	
nas rugulosa var. obliqua Bo urrelly 1952: 185, pll. XXIII: ïgs 308, 309 Fig. 2F)	Colour	(33)	Dark yellow	Dark yellow	
	Collar		Apical pore with a short collar directly inside lorica	With a short collar directly inside loric	
	Flagellum		Twice long as lorica	Twice long as lorica	
	Habitat		Ponds, Puddles	Ponds, Paddy field	
	Habit		No mention	Plankton	
<i>Trachelomo- nas hirta</i> var. <i>duplex</i> Defland re 1928: 664, fig. 6 (Fig. 2G)	Shape	(31)	Broad- ellipsoidal with round anterior and posterior end	Broad- ellipsoidal, walls covered with short-blunt spines	
	Length		18-22 μm	19-22.3 μm	
	Width		14-15 μm	13-14 µm	
	Colour		Yellowish	Yellowish	
	Collar		Without collar	Without collar	
	Flagellum		No mention	Not seen	
	Habitat		Ditch, Pond, Moat	Ponds, Ditch	
	Habit		No mention	Free Floating	

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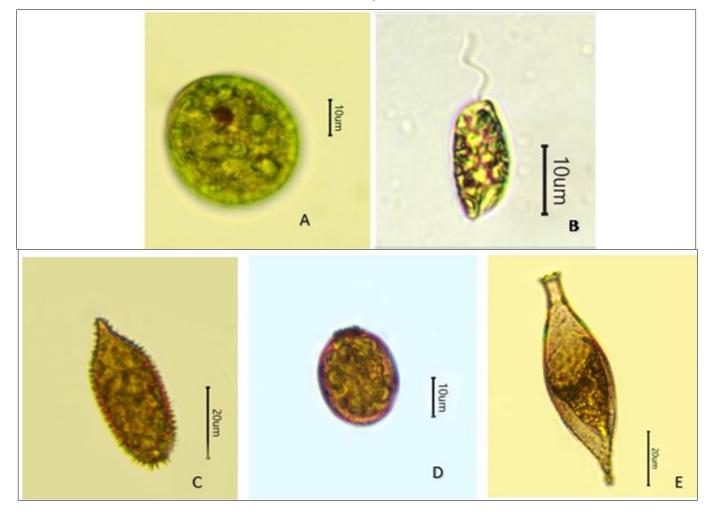
Table 2. Morphological characteristics of species new to India under genus Phacus with their comparison to already reported taxa.

Constant Inc.	F	Information available on the taxa with source		Duranutation
Species	Features	Source	Characters	<ul> <li>Present study</li> </ul>
<i>Phacus corculum</i> Pochmann 1942: 121, fig. 2 (Fig. 2A)	Shape		Heart shaped, cut in anterior end and posterior end narrow	Heart shaped
	Length		17-20 μm	17.8-22 μm
	Width		12-13.6 μm	13.2-14 μm
	Paramylon bodies	(33)	1-2, disc shaped	2 disc-shaped
	Chloroplasts		Numerous	Numerous
	Habitat		Ponds, Ditch, Puddles	Ponds, Ditch
	Habit		No mention	Free floating

Table 3. Morphological characteristics of species new to India under genus Menoidium with their comparison to already reported taxa.

<b>T</b> aux	Frations	Inform	ation available on the taxa with source	Provide the de	
Таха	Features	Source	Characters	<ul> <li>Present study</li> </ul>	
	Shape		Half-moon like bent,	Elongated, curved, with delicate striations	
	Length		18 µm	15-20 μm	
	Width Colour		6 µm	8-10 μm	
			Greenish	Greenish	
Menoidium tremulum Skvortzov 1924: 183, fig. A 9 (Fig. 2B)	Collar	(32)	Anterior end of cell usually protruded into a narrow neck	Protruded narrow neck not seen	
(1.6.20)	Flagellum		Flagellum long	One long emergent flagellum	
	Paramylon bodies Habitat		Large	Large and small	
			Ponds, lakes, rivers	Ponds, Paddy field	
	Habit		Plankton	Plankton	

## Systematic Account



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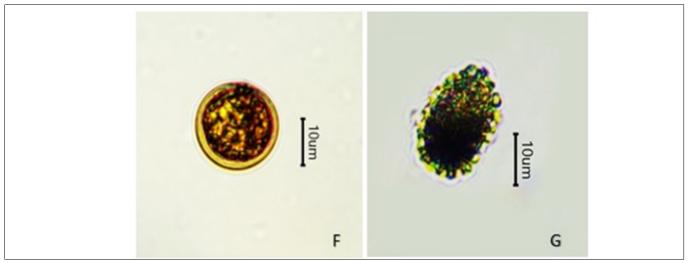


Fig. 2. Photomicrographs of Euglenoid species new to India. A. Phacus corculum Pochmann, B. Menoidium tremulum Skvortzov, C. Trachelomonas crispa Balech, D. Trachelomonas corniformis Y.V.Roll, E. Trachelomonas nadsonii Skvortzov, F. Trachelomonas rugulosa var. obliqua Bourrelly, G. Trachelomonas hirta var. duplex Deflandre.

Phylum: Euglenozoa

Class: Euglenophyceae

**Order: Euglenales** 

Family: Euglenaceae

Genus: Trachelomonas (Ehrenberg, 1834)

Cells are solitary and free swimming, protoplast thin walled and naked and enclosed within a fragile lorica, lorica shapes varies from globose to ovoid, spindle, cylindric, pyriform or fusiform, anterior end with or without distinct collar, sometimes collar coarse, elongated necklike, surrounding terminal flagellum pore, flagellum usually longer than body length, distinct cauda may be present at the posterior end, lorica wall smooth to rough, ornamented, dentate, punctate, granulate, scrobiculate, perforate, reticulate or spinose, spines thick or fine, blunt or sharp, distribution localized, sparse or dense, chloroplasts 2 to many, discoid to trough shaped, paramylon bodies round elongated or absent.

#### Family: Phacaceae

Genus: Phacus (Dujardin, 1841)

Cells are solitary and free swimming, almost flattened, pyriform, fusiform or orbicular, twisted sometimes, cells bear short or long, sharply pointed or blunt caudas at posterior ends, one flagellum at anterior end, pellicle ornamented with spiral or longitudinal striations, chloroplasts numerous in ovoid disc-band like, paramylon bodies circular plates or rings like, sometimes rods like, distinct stigma, some species may have eye spot.

Phylum: Euglenozoa Class: Peranemea Order: Natomonadida Family: Astasiidae Genus: *Menoidium* (Perty, 1852)

Cells are solitary, uniflagellate, compressed, curved,

long fusiform in face view, narrow long thin, cylindric in side view, anterior ends narrowed into a quadrate and beak like apex, posterior ends gradually narrowed into a rounded end, periplast rigid, with longitudinal, parallel striae, cytoplasm granulate, with 2 or several, long rod like or ring like paramylon bodies.

## Conclusion

The euglenoid species documented for the first time from India in the present endeavour could be a momentous contribution to the extensive record of the algal database of the state and the nation as well. As the recorded euglenoid species were from hydrocarbon contaminated water bodies, they are either resistant or adapted to grow in such harsh environment and hence, may be considered as a biomonitoring tool in the future.

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#### **Authors' contributions**

BB: Conceptualization, Visualization, Investigation, Writing original draft. PPB: Supervision, Conceptualization, Writing - reviewing and editing.

#### **Compliance with ethical standards**

**Conflict of interest**: The authors declare that they have no conflict of interest that could have appeared to influence the work reported in this paper.

Ethical issues: None.

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