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**RESEARCH ARTICLE**

# ORANGE WRAPPER AT MANGROVE: *TRENTEPOHLIA FLAVA* (W. J. HOOKER AND ARNOTT) CRIBB, A NEW DISCOVERY FROM BHITARKANIKA CONSERVATORY, INDIA

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**ABSTRACT**

The interesting Chlorophycean epiphytic algae *Trentepohlia* forming mostly yellow to bright orange or red-brown coloured covering on tree barks, rocks, walls etc. The features of the order Trentepohliales have been investigated in details from several decades and it has taken a great interest to the scientists for its form and diversity. However, the information available for this group in India is relatively limited and insufficient. The collections from Bhitarkanika Conservatory, a rich, sensitive mangrove eco-system lying in the estuarine region of Brahmani- Baitarani in the North-Eastern corner of Kendrapara district of Orissa, India which allowed the new discovery and characterization of some poorly known species possessing tremendous ecological and taxonomic value, such as *Trentepohlia flava* appeared as orange covering on the tree bark within the mangroves. The morphology and distribution of the species described and the taxonomic as well as bio-geographical implications of the new records are discussed in this paper.



## INTRODUCTION

The order Trentepohliales includes sub-aerial green algae widespread in tropical and temperate regions with humid climates, where they occupy a wide range of habitats and occur on a great range of substrata (Chapman 1984; Ettl & Gärtner 1995; Thompson & Wujek 1997). A large number of studies on the morphology of these algae have been published (e.g. Printz 1939; Cribb 1970;Chapman 1984-2001;Hariot 1989-91;John 2003;Islam 1962; Harvey 1860; etc). This order is distinguished from all other green algae by the unique combination of the following features: presence of β-carotene and haematochrome (which colour the thallus yellow, orange or red), absence of pyrenoids in the chloroplast, a unique flagella apparatus, transverse cell walls with plasmodesmata, and presence of a unique reproductive structure, the sporangiate lateral.

The sporangiate lateral is a highly modified branch; it consists of an apical cell (the suffultory cell) that is swollen basally and tapers apically into a short neck, on which a spherical or oval zoosporangium is borne. To date, species-level identification in the Trentepohliales is almost entirely based on gross morphology, which in some species is known to encompass considerable variation. Due to such unusual combination of features, position of the Trentepohliales at the class level has been long uncertain (Chapman *et al.*, 2001; Lopez-Bautista *et al*., 2002) . Within the order the genus Trentepohlia would not, at first glance, be taken as a green alga as the species are mostly yellow to light orange or red-brown in colour due to the presence of haematochrome which usually hides the green of the chlorophyll and protects the alga's chlorophyll in the sub-aerial habitats (Mondal and Mondal [Parui], 2008) However, the information available for this group in India is relatively limited and insufficient. The new collections from Vitarkanika National Park, the rich, sensitive mangrove eco-system lying in the estuarine region of Brahmani-Baitarani in the North-Eastern corner of Kendrapara district of Orissa, India which allowed the rediscovery and characterization of some poorly known species, of dubious taxonomic validity, such as *Trentepohlia flava.* Mangroves are salt tolerant, complex and dynamic eco-system that occur in tropical and subtropical inter-tidal regions. Bhitarkanika is one such location of rich, sensitive eco-system lying in the estuarine region of Brahmani- Baitarani in the North-Eastern corner of Kendrapara district of Orissa, India (Figure1).

The area is intersected by a network of creeks with Bay of Bengal on the East. The alley between the meandering creeks and rivers, houses the second largest viable mangrove eco-system of India. Its 672 sq.kms. of mangrove forest & wetland, provides home to well over 215 species of birds including winter migrants from central-Asia and Europe. Giant salt water crocodiles and variety of other Wildlife inhabitation in this eco-system which form Asia's one of the most spectacular Wildlife area. Areas of 145 Sq.kms. have been notified as Bhitarkanika National Park vide Notification No.19686/F & E dated 16.9.1998 of Forests & Environment Department, Govt. of Orissa. It has much significance with regard to ecological geo-morphological and biological

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background which includes mangrove forests, rivers, creeks, estuaries, back water, accreted land and mud flats. Bhitarkanika National Park is the core area of Bhitarkanika Sanctuary. Bhitarkanika Wildlife Sanctuary was declared vide notification No.6958/FF AH Dated. 22.04.1975 over an area of 672 square kilometres. The Sanctuary comprising Mangrove Forests meandering rivers, innumerable criss-crossed tidal inundated creeks provide last refuge to the already endangered salt water Crocodile (*Crocodile porosus*). During 2002 the Bhitarkanika mangroves having an area of 2672 sq-km. been declared as a Ramsar site being a wetland of international importance.

## MATERIALS AND METHODS

**Field studies:** During the period of December 2009 to January 2012 ( Except the period the conservatory remain closed), the collections of Trentepohlia flava were made at Bhitarkanika National Park including the sea shore of Bay of Bengal where orange or red-brown patches are preferable to this algae could be recognized by the unaided eye. The pH of the soil samples were also measured by pH meter. Data collection and data analysis through extensive survey at different seasons of the year and estimated using statistical methods.

**Pattern of Survey:** The large area that is 10m X 10m patches of sandy plant were chosen following the Quadrate method for study of species composition and relative cover. The plant samples were randomly collected using quadrate ($1m^2$) along transects perpendicular to the shore.

**Identification and preservation of specimen:** A literature survey was carried out for compilation of existing information on the morphology and others description of this particular genus along with the distribution pattern. Each of the sample material was assigned a field note books and documented, the voucher specimens were collected and identified by referring to BSI, Shibpur, Howrah, India. The voucher specimens were maintained in the herbarium at Department of Botany and Forestry, Vidyasagar University, Midnapur, India. Microphotographs were taken by Nikon (D90) digital camera and pasted with Adobe Photoshop in jpg format.

**Morphological Study:** Morphological study of different plant parts determining the variation in structure and presence of any special character using light microscope and compound microscope (Olympus-CH20i BIMF) and Leica-DM-1000.

## RESULT AND DISCUSSION

**A. Places of Collection:** This species formed a velvety coating on stems and leaves of shrubs and trees in the coastal region of Vitarkanika, Orissa, India (Figure 1 and 2). It was also collected from the painted concrete wall of a cottage-like building at Havelikothi Forest rest house.

**B. Climatic factor:** In summer, temperature is as high as 40°C, in winter it goes down at 10-15°C. May & June are the hottest months of a year. Average annual rainfall is 200-220mm, it has been also seen that naturally growing mangrove species are frequently damaged by heavy air blow during summer season.

**C. Edaphic factor:** There are mainly two types of soil in this region of the coastal zone of Bay of Bengal namely sandy soil & sandy loan. The pH of different soil type is shown in table no.1.

**D. Morphological description of vegetative and reproductive structure:** The alga consisted of erect axes, up to 700-800 µm tall, arising from a well-developed system of prostrate filaments. The erect filaments were straight or curved, poorly branched or unbranched. When present, branches mainly occurred in the lower parts, without a regular arrangement; New branches were issued as protrusions from the top corner of the cells. In the material from Ekakulum and Havelikothi the prostrate filaments were compact, almost pseudoparenchymatous layer (Table 4). In the erect parts, the cells were cylindrical or slightly inflated, 12-16 µm wide (mainly 13-15 µm for Havelikothi, 14-15 µm for Dhamra, Dangamal 12-15 µm, Ekakulum 13-15 µm), 1-3 times as long as wide. Moreover the length and breadth of the vegetative cells of the collected samples were examined using statistical method (Table 2 and Figure4 ) and the results (R=0.69833) also proved that there was no significantly difference in between the measurements of the vegetative cells which allow detecting the sample as a particular species distributed at different zones of this mangrove eco-system. Most of the cells of the erect axes had a thick cell wall (4-5 µm) (Figure 3), heavily grooved by spiral strands .Apical cells were either sharpened, with a variably developed cap.

Whereas in the sample from the cells of the prostrate parts were similar in shape and size to those of the erect parts, in the material from they were mostly globular or elliptical, 13-20 µm in diameter, for both samples, the grooved parts observed in the erect parts was either absent or weakly developed. Presumptive gametangia were observed in the sample from Ekakulum. They were globular or urn-shaped, 20-23 µm in diameter, and occurred either terminally at the top of the erect axes. Some sporangiate laterals, produced either on apical cells or at the top of short lateral branches, were observed in the sample from Dangamal. The zoosporangia were globular or subglobular, 20-30 µm in diameter, with the ostiole opposite to the attachment. The neck of the suffultory cell was straight. Considerable uncertainty has long surrounded the exact circumscription of this species, which is mostly reported in the literature as *Trentepohlia polycarpa* Nees & Montagne or *Trentepohlia aurea* var. *polycarpa* (Nees & Montagne) Hariot. By examination of a large number of relevant herbarium specimens, Cribb (1970) reassessed its taxonomic position and geographical distribution, and renamed it *Trentepohlia flava*. Our specimens are generally in good agreement with the morphological characterization provided by this author and we largely agree with his conclusions. Our records, however, show that this species is not strictly confined to America, as believed by Cribb.

**Identified Host Plants:** The tree species like *Avicennia alba*, *Avicennia officinalis*, *Ceriops decandra* ,*Heritiera fomes*, *Rhizophora apiculata* etc are found as the major hosts of *Trentepohlia flava* and in some area such as Havelikothi sea beach, a complete patch of forest belonging to *Avicennia alba* are almost covered by this algal Species (Table.3 and Figure 6-10).

**Table 1: pH of the soil samples**

| Study Area | Place of Collection | Quality of Soil | pH | Organic matter (%) |
|---|---|---|---|---|
| Bhitarkanika National Park (Bhadrak district), Orissa, India | Dangamal | Medium to Fine | 7.5-7.6 | 0.08-0.21 |
| | Ekakulum | Medium | 6.8-7.0 | 0.07-0.15 |
| | Havelikothi | Coarse | 7.0 – 7.1 | 0.01-0.07 |

**Table 2. Independent t-Test on Data Sample A and Sample B**

| Data | Breadth of cell of collected sample in μ | Mean | SE | Variance | N | Result |
|---|---|---|---|---|---|---|
| A | 13 | 14 | ±0.7 | 1 | 3 | At the 0.05 level for p=0.64333 |
| (Sample collected from Havelikothi) | 14 | | | | | The two means are NOT |
| | 15 | | | | | significantly different |
| B | 14 | 14.3 | ±0.715 | 0.33 | 3 | |
| (Sample collected from Dhamra) | 14 | | | | | |
| | 15 | | | | | |

**Table 3. List of identified plant species at Bhitarkanika National Park indicating the host of *Trentepohlia flava* as (*)**

| Sl.No. | Scientific Name | Common Name | Habit | Family |
|---|---|---|---|---|
| 1. | *Acanthus ilicifolius* | Harakancha | Herb | Acanthaceae |
| 2. | *Acanthus volubilis* | Harakancha | Herb | Acanthaceae |
| 3. | *Acrostichum aureum* | Kharkhari | Fem | Polypodiaceae |
| 4. | *Aegialitis rotundifolia* | Banarua | Tree | Plumbaginaceae |
| 5. | *Aegiceras corniculatum* | Kharsi | Tree | Plumbaginaceae |
| 6. | *Amoora cucullata* | Ooanra | Tree | Meliaceae |
| 7. | *Avicennia alba** | Kala bani | Tree | Avicenniaceae |
| 8. | *Avicennia marina** | Singala bani | Tree | Avicenniaceae |
| 9. | *Avicennia officinalis** | Bada Bani | Tree | Avicennieceae |
| 10. | *Brownlowia tersa* | Lati sundari | Herb | Tiliaceae |
| 11. | *Bruguiera cylindrica* | Dot | Tree | Rhizophoraceae |
| 12. | *Bruguiera gymnorrhiza* | Bandari | Tree | Rhizophoraceae |
| 13. | *Bruguiera parviflora* | Kaliachua | Tree | Rhizophoraceae |
| 14. | *Bruguiera sexangula* | Bandari | Tree | Rhizophoraceae |
| 15. | *Caesalpinia crista* | Nentei | Creeeper | Caesalpiniacea |
| 16. | *Caesalpinia crista* | Gilo | Creeper | Caesalpiniacea |
| 17. | *Cerbera manghus* | Pani amba | Tree | Apocynaceae |
| 18. | *Ceriops decandra** | Garani | Tree | Rhizophoraceae |
| 19. | *Ceriops tagal** | Garani | Tree | Rhizophoraceae |
| 20. | *Clerodendron inerme* | Chiani | Creeper | Verbenaceae |
| 21. | *Crinum asiaticum* | Pani kenduli | Herb | Amaryllidaceae |
| 22. | *Crinum defixum* | Pani kenduli | Herb | Amaryllidaceae |
| 23. | *Cynometra ramiflora* | Singada | Tree | Leguminosae |
| 24. | *Cynometra iripa* | Singada | Tree | Leguminosae |
| 25. | *Cyperus compactus* | Tianshi ghasa | Grass | Cyperaceae |
| 26. | *Cyperus corymbosis* | Keutia ghasa | Grass | Cyperaceae |
| 27. | *Dalbergia spinosa* | Goera kanta | Creeper | Papilionaceae |
| 28. | *Derris hetrophylla* | Katira nai | Creeper | Leguminosae |
| 29. | *Derris scandens* | Katria nai | Creeper | Leguminosae |
| 30. | *Dolichandrone spathacea** | Gosiga | Tree | Bignoniaceae |
| 31. | *Excoecaria agallocha** | Guan | Tree | Euphorbiaceae |
| 32. | *Fimbristylis ferruginea* | Luni ghasa | Grass | Cyperaceae |
| 33. | *Finlaysonia obovata* | Lata rai | Creeper | Peripocaceae |
| 34. | *Flageliaria indica* | Bahumruga | Crepper | Flagellariaceae |
| 35. | *Heritiera fomes** | Bada Sundari | Tree | Sterculiaceae |
| 36. | *Heritiera kanikensis** | Kanika sundari | Tree | Sterculiaceae |
| 37. | *Heritiera littoralis** | Dhala sundari | Tree | Sterculiaceae |
| 38. | *Hibiscus tiliaceus* | Bania | Herb | Malvaceae |
| 39. | *Intsia bijuga* | Massitha | Tree | Leguminosae |
| 40. | *Kandelia candel* | Sindhuguan | Tree | Rhizophoraceae |
| 41. | *Lumnitzera racemosa* | Churanda | Tree | Combretaceae |
| 42. | *Lumnitzera littorea* | Churanda | Tree | Combretaceae |
| 43. | *Merope angulata* | Bana lembu | Herb | Rutaceae |
| 44. | *Myriostachya wightiana* | Nalia ghasa | Grass | Poaceae |
| 45. | *Nypa fruticans* | Nypa | Tree | Palmae |
| 46. | *Pandanus fascicularis* | Luni kia | Herb | Pandanaceae |
| 47. | *Phoenix paludosa* | Hentala | Tree | Palmae |
| 48. | *Phragmites karka* | Nala | Grass | Gramineae |
| 49. | *Porteresia coarctata* | Dhani dhana | Grass | Poaceae |
| 50. | *Rhizophora apiculata** | Rai | Tree | Rhizophoraceae |
| 51. | *Rhizophora mucronata** | Rai | Tree | Rhizophoraceae |
| 52. | *Rhizophora stylosa** | Rai | Tree | Rhizophoraceae |
| 53. | *Salacia prinoides* | Batra lata | Creeper | Hippocrateaeceae |
| 54. | *Salicornia brachiata* | - | Herb | Chenopodiaceae |
| 55. | *Salvadora persica* | Miriga | Herb | Salvodoraceae |
| 56. | *Sapium indicum* | Batula | Shrub | Euphorbiaceae |
| 57. | *Sarcolobus carinatus* | Raigidi | Shrub | Asclepiadaceae |
| 58. | *Sesuvium portulacastrum* | - | Herb | Aizoaceae |
| 59. | *Sonneratia apetala** | Keruan | Tree | Sonneratiaceae |
| 60. | *Sonneratia caseolaris** | Orua | Tree | Sonneratiaceae |
| 61. | *Sonneratia griffithii** | Orua/Chakada | Tree | Sonneratiaceae |
| 62 | *Spinifex littoreus* | Balukata | Herb | Poaceae |
| 63. | *Suaeda maritima* | Giria saga | Herb | Chenopodiaceae |
| 64. | *Suaeda monoeca* | Giria saga | Herb | Chenopodiaceae |
| 65. | *Suaeda nudiflora* | Giria saga | Herb | Chenopodiaceae |
| 66. | *Tamarix ericoides* | Jagula | Herb | Tamaricaceae |
| 67. | *Tamarix dioica* | Jagula | Herb | Tamaricaceae |
| 68. | *Tamarix troupii* | Jagula | Herb | Tamaricaceae |
| 69. | *Thespesia populnea* | Habali | Tree | Malvaceae |
| 70. | *Xylocarpus granatum** | Shishumar | Tree | Meliaceae |
| 71. | *Xylocarpus mekongensis** | Pitakorua | Tree | Meliaceae |
| 72. | *Xylocarpus moluccensis** | Pitamari | Tree | Meliaceae |

**Table 4. Measurements of erect axis, prostrate part of the algal samples collected from different places around Bhitarkanika, Orissa, India**

| Place | Length of Erect axis ( µ) | Mean(µ) | Breadth of the cells of erect axis( µ) | SE($y^{Er\pm}$) | Mean(µ) | Breadth of the cells of prostrate part (µ) | SE($y^{Er\pm}$) | Mean(µ) |
|---|---|---|---|---|---|---|---|---|
| Havelikothi | 605 | 602.33 | 9 | 0.33 | 9.33 | 13 | 0.57735 | 14 |
| | 600 | | 9 | | | 14 | | |
| | 602 | | 10 | | | 15 | | |
| Dangamal | 600 | 602.33 | 9 | 0.66 | 10.33 | 12 | 0.88 | 13.66 |
| | 606 | | 11 | | | 14 | | |
| | 601 | | 11 | | | 15 | | |
| Ekakulum | 600 | 602 | 10 | 0.5773 | 10 | 13 | 0.66 | 13.66 |
| | 602 | | 9 | | | 13 | | |
| | 604 | | 11 | | | 15 | | |

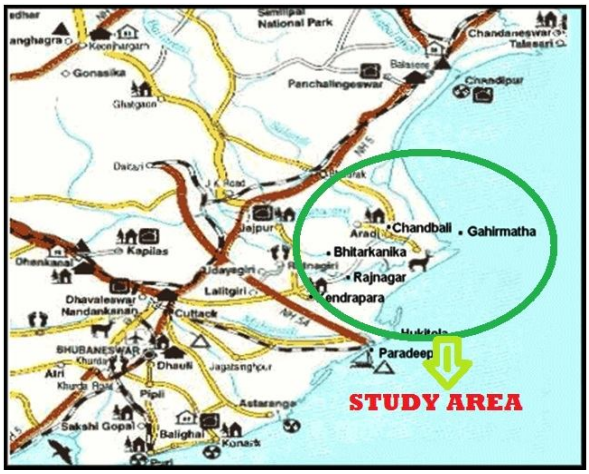


**Figure 1: Study area**


**Figure 2: Entry point of Bhitarkanika Ramsar site**

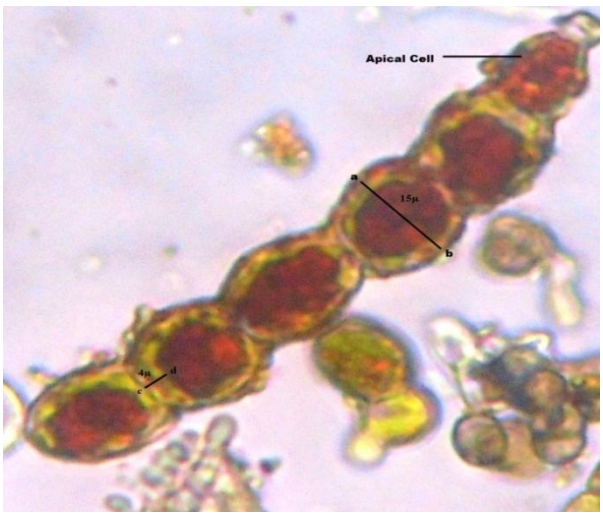


**Figure 3: Vegetative cells under microscope**

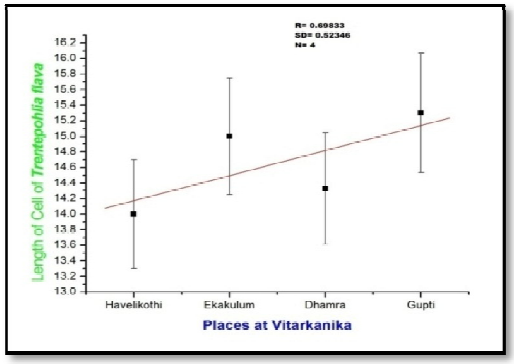


**Figure 4: Measurements of vegetative cells of prostrate part of the algal samples collected From different parts of Bhitarkanika conservatory showing statistical data analysis**

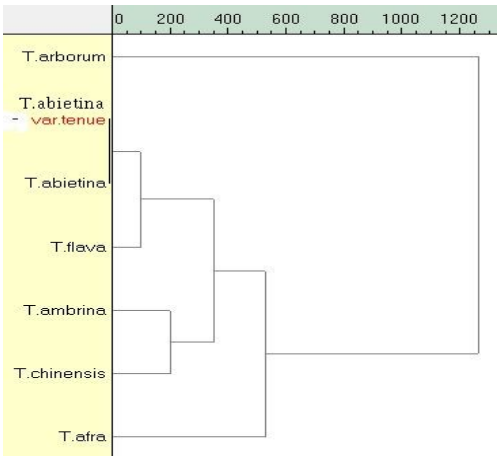


**Figure 5: Dendrogram showing Cluster analysis (Average linkage method) of 7 species of *Trentepohlia* (*T.abietina, T.arborum, T.afra, T.flava, T.chinensis, T.ambrina, T.abietina* var. *tenue*) using the morphological data based on 2 variables-the size (µ) of the prostrate cell and the length of the erect axis**

**Figure 6**

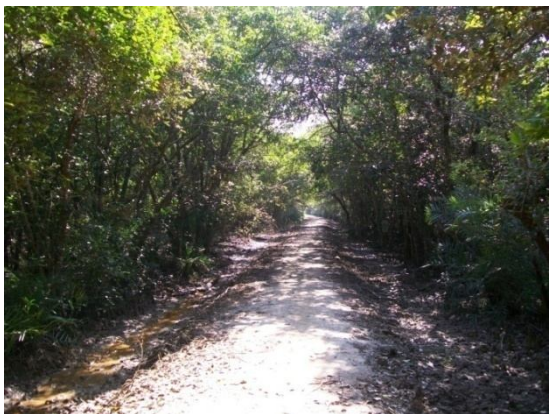

**Figure 7**


**Figure 8**

**Figure 9**

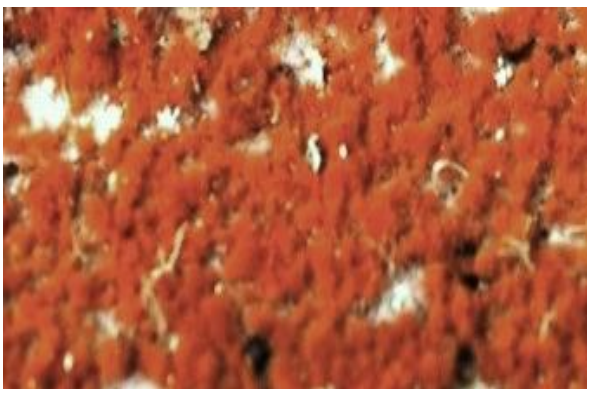

**Figure 10**

**Figure 6 to Figure 10: Different host plants and covering of the samples throughout the forest area**

**Taxonomy:** Trentepohlia flava was proposed as a new combination based on *Mycinema flava* W.J.Hooker & Arnott (1832:54) by Crib (1970), who showed that this is the correct name for the species most frequently reported as *Trentepohlia polycarpa* Nees and Montagne. The taxonomic position of this species is in doubt. After examination of the type specimens of *M.flava*, *T.polycarpa* and other relevant specimens, Cribb (1970) provided a detailed characterization of *T.flava* concluding that it is must be regarded as a species independent of *T.aurea*. Our study also reveals the superficial corrugation of the wall and the specific thickness (5µ-6µ) of the cell wall which supports the view Cribb (1970) to place *Trentepohlia flava* as a different species. The similarity matrix obtained in the present study was used to construct a Dendrogram by the Average linkage method comparing the morphological data (size of the prostrate cells and length of the erect axis) of seven species *T.abietina*, *T.arborum*, *T.chinensis*, *T.flava*, *T.ambrina*, *T.afra* and *T.abietine var.tenue*) reveals the dendrograms generated were in broad agreement with each other and also with accepted taxonomy; two major groups were obtained and most of the related species were found to be grouped together. Two cluster in which *T.arborum* belonging to a different individual cluster than the rest of the species which having greater resemblance (Figure 5). The result also denotes the close resemblance of *T.abietina*, *T.abietine var.tenue*, *T.flava* base on their vegetative cell size and characteristic features.

**Ecology**: Ecology of the species recorded in this particular mangrove area is in general agreement with reports for other regions. The species collected from all parts of the area was only on the bark of the trees like *Avicennia alba*, *Avicennia officinalis* etc. So tree bark was the only type of substratum colonised by the algal species. This genus does not show any evident association with building surfaces as found for other tropical and temperate regions. This type of association indicates the relation with biodiversity and conservation. The association of *Trentepohlia flava* with the tree bark observed strictly in our study is in generalization with other tropical areas in which these algae have been studied in detail. This association is also highlighted than for temperate regions where this alga is frequently more common on rock or other non-living substrata (Mondal *et al.*, 2008). So in the tropical areas the highest diversity and distribution of this species along with other species of the genera *Trentepohlia* can be expected to occur in forest area. This association will be even stricter for the other genera of Trentepohliaceae which are mostly foliicolous organisms. So this is general consensus that the Trentepohliaceae have their highest diversity in the tropics and the future taxonomic work on this group should focus on tropical areas (Thompson and wujek 1997; Lopez-Bautista *et al.*, 2002; Rindi *et al.*, 2004), where new undescribed species can be expected to occur. So the rapid urbanisation with huge deforestation at the tropical area particularly at the coastal belt of Indian subcontinent produces a serious threat to the biodiversity and if the deleterious effect of anthropogenic activity will be continued as such then almost a huge population of *Trentepohlia* will be destroyed before exploration.

## Conclusion

The investigations of the taxonomy, ecology and distribution of *Trentepohlia* must still be considered very valuable,

especially for geographical areas for which the information available is limited. In India detailed information available on the fresh water algal flora but information published on subaerial algae is much more limited and, in the case of *Trentepohlia*, almost nonexistent (Rindi *et al*., 2005). Our observation based on the morphological data clearly supports the view to determine the algal species *Trentepohlia flava* having a distinct identity from the closely related taxa *Trentepohlia abietina* and the variety *Trentepohlia abietina* var. *Tenue* (Zeller) Cribb. Cribb (1970) described two varieties of *Trentepohlia abietina* characterised by a spiral wall ornamentation, *Trentepohlia abietina* var. *Tenue* and *Trentepohlia abietina* var. *Corrugate*. It is reasonable to characterise and determine the two samples *Trentepohlia abietina* var. *Tenue* and *Trentepohlia flava* as two different species (Rindi *et al*., 2005) on the basis of differences in colour (dark red in T.flava and bright orange in T.abietina var.tenue), substratum and cell size and the cluster analysis of 7 species of *Trentepohlia* (*T.abietina*, *T.arborum*, *T.afra*, *T.flava*, *T.chinensis*, *T.ambrina*, *T.abietina* var. *tenue*) using the morphological data based on 2 variables-the size (μ) of the prostrate cell and the length of the erect axis also suggests the close resemblance of *Trentepohlia abietina* var. *Tenue* and *Trentepohlia flava* though only molecular data will provide a clear picture about the taxonomic relationship.

In this paper a framework about the taxonomic relationship with some members of the group has been discussed but detailed taxonomic assessment at species and genus level, based on a combination of morphological, molecular and physiological data and a careful examination of many herbarium specimens, is essential to clarify the phylogeny and phylogeographic pattern and taxonomic relationship in this order. The ecological study of the species indicated the dominant presence of this genus on the substratum like tree bark in the tropical area. The rapid and vigorous growth within the mangrove forest as recorded in Bhitarkanika Conservatory also evoked the possibility of highest diversity and distribution of this species along with other species of the genera *Trentepohlia* to occur in forest area. So new, unexplored, undescribed species can be expected to be found at the forest area of tropics. It is therefore concluded that destruction of tropical forest and other forms of habitat loss in the tropics alarming a major threat to the diversity of Trentepohliaceae and as example it should be mentioned that rapid destruction of mangrove forests in Sundarban biosphere reserve, West Bengal, India may lead to the loss of diversity of this group continuously.

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