

## ***ACHNANTHIDIUM CHITRAKOOTENSE* SPEC. NOV. FROM RIVERS OF NORTHERN AND CENTRAL INDIA\***

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**Abstract.** The group of diatoms morphologically similar to *Achnantheidium pyrenaicum* (Hustedt) Kobayasi includes a number of ecologically important taxa. The restricted distribution patterns of some *Achnantheidium* Kützing species have become more obvious after detailed studies of this relatively small group. It has been characterized as having terminal raphe fissures curved to the same side, slit-like external areola openings, and internal hymenate areola occlusions. Unlike the rest of *Achnantheidium*, the new species *A. chittrakootense* possesses areola occlusions of an unusual type. It is also characterized by a raphe with terminals externally curved towards the same side of the valve, crossing the hyaline rim between the valve face and valve mantle, and internally undulate raphe terminals. The new member of the *Achnantheidium pyrenaicum* group is described from rivers of northern and central India. The species is documented with LM and SEM micrographs, and the frustule morphology of this species and similar taxa is compared and briefly discussed.

**Key words:** *Achnantheidium*, taxonomy, morphology, biogeography, India

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### INTRODUCTION

Representatives of the genus *Achnantheidium* Kützing are common and abundant, living attached to various kinds of substrate in diverse freshwater habitats (e.g., Nautiyal *et al.* 2004; Potapova & Ponader 2004; Potapova & Hamilton 2007). Several of them have clearly defined environmental ranges and therefore are used as bio-indicators. Diatoms of the genus *Achnantheidium* belong to two groups around their flagship species: *A. minutissimum* (Kütz.) Czarnecki with slightly deflected raphe terminals and round external areola openings, and *Achnantheidium pyrenaicum* (Hustedt) Kobayasi with terminal raphe fissures curved to the same side of the valve and slit-like external

areola openings. Besides the cosmopolitan species, several representatives of these groups have a more limited geographical distribution (e.g., Kobayasi 1987; Moser *et al.* 1998; Potapova & Ponader 2004; Potapova 2006).

The taxa around *A. pyrenaicum* are known from the Northern Hemisphere (e.g., Potapova & Ponader 2004) but the distribution of some of them is related to Asia and Oceania (Hustedt 1938; Kobayasi *et al.* 1986; Kobayasi 1987). The Indian region undoubtedly possesses a rich and specific diatom flora with many yet-undescribed taxa (Jütner *et al.* 2000, 2004) including unknown *Achnantheidium* taxa, which are common in rivers in the West Himalayas and Central India (e.g., Cantonati *et al.* 2001; Nautiyal *et al.* 2004, in press; Verma *et al.* in press).

\* Dedicated to Dr. Kurt Krammer on the occasion of his 85<sup>th</sup> birthday

The present study examines the distribution of *Achnantheidium* species in different ecoregions, the West Himalayas and Central Highlands, separated by the Indo-Gangetic Plains. This is a part of a larger study conducted to compare the diatom biodiversity in these two highland ecoregions.

#### STUDY AREA

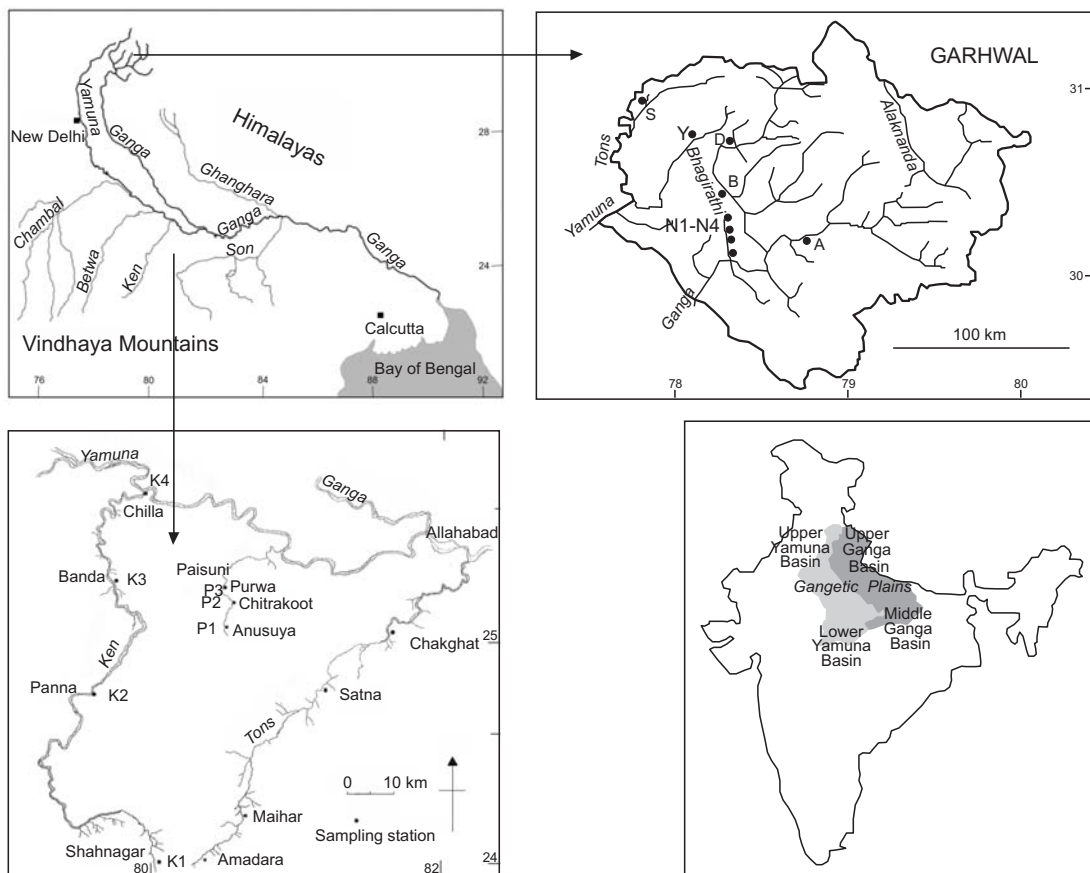
Diatoms were collected from rivers in central India (Central Highlands Ecoregion) and northern India (West Himalaya Ecoregion) (Fig. 1, Table 1). The central India rivers (the Ken, the Paisuni, the Tons) are situated on the Bundelkhand Plateau (23°30' to 26°N, 78°30' to 82°30'E, elevation 86–365 m a.s.l.), but arise in the Kaimur hills, Vindhyan ranges, north of the Narmada River. The Ken and the Paisuni flow north to northeast and join the lower stretch of the Yamuna River 200 and

100 km respectively, before the confluence with the Ganga at Allahabad, while the Tons River joins the middle stretch of the Ganga River (~100 km downstream of Allahabad, in the Gangetic Plains). The north Indian rivers (the Yamuna, the Supin in the Himalayan stretch of the Yamuna River, the Bhagirathi, the Alaknanda in the Himalayan stretch of the Ganga River; all glacier/snow-fed), the Dharasu Gad and the Nagni Gad are situated in the Garhwal region, Uttarakhand State, India (29°31' to 31°20'N, 77°33' to 80°6'E, elevation 550–2200 m a.s.l. (Table 1).

Sandstones are the most common rock throughout the Vindhyan division with the exception of the lower Bhandar stage, which is for the most part calcareous (Wadia 1983). Granulite with marble bands, basic metavolcanic rocks, dolomite slate and sheeny phyllite are the main types of rocks and sediments in the catchment of

**Table 1.** Geographical coordinates of the sampling locations in the rivers Ken, Paisuni and Tons from the Central Highlands, and Supin, Yamuna, Bhagirathi, Alaknanda, Dharasu and Nagni Gad from the West Himalayas Ecoregion.

| River system                   | River/Station with Acronym | Latitude (N) | Longitude (E) | Altitude (m.a.s.l.) |
|--------------------------------|----------------------------|--------------|---------------|---------------------|
| Yamuna<br>(Lower stretch)      | Ken River                  |              |               |                     |
|                                | Shahnagar K1               | 23°59'28.92" | 80°18'1.77"   | 365                 |
|                                | Panna K2                   | 24°44'17.38" | 80°0'41.16"   | 200                 |
|                                | Banda K3                   | 25°28'38.25" | 80°18'51.62"  | 95                  |
|                                | Chilla K4                  | 25°46'15.49" | 80°31'36.99"  | 86                  |
|                                | Paisuni River              |              |               |                     |
|                                | Anusuya P1                 | 25°04'25"    | 80°52'05"     | 180                 |
|                                | Chitrakoot P2              | 25°10'25"    | 80°52'12"     | 135                 |
|                                | Purwa P3                   | 25°13'01"    | 80°54'09"     | 131                 |
| Ganga<br>(Middle stretch)      | Tons River                 |              |               |                     |
|                                | Amdara T1                  | 24°6'30.83"  | 80°36'11.34"  | 360                 |
|                                | Maihar T2                  | 24°16'14.13" | 80°48'18.11"  | 326                 |
|                                | Satna T3                   | 24°33'42.88" | 80°54'26.34"  | 290                 |
|                                | Chakghat T4                | 25°2'1.06"   | 81°43'51.75"  | 94                  |
| Yamuna<br>(Upper stretch)      | Supin (Tons) S             | 31°45'2"     | 78°10'29"     | 1500                |
|                                | Yamuna Y                   | 30°55'35"    | 78°23'8"      | 1800                |
| Bhagirathi<br>(Middle stretch) | Dharasu D                  | 30°37'17"    | 78°19'17"     | 800                 |
|                                | Bhagirathi B               | 30°24'48"    | 78°27'25"     | 620                 |
| Ganga<br>(foothill)            | Nagni N                    |              |               |                     |
|                                | Khiret N1                  | 30°23'25"    | 78°19'30"     | 2200                |
|                                | Nagni N2                   | 30°19'15"    | 78°21'10"     | 1400                |
|                                | Jajal N3                   | 30°18'15"    | 78°20'40"     | 1200                |
|                                | Shivpuri N4                | 30°8'15"     | 78°23'30"     | 375                 |
| Alaknanda (Middle stretch)     | Alaknanda A                | 30°13'10"    | 78°48'32"     | 550                 |



**Fig. 1.** Location of study area in India, showing the extent of the Yamuna and Ganga basins. Their upper basins are in the West Himalayas. The middle basins of both rivers drain the Gangetic Plains, and the lower basin of the Yamuna drains the northern slopes of the Vindhyan ranges (Upper). The middle reaches of the Ganga also receive flows from the rivers emerging from the Vindhyan hills (Lower). The map shows major drainages in the mountain stretch of the Ganga in the Himalayas and the sampling localities for the study in the West Himalayas. Map shows the rivers and sampling localities in the study area in the Central Highlands Ecoregion.

the Alaknanda (Valdiya 1962). The Vindhya has a monsoonal climate and is characterized by long dry periods with high air temperatures (43°C). Population density is low in the area studied and land use is predominately agriculture.

**MATERIALS AND METHODS**

The material was collected from November 2004 to January 2005 at 11 locations on three Central Highland rivers (4 at the Ken, 3 at the Paisuni, 4 at the Tons) and at 9 locations on 3 rivers and 2 streams in the Himalayas (1 each at the Supin, Yamuna, Dharasu Gad, Bhagirathi,

Alaknanda; 4 in the Nagni Gad; Fig. 1, Table 1). The samples were scraped from hard substrate such as rocks, boulders and cobbles (Fig. 1, Table 1). A replicate was obtained at each station. Owing to the great distances in the Central Highlands and difficult access along the length of the river, each station was sampled only once in this period. The physicochemical characteristics of the water are presented as ranges (minimum and maximum values) in Table 2.

The samples for diatom analysis were treated with 10% HCl, washed several times with distilled water and then boiled in concentrated acids (H<sub>2</sub>SO<sub>4</sub>, HNO<sub>3</sub>) to remove organic matter. After washing several times with distilled water the cleaned diatom material was air-dried

**Table 2.** Some physicochemical characteristics of water: range (minimum and maximum values) during winters is based on the diurnal and longitudinal (downstream of the source) profile of these parameters for the Central Highland Ecoregion rivers and the Nagni Gad (West Himalayas), and only diurnal variations for other rivers from the West Himalayas.

| Stream/River | Air temperature (°C) | Water temperature (°C) | Water current (cm · s <sup>-1</sup> ) | pH      | Conductivity (μS · cm <sup>-1</sup> ) | Dissolved oxygen (mg · l <sup>-1</sup> ) |
|--------------|----------------------|------------------------|---------------------------------------|---------|---------------------------------------|--|
| Ken          | 11–32                | 15–31                  | 0–42                                  | 7.0–7.5 | 165–420                               | 8.6–10.7                                 |
| Paisuni      | 10.0–21.5            | 16.0–22.5              | 2.8–30.9                              | 7.0–7.7 | 170–440                               | 8.0–11.5                                 |
| Tons         | 17–33                | 18–33                  | 1.5–35.0                              | 7.0–7.8 | 160–420                               | 8.2–10.5                                 |
| Supin        | 11                   | 9                      | 90                                    | 7.0     | 62                                    | 10.0                                     |
| Yamuna       | 12                   | 8                      | 100                                   | 7.0     | 59                                    | 10.8                                     |
| Dharasu Gad  | 13                   | 12                     | 30                                    | 7.2     | 60                                    | 10.7                                     |
| Bhagirathi   | 15                   | 11                     | 110                                   | 7.3     | 74                                    | 10.7                                     |
| Nagni Gad    | 9–15                 | 8–13                   | 2–45                                  | 7.1–7.8 | 80–250                                | 7.5–12.5                                 |
| Alaknanda    | 11                   | 10                     | 90                                    | 7.4     | 150                                   | 110                                      |

on coverslips and mounted in Naphrax®. The diatoms were observed with a Nikon Eclipse 600 light microscope with Nomarski phase contrast. The micrographs were taken with a Nikon DS-Fi 1 camera. For SEM analysis the material was pipetted onto cover glasses, air-dried and affixed to an aluminum stub with double-sided transparent tape. The stubs were sputter-coated with gold and viewed with a Hitachi S-4700 scanning electron microscope. SEM micrographs were taken in the Laboratory of Field Emission Scanning Electron Microscopy and Microanalysis at the Institute of Geological Sciences of the Jagiellonian University.

## RESULTS AND DISCUSSION

*Achnantheidium chitrakootense* Wojtal, Lange-Bertalot & Nautiyal P., *sp. nov.* Figs 2–32

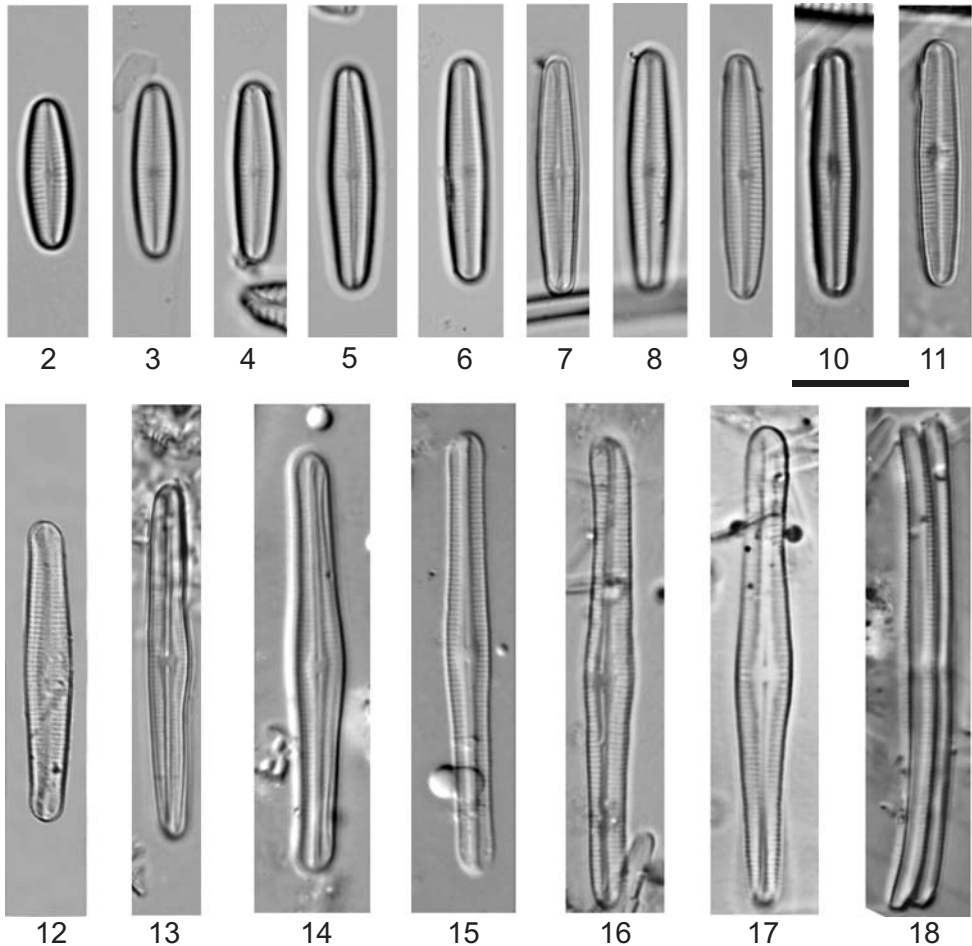
*Aspectus microscopico photonico. Frustula aspectu cinguli leniter arcuata apicibus non retroflexis, externe cum raphovalva biconcava areovalvae paulo biconvexa. Valvae lineares plus minusve modice inflatae in medio quoad individua media et maiora sed lineari-ellipticae marginibus modice convexae quoad individua minora. Apices simpliciter obtuse rotundati vel aliquid subcapitati quoad individua maiora. Longitudo 13–43 μm, latitudo 3.4–4.2 μm in mediis partibus, cellulae primae usque ad 5 μm. Raphovalva: area axialis linearis angustissima. Area centralis non vel vix separata. Raphe filiformis extremis centralibus rectis apparentibus fere dense positae inter se. Striae transapicales parallelae*

*26–30 (raro 32) in 10 μm. Areovalva: Areae et striae non significanter differunt. Areolae non spectabiles. Aspectus ultramicroscopicus externus. Extrema centralia raphis cum poris dilatatis guttiformibus parum deflexis ad latera opposita fissuris terminalibus valde curvatis unilateraliter in limbum valvae ubi area terminalis etiam unilateraliter extendens. Striae transapicales cum foraminibus areolarum distincte elongatis circiter 40 in 10 μm, interruptae linea hyaline apicali ad iuncturam inter faciem et limbum valvae. Aspectus internus. Extrema centralia interna raphis non coaxialia nec dilatata. Extrema distalia sub polos distincte undulata tum in helictoglossis conspicuis terminantes. Areolae singulae oclusae in forma speciosa generaliter incognita in genere Achnantheidium.*

TYPE: INDIA, Chitrakoot, Paisuni River 25°10'25"N, 80°52'12"E, 135 m a.s.l., November 2004, *leg. P. Nautiyal* [HOLOTYPE: slide no AS-SAZ-35 in Department of Phycology, W. Szafer Institute of Botany, Polish Academy of Sciences, Kraków, Poland (Figs 10 & 11); ISOTYPES: Frankfurt am Main, Germany, access no. AS-101 (FR), Academy of Natural Sciences, Philadelphia, U.S.A., access no. ANSP GC36339; Institute of Marine Sciences, University of Szczecin, access no. 15659].

ETYMOLOGY: the species name corresponds to the type locality, Chitrakoot, Paisuni River.

LM. Frustules with slightly bent apices in girdle view. Raphid valves externally concave, and rapheless valves are slightly convex. Large valves linear with moderate inflation in middle portion of valve,



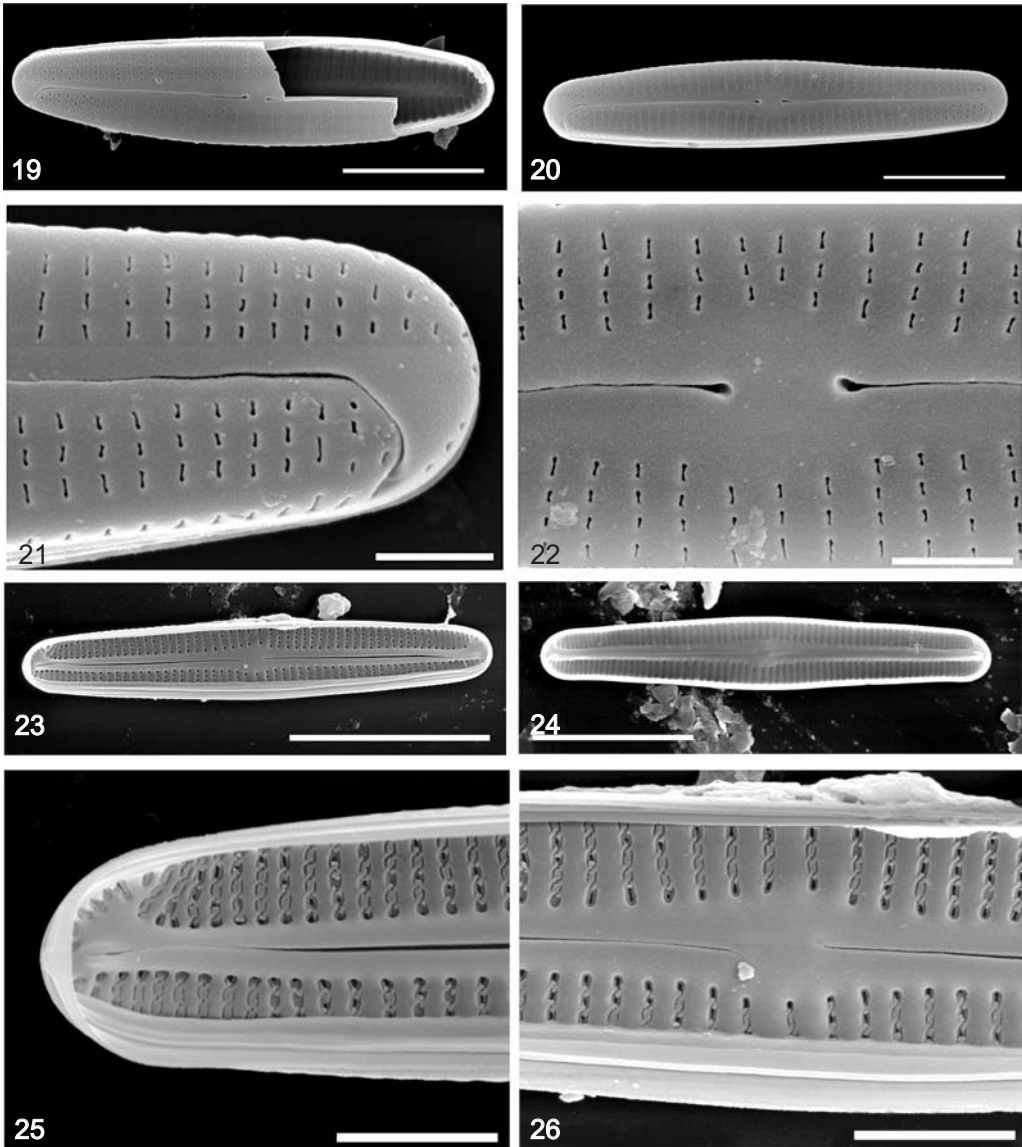
**Figs 2–18.** *Achnanthyidium chitrakootense* Wojtal, Lange-Bertalot & Nautiyal P., *sp. nov.* 7, 8, 10 & 11 – micrographs of the same frustules at different focus; 10 & 11 – raphid and rapheless valves of holotype specimen; 18 – two frustules in girdle view. All LM. Scale bar = 10  $\mu$ m.

and somewhat subcapitate apices. Smaller valves of linear-elliptical outline with broadly rounded (blunt) apices. Valve length from 13 to 42  $\mu$ m, and valve width from 3.4 to 4.2  $\mu$ m in the middle portion of the valve. Width of post-initial cells up to 5  $\mu$ m. Raphid valves with very narrow axial area, without distinct central area. Raphe filiform, straight from external view with relatively closely located proximal raphe pores. Transapical striae parallel, 26 to 30 (rarely 30) in 10  $\mu$ m. Areolae indistinct.

SEM. External view. Proximal raphe ends drop-shaped, slightly deflected towards opposite sides

(Fig. 22). Distal raphe terminals strongly curved to the same side, running into the mantle (Fig. 21). Transapical striae consist of areolae which have transversely elongated external foramina, ca 40 in 10  $\mu$ m (Figs 21, 22 & 31), interrupted by hyaline area located at junction of mantle and valve face (Fig. 21).

Internal view. Proximal raphe terminals undifferentiated, slightly deflected to opposite sides (Fig. 26). Raphe branches internally distinctively undulate near ends into helictoglossae (Fig. 25). Internal areolae foramina are of volate-type occlusion (Figs 25–27 & 29).

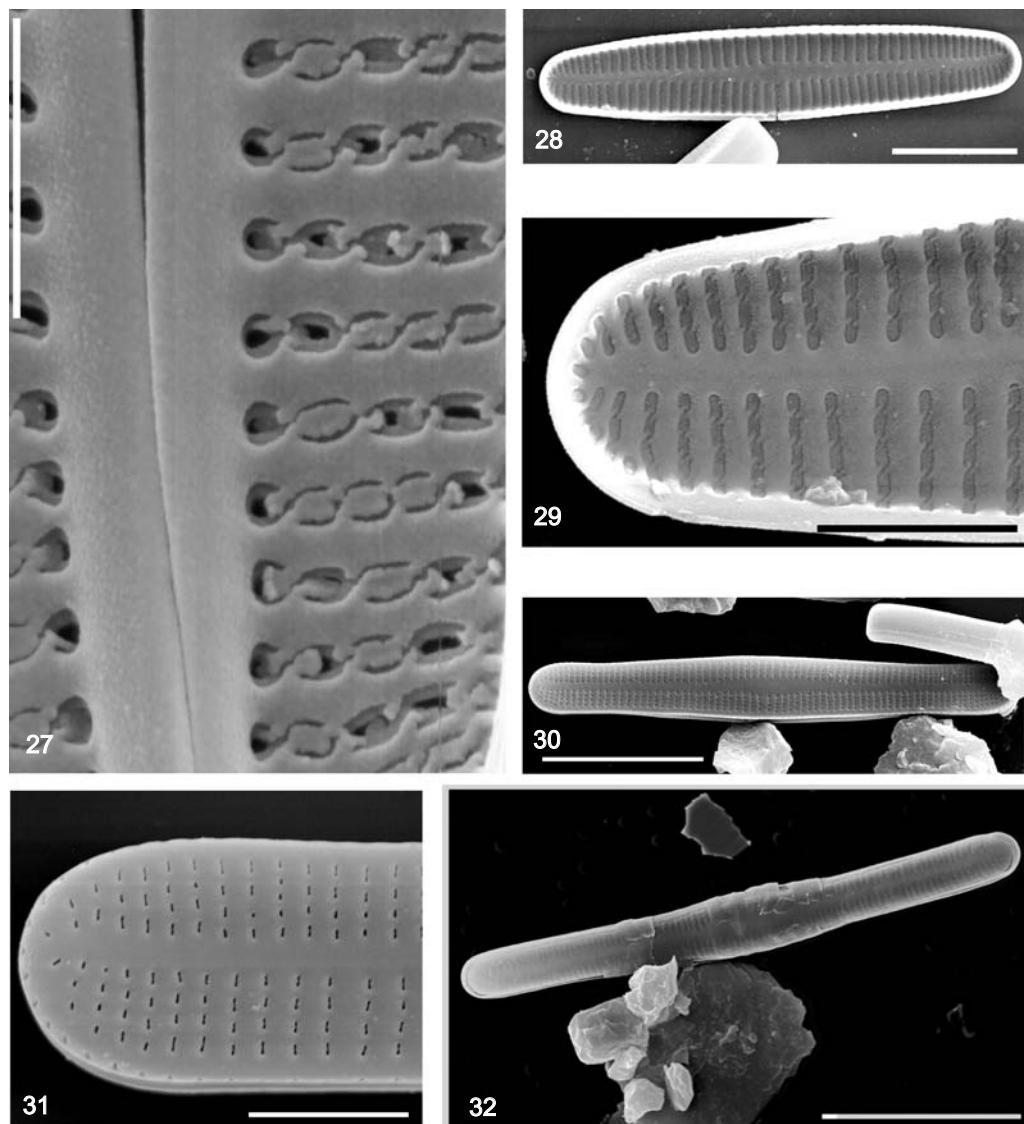


**Figs 19–26.** *Achnanthisdium chitrakootense* Wojtal, Lange-Bertalot & Nautiyal P., *sp. nov.* Raphid valves, external (19–22) and internal (23–26) view. 21 – raphe terminals strongly curved to the same side, running into the mantle; 22 – drop-formed raphe ends, slightly deflected towards opposite sides; 25 – distal raphe ends in helictoglossa; 26 – undifferentiated proximal raphe ends, slightly deflected to opposite sides. All SEM. Scale bars: 19 & 20 = 5  $\mu\text{m}$ ; 21 & 22 = 1  $\mu\text{m}$ ; 23 & 24 = 10  $\mu\text{m}$ ; 25 & 26 = 2  $\mu\text{m}$ .

**AUTECOLOGY.** Circumneutral to slightly alkaline waters of moderate conductivity (Table 2).

**DISTRIBUTION.** *Achnanthisdium chitrakootense* was common along the upper course of the Paisuni (P1 to P3) and Tons (T1 to T3) rivers, especially

at Anusuya and Chitrakoot in the former, and at Amdara and Maihar in the latter. It was not found in the Ken (Table 1). It is probably more widespread, but not separated from similar, commonly reported diatoms.



**Figs 27–32.** *Achnanthydium chittrakootense* Wojtal, Lange-Bertalot & Nautiyal P., *sp. nov.* 27 – raphid valve, internal view. 28–31 – raphidless valves, internal (28 & 29) and external (30 & 31) view; 32 – initial cell. 27 – internal areola openings with volate occlusions; 31 – transversely elongated external foramina; 32 – initial cell with transverse perizonial bands. All SEM. Scale bars: 27 = 1  $\mu\text{m}$ ; 28 = 5  $\mu\text{m}$ ; 29 & 31 = 2  $\mu\text{m}$ ; 32 = 20  $\mu\text{m}$ .

Similar species are *Achnanthydium convergens*, *A. crassum* and *Achnanthes trigibba*, but the most morphologically similar taxon is *A. deflexum* (Table 3). *Achnanthydium chittrakootense* and *A. deflexum* overlap considerably in frustule size and striae pattern, but differ in valve proportion, outline, raphe and areola morphology. Valves

of *A. deflexum* are more elliptic in outline and the internal areola openings are with hymenate occlusions. Additionally, its distal raphe ends are externally deflected in the subterminal position, whereas internally the proximal raphe ends are distinctly deflected in opposite directions (Potapova & Ponader 2004, Figs 97–103). All *Achnanthydium*

Table 3. Comparison of morphologically similar *Achnanthydium* species.

|                                       | <i>Achnanthydium chitrakootense</i><br>spec. nov.  | <i>Achnanthydium deflexum</i> (Reimer)<br>Potapova & Ponader   | <i>Achnanthydium convergens</i> Kobayasi   | <i>Achnanthydium</i><br><i>crassum</i> (Hustedt) Potapova<br>& Ponader   |
|---------------------------------------|--|--|--|--|
| Valve length                          | 12–28 $\mu\text{m}$ (initial cells up to 43 $\mu\text{m}$ )  | 7–25 $\mu\text{m}$<br>7.4–28.3 $\mu\text{m}$<br>(initial cells 24.2–26.2 $\mu\text{m}$ )   | 10–25 $\mu\text{m}$ (Ponader & Potapova 2004)  | 7.3–19.6 $\mu\text{m}$ (Ponader & Potapova 2004)   |
| Valve width                           | 3.4–4.2 $\mu\text{m}$ (initial cells up to 5 $\mu\text{m}$ )   | 3.8–4.5 $\mu\text{m}$<br>3.1–5.3 $\mu\text{m}$<br>(initial cells 3.8–4.2 $\mu\text{m}$ )   | 4.0–4.5 $\mu\text{m}$ (Ponader & Potapova 2004)  | 3.0–4.5 $\mu\text{m}$ (Ponader & Potapova 2004)  |
| Outline                               | Linear to linear-elliptical  | Linear to elliptical with subcapitately protracted to obtusely rounded, not protracted apices  | Linear-lanceolate with drawn-out apices to elliptical (Ponader & Potapova 2004)  | Linear-elliptical to elliptical with rounded, sometimes slightly drawn-out apices  |
| Raphe morphology                      | Externally – arched terminals towards the same side of the valve, crossing the hyaline rim between valve face and valve mantle. Internally raphe ends swollen. Central raphe ends only slightly deflected towards opposite sides | Proximal raphe ends close, distal ends curved or deflected in the same direction, subterminal (Patrick & Reimer 1966). Externally – filiform with curved terminals towards the same side of the valve, at the valve face (not crossing the hyaline rim). Internally raphe ends only deflected towards opposite sides | Externally – terminal raphe ends hooked towards the same side of the valve, at the valve face (not crossing the hyaline rim). Internally raphe ends straight. Central raphe ends only deflected towards opposite sides   | Externally – distal ends deflected in the same direction with curved terminals towards the same side of the valve, at the valve face (not crossing the hyaline rim). Central raphe ends not deflected (Ponader & Potapova 2004, figs 46, 48) |
| Striae density (in 10 $\mu\text{m}$ ) | 26–30(32)  | 20–22 at the center, 28–30 near the poles (Patrick & Reimer 1966) 15–26 at center 30–35 near the poles (Ponader & Potapova 2004)   | In raphid valves about 18 at the center, about 36(40) near the poles. In rapheless valves striae equidistant at most part of the valve (Kobayasi <i>et al.</i> 1986). 20–22 at the center, 20–24 near the poles (in SEM 40–45 near apices) (Ponader & Potapova 2004) | 17.5–24 at the center, 19–25 near the poles (in SEM 35–42 near apices) (Ponader & Potapova 2004)   |
| Striae orientation                    | Parallel   | Parallel or very slightly radiate  | Strongly convergent near poles of raphid valves and slightly radiate near poles of rapheless valves (Kobayasi <i>et al.</i> 1986)  | Radiate  |
| Areola morphology                     | Externally transversely oriented slit-form areolae openings and internally each poroid contains a single volute occlusion  | Externally transversely oriented slit-form areolae openings and internally each poroid contains a hyeminate occlusion  | Externally linear to elliptic areolar openings, internally each poroid contains a hyeminate occlusion  | Externally round or slightly transversely elongated, internally each poroid contains a hyeminate occlusion   |



taxa known until now possess a hymenate pore occlusion. The hymen occupies the inner areola openings, while the outer openings are slits or circular pores without any occlusion. The inner areola openings of *Achnantheidium chittrakootense* possess siliceous occlusions (vols?) located near the external apertures of the foramina. *Achnantheidium chittrakootense* probably has been observed several times in Indian materials, but lumped with similar species regarded as widely distributed, such as *Achnanthes minutissima* or *A. microcephala*. It seems likely that *Achnanthes trigibba* Hustedt, reported in 1970 by Gandhi from Jog-Falls in Mysore State (p. 759, Pl. 1: figs 11 & 12) belongs to *Achnantheidium 'chittrakootense'*. Despite Gandhi's opinion that the observed specimens correspond very well morphologically with *Achnanthes trigibba* Hustedt, his drawings (Pl. 1: figs 11 & 12) as well as the description differ from the type material of *A. trigibba* (Simonsen 1987).

Other morphologically allied specimens were observed by Sarode & Kamat (1980) in Nagpur and identified as *Achnanthes minutissima* (Pl. 1, figs 14 & 15), as well as in Maharashtra (Sarode & Kamat 1984, Pl. 5, fig. 121a, b) and identified as *Achnanthes microcephala* (Pl. 5, fig. 120a, b) and *A. microcephala* var. *gracillima* (Pl. 5, fig. 121a, b). They may have also been observed in the River Moosi, Hyderabad by Venkateswarlu (1983), from where similar specimens have been depicted (Pl. 1, fig. 12a, b) as belonging to *Achnanthes microcephala*. Other morphologically allied specimens were observed by Sarode and Kamat (1980) in Nagpur and identified as *Achnanthes minutissima* (Pl. 1, figs 14 & 15), as well as in Maharashtra (Sarode & Kamat 1984, Pl. 5, fig. 121a, b) and identified as *Achnanthes microcephala* (Plate 5, fig. 120a, b) and *A. microcephala* var. *gracillima* (Pl. 5, fig. 121a, b). It may have also been observed in the River Moosi, Hyderabad by Venkateswarlu (1983), from where similar specimens were depicted (Pl. 1, fig. 12a, b) as belonging to *Achnanthes microcephala*.

The distribution of *Achnantheidium chittrakootense* may be broader, as similar valves (here regarded as initial cells) were reported by Foged (1959) from Afghanistan as *Achnanthes* sp.

The use of a narrower species concept based on more detailed frustule morphology revealed the existence of several taxa which until recently remained hidden in broader species concepts. Amongst them are several *Achnantheidium* representatives belonging to the group around *A. pyrenaicum* (e.g., Kobayasi *et al.* 1986; Kobayasi 1987; Potapova & Ponader 2004; Potapova 2006). In the material studied, such widespread diatoms as *Achnantheidium pyrenaicum* Hustedt followed by *A. minutissimum* were abundant and common in the rivers studied. Other species of limited distribution, such as *A. cf. neomicrocephalum* or those known mainly from Asia [*A. japonicum* (Kobayasi) Kobayasi, *A. latecephalum* Kobayasi, *A. crassum* (Hustedt) Potapova *et* Ponader] were quite common in the area, though less abundant. The new species *Achnantheidium chittrakootense* was identified in several rivers studied, but it was most common in the Tons and Paisuni. In these rivers the diatom attained over 5% relative abundance, and initial cells (Figs 12–18) were observed.

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