# ON GENUS *CHLOROTYLIUM* KÜTZING—A NEW ADDITION TO THE INDIAN FLORA

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## ABSTRACT

The rare alga *Chlorotylium* Kützing has been recorded for the first time from India, found growing on banana logs floating in the stagnant back-waters in the river Gomati, in Lucknow. It grows in untreated sewage and polluted water. The alga forms bright green irregularly pulvinate crusts on the supporting substratum and is made up of branched filaments differentiated into longer and shorter cells which emerge from a basal cluster of cells and prostrate filaments. The thallus is encrusted with lime. Chemical analyses of water in which the alga was growing have been given and tolerance of the alga to heavy organic pollution has been recorded. Growth and development of the thallus has been traced from the observations on young stages of growth of the thallus. Frequent formation of akinetes and palmella stages from the smaller cells has been observed and differentiation into longer and shorter cells has been visualized as a means of adaptation to an epiphytic habit—the longer cells serving to space apart the smaller cells on the surface while the shorter cells alone are reproductive.

## INTRODUCTION

The genus *Chlorotylium* was established by KÜTZING from the European continent and has, since, been also recorded from the United States of America (COLLINS, 1909; SMITH, 1933). Three species of the genus were recognized by KÜTZING while certain details of morphology and life-history have been given by HANSGIRG (1886), REINKE (1879) and PRINTZ (1927). It is a rare fresh-water alga which forms hemispherical or irregularly pulvinate crusts on rocks or twigs and stems of aquatic plants. The thallus is generally incrusted with lime. A sexual reproduction occurs by the formation of quadriflagellate and biflagellate swarmers but sexual reproduction has not been observed. There is no record of the occurrance of this genus from the Indian sub-continent. It is, therefore, proposed to record it in our flora and to describe the Indian plant.

## MATERIAL AND METHODS

The alga was observed growing on discarded banana logs floating in stagnant waters in a polluted zone of river Gomati, in Lucknow (U. P.), in March 1975. The thallus of the alga formed bright green, irregularly pulvinate crusts on the surface of the logs and was incrusted with carbonate of lime. It was collected and put in cultures but attempts at growing it in the laboratory did not succeed. The material was preserved in 4 per cent formalin solution and decalcified for morphological observations. Decalcification was done by treating the material with 0.01 N HCl for five or six hours followed by repeated rinsing and washing in warm distilled water which preserved the alga without appreciable damage to the thallus organization. In order to assess the degree of pollution, chemical analysis of the water in which the alga was growing was also done. The values obtained have been given in Table 1.

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Table 1*

| Constituent factors | Conc. in mg/litre. |
|---|---|
| 1. pH | 8.4 |
| 2. Carbonates | 13.9 |
| 3. Bicarbonates | 424.6 |
| 4. Chlorides | 12.2 |
| 5. Total hardness | 314.0 |
| 6. Free and saline ammonia | 12.4 |
| 7. Albuminoid ammonia | 12.3 |
| 8. Nitrate-nitrogen | 2.7 |
| 9. Calcium | 60.1 |
| 10. Magnesium | 39.9 |

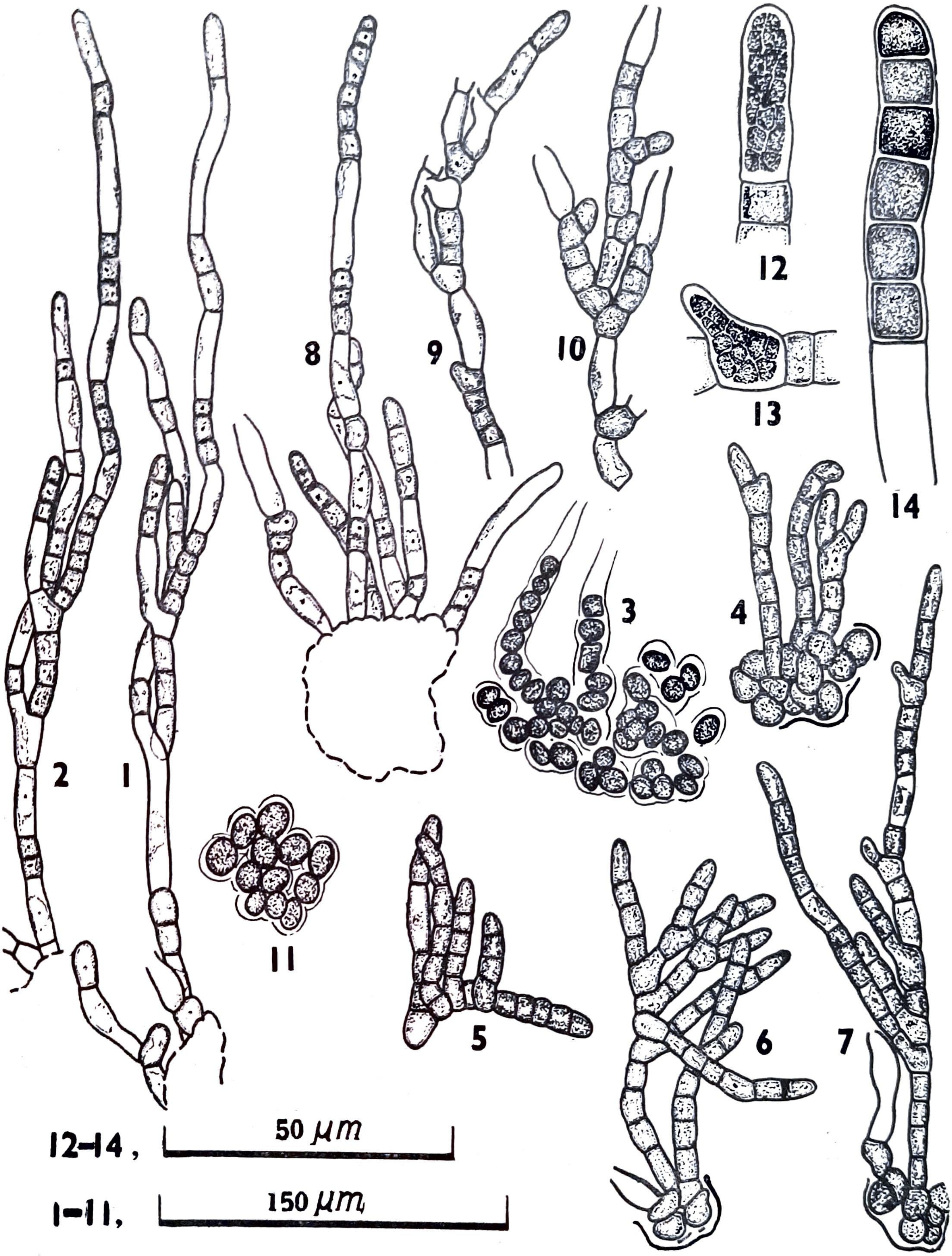
*The methods followed for the chemical analyses were those in "*Standard methods for the examination of water and waste water*" (1965) published jointly by American Public Health Association, American Water Works Association and Water Pollution Control Federation.

## OBSERVATIONS

The vegetative thallus of the alga is made up of uniseriate branched filaments. Cells in the filaments are of two kinds—longer and shorter. Generally, one or two longer cells alternate with a series of 1-8 shorter cells (Text-figs. 1, 2, 8). However, a definite pattern in the arrangement of longer and shorter cells may not be evident in relatively younger filaments (Pl. 1, Fig. 1; Text-figs. 6, 7). The differentiation into long and short cells does not take place at the same level in all the closely apposed threads, as a result of which the thallus lacks the distinct zonation as is characteristic of some species, e.g. *C. cataractarum*.

The longer cells are poor in contents and are very pale green in colour. There is a small pale green parietal, laminate chloroplast which occupies, in most cases, only the middle region; the remaining parts of the cell appear colourless. These cells measure 8-12 μm in diameter and are 2-6 times longer than broad. A single pyrenoid is present in each plastid, but it is often difficult to observe. The smaller cells, on the other hand, have the same diameter or width but are only 1 to 1½ times longer than broad and possess dense contents. These, likewise, contain a parietal, laminate, girdle-shaped chloroplast with a single pyrenoid, which fills the entire cell and has a deep green colour. The filaments emerge from rounded cells at the base of the thallus (Text-figs. 4, 5-7), a region which is difficult to observe because of the heavy calcification. Decalcification reveals clusters of rounded cells and filaments, irregularly disposed and packed together in a comparatively compact mass (Pl. 1, Fig. 7; Text-fig. 3). These cells possess contents which may be very pale green, slightly brownish or reddish in colour.

The pattern of growth and development of the thallus were studied from the young stages of the plant. The clusters of cells found at the base of the thallus form small filaments composed exclusively of short cells by successive divisions in one plane only (Text-fig. 4). These cells divide actively and the filaments grow in size and length. Certain cells of the young filament now stop dividing but continue to elongate and develop into the longer cells while other cells keep dividing and appear as smaller cells. This ultimately causes




Text-Figs. 1-14. *Chlorotylium mammiforme* Kützing—1-2. Vegetative filaments showing differentiation into longer and shorter cells; 3. Clusters of cells at the base of the thallus as revealed after decalcification; 4-7. Young stages of growth of the thallus; 8. Young unbranched filaments; 9. A portion of filament with empty intercalary cells; 10. A portion of filament showing empty terminal cells; 11. Cells of '*Gloeocystis*-generation'; 12, 13. Terminal and intercalary cells showing divided cell-contents; 14. A portion of the filament showingthickwalled perennating structures or akinetes.

the differentiation of the filaments into longer and smaller cells (Text-figs. 6-8). The longer cells after developing to a certain stage in elongation, seem incapable of cell division, while this faculty is retained by the smaller cells which further contribute to the growth of the filaments by their active divisions followed by the differentiation into longer and shorter cells among them. Branching of the filament is almost entirely unilateral and monopodial. In young filaments, branches begin as a lateral protuberance usually near the upper end of the smaller cells. Development of this projection and subsequent formation of septum results in a small branch-cell. This is analogous to the smaller cells mentioned above and further divisions finally produce a lateral branch (Text-fig. 7). The cells at the base of branches often get elongated.

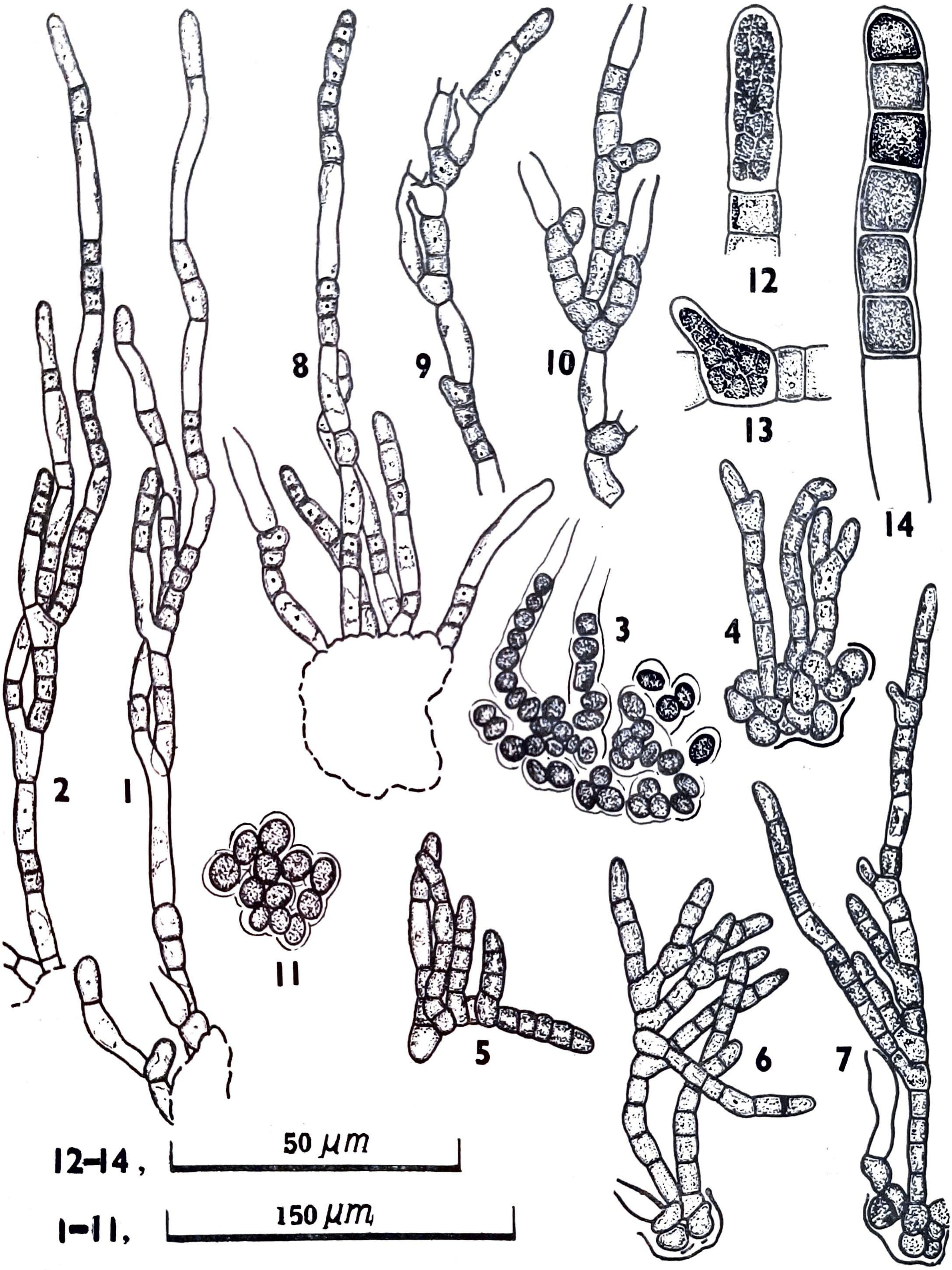
Though the formation and release of zoospores could not be observed, many cells with divided contents were observed in our preserved material (Pl. 1, Figs. 2-4, 6). Such cells occur both in terminal as well as intercalary positions (Text-figs. 12, 13). Intercalary cells with divided contents often possess a bluntly conical or slightly elongate sac-like projection on one side (Pl. 1, Fig. 2; Text-fig. 13). Empty cells with a distinct aperture or pore at the end of such projections or at the tips when terminal in position, were also seen (Pl. 1, Fig. 5; Text-figs. 9, 10). These seem to be pores through which the swarmers (or aplanospores) might have escaped out. These divided contents seem to be of two kinds, distinguishable by their number per cell and size. Some cells possess larger and fewer bodies (Pl. 1, Fig. 6) while others had more and smaller units (Pl. 1, Figs. 3, 4). Since their behaviour could not be observed in our preserved material it can not be said whether these are macro- and micro-zoospores or heterogametes. The smaller cells may also develop into thick-walled perennating structures or akinetes which are frequent in our material (Text-fig. 14). Further fate of the akinetes could not be traced.

The so-called "*Gloeocystis*-generation" (cf. KÜTZING, 1843; HEERING, 1914; PRINTZ, 1927) was also observed (Pl. 1, Fig. 8; Text-fig. 11). Such stages develop from the shorter cells by frequent cell-divisions in irregular planes accompanied by the development of mucilaginous layers around each of the daughter cells. Mucilaginous clusters of more than a dozen cells each, however, could not be observed. The clusters of cells found at the base of the thallus show a close resemblance to the cells of the palmella stages but for the lack of mucilage and the presence of calcification.

It is interesting to note that the results of chemical analyses of the water in which the present alga was found growing, indicate a relatively higher concentration of almost all the chemical constituent factors estimated (cf. Table 1). Relatively higher concentrations of carbonates and bicarbonates tend to raise the pH while a higher chloride value is to be normally expected in the city's untreated waste waters. A comparatively higher concentration of free and saline ammonia and the presence of nitrates are suggestive of active microbial activity while the greater amount of albuminoid ammonia is indicative of the fact that the water is heavily polluted.

## DISCUSSION

The systematic position of *Chlcrotylium* is somewhat controversial. While FRITSCH (1935) places it among the Gongrosireae in the family Trentepohliaceae, SMITH (1933) places it in family Chaetophoraceae. PRINTZ (1964) also puts it in the Chaetophoraceae and not in the Trentepohliaceae but this author does not recognise Gongrosireae as a tribe and has included the genus *Gongrosira* itself in the Chaetophoraceae although SMITH (1933) has placed it (*Gongrosira*) in the Trentepohliaceae. BOURRELLY (1966) also places it in

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The alga shows a high degree of differentiation into smaller and longer cells in the erect filaments and in this feature the thallus organization is reminiscent of those species of *Stigeoclonium* in which the main axis is differentiated into shorter and longer cells (ISLAM, 1963) or of *Draparnaldiopsis* in which the smaller nodal cells are the points wherefrom lateral branches are given out (BHARADWAJA, 1933). In the present alga, the shorter cells are adapted for an entirely different objective as evidenced by the division of their contents, formation of palmella-stages and akinetes. The scanty contents in the longer cells and the unilateral branching of the threads, seem to suggest that the longer cells serve merely to avoid overcrowding of the smaller cells within a small area by spacing these cells apart and thus ensuring even distribution of the thallus on the surface of the supporting substratum while the smaller cells help in reproduction of the plant. Therefore, such a differentiation into two kinds of cells appears to be an adaptation to an epiphytic habit. The longer cells which have nearly lost their contents form weaker links in the filaments which may be helpful in fragmentation of the thallus.

The present alga has been recorded as occurring in running as also standing waters (SMITH, 1933). However, no records have been made on the chemical environment in which the alga flourishes. The chemical analyses (cf. Table 1) show that the alga can tolerate a high degree of organic pollution.

The features of the present alga tally closely with the description of *Chlorotylium mammiforme* Kützing and, therefore, it has been identified as such.

## REFERENCES

BHARADWAJA, Y. (1933). A new species of *Draparnaldiopsis* (*D. indica* sp. nov.). *New Phytol.* **32:** 165-74.

BOURRELLY, P. (1966). *Les algues d' eau douce. Initiation à la systématique.* **1:** Les Algues Vertex. N. Boubee & Cie.

COLLINS, F. S. (1909). The green algae of North America *Tufts College Studies. Sci. Ser.* **2:** 79-480.

FRITSCH, F. E. (1935). *The structure and reproduction of Algae*, I. Cambridge Univ. Press.

HANSGIRG, W. (1886). *Prodromus der Algenflora von Böhmen*. **1.** Prag.

HEERING, W. (1914). In Pascher, A. *Die Süsswasserflora Deutschlands, Osterreichs und der Schweiz.* **6.** Jena.

ISLAM, N. (1963). A revision of genus *Stigeoclonium. Nova Hedwigia,* **10.**

KÜTZING, F. T. (1843). *Phycologia generalis, oder Anatomie, Physiologie und Systematik der Tange.* Leipzig.

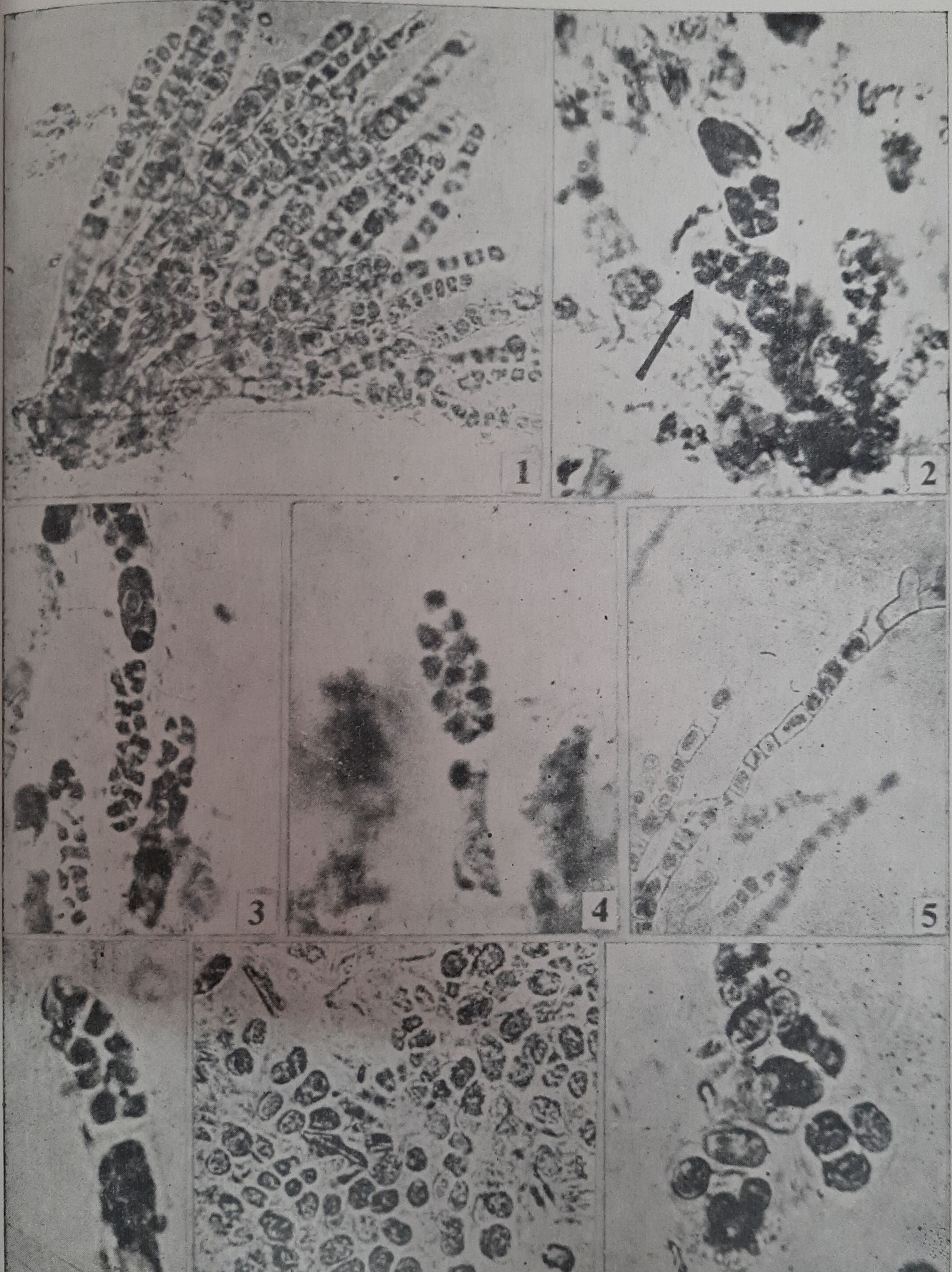
PRINTZ, H. (1927). Chlorophyceae In A. Engler & K. Prantl, *Die natürlichen Pflanzenfamilien.* **3 :** 1-463

PRINTZ, H. (1964). Die Chaetophoralen der Binnengewasser. *Hydrobiologia.* **24 :** (1-3). 1-376.

REINKE, J. (1879). Zwei parasitische Algen. *Bot. Zeitg.* **37 :** 473-478.

SMITH, G. M. (1933). *Fresh Water algae of United States.* McGraw Hill & Co.

Plate 1. *Chlorotylium mammiforme* Kützing—1. A young thallus, (×450); 2. An intercalary cell with divided contents showing a pouch-like lateral protuberance, (×900); 3. Cells with divided contents, (×700); 4. A terminal cell with divided contents, (×1000); 5. Empty intercalary and terminal cells, (×350); 6. A terminal cell with divided contents which are larger and fewer than those in Figs. 3 & 4, (×1200); 7. Clusters of cells and filaments at the base of the thallus as revealed after decalcification, (×450); 8. Cells of "*Gloeocystis*-generation", (×600).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