Contributions to our Knowledge of the Myxophyceae of India.[1]

BY

YAJNAVALKYA BHARADWAJA.

(*Department of Botany, East London College.*)

With eight Figures in the Text.

1. *On a Form of Cylindrospermum with Heterocysts at both ends of the Filaments* (Cylindrospermum muscicola *Kütz. var.* kashmirensis *var. nov.*)[2]

THE alga, forming the subject of this communication, was collected in May, 1931, from a shallow pond in Srinagar, Kashmir, at a height of 5,190 feet above sea-level, growing as an epiphyte on *Myriophyllum*. It forms a thin, velvety, shining, bluish-green irregular layer, very soft and mucilaginous to the touch, attached closely to the substratum; the individual strata are of limited extent, the maximum width of a stratum being about 1·5 cm. The mucilaginous mass comprises many layers of unbranched filaments, which in the upper part of the stratum are more or less straight and parallel and form a compact layer, whilst in the lower part adjacent to the substratum they are irregularly bent and more or less entangled.

The blue-green trichomes are of considerable length (from $\frac{1}{8}$ to $\frac{1}{2}$ mm. long) and are slightly constricted at the joints. The cells are more or less barrel-shaped, and may be twice as long as broad, though occasionally length and breadth are equal; the dimensions are 2·6–3·9 μ broad and 2·6–8·4 μ long. The septa are very distinct. When a filament is stained with an alcoholic solution of iodine, the longitudinal walls, representing the outer investments of the cells, become dark brown and appear moderately thick (Fig. 1, B and C, *o*). The longitudinal walls of adjacent cells are interrupted by the transverse septa which remain colourless (Fig, 1, B and C, *t.*). Internal to the outer investment lies a narrow colourless strip, not stained with alcoholic iodine; this is continuous with the septum and represents the inner investment (Fig. 1, B and C, *i.*). The two envelopes

[1] From the Department of Botany, East London College, University of London.

[2] Latin diagnoses of the new forms described in this paper will be published elsewhere.

[Annals of Botany, Vol. XLVII. No. CLXXXV. January, 1933.]

of the cells thus show the features described by Fritsch (11) in *Anabaena*, and later by Spratt (24) in *A. Cycadeae*. After solution of the cell-contents with 33 per cent. chromic acid the walls exhibit considerable contraction.

Granules, which are angular in the preserved material, are always found in the peripheral cytoplasm; they are larger in old than in young cells and specially large in the spores. The cell contents take up all the stains mentioned by Geitler (16) and Olive (22). Long immersion in aqueous methyl green, however, turns the cell contents violet. Alcoholic eosin stains the cytoplasm lightly and the granules very deeply (cf. Fritsch (10) and Spratt (24)). Alcoholic brilliant green colours the cytoplasm without affecting the granules. The granules disappear in Eau de Javelle, dilute acids and caustic potash.

A single heterocyst is situated at each extremity of the mature filament (Fig. 1, A, *h.*), although in some of the younger filaments there may be a heterocyst only at one end. The heterocysts are oval or ellipsoidal (Fig. 1, B, D–G) and have homogeneous contents of a greyish colour in the preserved material. They are 3·9–5·2 μ broad and 5·2–10·5 μ long, and are provided with two walls.

In the development of a heterocyst from an ordinary vegetative cell, the large granules first disappear and the cell contents gradually become homogeneous. At the same time the cell enlarges and its inner investment thickens and separates from the outer on the side adjoining the next vegetative cell, where a shorter or longer pore is formed (Fig. 2, A). Through this pore communication between the developing heterocyst and the adjoining vegetative cell is maintained. Before the granules disappear the two envelopes of the developing heterocyst are sometimes clearly visible (Fig. 2, B), the inner one being somewhat thickened, most markedly around the pore. The relation of these two envelopes with those of the vegetative cell is not clear. The outer wall also thickens, and in mature heterocysts becomes equally prominent. After long immersion in chlor-zinc iodide the inner wall gives the violet reaction of cellulose (cf. Geitler (13)) and the pore stands out clearly. According to Geitler (13, p. 227) the outer wall of the heterocyst is of a pectic nature, but I have failed to obtain the reaction for pectin substances when my material was treated with ruthenium red (cf. Tunmann (25)). At a later stage there appears opposite the pore a bright refractive granule which seems to enlarge as the heterocyst matures, so that when the pore has closed through the thickening of its walls a large angular granule lies close behind it (Fig. 1, D and E, *g.*), occasionally a little to one side. These granules show the same reactions as those found in the vegetative cells. In some cases a few further granules, apparently of the same nature, have been observed (Fig. 1, E, *g'.*). In heterocysts adjoining mature spores there are no granules (Fig. 1, F and G). In some cases the contents of the mature heterocysts had contracted away

from the inner wall at the distal end (Fig. 1, F and Fig. 2, C), but this may have been due to the action of the preservative. The contents assume a deep colour after long immersion in aqueous methyl green or in Heiden-

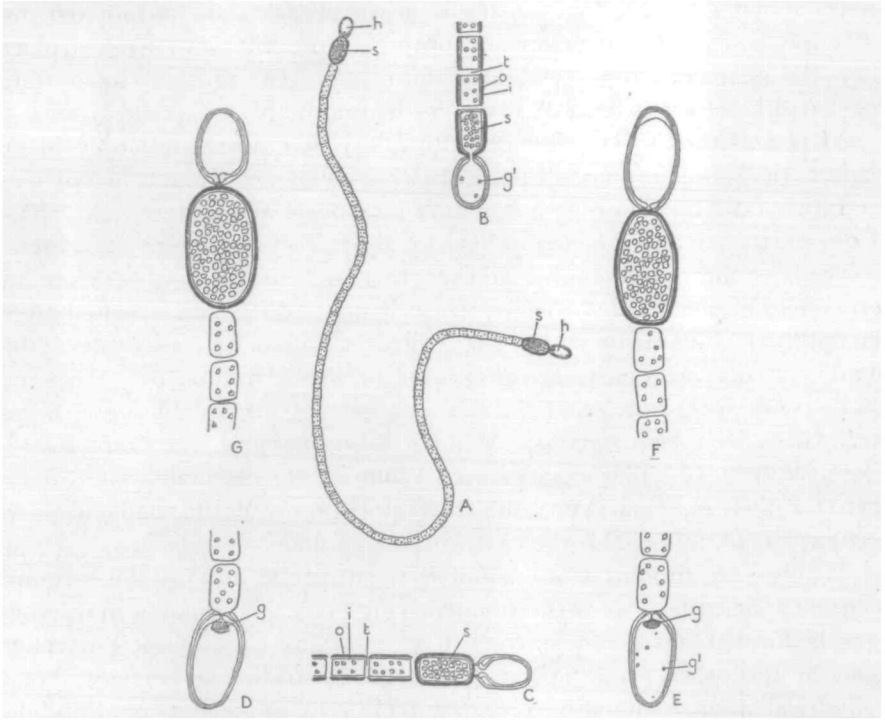

FIG. 1. *Cylindrospermum muscicola* Kütz. var. *kashmirensis*, var. nov. A, mature filament with heterocysts and spores; B and C, portions of filaments stained with iodine, showing the investments of the cell and the development of the spore; D and E, mature terminal heterocysts; F and G, heterocysts with mature spores. *g.*, refractive granule of heterocyst; *g'.*, other granules; *h.*, heterocyst; *i.*, inner investment; *o.*, outer investment; *s.*, spore; *t.*, septum. A × 440; B–G × 1,475.

hain's haematoxylin. In some cases the heterocysts were surrounded by a layer of mucilage which stained with methylene blue and bismarck brown. To this mucilage were attached a number of rod-like bacteria either singly or in linear chains (cf. Borzi (3)).

Intercalary heterocysts, which either occur singly (Fig. 2, D and E, *h.*) or in pairs (Fig. 2, G, *h.*) and are always in an early stage of development, are sometimes found in the middle of a filament. Such heterocysts develop from vegetative cells in the way just described. When these heterocysts occur in pairs, they are usually shorter than the ordinary vegetative cells and appear to have originated from a cell (Fig. 2, F) which has recently divided. Elongate incipient intercalary heterocysts, with a delicate transverse septum dividing them into two (Fig. 2, F, *h.*), are occasionally observed, and it is possible that pairs of intercalary heterocysts always arise in this way, viz. by division of a heterocyst rudiment. At

a time when the young heterocyst only differs from a vegetative cell in the paucity of granules and the homogeneous character of the cell contents, but before the wall thickens it rounds off on one side and the filament breaks into two pieces, one or both of which are thus terminated by a heterocyst. Further development of the young heterocysts takes place after the filaments have separated into two. In no case have fully developed heterocysts been observed in the middle of a filament.

When mature, these originally intercalary heterocysts resemble in all respects the terminal ones. Filaments with young intercalary heterocysts are rather rare, but those with a mature heterocyst and an adjacent spore at one extremity and a young undeveloped heterocyst or merely a vegetative cell, as the case may be, at the other are easily found. Filaments terminated at one end by a vegetative cell may either have resulted from the splitting of filaments with a single median heterocyst or by the dying of an occasional vegetative cell (Fig. 2, H, *d.*) in the middle of a filament. Woloszyńska (27) states that the terminal heterocysts in *Anabaena circularis* G. S. West var. *javanica* Woloosz. (*Anabaenopsis circularis* (G. S. West) Woloosz. et Miller var. *javanica* Woloosz.) are originally intercalary, their terminal position being due to fragmentation of the filaments. As just explained, this is also the case in the alga under discussion.

Spores are produced later, and frequently one adjoins the terminal heterocyst at either end of the filament (Fig. 1, A, *s.*). Spores may, however, be formed only at one extremity, without any trace of spore-development at the other. The spores are larger than the heterocysts, being cylindrical when young (Fig. 1, C, *s.*) and either barrel-shaped or ellipsoidal when mature (Fig. 1, F and G). The mature spores measure 5·2–7·8 μ broad and 9·4–13·6 μ long. Each spore has two walls, the outer very thick, smooth and brown in colour, the inner thin and transparent. The two walls are developed from the two investments of the vegetative cell from which the spore differentiates (Fig. 1, B, *s.*). The contents are bluish-green with numerous large granules which occupy the whole protoplast.

During the early stages of spore-development the wall adjacent to the heterocyst exhibits a distinct pore, by means of which the contents of the sporogenous cell communicate with those of the heterocyst, whose canal-like pore is wide open (Fig. 2, C, *s.*). The granules in the cytoplasm of the sporogenous cell increase in number till they fill the whole cavity. At the same time the outer investment gradually closes in round the open ends and ultimately forms a complete envelope round the mature spore (Fig. 1, B, *s.*). As this happens, part of the transparent intercellular septum is enclosed to form the inner wall at the ends of the spore. The communication between the developing spore and the heterocyst is thus closed (Fig. 1, C, *s.*). The outer wall gradually undergoes uniform thickening, except on the side remote from the heterocyst, where the thickening takes place a little later

(Fig. 1, B, *s.*). The development of the spores is thus in close agreement with that described by Fritsch (11) for *Anabaena*, except for the delayed thickening of the wall on the side remote from the heterocyst. The spores do not contain any oil, as treatment with osmic acid gives no black colour.

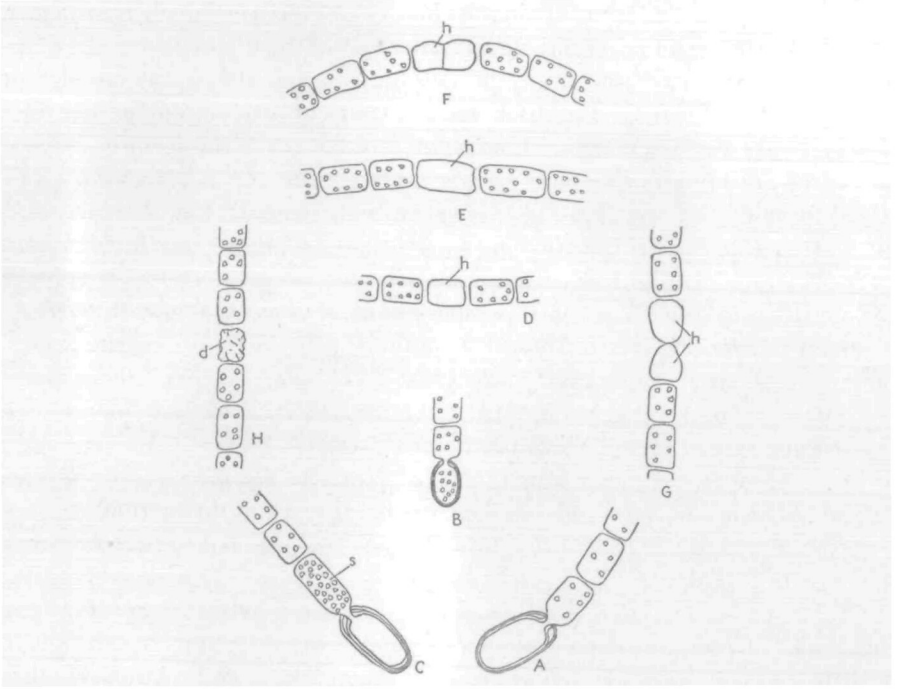

FIG. 2. *Cylindrospermum muscicola* Kütz. var. *kashmirensis* var. nov. A and B, young terminal heterocysts; C, development of spore; D and E, intercalary heterocysts; F, formation of a pair of intercalary heterocysts by the division of a heterocyst-rudiment; G, pair of intercalary heterocysts about to separate; H, filament with a dead cell. *d.*, dead cell; *h.*, heterocyst; *s.*, spore. All × 3,475.

This alga resembles *Cylindrospermum* in possessing terminal heterocysts, each with an adjoining spore, but it differs in the usual presence of a heterocyst at each end of the filament. This last character is distinctive of the genus *Anabaenopsis*, established in 1923 by Miller (21) who raised Wołoszyńska's section *Anabaenopsis* of *Anabaena* to generic rank. *Anabaenopsis* includes those anabaenoid forms which exhibit a terminal heterocyst at each end of the filament. Miller included four species in this new genus: viz. two African ones described by West (26), *A. circularis* (G. S. West) Wołosz. et Miller (*A. flos-aquae* (Lyngb.) Bréb. var. *circularis* West) and *A. Tanganyikae* (G. S. West) Wołosz. et Miller (*A. Tanganyikae* West); one from Java, *A. Raciborskii* Wołosz.; and a Russian species, *A. Elenkini* v. Miller. The genus is recognized by Frémy (8) and Geitler (14, 15, 16); the latter (16) includes three further species, namely *A. Nadsonii* and *A. Milleri* described by Woronichin from Siberia and *A. Arnoldii* described

by Aptekarj from Russia. The alga here described resembles *A. Arnoldii* and *A. circularis* var. *javanica* in the presence of intercalary heterocysts, though they do not mature until the fragmentation of the filament is accomplished.

Frémy gives as the distinguishing characteristics of *Anabaenopsis*, apart from the terminal heterocyst at each end of the filament, the development of the spores remote from the heterocysts and the usual circular or spirally coiled form of the trichomes. Geitler (16) similarly describes *Anabaenopsis* as possessing trichomes, mostly coiled in the form of a spiral or a screw, rarely straight, and spores remote from the heterocysts. The coiled form of the trichomes is, however, not general for *Anabaenopsis*, since in *A. Raciborskii* the trichomes are usually straight and in *A. circularis* occasional straight ones are met with. Moreover, coiled trichomes are met with in quite a number of species of *Anabaena* (e.g. *A. spiroides* Klebahn, *A. Bolochonzewii* Meyer). Similarly, the position of the spores cannot be taken as a general characteristic of *Anabaenopsis*, since spores are unknown in one species and are only known in a variety of another. Intercalary spores remote from the heterocysts and either occurring singly or in pairs, moreover occur in many species of *Anabaena* (e.g. *A. elliptica* Lemm., *A. macrospora* Klebahn, &c.). The only valid distinction between *Anabaenopsis* and *Anabaena* therefore lies in the presence of heterocysts at the ends of the filaments.

The species of *Cylindrospermum* usually have a heterocyst only at one end of the filament. Glade (18), in *C. minutissimum* Collins, has, however, recorded filaments with heterocysts at each end, while Drew (7) has described similar cases in *C. licheniforme* Kütz. and *C. maius* Kütz., spores sometimes adjoining each terminal heterocyst in her material. Glade's material had been grown in culture solutions, while that of Drew had been kept in a damp chamber for a few days. Their forms thus occurred under artificial conditions and could therefore be regarded as exceptional cases, were it not that Frémy (9) has recently in material collected from natural habitats recorded heterocysts at both ends of the filaments, not only in *C. maius* Kütz. and *C. licheniforme* Kütz. but also in *C. catenatum* Ralfs. Brühl and Biswas (5) likewise report the finding of such cases in a *Cylindrospermum*[1] from a natural habitat, while Borge (2) has described two African varieties (*C. Goetzei* Schmidle var. *binum* and *C. muscicola* Kütz. var. *variabilis*) in which heterocysts normally occur at both ends of the trichomes, although in the second case this is only an occasional condition. Borge points out that this recalls *Anabaenopsis Raciborskii* and remarks that, if Geitler's (15) diagnosis of *Anabaenopsis* be followed, the two plants referred by him to *Cylindrospermum* would have to be included in the

[1] They describe the form as *C. doryphorum* Brühl and Biswas, but, as Geitler (16, p. 814) points out, so inadequately that the species must be dropped.

former genus. Owing to the position of the spores adjacent to the heterocysts Borge, however, adopts a reference to *Cylindrospermum*. About one-third of the described species of *Cylindrospermum* are thus at the present time known to be capable of occasionally forming heterocysts at both ends of their filaments. It would, however, scarcely be justifiable to create another new genus on this basis alone for the accommodation of these species.

Skuja (23), moreover, has in *Anabaena echinospora* Skuja recorded both intercalary and terminal heterocysts in the mature filaments, while the young ones have heterocysts only at the two ends. Heterocysts may thus be developed at both ends of a filament, both in *Cylindrospermum* and *Anabaena*. In view of these facts *Anabaenopsis* can scarcely stand as a separate genus, since the only valid feature, the presence of a heterocyst at either end of the filament is one that can occur both in anabaenoid and in *Cylindrospermum*-types. It would consequently seem best to discard the genus *Anabaenopsis* and to re-establish the section *Anabaenopsis* (as distinct from the section *Euanabaena*) of *Anabaena* to include those species of the latter which possess heterocysts at both ends of the filaments. In the same way a section *Cylindrospermopsis* of *Cylindrospermum* might be created for the Indian form here described and other species of the genus with heterocysts at both ends.

According to the key given by Geitler (16) the alga described above agrees in essentials with *C. muscicola* Kütz., except in the blue-green colour of the thallus, the more or less barrel-shaped comparatively narrow vegetative cells which are longer than broad, the oval or ellipsoidal heterocysts which are larger, and the ellipsoidal or barrel-shaped spores which are much smaller. The epiphytic habit of the Indian plant is also very distinctive. The alga is thus a new variety of *C. muscicola* Kütz. which may be named var. *kashmirensis*.

2. *Formation and Germination of Spores in* Aulosira Fritschii *sp. nov.*

The alga described below was found in July 1931, together with certain other algae, on dead leaves that had fallen into the water of a shallow pond near the Benares Hindu University grounds. The alga itself bore numerous epiphytes, including narrow species of *Oedogonium* and *Bulbochaete*, as well as unicellular forms. The material, on which the following description is based, was preserved on the spot in weak formalin. The pond contained plenty of water, and there was no reason to assume that the alga was approaching the end of its period of abundance.

The thallus forms a dirty blue-green or dark green woolly felt-like covering on the surface of the leaves. It consists of unbranched filaments which reach a length of about 7 mm. and are more or less loosely

entangled; the filaments are straight or occasionally irregularly bent. The blue or bluish-green trichomes are constricted at the joints and enclosed in a thick sheath in which two portions can be distinguished (Fig. 3, A–C). The outer hyaline portion (*o*) is stratified with parallel layers, its outer edge being irregular and diffluent. The stratification is clearly brought out by Heidenhain's haematoxylin. This outer sheath is well defined in the purely vegetative trichomes, but in the sporogenous ones it becomes more diffluent and partly dissolved (Fig. 3, D; Fig. 4, C, D, and F, *o*.). The inner sheath is of a denser consistency and rather thinner, unstratified, and of more or less uniform thickness (Fig. 3, A–C, *i*.), except around the spores where it is generally compressed and appears very thin (Fig. 4, A–C, *i*.), sometimes being only recognizable as a faint line (Fig. 3, E and F, *i*.). In such filaments, however, it exhibits annular ingrowths, conical in optical section, which occur opposite the constrictions between adjacent spores (Fig. 3, D–F; Fig. 4, A–E, *a.i*.). The maximum thickness of the double sheath is 3·1 μ. In the vegetative trichomes both the inner and outer sheaths are firm and retain their form, when the contained cells die (Fig. 3, A; Fig. 4, G). The sheath readily takes up all kinds of stains (aqueous methyl green, ruthenium red, alcoholic safranin, &c.), the inner sheath in all cases staining deeper than the outer. These reactions show an affinity of the sheath for basic aniline dyes and indicate its pectic nature (cf. Lemaire (19) and Geitler (16)). Chlor-zinc iodide stains the sheath violet, with or without previous treatment with Eau de Javelle, and this shows that it also contains cellulose, probably in a combined state (cf. Lemaire (19)). The sheaths of sporogenous trichomes stain less deeply than those of the purely vegetative ones. There is generally a narrow space between trichome and sheath.

The cells are 8·4–10·5 μ in diameter, generally slightly longer than broad, but sometimes quadratic in optical section; more rarely they are elongate, two, three, or even four times as long as broad. The cells on either side of the heterocyst are generally drawn out towards it in a characteristic way (Fig. 3, B and C). A similar feature has been recorded by Carter (6) in *Michrochaete uberrima* Carter and by Lemmermann (20) in *Aulosira Schauinslandii* Lemm. The cell-walls are of some thickness. The cell contents are blue or deep blue-green and finely granular. They show practically the same reactions to various stains and reagents as in the *Cylindrospermum* above described. With Heidenhain's haematoxylin the stain is lighter, but the granules become very prominent.

The heterocysts are single and intercalary (Fig. 3, B and C), generally cylindrical or rarely elongate-ellipsoidal in form and usually slightly longer than broad, or sometimes about twice as long as broad; the dimensions are 9·4–11·5 μ broad and 14·7–23·5 μ long. A single heterocyst was found which was four times as long as broad (9·4 $\mu \times$ 37·6 μ). Young heterocysts

have deep blue-green, rarely light violet, finely granular contents like those of the vegetative cells, but in the mature ones the contents become vacuolated and assume a yellow or orange colour, although the granules remain as before. The heterocysts extend right up to the inner sheath with which they are in close contact. The outer wall is thick, while the inner one is thin, being clearly evident in those rare cases in which it has receded from the outer one (Fig. 3, C). The wall around the distinct terminal pores is not specially thickened. The conspicuous granules internal to the pores sometimes reach a considerable size in old heterocysts (Fig. 4, G, *g*.). They are sometimes of irregular shape, though often either plano-convex or concavo-convex, with the convex side away from the pore. Vacuolization commences rather early, so that by the time a heterocyst has reached its full size it possesses a large central cavity with the granular contents occupying the periphery. The oldest heterocysts show practically no contents (Fig. 4, D and G, *h*.). The outer wall of the heterocyst is very persistent, being evident at regular intervals in old filaments in which the vegetative cells have perished. The contents of the young unvacuolated heterocysts stain much more deeply with 1 per cent. aqueous methylene blue than do those of the vegetative cells, but the outer wall is not affected. The cellulose inner wall stains violet with chlor-zinc iodide. With Ehrlich's haematoxylin the granules adjacent to the pores assume a dark colour like those in the vegetative cells.

The *spores* are formed centrifugally in long chains, their formation commencing about midway between two heterocysts and gradually advancing towards them. In one filament sixty-five spores occurred in a continuous chain between two heterocysts, but usually the latter are much nearer to each other so that the chains of spores are considerably shorter. Not uncommonly they are interrupted by vegetative cells. Sometimes the whole of a long filament may consist solely of spores, with intervening heterocysts at more or less regular intervals. The spores are usually roughly quadratic in optical section, varying between 10·5 μ and 12·6 μ in width. Some are cylindrical, up to twice as long as broad.

Each spore has a thick pale brown exospore and a thin hyaline endospore (Fig. 3, D-F, *Ex.*, *End.*); in the preserved material the latter had, in most cases, contracted slightly away from the former. In the mature condition the adjacent parts of the exospore of adjoining spores are closely adpressed to each other, and the appearance is obtained as though all the spores of a chain were joined together and inseparable. Even when sporogenous filaments were placed in strong sulphuric acid for more than a fortnight and subjected to subsequent teasing, the spores could not be separated. The mature exospore, though uniformly thickened over most of its surface, exhibits a special thickening at the corners, which is more marked towards the inside than towards the outside; as a result the inner

surface of the membrane at the four corners is protruded, giving the inner contour of the spore a somewhat octagonal outline in optical section (Fig. 3, D–F). In the contents of the mature spores one can distinguish an extensive central region containing large refractive angular granules and a peripheral region of varying width occupied by fine granules, like those found in the vegetative cells (Fig. 3, D–F, *l.g.*, *f.g.*). The mature spores are thus characterized by the thick closely adpressed transverse walls, the sub-octagonal contour of the contents and the large granules occupying the greater portion of the latter.

Stained with 1 per cent. aqueous methylene blue the exospore becomes light blue, and is difficult to distinguish from the similarly coloured inner sheath which is closely adpressed to it. In the same way alcoholic safranin and aqueous ruthenium red stain the exospore red. The contents, including the granules, are unaffected by any of these stains, as well as by eosin. On the other hand, methyl green colours the exospore light violet, while the inner sheath becomes blue, and thus the latter stands out clearly between the exospore and the outer sheath which also stains violet. With iodine reagents the contents of a mature spore become brown or yellowish-brown, the granules appearing slightly darker. The granules are dissolved by Eau de Javelle, potassium hydroxide, and dilute acids. There appears to be no fat present. In chlor-zinc iodide or strong salt solution the contents of most of the mature spores contract into a dumb-bell shaped mass; the cause of this contraction is not apparent.

The spores develop from the vegetative cells by increase of size and enlargement of the granules, while the colour of the contents changes to light blue-green or yellowish-green. The walls thicken and the exospore and endospore soon become distinguishable. The immature spores thus formed are light blue-green or yellowish-green with a thick pale brown exospore, while large granules are scattered throughout the contents; the walls of adjoining spores appear separated at their middle, while in contact at their edges (Fig. 4, A). At a slightly later stage the inner contour of the spore-wall becomes somewhat protruded at the corners (Fig. 4, B and F) and subsequently, when the contents may acquire a yellow colour, they differentiate into a central region with large granules and a peripheral zone with minute granules; the adjoining walls of adjacent spores are still recognizable as separate entities (Fig. 4, C). This leads over to the mature state in which the inner contour of the spore-wall shows the typical form, and the adjoining walls between the spores are so closely adpressed to each other throughout their length that they appear to form a common transverse septum (Fig. 3, D–F, *t.*).

Certain of the unripe spores of various ages show vacuoles in their contents, and these are very distinct at the stage when only small granules are present (Fig. 4, A, *v.*). The vacuoles are at first small and gradually

merge to form larger ones. Short chains of unripe spores exhibiting these phenomena have been commonly observed. In most of these cases vacuolization proceeds to such an extent that practically the entire contents

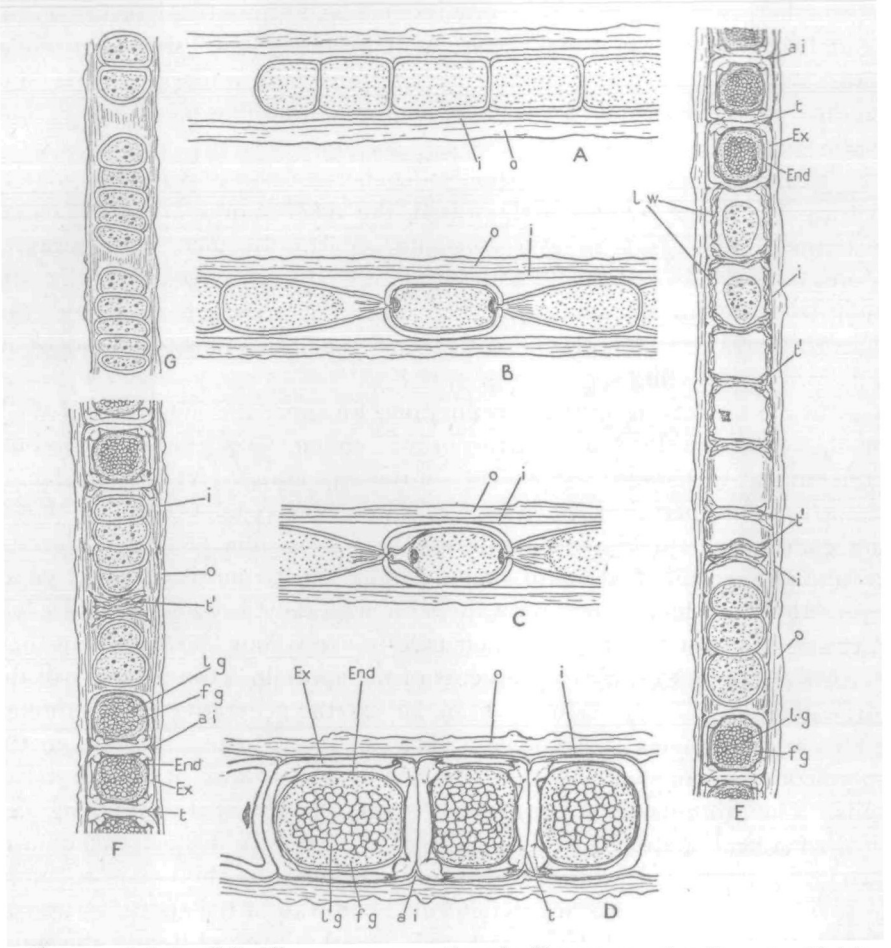

FIG. 3. *Aulosira Fritschii* sp. nov. A, portion of a filament; B and C, filaments with intercalary heterocysts; D, filament with mature spores; E and F, germination of mature spores; G, three new trichomes formed by the germination of mature spores, embedded in a linear series within a tubular mass of mucilage; *a.i.*, annular ingrowth of the inner sheath opposite the constrictions between adjacent spores; *End.*, endospore; *Ex.*, exospore; *f.g.*, fine granules; *i.*, inner sheath; *l.g.*, large granules; *l.w.*, longitudinal diffluent walls of germinating spores; *o.*, outer sheath; *t.*, common septum between adjacent spores; *t'.*, septa left by dead spores. A–C and E–G × 685; D × 1,475.

are obliterated and such spores ultimately degenerate and die (Fig. 4, A and B, *d.s.*). After the contents have died the transverse walls become dissolved, while the longitudinal walls remain intact for some time, so that such stretches of a filament appear as a hollow tube, the wall of which consists of the two sheaths and of the longitudinal walls of the dead spores

(Fig. 4, A, *h.t.*). The solution of the transverse wall starts at its centre and proceeds towards the periphery and generally leaves a narrow peripheral strip attached to the longitudinal walls. Such strips appear as ring-like ingrowths jutting in from the longitudinal walls into the interior of the dead filament (Fig. 4, A and E, *i.d.s.*). Ultimately both the longitudinal walls and the ring-like remains of the transverse walls gelatinize and gradually disappear (Fig. 4, F and G), the latter vanishing much earlier than the former.

Not uncommonly mature spores degenerate in the same way, but in this case only the longitudinal walls of the dead spores are transformed into mucilage, whilst the transverse septa persist. By such dying away of spores gaps are formed which make it possible for other spores in the filament to germinate *in situ* (Fig. 3, E). The transverse septa, left by dead mature spores, are then gradually pushed forwards, as new trichomes grow out from germinating spores (Fig. 3, E, *t'*.).

In the first stages of the germination of a spore the longitudinal walls of the exospore lose their pale brown colour and gradually become diffluent; at this stage they present a wavy appearance and become deeply stained with 1 per cent. aqueous methylene blue (Fig. 3, E, *l.w.*). Later the endospore also becomes mucilaginous. The mucilage thus formed around the germinating spore shows parallel strata and is coloured violet by methylene blue. The end-walls, although they become more or less shrivelled and deformed, retain their pale brown colour and remain in their places. In the meantime the contents of the spore lose the majority of the large granules and become finely granular like the vegetative cells, although a few larger granules are to be seen here and there. In this condition the spore contents are stained with methylene blue like those of the vegetative cells. The protoplast now commences to lengthen parallel to the long axis of the filament, and becomes divided by septa to form a short trichome of two or more cells, the walls of which soon thicken. As the young trichome grows, it pushes the pale brown deformed end-wall of the spore, as well as the persisting end-walls of disintegrated spores, in front of it, and the number of the latter indicates the number of original spore-cavities which it has occupied during its growth (Fig. 3, E, *t'*.). When two such trichomes develop from opposite sides, at some little distance away from one another, they gradually push the persisting remnants of the transverse walls together into a central group (Fig. 3. F, *t'*.). Later, the displaced remnants of the transverse walls become transformed into mucilage and dissolved. The two sheaths of the original trichome also become mucilaginous and the new trichomes, which are then usually from 2–5 cells in length, are found embedded in a linear series within a tubular mass of mucilage (Fig. 3, G) in which they continue to lengthen with the formation of new sheaths till they become typical filaments.

The spores of this plant germinate soon after they are mature, although even immature spores may germinate (cf. below). There is there-

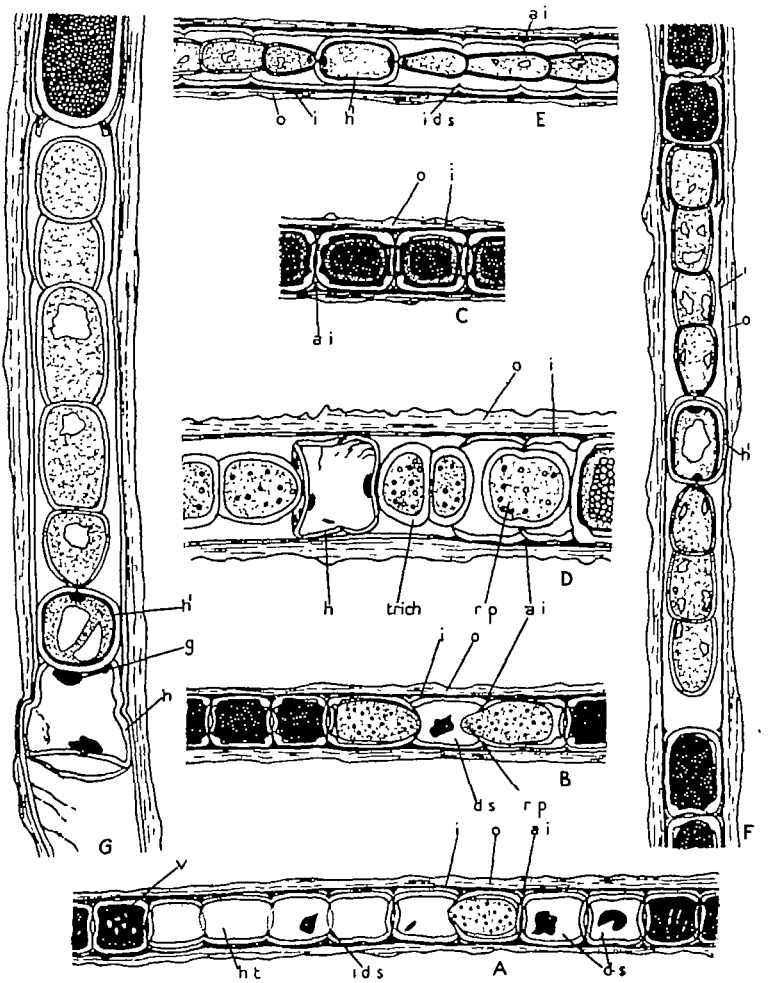

FIG. 4. *Aulosira Fritschii* sp. nov. A and B, filaments with living spores and various stages in their degeneration, also germinating spores; C, filament with immature spores, the end walls of adjacent spores still recognizable; D, germination of immature spores, like those in C; E, new trichome formed by the germination of an immature spore and surrounded by the persisting longitudinal walls of disintegrated spores and the sheaths of the parent-filament; F and G, new trichomes formed by the germination of immature spores within the sheaths of the parent filaments. *a.s.*, annular ingrowth of the inner sheath; *d.s.*, dead spore; *g.*, granule; *h.*, heterocyst of parent-trichome; *h'.*, heterocyst of new trichome; *h.t.*, tube formed by the death of the contents of immature spores; *i.*, inner sheath; *i.d.s.*, peripheral strips of incompletely dissolved septa; *o.*, outer sheath; *r.p.*, rejuvenated protoplast; *trich.*, trichome; *v.*, vacuole. A–C and E and F × 685; D and G × 1,475.

fore, so far as my material shows, no resting period. Similar cases have been described by Fritsch (12) and Bristol (4). Since the end-walls of the spores at first remain unaltered, we have a partial gelatinization of the

spore-walls, such as has been reported by Fritsch in *Anabaena Azollae* (loc. cit., Fig. 6). The germination of a number of spores *in situ*, with the production of a linear series of young trichomes embedded in mucilage, recalls the condition figured by Fritsch (loc. cit., Fig. 11).

Quite frequently immature spores likewise germinate *in situ*, forming short trichomes which grow into a gap created by the death of one or more of such immature spores. In germination the contents of the immature spores undergo the same changes as were above described for the mature ones and the endospore becomes mucilaginous. That half of the common transverse wall, which belongs to the adjoining dead spore, becomes dissolved, while the other half becomes protruded into the adjoining space (Fig. 4, A and B). The central part of this protuberance becomes ruptured, and through the aperture the rejuvenated protoplast emerges (Fig. 4, B, *r.p.*), usually appearing compressed at the point of exit, although sooner or later the aperture widens and the narrowing of the protoplast disappears (Fig. 4, D, *r.p.*). This method of germination agrees with observations of Borzi (3), Fritsch (12), and Spratt (24) (cf. Fritsch, Fig. 25, and Spratt, Fig. 17). As soon as it emerges, the protoplast becomes divided by a septum and forms a trichome of two cells (Fig. 4, D, *trich.*), which, provided sufficient space is available, may grow into a fairly long thread. In such cases the trichomes are enclosed in a tubular envelope composed of the two sheaths and the undissolved longitudinal walls of the disintegrated spores, bearing as ingrowths the incompletely dissolved transverse septa which usually in no way correspond in position to the septa of the new trichome (Fig. 4, E). The appearance thus obtained is highly peculiar, since the envelope of the trichome bears at regular intervals annular ingrowths which, without a knowledge of their mode of origin, would be regarded as part of the sheath. As previously stated, the incompletely dissolved transverse septa ultimately get dissolved, and sooner or later they are followed by the longitudinal walls of the disintegrated spores (Fig. 4, F and G).

Heterocysts are formed quite early (cf. Fritsch (12) and Bristol (4)), being found usually already in trichomes of only 6–8 cells (Fig. 4, F and G, *h'*.). These heterocysts in the young trichomes are generally intercalary (Fig. 4, E and F, *h'*.), sometimes terminal (Fig. 4, G, *h'*.). The trichomes originating from immature spores always remain inside the sheaths of the parent-filament and, though they sometimes attain considerable length, they appear not to live long as vacuolization commences at an early stage, both in the cells and heterocysts. Even in short trichomes of a few cells only, practically all the cells and heterocysts exhibit vacuolization and the contents usually show signs of degeneration. In long trichomes, which are generally found at the ends of the parent-filaments, a number of the terminal cells often assume a rounded shape, but such cells are frequently

practically devoid of contents. When such a long trichome, formed either in the middle of a filament or at one end, possesses a terminal heterocyst adjoining an empty space, the parent-filament sometimes breaks away from the heterocyst, so that the trichome enclosed within the sheaths of the parent-filament is separated off.

The mode of persistence of this alga thus remains obscure. It is probable, however, that some of the spores do not germinate *in situ*, but survive after the death of the rest and lead to a fresh development during the next period of activity.

This alga undoubtedly belongs to the genus *Aulosira* of the family Microchaetaceae, as shown by the unbranched filaments, without differentiation between base and apex, the intercalary heterocysts and the thick firm sheath. It does not, however, agree with any of the species so far described, and there are several respects in which it differs from all previously established species of the genus. These are the double sheath, the presence of granules in the contents of even old heterocysts, the remarkable thickening in the corners of the exospore of the mature spore, and the degeneration of a large number of spores at an earlier or later stage combined with germination of others *in situ*. Like *Aulosira implexa* Born. et Flah., *A. fertilissima* Ghose, and *A. africana* Frémy, the spores are formed in chains, but their shape and dimensions are different to those of any of these species; *A. implexa* and *A. fertilissima* have elongate-ellipsoidal spores with rounded ends, while those of *A. africana* are slightly constricted in the middle. Nor do any of these species develop the remarkably thick spore-envelopes of the form here described. The threads are never associated in bundles, as is characteristic of *A. implexa*. The mode of occurrence on dead leaves is similar to that of *A. fertilissima*.

The alga must therefore be regarded as a new species to be named *Aulosira Fritschii*, sp. nov.

3. *On a Species of Aulosira* (A. prolifica *sp. nov.*) *exhibiting only Vegetative Reproduction.*

In August, 1931, a dense mucilaginous scum formed on the surface of the water of a shallow pond near the Benares Hindu University grounds. When not producing hormogones this consisted of a mass of unbranched parallel filaments, straight or slightly curved, but not agglutinated. The colour of such strata ranged from pale brownish-green to blue-green or green, whilst older strata with abundant hormogone-formation were brownish- or greyish-yellow.

The blue-green trichomes, which are markedly constricted at the joints and exhibit fairly distinct septa, are provided with a thick mucilage-sheath. The latter is at first homogeneous (Fig. 5, A, *s.*), but in the older filaments

becomes differentiated into two parts. The outer sheath gradually becomes diffluent, ultimately leaving only the inner one surrounding the trichome (Fig. 5, B, *o.*, *i.*). The inner sheath is of roughly uniform thickness (1 μ), while the outer one does not exceed 1·75 μ. The outer sheath is soft and colourless, while the inner one possesses some degree of firmness, generally retaining its cylindrical shape when hormogones have escaped from it (Fig. 5, L and M; Fig. 6, I and J), and also occasionally at points where a number of the cells of the trichome have died (Fig, 5, G and I). The empty sheaths persist for some time after the emergence of hormogones, but ultimately become diffluent, and are dissolved. The sheaths are pectic in nature (cf. Lemaire (19) and Geitler (16)), as shown by the stains they take up. The inner sheath always assumes a slightly deeper shade than the outer.

The cells are cylindrical and generally longer than broad (3·1–5·2 μ broad and 6·3–21·0 μ long), rarely quadratic in optical section. The terminal cells (Fig. 5, G), or in young trichomes, rarely also one or two of the subterminal cells (Fig. 5, H), generally taper towards the rounded apex. The cells have coarse granular contents, the granules sometimes being larger in escaped hormogones than in older trichomes. The cell contents show the same reactions as in the forms previously discussed. With stains such as aqueous methylene blue, safranin, Heidenhain's haematoxylin, &c., the central portion of the protoplast becomes deeply stained (cf. Geitler (16) and Olive (22) (Fig. 5, E and F, *c.*). With chlor-zinc iodine the contents contract. Small irregular vacuoles are found in many of the cells (Fig. 5, A, B, and E, *v.*), and in some they develop to such an extent that the protoplast is practically obliterated and ultimately dies. Gaps due to such dying away of cells occur here and there in the filaments. When they are short the inner sheath often becomes bent or distorted at such points (Fig. 6, B); around long gaps it usually contracts to a narrow string (Fig. 6, E), which often breaks across, leaving two filaments bearing at their ends the shrivelled remains of the inner sheath (Fig. 5, I; Fig. 6, D and J). A filament thus generally breaks into a number of pieces containing non-vacuolated cells, due to the death of cells along certain stretches. In some cases the fragments are very short, sometimes consisting of only two cells (Fig. 5, J).

The heterocysts are intercalary or terminal, and are considerably wider than the vegetative cells, so that the sheath is often bulged out opposite the points where they occur. The intercalary heterocysts are generally single (Fig. 5, A, C, and D), rarely in pairs, and are placed at more or less regular intervals throughout the length of the filament. Terminal heterocysts (Fig. 6, B–E) arise at the ends of the fragments of a trichome, produced in the way just described, and also adjacent to biconcave cells (Fig. 6, A) (cf. below). The intercalary heterocysts are always ellipsoidal

(Fig. 5, A, C, and D), whilst the terminal ones are ellipsoidal (Fig. 6, A–C),

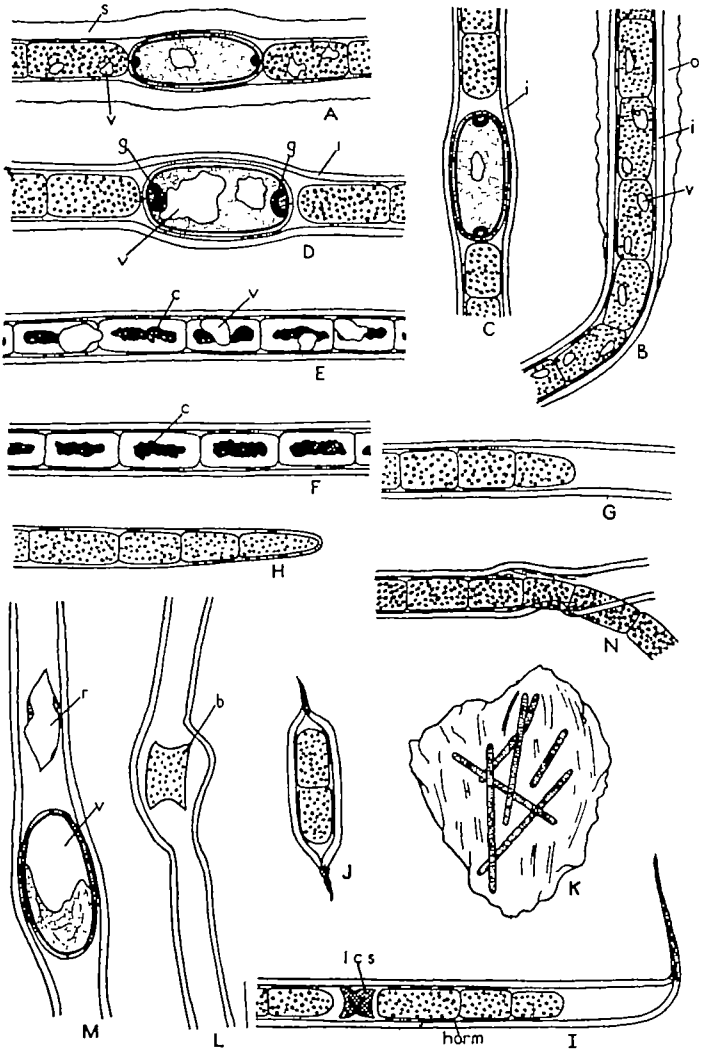

FIG. 5. *Aulosira prolifica* sp. nov. A, filament with homogeneous sheath; B, filament with sheath differentiated into two regions; C and D, filaments with inner sheath only; E and F, filaments stained with methylene blue, showing the central body; G, terminal portion of an old, and H, of a young trichome; I, trichome with a pad of intercellular substance; J, small fragment of a filament; K, mucilage formed from the sheaths intermingled with escaped hormogones; L, empty sheath with biconcave cell left behind on the escape of hormogones; M, empty sheath with old heterocyst, showing an irregular rupture for the emergence of a hormogone; N, escape of a hormogone by longitudinal splitting of the sheath. *b.*, biconcave cell; *c.*, central portion of the protoplast; *g.*, granule; *horm.*, hormogone; *i.*, inner sheath; *i.c.s.*, pad of intercellular substance; *o.*, outer sheath; *r.*, rupture in the sheath; *s.*, sheath; *v.*, vacuole. A–J and L–N × 1,475; K × 320.

oval 6, D) or sub-conical (Fig. 6, J); they are always longer than broad (4·2–8·4 μ broad and 6·4–23·5 μ long). The contents are finely granular,

and show the same behaviour towards stains as do those of the vegetative cells. The outer wall is uniformly thick all the way round, while the inner cellulose layer is very thin and rather indistinct, except around the terminal pores where it is slightly thickened. The outer wall did not show any of the reactions for pectic substances given by Tunmann (25). The terminal pores are usually very narrow and indistinct, but in rare cases they were wider, and exhibited a distinct communication between a young heterocyst and the adjoining vegetative cell (Fig. 6, B). As the heterocyst matures, this connexion disappears, and there is usually a small space between it and the vegetative cell (Fig. 5, C and D; Fig. 6, A). As a general rule a granule lies opposite each pore, and these granules appear to increase in size in the older heterocysts, sometimes being very large (Fig, 5, D, *g.*). They may be plano-convex (Fig. 5, A; Fig. 6, E), biconvex (Fig. 6, A and I), kidney-shaped or semicircular (Fig. 5, D, *g.*), with the convex side away from the pore. The granules, like those of the vegetative cells, take up a dark colour with Ehrlich's haematoxylin. The older heterocysts contain one or two large vacuoles (Fig. 5, D, *v.*), and in later stages there is very little cytoplasm, the whole cavity being practically occupied by one large vacuole (Fig. 5, M; Fig. 6, I, *v.*). The outer wall of the heterocyst is very persistent, the heterocysts or their remains being recognizable at regular intervals within otherwise empty sheaths (Fig. 5, M; Fig. 6, I and J).

In the formation of a heterocyst the adjacent cell or cells round off on the side towards the heterocystous cell (Fig. 6, F, *h.c.*), so that the latter is distinctly marked out. Later the end-walls of the heterocystous cell also round off, and at the same time the granules in the protoplast become smaller (Fig. 6, G and H, *h.c.*). The heterocyst then enlarges to its full size, while the two walls and the pores are differentiated. If a terminal heterocyst dies, the adjoining vegetative cell usually develops into a heterocyst (Fig. 6 D), so that separated portions of a trichome nearly always possess heterocysts at their free ends. The intercalary heterocysts are formed late as they are wanting in young sheathed trichomes of some length. This fact, combined with the ultimate severance of the connexion between older heterocysts and the adjoining vegetative cells, signifies that the formation of heterocysts conditions a breaking up of the trichomes into hormogones, and that intercalary heterocysts probably only arise either a little before or during the formation of the latter.

Spore-formation has not been observed, and the only method of multiplication of this alga in the present material is by means of hormogones, similar to those found by the writer (1) in *Scytonema Malaviyaensis*. These hormogones may consist of many cells or only of one (Fig. 6, C, *horm.*), two (Fig. 6, A, *horm.*), or three (Fig. 5, I, *horm.*). Such hormogones, as already described, often arise as a result of the dying of series of cells at certain points in the trichomes (Fig. 6, B, C, and E). They are also

occasionally formed by the secretion of dark green intercellular substance between two cells, such secretions often taking the form of a thick biconcave

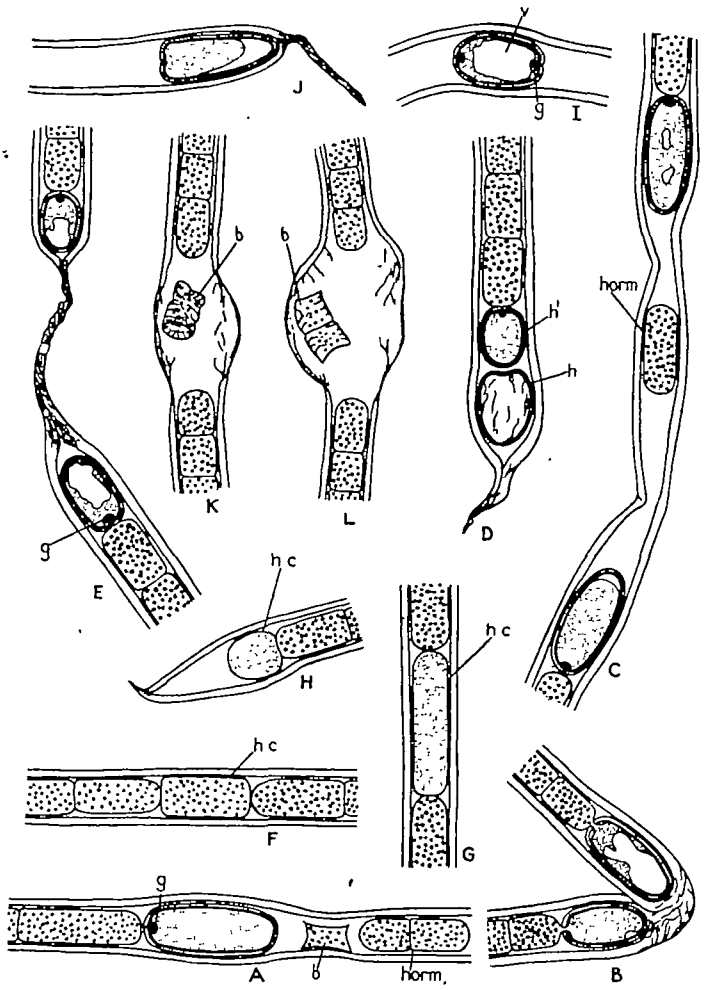

FIG. 6. *Aulosira prolifica* sp. nov. A, trichome interrupted by a biconcave cell with a terminal heterocyst at the end of one of the fragments; B–E, filaments showing the formation of heterocysts at the ends of the fragments produced by the dying of cells; F, G, and H, development of heterocysts; I and J, empty sheaths with persisting heterocysts; K and L, filaments breaking adjacent to biconcave cells where the sheath becomes dilated and more or less diffluent. *b.*, biconcave cell; *g.*, granule; *h.*, terminal dead heterocyst; *h'.*, second terminal heterocyst; *h.c.*, heterocystous cell; *horm.*, hormogone; *v.*, vacuole. All × 1,475.

pad (Fig. 5, I, *i.c.s.*), similar to those met with in many filamentous Cyanophyceae. A third method of hormogone-formation is initiated by the development of intercalary heterocysts, and has already been referred to above. They are also formed by the production of single or paired biconcave cells with granular contents, such as were described by Ghose (17) in

Aulosira fertilissima, and have been commonly recorded in Cyanophyceae. At the points where such biconcave cells are formed, the sheath sometimes becomes dilated and more or less diffluent (Fig. 6, K and L). Later the filament breaks into two portions, and the biconcave cell, which has generally shrivelled owing to the disorganization of its contents, is thrown out. In other cases, however, the sheath remains firm (Fig. 6, A) and the biconcave cells are left behind in the empty sheath as the hormogones escape (Fig. 5, L, *b.*), although they ultimately get dissolved. The escape of the hormogones sometimes takes place through the open end of the sheath, but often by a lateral rupture of the sheath (Fig. 5, M, *r.*, and N), the heterocysts or biconcave cells, as the case may be, remaining behind, and persisting for some time within the empty sheath (Fig. 5, L, *b.*, M, *h.*; Fig. 6, I and J, *h.*). The bulk of the material consisted of loosely entangled filaments in which the trichomes were either entire or broken up into hormogones and enveloped only by the inner sheath; at other points, however, there was merely an irregular mass of empty sheaths or a mass of mucilage formed from these sheaths intermingled with escaped hormogones (Fig. 5, K).

Perennation is accomplished either by entire filaments or by fragments (sometimes only two-celled) of the filaments, or by hormogones remaining dormant inside the persistent sheaths. On the re-occurrence of favourable conditions the hormogones slowly emerge either from the open ends of the sheaths or by longitudinal irregular splitting of the latter, which are now fragile, and often exhibit a rather distorted form at these places (Fig. 5, M, *r.*, and N). The hormogones then lie intermingled with empty sheaths, which ultimately become mucilaginous and get dissolved (Fig. 5, K). Later, the hormogones secrete new sheaths and develop into the young filaments (Fig. 5, H).

The alga just described must be referred to the genus *Aulosira* in view of its possession of unbranched filaments without differentiation between base and apex, of intercalary heterocysts, and of a thick firm sheath. It differs, however, from all previously described species in (*a*) the possession of a double sheath, (*b*) the enlargement of the latter opposite the heterocysts which are considerably wider than the vegetable cells, (*c*) the exceptionally great length of the heterocysts and the presence of granular contents, even in old ones, and (*d*) the formation of terminal heterocysts at the ends of the fragments of a trichome separated by the formation of biconcave cells or by the death of intervening vegetative cells. It is also possible that the absence of spores is characteristic of this species which reproduces so abundantly by vegetative means. It may suitably be called *Aulosira prolifica* sp. nov.

4. *On the False Branching of a Species of Aulosira* (A. pseudoramosa *sp. nov.*)

The peculiar blue-green alga described below formed a flat compact bluish-green stratum with an uneven surface showing rounded prominences among mosses and liverworts on the wall of a house near the famous temple of Shri Vishvanâtha at Benares; it was collected in August, 1931.

In the ordinary vegetative state the stratum consists of unbranched filaments which are 9·5–14·7 μ broad and up to 2 mm. long, and are generally irregularly bent and densely entangled with each other. The blue-green trichomes only occasionally show constrictions at the joints. The sheath is thick, hyaline, and stratified with parallel strata (Fig. 7, A, *s.*). The outer surface is somewhat uneven, while the inner surface is quite smooth except where the trichomes are constricted. At these points the sheath shows slight ring-like projections opposite the septa (Fig. 7, B, *s.*). The sheath is quite firm, since it retains its cylindrical form after parts of the trichomes have perished (Fig. 7, B and G), or after hormogones have escaped (Fig. 7, K). The thickness of the sheath varies between 0·75 and 2·6 μ, according to the age of the filament, but in mature threads it is usually 2·1 μ. In young filaments the sheath is closely adpressed to the trichome, but in mature ones the two are often separated by a narrow space. In older filaments the sheath in most cases gradually acquires a yellow colour, and ultimately becomes deep yellow or golden yellow. At the same time it becomes thinner, hard, and brittle. Even the occasional hyaline sheaths of mature filaments are more or less brittle, as they become irregularly broken after teasing, but the coloured sheath is always very brittle, rupturing very readily (Fig. 7, C), and even breaking of itself under natural conditions, as will be described later. The hyaline sheath is readily stained by the usual pectic stains, but as its colour changes it gradually loses the capacity to take up stains, and the deep yellow or golden yellow sheaths take practically no stain.

The cells are generally cylindrical, rarely barrel-shaped, usually slightly longer than broad, but sometimes as much as twice as long as broad; in rare cases they are shorter than broad. They are 6·3–10·5 μ in diameter. The cell-contents are blue-green and finely granular, and react towards stains and reagents like those of the other forms previously discussed.

In old trichomes occasional vegetative cells, occurring either singly or in short or long chains, show a paling of the contents; ultimately these cells die and disappear, so that gaps are formed in the filaments. In other cases such cells persist, their contents becoming yellowish-brown or brown, and the granules usually less distinct, and eventually disappearing. These cells separate from the other living cells and, when single, appear as contracted (plano-convex, biconvex, concavo-convex, &c.) structures (Fig. 7, D,

E and I, *d*), while, when a number lie together, the whole shrinks (Fig. 7, F, *c.d.*), and becomes distorted in various ways by the growth of the adjacent living cells (Fig. 7, G and H; Fig. 8, A, *c.d.*). These dead cells take up the same stains as the living ones. Dark green biconcave discs of intercellular substance are quite commonly secreted between adjacent cells (Fig. 7, B, E, I, and J, *i.c.s.*), and are sometimes formed quite early in young trichomes and germinating hormogones. Ultimately they become brown, and stain like the living cells, being coloured very deeply by aqueous methylene blue. In some cases the cells adjacent to these intercellular discs degenerate one after the other (Fig. 7, I, *d.*). The trichomes of older filaments are thus broken into separate portions by the dying of intermediate cells, or by the formation of intercellular substance.

Heterocysts are absent from young trichomes, and are formed rather late, and only in small numbers in the mature ones. They are rarely found in trichomes with a hyaline sheath (Fig. 7, A), and are even lacking in a considerable number of those with coloured sheaths. The heterocysts are for the most part intercalary (Fig. 7, A and B), and usually occur singly, though in a few cases in pairs. Terminal heterocysts (Fig. 7, B, *h.*) are formed from the end cells of segments of a trichome separated by intercellular substance. The heterocysts are cylindrical, with rounded ends or ellipsoidal. They are generally of the same width as the vegetative cells, and are commonly slightly longer than broad, rarely twice as long as broad (6·3–10·5 μ broad and 6·3–18·9 μ long). They have fine granular bluish-green contents which stain like those of the vegetative cells. The outer wall of the heterocyst is slightly thicker than the inner. The pores are not clearly visible, since there is no special thickening around them, but a granule is located opposite each. Mature heterocysts are generally isolated from the adjoining vegetative cells, so that the development of a heterocyst likewise causes a trichome to fragment. Sometimes the heterocysts become isolated by the dying of the cells on either side (Fig. 7, B). In old heterocysts the contents degenerate, and the heterocysts often become compressed (Fig. 8, C). In this condition they may be difficult to distinguish from dead cells and pads of intercellular substance, until treated with chlor-zinc-iodide, when the inner wall gives the usual deep violet reaction.

Spore-formation has not been observed, and multiplication in the present material is effected solely by hormogones, formed in the way above described. The hormogones may consist of only one, two, or three cells (Fig. 7, D and E, *horm.*) or may attain some length. By the time hormogone-formation takes place the sheath is generally coloured deep yellow or golden yellow, and within it hormogones can pass through a dormant period, their cells sometimes assuming a rounded or barrel-shaped form (Fig. 8, G, *horm.*). Owing to the brittle nature of the sheath, however, the filament may break into pieces, each enclosing one or more hormogones.

When conditions become favourable for growth, the hormogone secretes a new hyaline sheath, which is very thin and closely adpressed (Fig. 7, F, G, and I; Fig. 8, B, D, and E, *horm.*). It appears that the hormo-

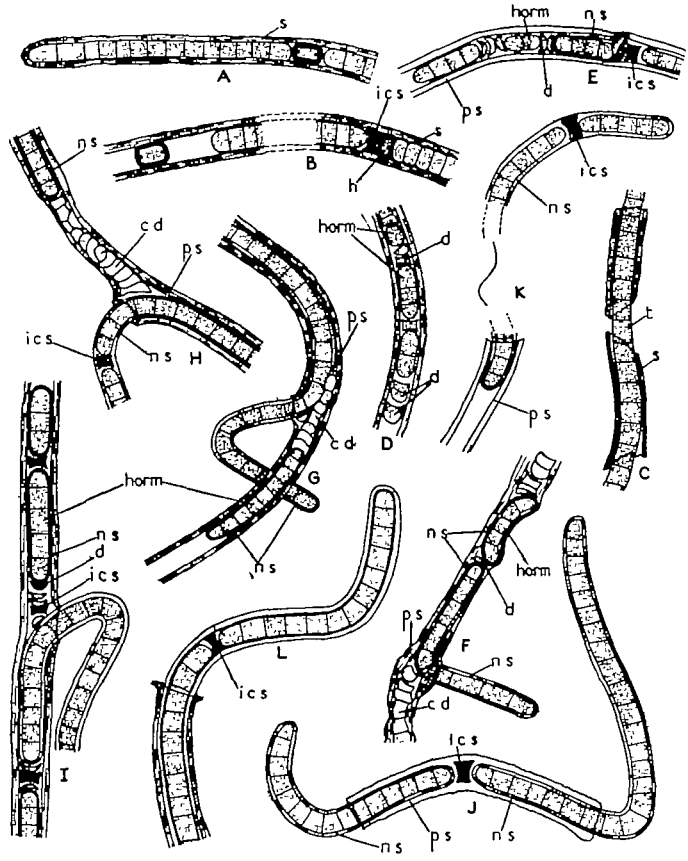

FIG 7. *Aulosira pseudoramosa* sp. nov. A and B, portions of filaments; C, filament showing rupture of the brittle sheath; D and E, breaking up of trichomes into hormogones; F–L, germination and emergence of hormogones after secretion of a new sheath. *c d.*, chain of dead cells; *d.*, dead cell; *h.*, terminal heterocyst; *horm.*, hormogone; *i.c.s.*, intercellular substance; *n.s.*, new sheath of hormogone; *p.s.*, sheath of parent filament; *s.*, sheath; *t.*, trichome. All × 320.

gones do not all start to grow at the same time (Fig. 7, E). The cells elongate, assume a cylindrical form, and commence to divide. When there is plenty of room inside the old sheath, the hormogone may grow to a considerable length before one of its ends penetrates the latter. The new sheath of the hormogone thickens, and becomes stratified like that of the parent filament, which by this time is often very thin and brittle; the combined thickness of the two sheaths is about 3·1 μ. By staining with 1 per cent. aqueous methylene blue the two sheaths can be distinguished (Fig. 8, J) by the deep coloration of the inner one, while the outer remains

unstained. The mode of emergence of the germinating hormogones from the old sheath varies. When they are situated near the open end of the old sheath they grow out direct (Fig. 7, L and K; Fig. 8, D), whilst when there is no such opening at hand they sometimes elongate alongside the adjoining rows of dead cells (Fig. 8, A).

When there is no other room for growth, one end of the hormogone breaks through the sheath of the parent filament (Fig. 7, E and G). Such germinating hormogones, enclosed in their own sheaths, are found emerging adjacent to dead cells (Fig. 7, F–I; Fig. 8, F, H, and I), heterocysts (Fig. 8, C) or discs of intercellular substance (Fig. 8, B, E, and G). When a hormogone penetrates the old sheath in the vicinity of a heterocyst, an appearance very similar to that of a branching *Tolypothrix* is obtained (Fig. 8, C and F). On the other hand, when adjacent ends of germinating hormogones grow out together, an appearance resembling that of a geminate branching of a *Scytonema* is realized (Fig. 8, H), especially in the rare cases in which the dead cells or intercellular discs separating the hormogones have already disintegrated (Fig. 8, I). Without adequate knowledge of the development of the alga, as here described, one might readily regard it as a branching form.

When the hormogones are short, they usually escape wholly from the coloured sheaths of the parent (Fig. 7, J and K; Fig. 8, C, D, and H), so that when a particular trichome is divided up into short hormogones only its sheath may be left altogether empty. Empty sheaths have been commonly observed, though always broken irregularly on account of their brittle nature. Escaped hormogones of this kind are more or less straight, and remain distinct from one another for some time. As they lengthen they become entangled, and the sheaths gradually change colour. Long hormogones or short ones, which have grown to a considerable length owing to the available space within the parent filament, are much delayed in their emergence. As a consequence such hormogones enveloped in their own sheath may, while some part of them is still encased within that of the parent filament, become divided up into fresh hormogones as a result of the formation of biconcave discs (Fig. 7, H, *i.c.s.*); moreover, their sheath may turn yellow before they are set free. In still other cases, which were rarely observed, hormogones which had grown to a considerable length, and were for the most part or entirely enclosed within the sheath of the parent filament, failed to emerge, and remained permanently within the latter. When the sheaths of such hormogones turn yellow, they are difficult to distinguish from the outer sheath of the parent, which is now very thin and papery. Their trichomes may in their turn become divided into hormogones, which in due course again germinate *in situ*. The old sheath of the parent is, however, ultimately thrown off, either throughout its whole length, or only at certain points; in the latter case remnants of it

are sometimes distinctly visible (Fig. 8, K, *r.*). In one exceptional case a sheathed hormogone had a considerable portion of its body inside the parent-filament, whilst the free part outside had grown to a great length;

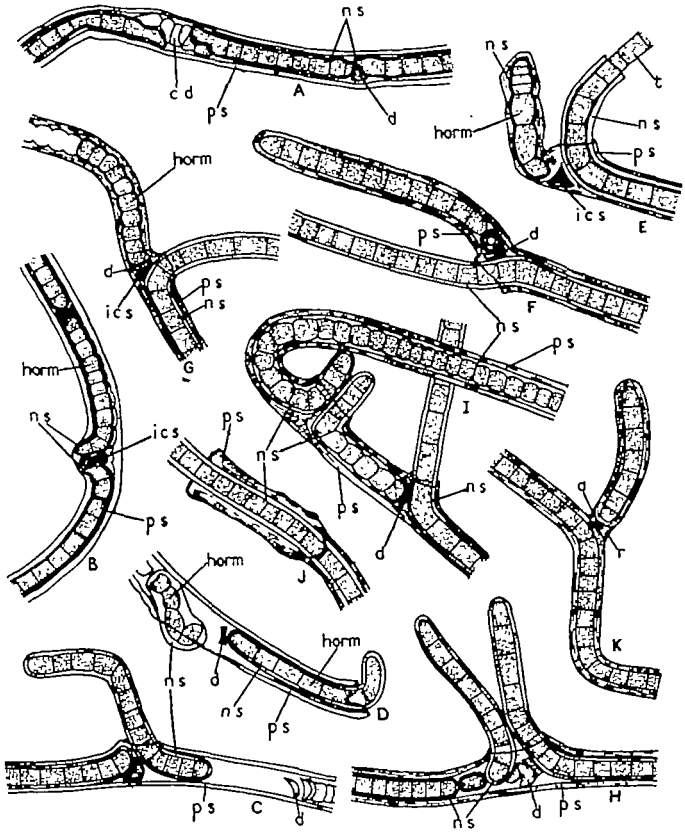

FIG. 8. *Aulosira pseudoramosa* sp. nov. A, germination and elongation of hormogones within parent-sheath; B, two germinating hormogones; C and D, emergence of short germinating hormogones; E–G, emergence of long germinating hormogones, most of which are still enclosed within the parent-sheath; H and I, two germinating hormogones emerging side by side; J, filament with the outer sheath teased out to show the new sheath enclosing a long hormogone; K, two hormogones, enclosed within thick coloured sheaths, joined together by a small remnant of the parent-sheath. *c.d.*, chain of dead cells; *d.*, dead cell; *horm.*, hormogone; *i.c.s.*, intercellular substance; *n.s.*, sheath of hormogone; *p.s.*, sheath of parent-filament; *r.*, remnant of parent sheath; *t.*, trichome. All × 320.

the enclosed portion and the lower part of the free one had a deep yellow sheath, while the terminal part of the free portion which was still elongating had a hyaline sheath.

In view of the marked resemblance of this form to one of the Scytonemataceae, when abundant germination of hormogones is taking place, it may be well to mention that in the false branching of the family just named there is no envelopment of the branch-trichome by a complete new sheath.

The branch-trichome secretes a new sheath of its own which can be traced back for a short distance into the main filament, but does not extend for any considerable distance backwards, terminating blindly. This has been established by examining a number of Scytonemataceae, and the writer hopes to deal with such false branching in greater detail in a later communication. Ghose (17) has described the occurrence of rare branches in *Aulosira fertilissima*, but his data do not enable one to determine their exact nature.

Since the branching of the vegetative filaments has been shown to be solely due to germination of hormogones *in situ*, the absence of differentiation between base and apex, the presence of intercalary heterocysts and the thick firm sheath show that we are dealing with a species of *Aulosira*. There is some resemblance in the possession of a thick coloured sheath to *A. fertilissima* Ghose and in the cylindrical cells and heterocysts to *A. implexa* Born. et Flah. ; but it differs from both in the irregularly bent and densely entangled filaments, and from the former in the cylindrical heterocysts with rounded ends, while the latter has a colourless sheath. The alga under discussion is also peculiar in its habitat (on walls), in its false branching, and in the absence of spores. Since the second of these features is the most distinctive, it may be named *Aulosira pseudoramosa* sp. nov.

In conclusion, I have pleasure in expressing my gratitude to Professor F. E. Fritsch for his guidance and criticism throughout this work. I am also indebted to Dr. N. Carter and Miss F. Rich for many useful suggestions and advice.

April, 1932.

Literature Cited.

1. Bharadwaja, Yajnavalkya : *Scytonema Malaviyaensis*. Rev. Algol., v. 223, 1930.
2. Borge, O.: Süsswasseralgen (Zellpflanzen Ostafrikas gesammelt auf der Akademischen Studienfahrt, 1910). Hedwigia, lxviii. 93, 1928.
3. Borzi, A. : Note alla morfologia e biologia delle alghe Ficocromacee. Nuov. Giorn. Bot. Ital., x. 236, 1878.
4. Bristol, B. M. : On the Alga-Flora of Some Desiccated English Soils : an Important Factor in Soil Biology. Ann. Bot., xxxiv. 35, 1920.
5. Brühl, P., and Biswas, K. : *Cylindrospermum doryphorum*. Journ. and Proc. Asiatic Soc., Bengal, New Ser., xviii. 577, 1922.
6. Carter, N. : Freshwater Algae from India. Rec. Bot. Survey, India, ix, no. 4, 263, 1926.
7. Drew, M. : The Occurrence of Heterocysts and Spores at both Ends of the Filament in the Genus *Cylindrospermum* Kütz. Rev. Algol., v. 143, 1930.
8. Frémy, P. : Les Myxophycées de l'Afrique équatoriale française. Arch. d. Bot., iii. Mem. 2, 1929.
9. ————: Les *Cylindrospermum* de la Normandie. Assoc. Franc. avanc. sciences, Le Havre, 407, 1929.

10. FRITSCH, F. E.: Studies on Cyanophyceae. I. New Phytol., iii. 85, 1904.
11. ———————: Ibid. II. Structure of the Investment and Spore-development in Some Cyanophyceae. Beih. Bot. Centralbl., xviii. 194, 1905.
12. ———————: Ibid. III. Some Points in the Reproduction of *Anabaena*. New Phytol., iii. 216, 1904.
13. GEITLER, L.: Versuch einer Lösung des Heterocysten-Problems. Sitzber. Ak. Wiss. Wien, Mat.-Nat. Kl., Abt. 1, cxxx. 223, 1921.
14. ———————: Synoptische Darstellung der Cyanophyceen in morphologischer und systematischer Hinsicht. Beih. Bot. Centralbl., xli, Abt. 2, 163. 1925.
15. ———————: Die Süsswasserflora Deutschlands, Osterreichs u. d. Schweiz. Heft 12, Cyanophyceae, 1925.
16. ———————: Cyanophyceae, in Rabenhorst's Kryptogamen-Flora von Deutschland, Osterreich und der Schweiz, xiv, 1930–2.
17. GHOSE, S. L.: A Systematic and Ecological Account of a Collection of Blue-green Algae from Lahore and Simla. Journ. Linn. Soc., London, Bot., xlvi. 333, 1923.
18. GLADE, R.: Zur Kenntnis der Gattung *Cylindrospermum*. Cohn's Beitr. Biol. d. Pflanzen, xii. 295, 1914.
19. LEMAIRE, A.: Recherches microchimiques sur la gaine de quelques schizophycées. Journ. de Bot., xv. 255, 302, and 329, 1901.
20. LEMMERMANN, E.: Die Algenflora der Sandwich-Inseln. Engler's Bot. Jahrb., xxxiv. 607, 1905.
21. MILLER, E.: Arch. Soc. russ. Protistol., ii. 125, 1923. (Cited after Geitler (16)).
22. OLIVE, E. W.: Mitotic Division of the Cyanophyceae. Beih. Bot. Centralbl., xviii. 9, 1905.
23. SKUJA, S.: Vorarbeiten zu einer Algenflora von Lettland, ii. Act. Hort. Bot. Univers. Latviensis, i. 149, 1926.
24. SPRATT, E. R.: Some Observations on the Life-history of *Anabaena cycadeae*. Ann. Bot., xxv. 369, 1911.
25. TUNMANN, O.: Pflanzenmikrochemie. Berlin, 1913.
26. WEST, G. S.: Report on the Freshwater Algae, including Phytoplankton of the Third Tanganyika Expedition conducted by Dr. W. A. Cunnington, 1904–5. Journ. Linn. Soc., London, Bot., xxxviii. 81, 1907.
27. WOŁOSZYŃSKA, J.: Das Phytoplankton einiger javanischer Seen mit Berücksichtigung des Sawa-Planktons. Bull. Internat. Acad. Sci., Cracovie, Ser. B, 649, 1912.