

Studies on the Chloroplasts of Desmids. IV.

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With Plates XIV-XVI.

CONTENTS.

	PAGE
XI. THE CHLOROPLASTS OF <i>Staurastrum</i>	303
XII. THE BEHAVIOUR OF THE CHLOROPLASTS DURING CELL-DIVISION	310

XI. THE CHLOROPLASTS OF *STAURASTRUM*.

THE chloroplasts of some of the smaller species of this genus are amongst the earliest and best known of all Desmids, since in so many cases their structure is quite clear from the examination of the living individual in the end view. Thus Ralfs (1848) figured the chloroplasts of *Staurastrum furcigerum* and others, whilst Nägeli (1849) and Delponté (1873) also illustrated the form of chloroplast in several species. The chloroplasts were also figured in several species by W. and G. S. West (1904-11), but in the case of the larger species of the genus the disposition of the chloroplasts is more obscure, and in many cases their structure is not known. It has often been assumed that these large species with very densely green cell-contents were provided with parietal chloroplasts, and Lundell (1871) separated several such species and placed them in a new sub-genus *Pleurenterium*, this being raised by Lagerheim (1888) to the rank of a genus. As in the case of *Pleurotaeniopsis*, however, such a division is undesirable, as explained by W. and G. S. West (1904-11), particularly in view of the fact that many of the species included by Lundell in *Pleurenterium* really possess axile chloroplasts just as the majority of species of *Staurastrum*. The only species of the genus found during this work to be provided invariably with parietal chloroplasts was *St. tumidum*.

The species examined included *St. aversum*, Lund.; *St. hirsutum*, (Ehrenb.) Ralfs; *St. orbiculare*, Ralfs; *St. punctulatum*, Bréb.; *St. aculeatum*, (Ehrenb.) Menegh.; *St. pyramidatum*, West; *St. Simonyi*, Heimerl.; *St. forficulatum*, Lund.; *St. Sebaldi*, Reinsch.; *St. Manfeldtii*, Delp.; *St. Cerastes*, Lund.; *St. gracile*, Ralfs; *St. margaritaceum*, (Ehrenb.) Menegh.; *St. paradoxum*, Meyen, var. *longipes*, Nordst.; *St. jaculiferum*, West; *St. furcigerum*, Bréb.; *St. Ophiura*, Lund.; *St. Arctiscon*, (Ehrenb.) Lund.;

St. anatinum, Cook and Wills; *St. sexangulare*, (Bulnh.) Lund.; *St. Brasiliense*, Nordst., var. *Lundellii*, W. and G. S. West; *St. grande*, Bulnh.; and *St. tumidum*, Bréb.

The first sixteen species agree with each other fairly well in having a simple axile chloroplast with a centrally placed pyrenoid such as was figured by Nägeli (1849). In all the remaining species with the exception of *St. tumidum*, the general plan of the chloroplast is quite similar to that of the first large group, but there are variations in the number and arrangement of the pyrenoids, and in the number of prolongations of the chloroplast running into each angle of the cell. *St. tumidum* has chloroplasts of quite different structure.

In the simplest type of chloroplast there is a fairly massive central axile mass containing typically a single pyrenoid, and from this a more or less definite lobe arises opposite each angle of the cell (Fig. 18). Each of these lobes forks sooner or later, so that there are two distinct masses in each angle of the cell. Very often this forking is so deep that there are apparently two masses or plates arising directly from the axis of the chloroplast opposite each angle (Figs. 2, 4, 6, 8, 10, 12, &c.).

The form of the plates often depends to some extent on the cell-wall, and if this is drawn out at the angles of the cell to form hollow processes or arms, then at corresponding points the plates of the chloroplasts are drawn out to form projections, often of considerable length, which are lodged in them (Figs. 7, 8, 24, 25, 34, and 35). Sometimes the lobes of the chloroplast in the angles of the cell are very thin and plate-like, in which case it not infrequently happens that each lobe forks near the periphery (Fig. 37), or that other smaller plates are given off from the axile mass towards the faces of the cell between the angles (Figs. 4 and 32). In other cases the chloroplast masses in the angles are so large that together they nearly fill the whole semi-cell, and in such chloroplasts the surface of the lobes is often ridged longitudinally, two such ridges being visible in the middle of each face of the semi-cell (Figs. 24, 25, 34, and 35). Such differences in the relative mass of the chloroplast are to be correlated with variations in the amount of stroma starch present in it.

Practically all the smaller species of *Staurostrum* examined have chloroplasts of this type, both those in which the angles of the cell are drawn out to form hollow processes, and those in which they are not.

S. Sebaldi.

Two varieties of this species were examined. The smaller, var. *ornatum*, Nordst., showed no deviations whatever from the simple chloroplast described above (Figs. 24 and 25). The larger one, however, var. *altum*, (Boldt) West, exhibited some remarkable variations in the number, size, and position of its pyrenoids. Unfortunately, it was only

possible to examine three specimens of the larger variety, and all these three differed in the arrangement and number of their pyrenoids, although the chloroplast in each case was of the ordinary type, being axile, with a pair of lobes running into each of the three angles of the semi-cell (Figs. 21 and 23). In practically every semi-cell there was also a single pyrenoid in the centre of the chloroplast (Fig. 21, lower semi-cell); in one, however, there was a group of three (Fig. 21, upper semi-cell), but that is not unusual. In all three specimens there were, however, in addition, other pyrenoids in one or more angles of the cell. Thus one specimen was quite normal in one semi-cell, but the other, in addition to a central group of three pyrenoids, had also a very small pyrenoid in one of its angles (Fig. 21, only two of the central group of three pyrenoids are visible). In the second specimen also the chloroplast was quite normal in one semi-cell, but in the other, besides the large central pyrenoid, there were also one or two very small pyrenoids in each angle of the semi-cell (Fig. 22). The third specimen also had one or two extra pyrenoids in each angle of the cell, but in this case the extra pyrenoids were of considerable size, being nearly as large as the central pyrenoids of the same cell (Fig. 23).

It may be that in the larger variety, as in var. *ornatum*, there is normally a single central pyrenoid in each semi-cell, but that, in the larger cells of var. *altum*, it is impossible in most individuals for all the accumulated products of photosynthesis to be stored in a single pyrenoid, because a pyrenoid cannot increase in size indefinitely. Therefore the original central pyrenoid may in some cases divide to produce a little group of pyrenoids in the middle of the semi-cell, or else new pyrenoids arise in the comparatively large masses of chloroplast in the angles of the cell. It seems natural that new centres of food-storage should be formed at these points, because the lobes of the chloroplast in the angles are in some cases very massive, and photosynthesis must be going on fairly extensively, whilst at the same time it would be a considerable distance for all food reserves to be transferred to the pyrenoid in the centre of the cell. The new pyrenoids are very small at first, increasing in size as stores of food accumulate until they are nearly as large as the older established central pyrenoid.

In the smaller cells of var. *ornatum* the single pyrenoid in the middle of the cell is quite sufficient for the storage of all food reserves, and the lobes of the chloroplast in the angles are neither very large, nor are they removed at any great length from the central pyrenoid, so that the formation of extra pyrenoids within them is unnecessary.

St. furcigerum.

In the form of *St. furcigerum* examined, the cell, which is triangular in the end view, is provided with two short hollow processes at each angle of the semi-cell, one above the other. The form of the chloroplast is, how-

ever, quite similar to that of the ordinary small species of *Staurostrum*. There is an axile mass of chloroplast in the centre of the cell containing typically one pyrenoid, and a biforked lobe running into each angle of the semi-cell. Each plate or lobe radiating from the central axile mass is shaped so as to project slightly into both the short processes of that particular angle.

St. Ophiura.

There is not much difference between the chloroplast of this species and that found in so many of the smaller species. In the latter, the lobes of the chloroplast given off from the central axile mass arise immediately opposite the angles of the cell into which they stretch, each one forking to form two plates, the two plates in any angle having been formed, therefore, from one single lobe (Fig. 8). In *St. Ophiura*, however, the lobes of the chloroplast do not arise opposite the arms of the cell, but between them (Figs. 26 and 27). The semi-cell is provided with a whorl of about eight hollow processes near its apex, and at the bases of these arms the large lobes of the chloroplast corresponding in number to the arms of the cell are drawn out at the apical region of each lateral edge to form a series of long attenuated projections which number twice as many as the massive lobes of the chloroplast given off from the central axis (Figs. 26 and 27). The projections enter the nearest hollow process of the cell-wall, and each arm, therefore, contains two projections, but these two have not been produced from one lobe of the chloroplast, but from two adjacent lobes (Fig. 27).

The chloroplast is very regular in shape, and constant in its form, showing practically no variation other than the occasional occurrence of more than one pyrenoid in the middle of the cell.

St. Artiscon.

The chloroplast in this species differs from the ordinary form in that only one projection is given off from the axile mass into each arm of the semi-cell instead of two (Fig. 29). Each half-cell is provided with fifteen hollow processes which are arranged in a definite way, and the form of the chloroplast naturally depends to some extent on this. The fifteen arms are arranged in two whorls, a whorl of six arms round the apex of the semi-cell, and a larger whorl of nine round the broadest part, lower down. The large central axile mass of chloroplast contains a single pyrenoid, and gives off a single projection to each of the arms, but the projections running into the two whorls seem to be related in a particular way. They broaden out from the arms towards the axis, and certain projections filling particular arms of the lower whorl seem to be connected with corresponding projections running into closely associated arms of the upper whorl, as if at these

points a single chloroplast plate given off from the axis forked into two horizontally, one part supplying an arm of the upper whorl, and the other a corresponding arm of the lower whorl (Fig. 28, *a, a'*; *b, b'*; *d, d'*; *e, e'*, &c.). But as there are nine arms in the lower whorl and only six in the upper, it is obvious that the arms cannot be associated in pairs, one belonging to each whorl, all round the semi-cell, and on closely examining the chloroplast it is found that there are two distinct kinds of plates given off from the central axis. The larger plates stretch nearly from end to end of the chloroplast and are drawn out at their upper and lower extremities to form two prolongations which enter corresponding arms of the upper and lower whorls (*a, a'*; *b, b'*; *d, d'*, &c.). There are also shorter ridges, mere projections which enter certain arms of the lower whorl only, and do not extend as far up as the upper whorl (*c, f, i*, &c.). Altogether there are six larger plates and three smaller ones in each semi-cell, and they alternate regularly, two of the former and one of the latter all round the semi-cell.

St. anatinum and *St. sexangulare*.

Here the chloroplasts differ considerably from those of the other species examined in the relative size of the axis and the plates which radiate into the angles. The central axis is very much reduced, and by far the greater part of the chloroplast is to be found in a more peripheral position in the angles of the cell. Consequently the pyrenoids are not, as before, in the centre of the cell, but in the much larger masses of chloroplast in the angles (Figs. 20 and 44).

St. anatinum is triangular in end view, and there are consequently three masses of chloroplast, one in each angle, which towards the centre of the cell become abruptly very much thinner, being all united by a very narrow surface in the middle of the cell. Towards the periphery each mass forks to form two projections which do not extend very far into the hollow processes of the cell-wall (Fig. 20). There are typically three pyrenoids in each semi-cell, one in each angle, but frequently one or more of them divide to form a little group in well-nourished cells.

The chloroplast of *St. sexangulare* corresponds in all important respects to that of *St. anatinum*. In the material examined each semi-cell was provided with ten hollow processes arranged in two whorls of five, and the cells in end view were pentagonal, corresponding arms of the two whorls being on top of each other (Fig. 44). As before, the greater part of the chloroplast is situated in the angles, and not in the centre of the cell, and there are accordingly five masses of chloroplast, one in each angle, and each containing a pyrenoid. All five masses are connected up in the middle of the cell, and towards the periphery each one forks to form two plates, which stretch out towards the angles (Fig. 44). But in this case there are two hollow processes at each angle, and so the two plates in each angle of the

semi-cell are drawn out at their upper and lower extremities to form four projections, the two upper entering the upper arm of the angle, and the two lower the lower arm (Figs. 43 and 44).

St. Brasiliense, var. *Lundellii*, and *St. grande*.

The chloroplast of the large species *St. Brasiliense*, var. *Lundellii*, differs in several ways from the ordinary type of chloroplast of the genus. The semi-cells are large, and are pentagonal in the end view, and there is a central axis of considerable size in the interior which gives rise to five much larger masses, one in each angle of the semi-cell. Each of these splits up towards the periphery to form four or five distinct though narrow lamellae, whose undulating margins lie against bounding walls of the angles (Fig. 45). The pyrenoids are extremely numerous, and are scattered throughout the central axis and the larger masses in the angles, occurring in six more or less distinct longitudinal rows (Figs. 45 and 46).

St. grande often has a chloroplast which is in some respects very similar to that of *St. Brasiliense*, var. *Lundellii*. The cell is triangular in end view, and there is a central axis in the middle of the semi-cell which gives rise to three fairly large masses of chloroplast, one in each angle. Each of these forks into two, but the resulting plates often cling closely to the walls of the angle after the manner of parietal chloroplasts (Fig. 42). The pyrenoids are probably in typical cases seven in each semi-cell, one in the central axis, and one in each of the plates in the angles. They are usually much more numerous, however, four or five being crowded together in a row in the middle of the cell, and as many as six or seven scattered in the plates of each angle (Figs. 41 and 42).

The chloroplast of this species shows a decided tendency to vary in its disposition, and not infrequently specimens in which the interior of the cell is quite free from chlorophyll-bearing substance are to be observed, the chloroplasts being confined to the angles (Fig. 39). In such cases the central axis of the chloroplast has entirely disappeared, the masses in the angles being thus isolated (Fig. 40). Sometimes these masses in the angles retain their form as single masses forking towards the periphery, but in other cases the angle may contain two distinct plate-like structures, which embrace the walls and are practically parietal bands (Fig. 39). In these chloroplasts the pyrenoids are often two in each angle, one on each side, but they are frequently much more numerous. Sometimes the chloroplast masses in the angles are very irregular in form, and contain numerous scattered pyrenoids (Fig. 40).

Thus when truly axile, the chloroplast of *St. grande* agrees with that of *St. Brasiliense*, var. *Lundellii*, in its massive form, and in the arrangement and frequently great number of its pyrenoids.

In the case of specimens showing parietal chloroplasts it is possible that the change in form occurred in the first place in young semi-cells during cell-divisions. For in *Staurostrum* it often happens that during cell-division the more peripheral lobes of the chloroplast bud into the young semi-cell from the old one much more quickly than the central axile part, streaming up over the quickly growing cell-wall and covering it in a parietal manner, as if the mantling of the whole surface of the cell-wall with photosynthetic material were the most important thing to get completed, and as long as this were accomplished the growth of the more central part of the chloroplast would be of secondary importance. Thus even in species with centrally placed pyrenoids it is frequently seen in young though quite full-sized semi-cells that the real axis of the chloroplast is exceedingly short, and that the plates radiating from this stretch up towards the apex and angles of the cell, almost completely enclosing a large colourless space in the upper region of the semi-cell. This is even more pronounced in *St. anatinum*, where the central axis contains no pyrenoids and is consequently, in any case, not of such vital importance to the young semi-cell. In young semi-cells of this species the central axis in some cases does not exist, and even in some fully grown semi-cells, after the division of the chloroplast at the isthmus between the old and the young semi-cells, it is only represented by an exceedingly short length near the nucleus, the radiating plates arising up from this and arching over the cell-wall of the angles in a parietal manner very suggestive of the parietal chloroplasts often observed in certain cells of *St. grande*. It seems possible that, in the very short semi-cells of *St. grande* particularly, the division of the chloroplast at the isthmus after cell-division may occasionally occur actually before the axis of the chloroplast has entered the new semi-cell at all, while yet the more peripheral parts of the chloroplast have already streamed in from the old semi-cell to the new, and well covered the young cell-wall. The shape of the semi-cell, with its bulging angles and extreme shortness, would seem to be very conducive to this, for it is quite possible that the peripheral parts of the chloroplast, whilst creeping up round the cell-wall of the angles of the young semi-cell, might be cut off from the more central part of the old chloroplast by the sudden division of the chloroplast at the isthmus. This idea is supported by the fact that in some individuals the chloroplasts in one semi-cell are parietal, whilst in the other there is only one chloroplast which is axile.

This strong tendency to variation in the disposition of the chloroplasts during cell-division makes it absolutely impossible for any reliable system of classification to be based on the characters of the chromatophore, such as was attempted by Lundell and Lagerheim.

St. tumidum.

This species differs from all the other species of the genus examined in having chloroplasts which are probably invariable in their parietal disposition. There are usually about twelve to fifteen or sometimes more chloroplasts in each semi-cell, and they are in the form of rather narrow bands, running longitudinally, each with two or three pyrenoids embedded in it (Fig. 48). Very often the bands are fairly even in width throughout their length, their outlines are often slightly irregular, but their structure does not seem to be complicated by the presence of projections from the edges and surface, as in the case of the parietal chloroplasts of some other genera. In other specimens the chloroplasts are drawn out at certain points throughout their length to form very thin strings between the large globular masses which alternate with them and contain the pyrenoids, whilst in other cells the bands are irregular in form and do not extend from end to end of the semi-cell (Fig. 47).

XII. THE BEHAVIOUR OF THE CHLOROPLASTS DURING CELL-DIVISION.

The general external appearance of cell-division in Desmids was first figured by Ehrenberg (1838) in *Cosmarium*, and later in other genera as well by Focke (1847), Ralfs (1848), Nägeli (1849), de Bary (1858), and Delponte (1873). In most cases, however, these investigators did not attempt to illustrate the origin of the chloroplast which eventually becomes apparent in the new semi-cell, and it is only in the figures of Focke (1847) for *Micrasterias*, and de Bary (1858) for *Cosmarium Botrytis*, that any clue is given to the behaviour of the chloroplast during cell-division. Amongst unconstricted Desmids the details of cell-division, including the division of the chloroplast, have been thoroughly investigated in *Closterium* by Fischer (1883) and Lutman (1911), and thus, although our knowledge of the processes accompanying cell-division in the Saccodermæ is fairly complete, in the much larger group of constricted Desmids nothing has been discovered since the time of de Bary, and there is no definite information concerning the division of the chloroplasts in these forms, with the exception of the very slightly constricted genus *Hyalotheca*, in which cell-division has been recently studied by Acton (1916).

In *Hyalotheca* and *Closterium*, the only two genera which have at present been investigated, the process of cell-division is not identical. In *Closterium*, as described by Lutman, the first intimation that cell-division is about to take place is seen in the chloroplasts, which show a pinching-in about one-third the distance from nucleus to apex. Several hours later the nucleus divides, and eventually a transverse wall is laid down between the daughter nuclei. The latter then become amoeboid and begin to move away in opposite directions, travelling towards the constrictions in the

chromatophores. Each nucleus finally takes up its position in the constriction, and the chloroplast then completes its division, the nucleus slipping in between the two newly-formed halves.

In *Hyalotheca* according to Acton (1916) it is the nucleus which divides first, and the transverse wall is then formed. As in *Closterium* the daughter nuclei then become amoeboid, and each one takes up a lateral position opposite the central pyrenoid of one of the chloroplasts. Under the influence of the nucleus the chloroplast and pyrenoid now divide, the nucleus slipping in between the two halves of the chloroplast as they separate.

The process in *Hyalotheca* differs from that in *Closterium* in that the chloroplast does not divide until after nuclear division has been completed, whereas in *Closterium* the chromatophores prepare for their division long before there are any other visible signs of cell-division. On the other hand the two genera agree in the amoeboid movements of the daughter nuclei.

During the present investigation the process of cell-division was studied in living examples of several species, including *Micrasterias rotata*, (Grev.) Ralfs; *M. denticulata*, Bréb.; *Cosmarium punctulatum*, Bréb.; *C. subtumidum*, Nordst.; *Euastrum Didelta*, (Turp.) Ralfs; *Eu. ansatum*, Ralfs; and *Staurastrum punctulatum*, Bréb., whilst the division process was investigated from stained preparations at various stages in several other species.

In *Netrium* and *Cylindrocystis* cell-division probably closely resembles that of *Closterium*. In Fig. 51 the first signs of cell-division are seen in *Netrium oblongum*, (de Bary) Lütkem., var. *cylindricum*, West, in the constriction of the chloroplast and pyrenoid about half-way between the nucleus and apex. (In *Closterium* this constriction occurs about one-third the distance from nucleus to apex, because the apices of the cell are usually so very attenuated, but in both cases the chlorophyll-bearing material is approximately halved.) In Fig. 53 (*Netrium Digitus*) the transverse wall has been formed, and the daughter nuclei have begun to migrate towards the gaps between the chloroplasts, the latter having apparently quite completed their division without the very close association of the nuclei. Fig. 52 shows the completed division.

A rather late division stage in *Cylindrocystis crassa*, de Bary, is seen in Fig. 50. The pyrenoids and nucleus have already divided, and the new cell-wall is beginning to form, whilst the daughter nuclei have migrated towards the constrictions in the chloroplasts.

In all the constricted species examined the process of cell-division is quite different from that in both *Closterium* and *Hyalotheca*. There is never any migration of the daughter nuclei, for these, in all the forms examined, naturally lie, as soon as formed, one at each end of the rapidly elongating isthmus between the two semi-cells of the dividing individual. Thus from the first they occupy their normal position in the isthmus, and as

the new semi-cells are formed there is no need for any change. In such forms it is rather the chromatophores which migrate.

Cell-division was very similar in all the Placodermæ investigated. The individual about to divide is usually very densely green, its chloroplasts are coarsely granular, and neither their definite structure nor the position of the pyrenoids can be distinguished. The reason for this is doubtless that the chloroplasts are very distended with stroma starch, which gives them the shapeless granular appearance, and by its great refractivity obscures the pyrenoids. The cytoplasm is sometimes crowded with numerous colourless oily-looking globules.

The division of the nucleus, accompanied by the elongation of the isthmus, and the subsequent formation of the transverse cell-wall are all completed before any visible changes take place in the chromatophores. The young semi-cells, still colourless, begin to round themselves off, and may even separate before anything further happens as far as the chromatophores are concerned. At this stage the protoplasm in the young semi-cells is becoming rather vacuolate, and its rapid streaming movements are clearly visible. Very often the daughter nuclei can be seen with high magnification as glistening bodies embedded in the protoplasm in the isthmus of each individual, and the colourless oily-looking globules may stream into the young semi-cells from the old one (Fig. 61). When the young semi-cells have attained a fair size, the chloroplast in each of the older half-cells begins to protrude slightly through the isthmus on each side (Fig. 62). This small part budded into the isthmus continues to increase in size as more and more of the chloroplast streams in from the old semi-cell into the rapidly growing young one. As the young semi-cell is gradually filled, the chloroplast contracts visibly from the wall of the older one (Figs. 62-5). Once the process has begun it continues very rapidly for a time; thus the period of time between the stages represented in Figs. 62 and 64 is only half an hour, and at the end of another half-hour the young semi-cells have begun to assume their characteristic form, and contain nearly as much of the chloroplast as the old ones (Fig. 65). The subsequent growth of the individuals was not so rapid, but in less than six hours after the beginning of the process the chloroplast had not only completed its budding, but in both old and young semi-cells had spread itself out so as to mantle completely the whole cell-wall, both individuals being now uniformly green (Fig. 67).

It now only remained for the chloroplast to divide at the isthmus. As observed in living specimens under the microscope this final process was very slow, and in many cases, in spite of the use of various arrangements to ensure a free supply of water between the slide and cover-glass, the individual often died before the chloroplasts had completed their division. In the specimen of *Euastrum Didelta* figured, the division was complete except for the final breaking of the thin strand connecting the two halves of

the chloroplast (Figs. 68–70), but the constriction of the chloroplast extended over two days. The complete division of the chloroplast was observed later in *Micrasterias denticulata*, in which the length of time between the stage represented in Fig. 69 and the final breaking of the drawn-out connecting thread was about sixteen hours. In their natural surroundings, however, it is most likely that the division of the chloroplast takes place far more rapidly than in specimens kept under observation under more or less unhealthy conditions. For in fixed and stained material one often encounters young individuals whose new semi-cells are not yet fully formed, yet whose chloroplasts have nevertheless completely divided at the isthmus.

It will be noticed that the actual division of the chloroplast differs somewhat from that previously described for *Closterium* and *Hyalotheca*. In both these genera there is a pinching-in of the chromatophore, and under the influence of the nucleus this furrow becomes deeper and deeper until the two halves are completely severed. In all the forms examined in this work such a pinching-in was not observed, the two halves of the chloroplast apparently pulling themselves apart, the connecting strand between them becoming thinner and thinner until it finally broke.

Owing to the dense nature of the cell-contents in most cases, and the large quantity of starch contained in the chromatophore, the behaviour of the pyrenoids could not usually be traced in the living condition, but in *Cosmarium subtumidum* it was possible under high magnification to distinguish the pyrenoids, and also to keep them under observation during the division processes.

In this species the single central pyrenoid, soon after the beginning of the budding of the chloroplast, begins to elongate slightly at its lower end (Fig. 71). The starch-grains rearrange themselves rapidly, and those at the lower end of the pyrenoid seem to form a small loop which doubtless contains a small globule budded off from the pyreno-crystal (Fig. 72). The latter cannot be seen, however, because of the great refractivity of the starch-grains. The small pyrenoid thus budded off from the original one apparently increases in size (Fig. 73), and the starch-grains arrange themselves so as to cut it off from the old one (Fig. 74). The process takes place very rapidly, ten to fifteen minutes sufficing for the complete division. During the formation of the new pyrenoid, the latter is gradually transported with the budding chloroplast into the young semi-cell (Figs. 71–4), but the two pyrenoids remain connected by means of starch-grains for a considerable time (Fig. 74).

In most of the species examined with more or less flattened cells, the chloroplast entered the young semi-cell through the isthmus as a somewhat bilobed protuberance (Fig. 62), although this form was sometimes quickly lost as the chloroplast streamed more and more into the new semi-cell. In the case of cells triangular or pentangular in the end view, and having nor-

mally a correspondingly three or five lobed chloroplast, the latter enters the new semi-cell from the old one as a three or five lobed mass (Figs. 49 and 55). Thus the definite form of the chloroplast is retained as far as possible during the process of budding, and, as seen in Fig. 55, the five-lobed form of the chloroplast is visible in the young semi-cells of *Staurastrum Brasiliense*, Nordst., var. *Lundellii*, West, at a very early stage, long before the characteristic shape of the cell-wall is apparent.

The nucleus of the cell is usually pushed to one side by the ingrowing chloroplast, especially where the latter is a single central one. In many cases it is very difficult, even in stained specimens, to distinguish the nucleus in newly divided individuals, in which the two halves of the chloroplast are not yet severed, but it can frequently be seen a little to one side (Figs. 49, 54, 56, 57, and 60). Occasionally it is carried away to some considerable distance into the new semi-cell by the intruding cell-contents, but whether such individuals survive in the end is doubtful.

The simple chloroplasts of the smaller species of *Euastrum* and *Cosmarium* offer no striking phenomena during cell-division. As in *C. sub-tumidum*, the single central pyrenoid constricts to form two during the process (Fig. 54). In the very numerous species of *Cosmarium* containing two chloroplasts in a semi-cell, each with one point of pyrenoid formation, the budding of the chloroplasts has already been figured by de Bary (1858) as typified by *C. Botrytis*. Each of the two chloroplasts bud into the new semi-cell through the isthmus, the pyrenoid of each meanwhile constricting to form two. The process is identical in *Euastrum verrucosum*, Ehrenb., and also in those species of *Xanthidium* having a similar chloroplast structure.

Where the axile chloroplast has more than one point of pyrenoid formation, as, for example, in *Staurastrum anatinum*, in which the cells, triangular in the end view, have typically one pyrenoid in each angle, all three pyrenoids constrict as the chloroplast projects as a trilobed mass into the young semi-cell (Fig. 58).

In the case of axile chloroplasts containing numerous scattered pyrenoids, e.g. *Tetmemorus*, *Micrasterias*, and *Staurastrum Brasiliense*, a number of pyrenoids are carried into the young semi-cell by the budding chloroplast (Figs. 56 and 60), and they do not conspicuously increase in number during the process.

With parietal chloroplasts, each one buds into the young semi-cell soon after its formation. Where each parietal plate contains a single pyrenoid, as in many species of *Xanthidium*, this constricts as the chromatophore projects through the isthmus, just as in the case of axile chloroplasts containing typically one pyrenoid; but if the pyrenoids are numerous and scattered, as in *X. armatum*, (Bréb.) Rabenh., a number of these pass into the new semi-cell along with the ingrowing chromatophore (Fig. 59).

In *Cosmarium Brebissonii*, Menegh., the complicated chloroplasts press into the young semi-cell as a comparatively shapeless mass, the pyrenoids thronging in at the same time (Fig. 57). In the figure the nucleus is seen pressed by the chloroplast to the right of the isthmus.

It was noticed that in the axile chloroplasts of many species there is a distinct tendency for the peripheral parts of the chromatophore to enter the young semi-cell very much more rapidly than the central part, as if there were an attempt on the part of the organism to cover the cell-wall with photosynthetic material as quickly as possible, no matter what happened in the interior of the cell. This is very pronounced in many species of *Staurostrum*, in which the angles of the young semi-cell may be quite filled by the chromatophore whilst yet the axial part of it has scarcely entered (Figs. 49 and 58). It is quite possible that very often the division of the chloroplast at the isthmus occurs before the axial part has entered the young semi-cell at all. Thus several distinct chloroplasts would be produced in the new half-cell instead of a single central one. In the next generation the entire cell would probably be provided with such chloroplasts in half the individuals.

This phenomenon has already been mentioned as being probably responsible for the frequent occurrence of wholly parietal chloroplasts in *St. grande* (Bulnh.), which normally possesses a single axile one, and also for the frequent shortening of the axis of the chloroplast in the median region in *Micrasterias truncata*, (Corda) Bréb., *M. oscitans*, var. *mucronata*, (Dixon) Wille, and *Cosmarium Ralfsii*, Bréb., and also for the production, in the extreme case, of two distinct chloroplasts in a semi-cell in the three latter species.

Further, in other species, one sometimes encounters odd specimens in which the chloroplasts, which normally should be axile, are more or less parietal. Such have been observed in *Tetmemorus Brebissonii*, (Menegh.) Ralfs, *T. granulatus*, (Bréb.) Ralfs, and others. The same explanation probably applies to these.

Again, it has been noted earlier that in the larger species of *Euastrum* a considerable proportion of individuals are found in which the axis of the chloroplast is either shortened, or else, together with the radiating plates, is altogether absent.¹ In extreme cases there may be several entirely parietal chloroplasts in a semi-cell. Unfortunately dividing specimens of the species concerned were not frequently met with in stained material, and so it was impossible to ascertain the real cause of the discrepancy, but it is not improbable that the abnormalities arose in young semi-cells during cell-division by the mantling of the cell-wall by the parietal plates long before the central axis and radiating plates had entered.

Finally, it is also suggested that in those species which normally possess

¹ Carter, N. : Studies on the Chloroplasts of Desmids. 1. Ann. Bot., vol. xxxiii, 1919.

parietal chloroplasts, these were originally derived in a similar manner, and during the course of ages have become permanent.

Thus the fact that the chloroplast, during cell-division, streams so rapidly through a usually very narrow isthmus from the older semi-cell into the new one, offers many opportunities for variation in its form, and it is really surprising that in so many species the structure of the chloroplast is characteristic and constant. Considering that every chloroplast is derived from an original small bud of green material squeezed through a narrow passage one would expect a large proportion of abnormalities, and for this reason, at any rate, it would seem unwise to make the form of the chloroplast in this group the basis of a classification.

SUMMARY OF THE SPECIAL CHARACTERS OF *STAURASTRUM*.

Most species of *Staurastrum* have axile chloroplasts. The only species examined which always has parietal chloroplasts is *St. tumidum*.

Many of the smaller species have a simple axile chloroplast consisting of a central axis which contains a single pyrenoid and a bilobed mass projecting into each angle of the semi-cell.

Amongst the larger species the general form of the chromatophore is often quite similar to that of the smaller ones, but there are variations in the number of plates in each angle and also in the number and arrangement of the pyrenoids.

Most of the species examined had one point of pyrenoid formation in the centre of the semi-cell, but in a few species the pyrenoids occur either in the angles only, or else in addition to those in the centre.

St. Brasiliense and *St. grande* differ from most of the other species examined in their very numerous pyrenoids.

In *St. grande* also some individuals show a tendency to the parietal disposition of the chloroplast by the total disappearance of the axis in the centre of the semi-cell, leaving the peripheral lobes of the chloroplasts isolated.

SUMMARY OF THE CHARACTERS OF CELL-DIVISION.

The chloroplasts of *Netrium* and *Cylindrocystis* probably behave during cell-division in a manner essentially similar to that already described by Lutman for *Closterium*.

In all the Placoderm Desmids examined the process of cell-division is rather different from that of the Saccodermæ. The nucleus of the cell completes its division, and the two new colourless semi-cells can readily be distinguished before there are any visible changes in the chromatophores. The latter then rapidly stream through the isthmus from the old semi-cell into the new one, so that by the time it is fully formed it is usually uniformly green. The process is completed by the division of the chloroplasts at the isthmus of each individual.

In those species in which the points at which pyrenoids may occur are fixed the young semi-cell is provided with a corresponding number of pyrenoids by the budding of those already existing in the old semi-cell. Where the pyrenoids are indefinite in number and scattered, a number of these enter the new semi-cell together with the budding chromatophore.

A striking feature of the ingrowth of the chloroplast in many species is the rapidity with which the cell-wall of the young semi-cell is completely mantled by the chloroplast, often at the expense of the more central parts of the semi-cell. This phenomenon is responsible for the formation of parietal chloroplasts in isolated specimens of species which normally possess axile ones, and probably also for their original production in species in which they have been permanently acquired.

In conclusion I have to acknowledge the invaluable help and advice which I received throughout the whole of this investigation from the late Professor G. S. West, and also my indebtedness to him for providing much of the material. My thanks are also due to the Birmingham Natural History and Philosophical Society for a grant from their Endowment of Research Fund to help in the cost of producing the plates illustrating this work, and also to the Royal Society for a further grant for the same purpose.

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DESCRIPTION OF PLATES XIV–XVI.

Illustrating Miss Nellie Carter's paper on the Chloroplasts of Desmids. IV.

During the prolonged processes of preparation the specific characters of Desmids often become obliterated, but all the species were identified, either in the living or carefully fixed condition, by Professor G. S. West. The figures are only intended to indicate the structure of the chromatophores, and only the outlines of the cells are given, surface markings being omitted.

PLATE XIV.

Figs. 1, 2. *Staurostrum forficulatum*, Lund. $\times 510$. Fig. 1, front view; Fig. 2, end view.
 Figs. 3, 4. *St. orbiculare*, Ralfs. $\times 510$. Fig. 3, front view; Fig. 4, end view.
 Figs. 5, 6. *St. Cerastes*, Lund. $\times 510$. Fig. 5, front view; Fig. 6, end view.
 Figs. 7, 8. *St. gracile*, Ralfs. $\times 510$. Fig. 7, front view; Fig. 8, end view.
 Figs. 9, 10. *St. Simonyi*, Heimerl. $\times 510$. Fig. 9, front view; Fig. 10, end view.
 Figs. 11, 12. *St. punctulatum*, Bréb. $\times 510$. Fig. 11, front view; Fig. 12, end view.
 Figs. 13, 14. *St. aversum*, Lund. $\times 510$. Fig. 13, front view; Fig. 14, end view.
 Figs. 15, 16. *St. hirsutum*, (Ehrenb.) Ralfs. $\times 510$. Fig. 15, front view; Fig. 16, end view.
 Figs. 17, 18. *St. jaculiferum*, West. $\times 510$. Fig. 17, front view; Fig. 18, end view.
 Figs. 19, 20. *St. anatinum*, Cook and Wills. $\times 510$. Fig. 19, front view; Fig. 20, transverse section.

Figs. 21–3. *St. Sebaldi*, Reinsch, var. *altum*, (Boldt) West. $\times 510$, showing variation in number, size, and position of the pyrenoids. Fig. 21, front view; Fig. 22, oblique end view; Fig. 23, from the end.

Figs. 24, 25. *St. Sebaldi*, Reinsch, var. *ornatum*, Nordst. $\times 510$. Fig. 24, oblique front view; Fig. 25, end view.

Figs. 26, 27. *St. Ophiura*, Lund. $\times 510$. Fig. 26, front view; Fig. 27, transverse section.

Figs. 28, 29. *St. Arciscon*, (Ehrenb.) Lund. $\times 510$. Fig. 28, front view; Fig. 29, oblique end view. In both cases the letters *a a'*, *b b'*, *c*, &c., show the relation between the processes of the cell-wall.

Figs. 30–2. *St. paradoxum*, Meyen, var. *longipes*, Nordst. $\times 510$. Fig. 30, front view; Figs. 31 and 32, end view.

Fig. 33. *St. aculeatum*, (Ehrenb.) Menegh. $\times 510$. End view.

Figs. 34, 35. *St. Manfeldtii*, Delp. $\times 510$. Fig. 34, front view; Fig. 35, end view.

Figs. 36–8. *St. pyramidatum*, West. $\times 510$. Fig. 36, front view; Figs. 37–38, end view.

PLATE XV.

Figs. 39–42. *St. grande*, Bulnh. $\times 510$. Fig. 39, oblique front view of specimen having several chloroplasts; Fig. 40, end view of a similar individual; Fig. 41, front view of individual having one chloroplast only; Fig. 42, end view of similar specimen.

Figs. 43, 44. *St. sexangulare*, (Bulnh.) Lund. $\times 510$. Fig. 43, front view; Fig. 44, end view.

Figs. 45, 46. *St. Brasiliense*, Nordst., var. *Lundellii*, West. $\times 510$. Fig. 45, front view; Fig. 46, transverse section.

Fig. 47, 48. *St. tumidum*, Bréb., front view. $\times 510$.

Fig. 49. *Staurostrum aversum*, Lund. $\times 510$. Showing the more rapid budding of the peripheral parts of the chloroplast into the young semi-cell as compared with that of the axis itself. The longitudinal stretching of the pyreno-crystal preparatory to its constriction to form two is also to be seen.

Fig. 50. *Cylindrocystis crassa*, de Bary. $\times 915$. The pyrenoid of each chloroplast and the nucleus of the cell have already divided, whilst in the peripheral part of each chloroplast a transverse cleft has appeared into which a daughter nucleus is pressing its way, and the transverse wall is beginning to form. A number of proteid granules or small pyrenoids can also be seen in the peripheral parts of each chloroplast.

Figs. 51, 52. *Netrium oblongum*, (de Bary) Lütkeim., var. *cylindricum*, W. and G. S.

West. $\times 510$. Fig. 51 shows the division of the pyrenoid and constriction of the chloroplast preparatory to cell-division; Fig. 52 shows the completed division.

Fig. 53. *Netrium Digitus*, (Ehrenb.) Itzigs and Rothe. $\times 510$. A stage in cell-division intermediate between Figs. 51 and 52, showing the travelling of the two daughter nuclei towards the clefts between the newly divided chloroplasts.

Fig. 54. *Euastrum bidentatum*, Näg. $\times 510$. Showing division of the pyrenoid during cell-division.

Figs. 55, 56. *Staurastrum Brasiliense*, Nordst., var. *Lundellii*, W. and G. S. West. $\times 510$. Fig. 55, very young semi-cell seen from the end, showing that the ingrowing chloroplast even at this early stage preserves its five-lobed structure, although the pentangular shape of the cell-wall itself is scarcely to be discerned; Fig. 56, front view of a much later stage, showing the entrance of the pyrenoids from the old semi-cell, and the nucleus displaced to one side.

Fig. 57. *Cosmarium Brebissonii*, Menegh. $\times 510$. Side view of dividing specimen, showing the ingrowth of the chloroplast and pyrenoids, and the nucleus pushed to one side.

Fig. 58. *Staurastrum anatinum*, Cook and Wills. $\times 510$. Front view of dividing individual, showing the division of the pyrenoids and the rapid growth of the more peripheral parts of the chloroplasts as compared with that of the more central part.

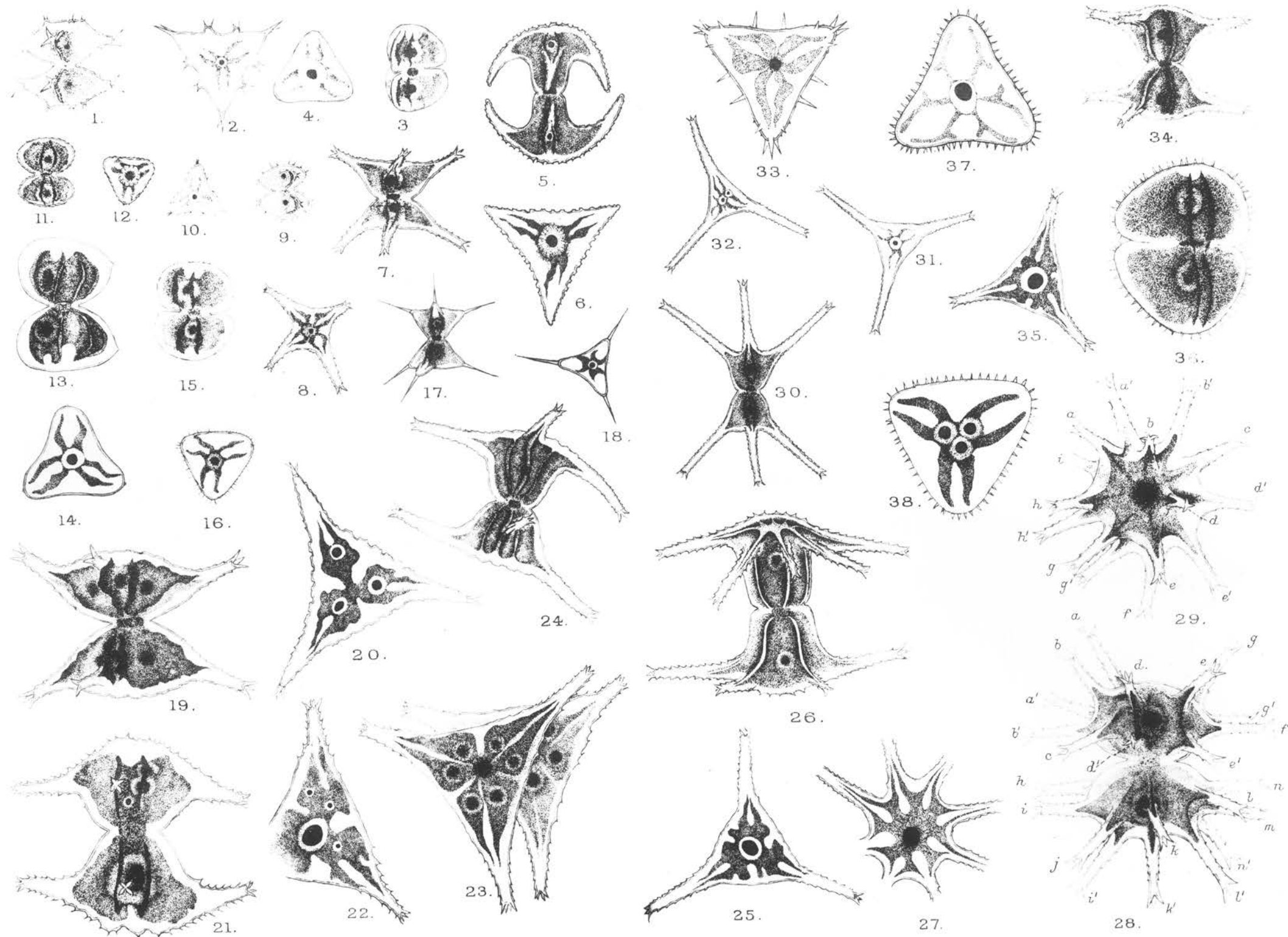
Fig. 59. *Xanthidium armatum*, (Bréb.) Rabenh. $\times 510$. Front view of a very early division stage, showing the beginning of the budding into the young semi-cell of the chloroplast together with its pyrenoids.

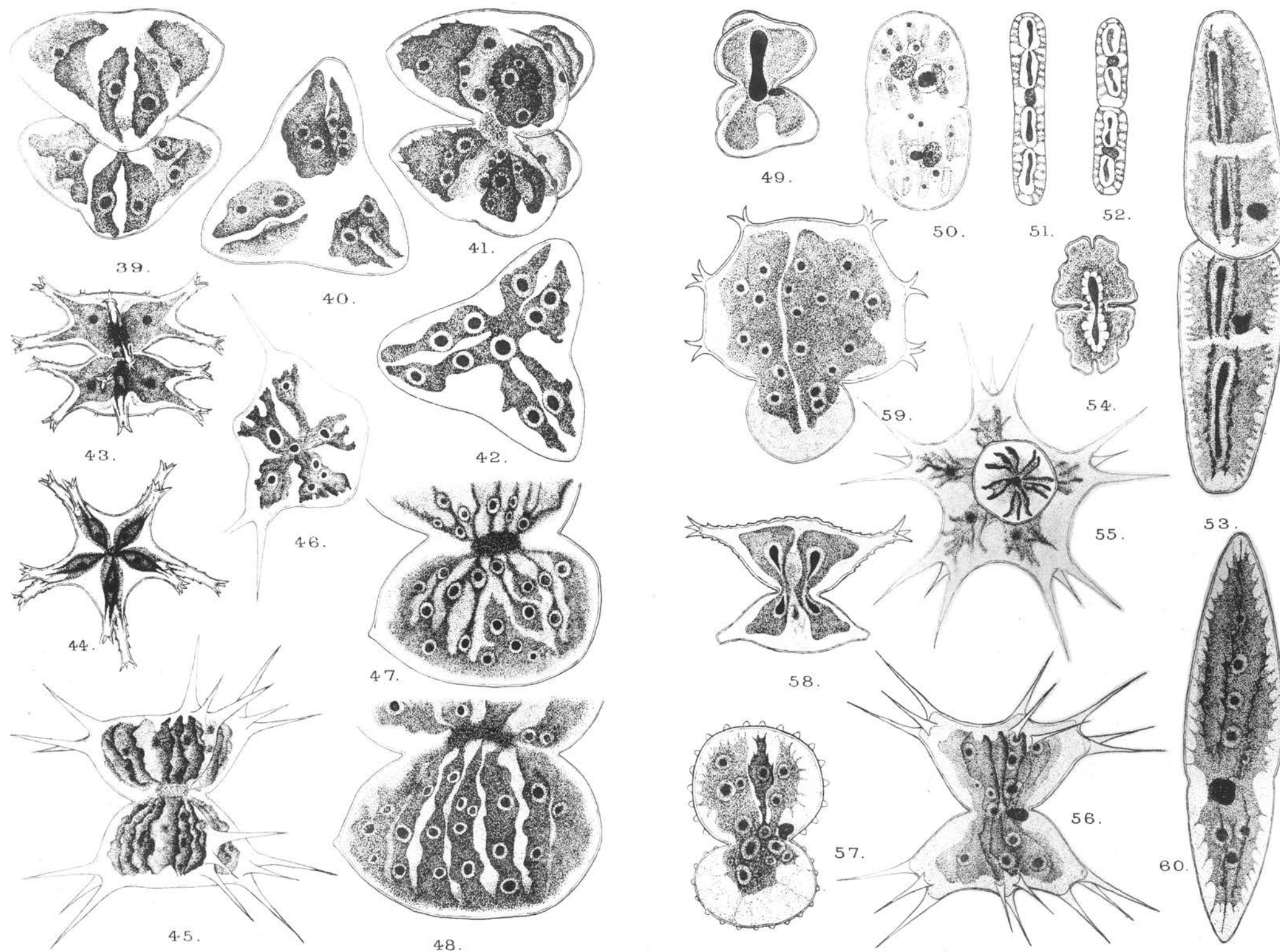
Fig. 60. *Tetmemorus granulatus*, (Bréb.) Ralfs. $\times 510$. Side view of a recently divided specimen in which the chloroplast has entered the young semi-cell, but division at the sinus has not yet taken place.

PLATE XVI.

Figs. 61-70. *Eu. Didelta*, (Turp.) Ralfs. $\times 350$. Process of cell-division followed in a single individual. Fig. 61, July 25th, 11.15 a.m.; Fig. 62, 11.30 a.m.; Fig. 63, 11.40 a.m.; Fig. 64, 12 noon; Fig. 65, 12.30 p.m.; Fig. 66, 2 p.m.; Fig. 67, 5 p.m.; Fig. 68, July 26th, 10.30 a.m.; Fig. 69, 11.30 a.m.; Fig. 70, July 27th, 10.30 a.m.

Figs. 71-4. *Cosmarium subtumidum*, Nordst. $\times 1,750$. Behaviour of the pyrenoid during cell-division. Fig. 71, 3.20 p.m.; Fig. 72, 3.25 p.m.; Fig. 73, 3.30 p.m.; Fig. 74, 3.37 p.m.

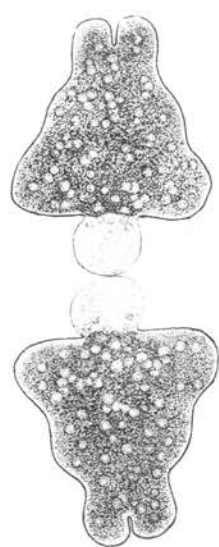




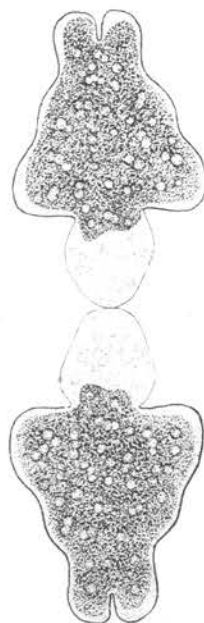
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CARTER-CHLOROPLASTS OF DESMIDS.



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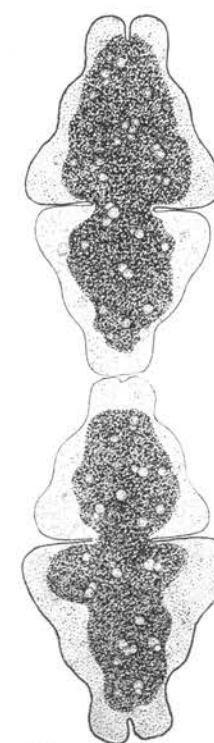
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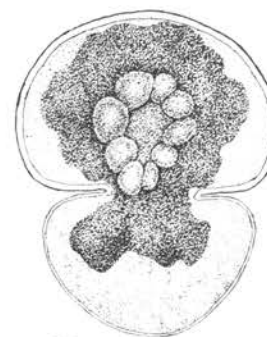
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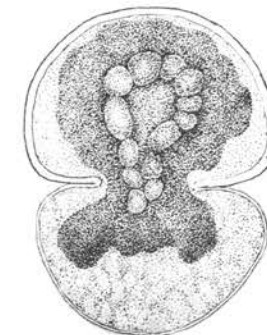
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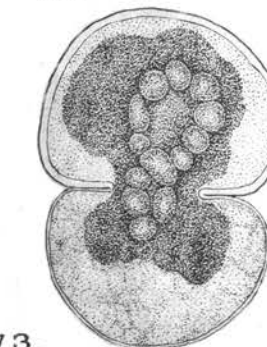
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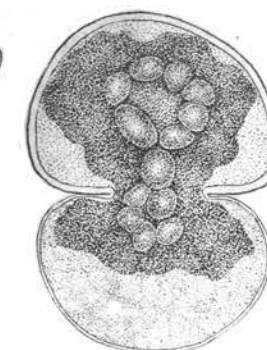
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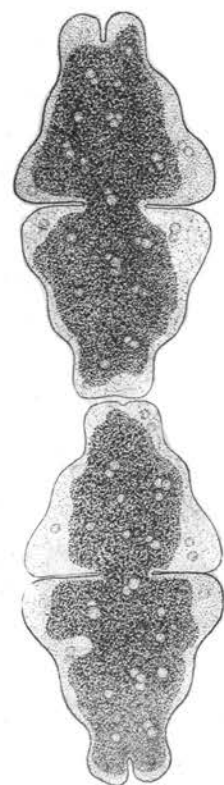
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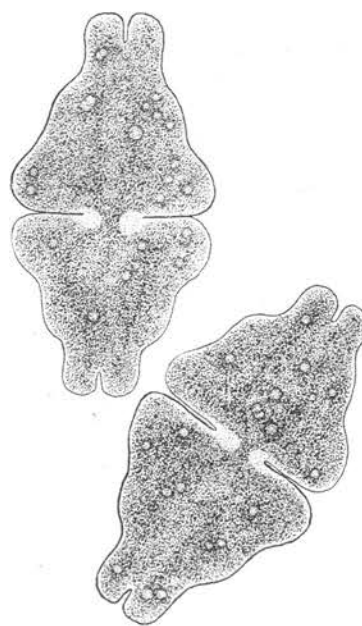
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