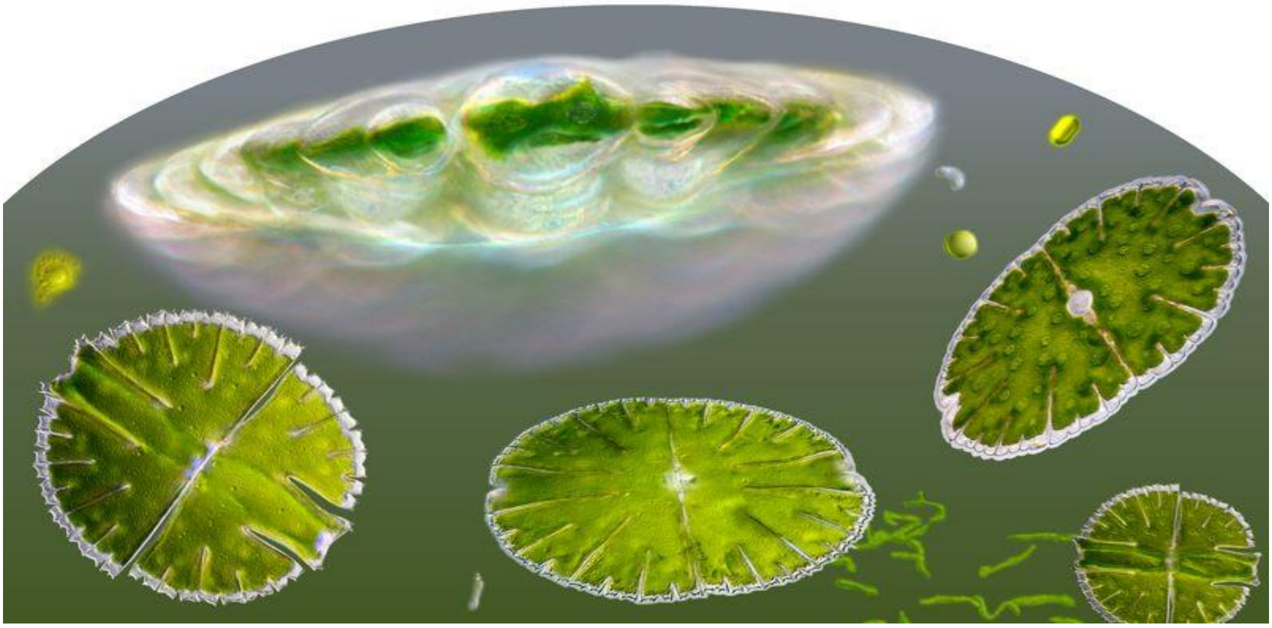

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Micrasterias – Little Stars

Part 1: Taxonomy, mitochondria

The species of the desmid genus *Micrasterias* are well known for their beauty. Their gracefully built cells in symmetry inspire the microscopist again and again anew, and their cell size facilitates the observation even with simpler microscopical equipment. *Micrasterias* means "little star".

The genus occurs worldwide and covers approx. 40 species. They populate oligotrophe, standing water bodies up to acidic and very nutrient-poor biotopes like bogs. These representatives of the unicellular, unflagellated green algae from the group of desmids (Zygnematophyceae/Streptophyta) are built up of large, flattened cells, whose lateral view is fusiform. A central constriction called "sinus" separates them into two halves. The nucleus is situated at the so-called "isthmus", the narrow junction zone of the two half-cells. Every half-cell owns its chloroplast with pyrenoids. Symmetrical cuts within the half-cells shape lobes, so the cells appear as small stars. Some species wear prickles on the cell surface.

After collecting the desmids, especially from a bog, they are much inured to treatment. It is simple to keep alive these algae in the collecting container without addition of nutrients for a longer period. It is important that the collection container is not exposed to the sun.

Classification

The taxonomic group called desmids is intensively investigated, maybe due to the attractiveness of its members. They occur in many biotopes around the globe and prefer nutrient-poor habitats. There are species, which occur in arctic regions (Lenzenweger and Lütz, 2006; Lenzenweger, 2007), others live in the tropics (Lenzenweger, 2003). Due to newest investigations from morphology, cytology and gene analyses the genus *Micrasterias* is still assigned to the group of the Desmidiales, which contains approx. 40 genera with overall 4,000–6,000 species. Within the superordinate group of the Streptophyta they are assembled in the clade Phragmoplastophyta (for details on the structure of the phragmoplast, see the next section, Fig. 1) within the Zygnematophyceae. The sister groups of them are the Charophyceae, the Choleochaetophyceae and the Embryophyta, which includes the higher plants (Adl et al., 2019).

The taxonomists refer to the term “clade”, if they assume that all group members descend from a common ancestor. An important distinguishing feature of all Streptophyta are plasmodesmata. During build-up of the separating structure in mitosis called phragmoplast, it is ensured that cell wall openings remain between the neighboring cells as communication channels, the plasmodesmata (see the next section, Fig. 2, for details).

The group of Phragmoplastophyta unites all genera of higher plants with the Zygnematophyceae, together with the Charophyceae and the Choleochaetophyceae. The paleobotanists assume that the higher plants developed approx. 450 million years ago on the basis of a common ancestor with the Zygnematophyceae. Therefore, the Desmidiales are classified as rather close relatives of all higher plants (Puttick et al., 2018).

Cytological details of the phragmoplast and plasmodesmata

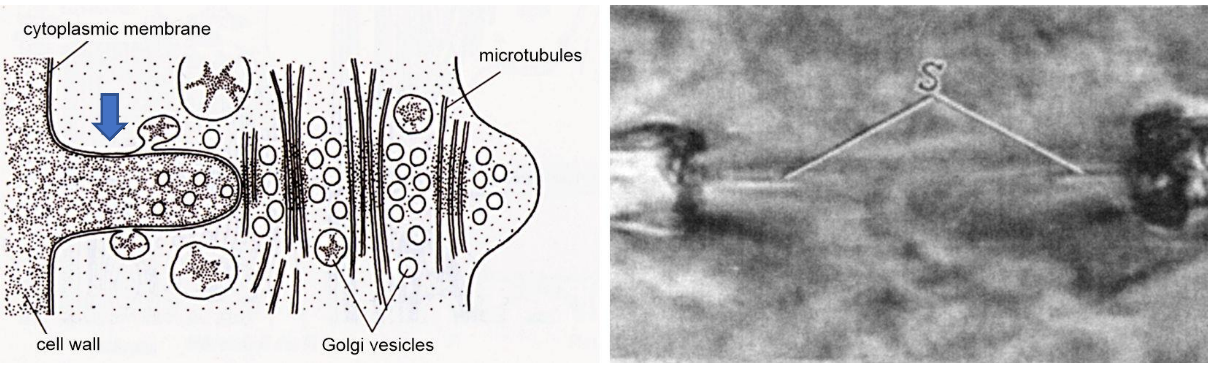


Fig. 1: The phragmoplast

a: Schematic drawing of a developing phragmoplast (arrow) in *Spirogyra* (genus in the Zygnematophyceae), based on transmission electron microscopy. In all Phragmoplastophyta species, the transverse wall (the phragmoplast) is formed in a similar manner following mitotic cell division. “Phragm” derives from ancient Greek and means lock, fence or wall. After the division of the cell nucleus, a new microtubule network forms in the middle of the cell, which is arranged parallel to the former spindle apparatus of the nuclear division. It guides the vesicles with wall substances formed by the Golgi apparatus to the site of the construction of the new transverse wall. The entirety of all dictyosomes in a cell is called the Golgi apparatus. Among other things, basic substances (pectins) for building cell walls are produced in the dictyosomes. Membrane-enclosed packages, the Golgi vesicles, are pinched off at the edge of the dictyosomes. Microtubules are fine (25 nm thick and up to 100 µm long) scaffolding tubes whose structure is the same in all eukaryotic cells and perform the support functions in the cytoplasm, but also represent guide rails for intracellular vesicle transport (Kleinig and Maier, 1999).

After Lecointre and Le Guyader, 2006, modified.

b: Formation of a structure analogous to the phragmoplast, called septum (S), during cell division of *Micrasterias denticulata* (after Kiermayer, 1966).

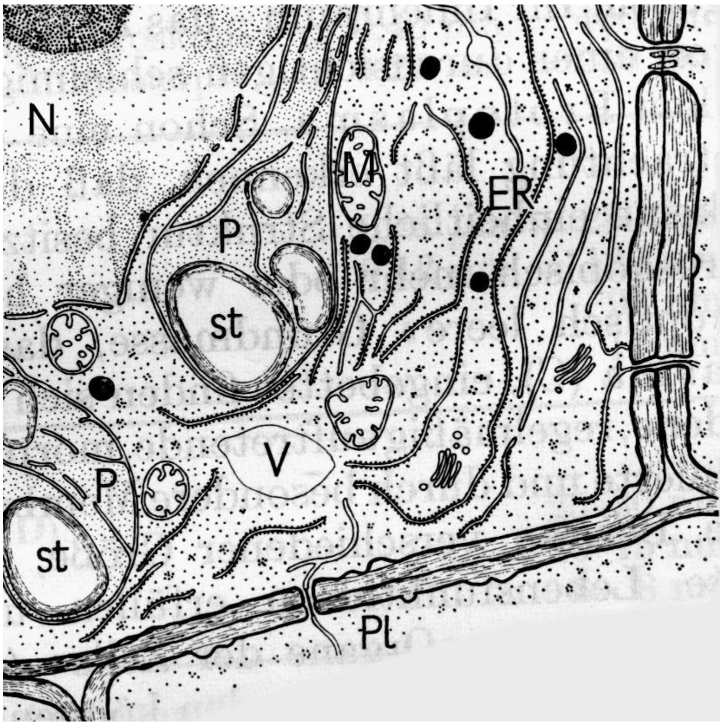


Fig. 2: The plasmodesmata

Schematic drawing of a young Streptophyta cell with developed plasmodesmata, based on transmission electron microscopy. In addition to cell organelles such as the cell nucleus (N), the plastids (P), vacuoles (V), mitochondria (M) and the endoplasmic reticulum (ER), the cell wall breakthroughs with the plasmodesmata (Pl) are shown. These openings allow the protoplasts of neighboring cells to connect by means of plasmodesmata (desmos from ancient Greek means connection, rope, shackle). Nutrients and messenger substances are exchanged via these plasma connections. If the cell walls are thicker, these cell wall breakthroughs can be seen in the light microscope as pit channels (after Strasburger, 1971, modified).

Light microscopy studies on *Micrasterias*

Micrasterias rotata is one of the most magnificent desmid stars concerning elegancy and also size (length 200–300 µm, width 190–270 µm). Since their cells are flat, it is not too difficult to take a photomicrograph exhibiting the outline of the cell together with the chloroplasts and the pyrenoids on them in focus.

Mitochondria

Looking into the microscope at higher magnification (oil immersion, aperture 1.0 and higher) with enhanced contrast (such as DIC), a whole set of structures is distinguishable in the cell plasma. Focusing slowly down, starting on the cell wall towards the chloroplast, scarcely underneath the cell wall an intense flow of tiny cytoplasm particles can be detected.

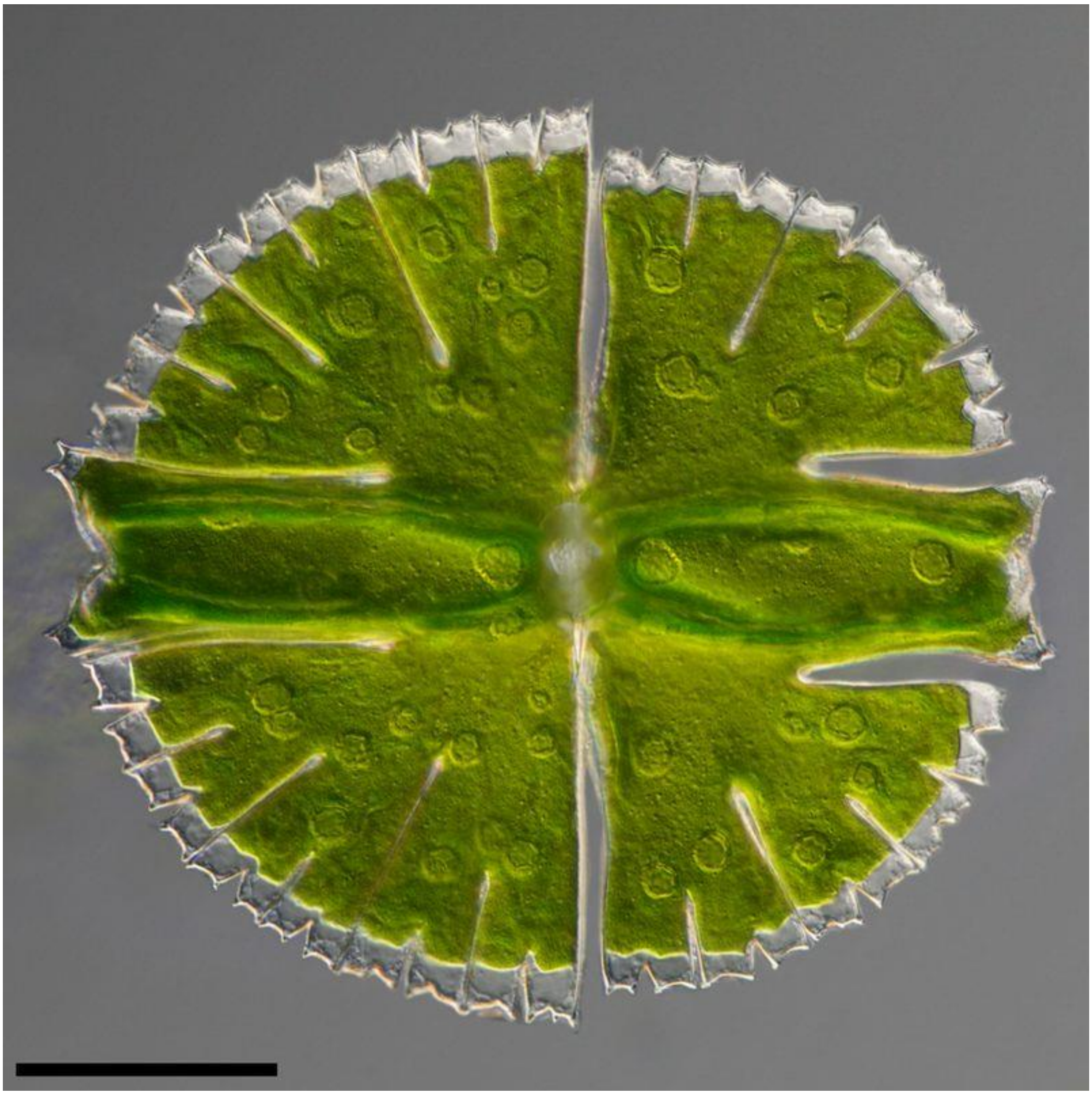


Fig. 3: *Micrasterias rotata*. Picture showing the outline together with the chloroplasts and the pyrenoids on them. Manually stacked using 8 micrograph frames (Zeiss Planapo 40/1.0, DIC). Scale bar indicates 50 µm.

During such an observation I had the ability to differentiate small rigid circular from oblong, form-variable components. The small circular ones with a diameter of app. 0.5 µm were swept away by the current of cytoplasm. On Figure 4 they are weakly recognizable as dimmed dots. The oblong parts however oscillated usually around a fixed point such as flags in the wind (see arrow heads on Fig. 4 and 5). Besides them again and again high-refractive spheres with diameters over 0.5 µm (probability oil droplet) were visible (arrows on Fig. 4).

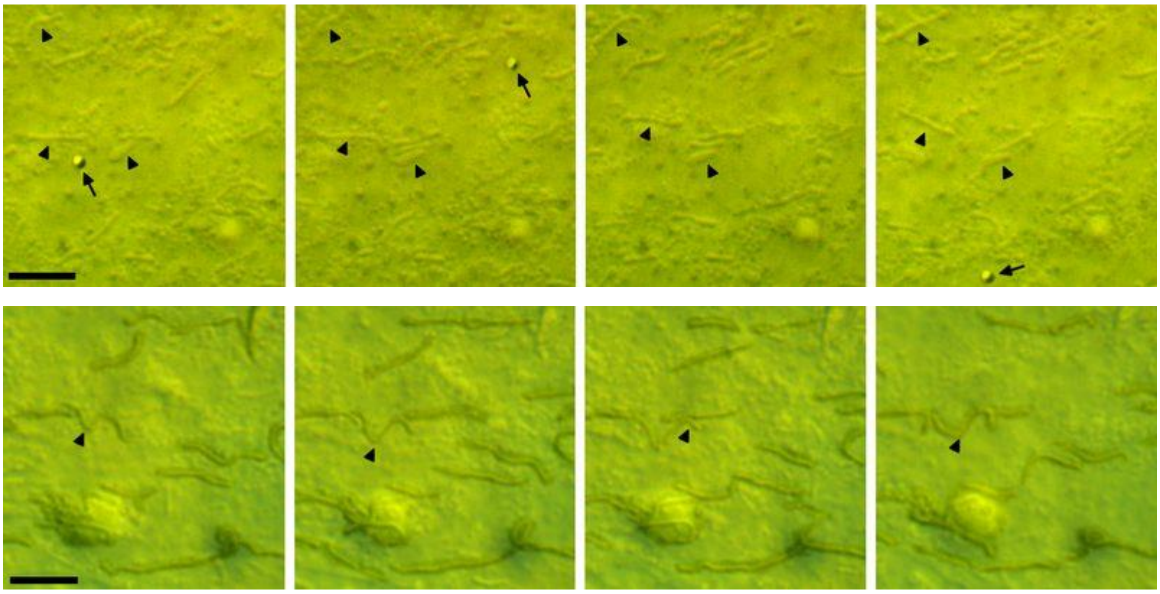


Fig. 4 and 5: Different particle types in the current of cytoplasm of *M. rotata*. The individual images of the two image series were recorded at 1-second intervals. Arrows = (probably) spherosomes, arrowheads = mitochondria. For reasons of clarity, only a few mitochondria's movement stages are marked. Scale bar indicates 5 µm.

By comparison with structures described by Drawert and Mix in their observations of *M. rotata* in 1961, I realized that the elongated particles were identical to mitochondria. In 1978 Bereiter-Hahn documented similar kinds of shaping and location with mitochondria on endothelium cells of tadpole hearts.

Studies on further *Micrasterias* species

After these discoveries with *M. rotata* I wanted to know whether similar observations would be possible also with other *Micrasterias* species with approximately the same size, for instance *M. denticulata* and *M. apiculata*. In much smaller species such as *M*. *truncata* the plasma stream is also visible. Using the light microscope, however, the particles in the plasma stream are too small for assigning them unequivocally to the above-mentioned groups.
Micrasterias denticulata (Fig. 6) often lives in similar habitats like *M. rotata*, this means acidic or moderate acidic waters of fens, swamp meadows, or pools in bog areas (Lenzenweger, 1996). This is also true for *M. apiculata,* but this species is rather rare.

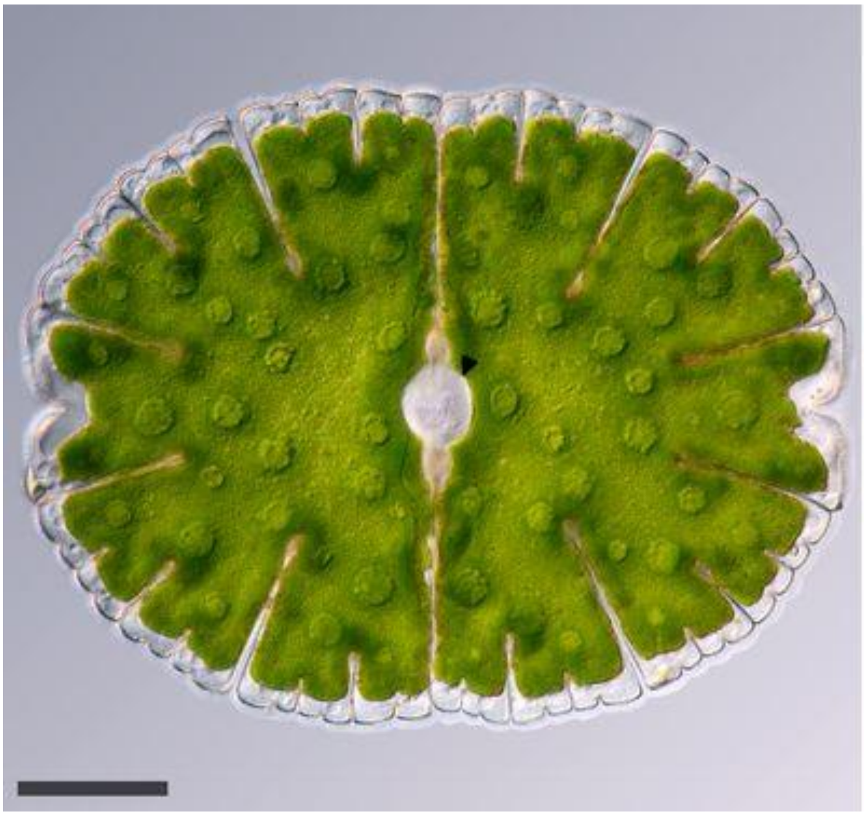
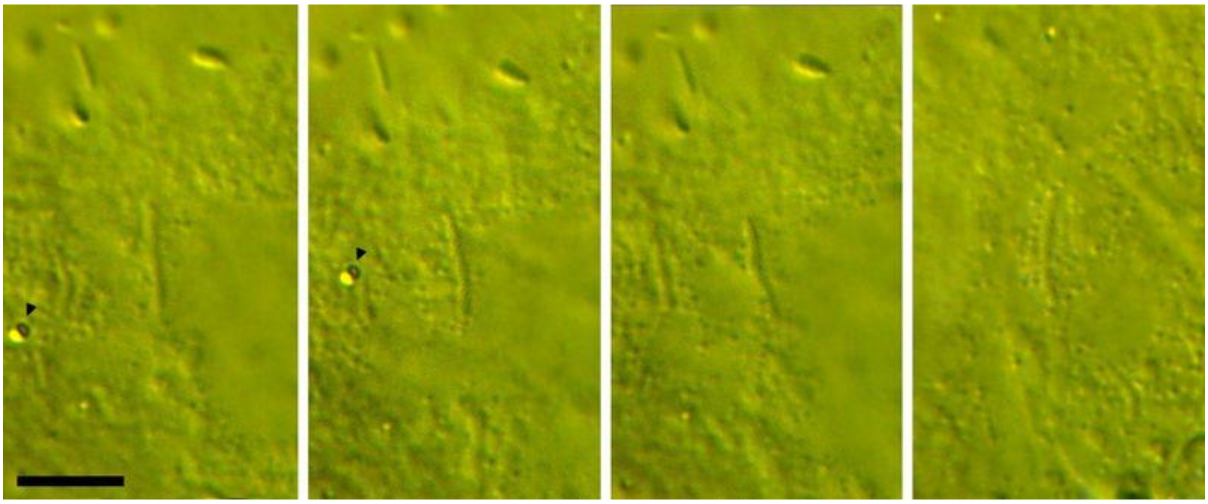


Fig. 6: *Micrasterias denticulata*, showing shape, chloroplasts with pyrenoids plus the central nucleus. Image composition from 20 individual frames, stacked manually. Scale bar indicates 50 µm.

In *M. apiculata* (Fig. 7) and *M. denticulata* (Fig. 8) just like in *M. rotata*, the mitochondria were permanent in oscillating motion. Whereas in *M. apiculata* the appearance of the mitochondria (size and place for observation) was very similar to those of *M. rotata*, the ones at *M. denticulata* were more voluminous but their number were substantially smaller and they were situated more deeply in the cell, in the proximity of the chloroplast.



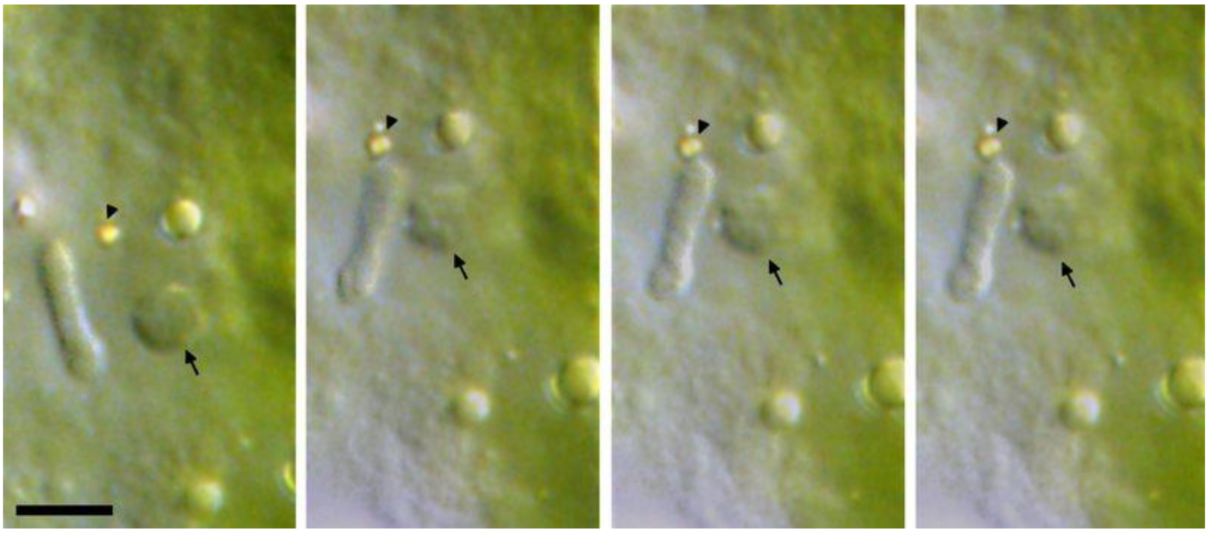


Fig. 7: A series of shots of *M. apiculata* showing a mitochondrion in oscillation together with a spherosome (arrowhead). Fig. 8: A series of shots of *M. denticulata* showing a mitochondrion in oscillation together with a dictyosome (arrow) and a spherosome (arrowhead). Scale bar indicates 5 µm.

According to the widely accepted serial endosymbiosis theory mitochondria originated from aerobic prokaryotes (bacteria). In the course of co-evolution within the eukaryotic cells the cell wall of the former bacteria was lost (Sagan, 1967; Bell, 2001; Plattner und Hentschel, 2006). The mitochondria still have their own (reduced) genome. They are mostly oblong organelles with dimensions of 0.2–1 µm by 2–8 µm, which corresponds to the size of most bacteria, and they divide independently. They are often variable in shape and are rapidly moved by the cytoplasmic current. The number of mitochondria per cell can vary significantly from cell type to cell type. Some oocytes contain more than 100,000 mitochondria, the trypanosomes (flagellates; one species is the causative agent of sleeping sickness) have only one, which runs through almost the entire cell body and occupies a very considerable volume.

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