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Serial Reconstruction of the Mitochondrial Reticulum in the Antarctic Flagellate, *Pyramimonas gelidicola* (*Prasinophyceae, Chlorophyta*)

Brief Report

G. I. McFadden* and R. Wetherbee

School of Botany, University of Melbourne

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Summary

A serial reconstruction of *Pyramimonas gelidicola* McFadden, Moestrup and Wetherbee has revealed a large reticulated mitochondrion branching throughout the cell. The possibility of single mitochondria in other members of the *Prasinophyceae* and the uniformity of the morphology of this organelle within the class is discussed.

Keywords: Prasinophyceae; Pyramimonas; Mitochondrion.

1. Introduction

Unicells from the green algal class *Prasinophyceae* range in size from the minute ultraplankton, which measure only 1.0 μm in length (*e.g.*, *Micromonas pusilla*, Manton 1959) up to the larger species from the genus *Pyramimonas* which can measure over 25 μm in length (Pennick 1978, Norris and Pienaar 1978). Fine structural studies on the smaller prasinophyte genera, including *Micromonas*, *Mantoniella*, *Mesostigma*, *Monomastix*, *Pedinomonas* and *Pseudoscourfieldia* (Norris 1980) have revealed the presence of a single, unbranched mitochondrion in each cell. While such observations are easily made on the smaller organisms, electron micrographs through larger cells often show numerous mitochondrial profiles, which investigators have interpreted as representing several mitochondria. The structure of the mitochondrion has been carefully investigated in only a few green algae, and in some organisms (*e.g.*, *Chlorella*, *Chlamydomonas* and the male gamete of *Bryopsis*) has been shown to be a single reticulated structure during at least part of the cell cycle (Atkinson *et al.* 1974, Arnold *et al.* 1972, Osafune *et al.* 1972, Burton and Moore 1974, Dempsey *et al.* 1980). This report describes the serial reconstruction of the mitochondrion of *Pyramimonas gelidicola*, one of the larger members of the *Prasinophyceae*.

* Correspondence and Reprints: School of Botany, University of Melbourne, Parkville, Victoria 3052, Australia.

2. Materials and Methods

Cultures of *Pyramimonas gelidicola* were grown, fixed and prepared for electron microscopy as described by McFadden *et al.* (1982). Approximately 150 serial sections were collected on formvar-coated slot grids. Three cells were reconstructed, two transversely sectioned and one longitudinally. Micrographs were projected onto styrofoam sheets so that the lateral magnification matched the ratio between the estimated section thickness and the thickness of the styrofoam. Outlines of mitochondrion, chloroplast and nucleus were traced and the shapes cut out by a heated wire and assembled.

3. Results

The fine structural features which characterize *Pyramimonas gelidicola* have been previously described (McFadden *et al.* 1981). Cells are up to 18 μm in length and 9 μm across and are conical in shape, with four flagellae emergent from an apical pit. The chloroplast is located

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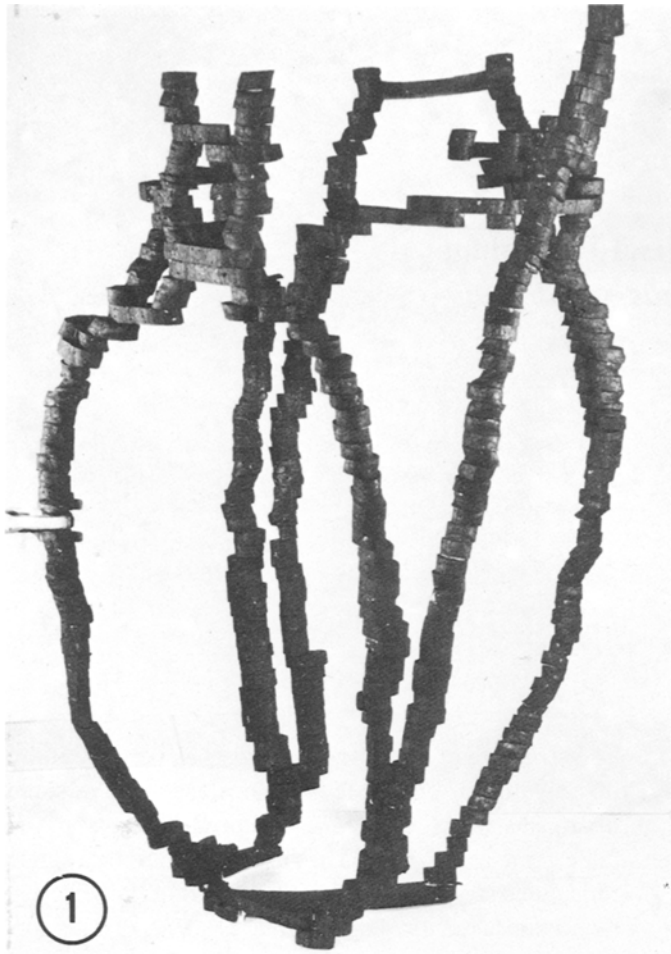

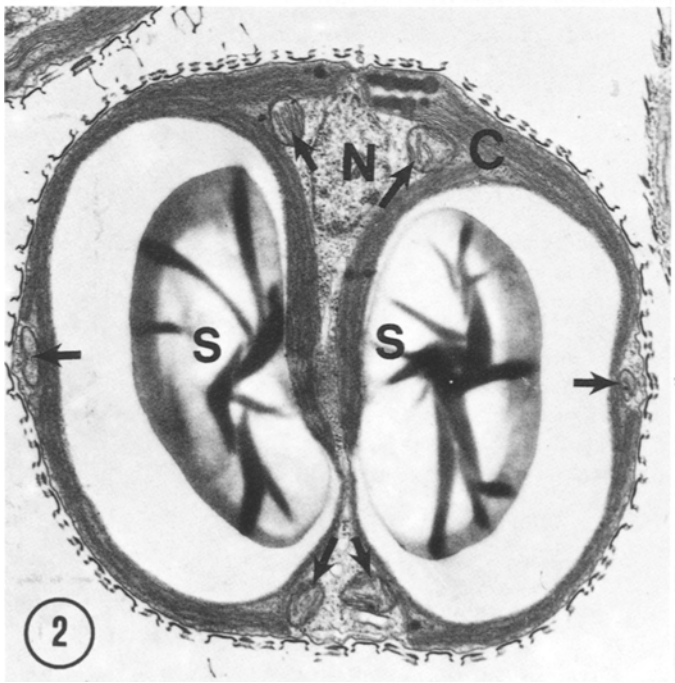
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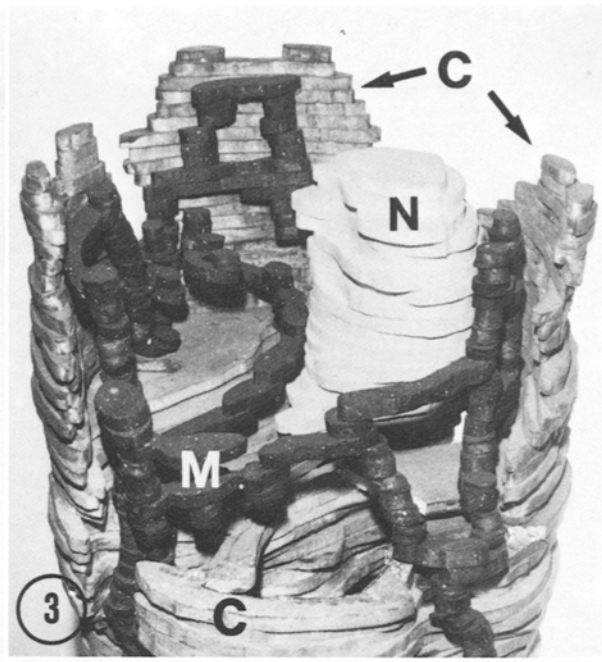
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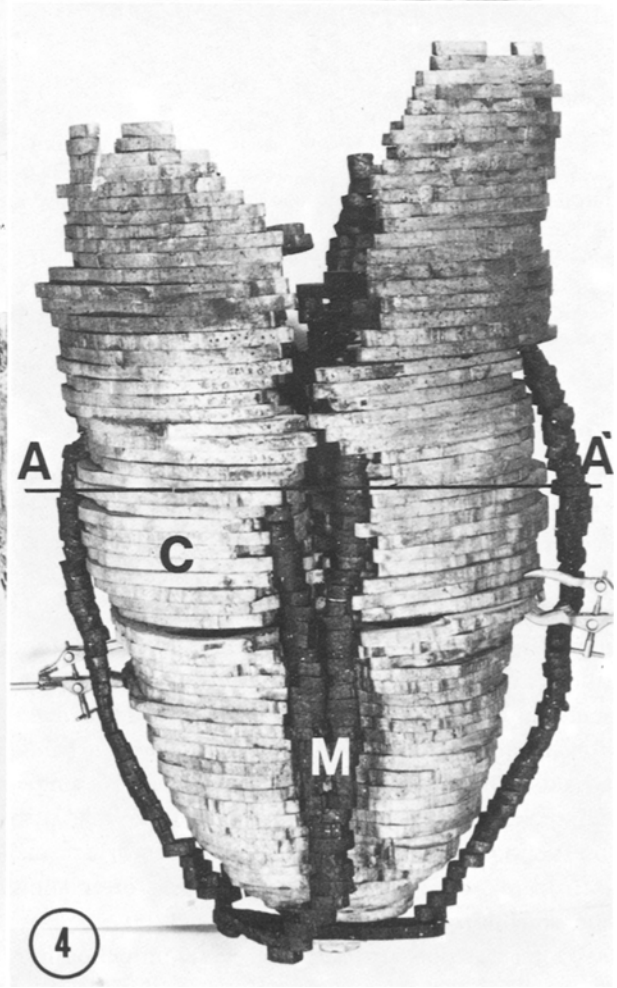
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directly beneath the plasmalemma, is cupshaped at the posterior end but divides into two, four and ultimately eight lobes towards the anterior end.

In the three cells examined, two in interphase and one in prophase, all proved to have a large, tubular, reticulated mitochondrion (Fig. 1). The mitochondrion is closely associated with the chloroplast in most regions of the cell, being so closely appressed that the mitochondrial and chloroplast envelopes almost touch (Figs. 2 and 3). In the posterior region of the cell, branches of the mitochondrion lie between the chloroplast and the plasmalemma (Fig. 4). Where the chloroplast divides into lobes, the mitochondrial branches pass through to the inner (cytoplasmic) side of the chloroplast. Further branches traverse the cell in the region of the basal bodies and fuse with opposite mitochondrial branches on the other side of the cell (Fig. 3). Lobes of the mitochondrion therefore appear closely associated with all the major organelles within the cell. The mitochondrial branches range in diameter from approximately 300-600 nm; the cristae are flattened and project straight into the lumen.

The nucleus is teardrop-shaped and is positioned in the junction of two chloroplast lobes (Fig. 3). There is a projection of the nucleus toward the basal bodies on one side and another projection between the two chloroplast lobes containing the eyespots on the other (Fig. 2).

4. Discussion

The extensive reticulation of the *Pyramimonas* mitochondrion and the relatively high surface to volume ratio maintained by the small diameter of the branches would appear to be both physiologically effective and even necessary for a large and complex cell, despite the possible restrictions imposed by a single mitochondrion. The unitary nature of the mitochondrion described here for *P. gelidicola* is similar to other reports on a number of flagellates and unicellular organisms of diverse affinities (for review see SANTORE and GREENWOOD 1977). A mitochondrion may be either simple or variously branched, but this feature is not necessarily related to the size of the cell. For example, most smaller cells would be expected to contain a simple, unbranched structure, though the mitochondrion of the small green alga *Chlorella minutissima*, which measures only 1.8-3.0 μm in size, is highly branched (DEMPSEY *et al.* 1980).

Considering the variety of smaller prasinophytes with a single mitochondrion and our observations on the highly branched mitochondrion in *Pyramimonas gelidicola*, it seems likely that a single and sometimes reticulated mitochondrion is present in most, if not all, prasinophytes. Mitochondrial profiles adjacent to the chloroplast and in somewhat similar positions as we have described in *P. gelidicola* are also observed in other larger prasinophytes, including *Nephroselmis olivacea* (MATTOX and STEWART 1977, MOESTRUP and ETTL 1979), *P. aff. plurioculata* (NORRIS and PIENAAR 1977) and *P. orientalis* (MOESTRUP and THOMSEN 1974), and we suggest that a unitary, branched mitochondrion may be a regular feature in these organisms.

It is not possible at present to predict whether criteria based on the number and structure of the mitochondrion will be of importance to systematics or phylogeny. In *Bryopsis*, for example, the male gamete has a single mitochondrion while the female gamete has several (BURR and WEST 1970). SANTORE and GREENWOOD (1977) observed a great uniformity in mitochondrial structure within the members of the *Cryptophyceae*, and it is possible that this feature will be of significance in distinguishing some classes of algae, including the *Prasinophyceae*.

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Fig. 1. Serial reconstruction of the mitochondrial reticulum in *Pyramimonas gelidicola*. ×7,600

Fig. 2. Electron micrograph of a transverse section through *Pyramimonas gelidicola* corresponding to *A A'* in Fig. 4. Two lobes of the chloroplast (*C*), starch grain (*S*) and the nucleus (*N*) can be seen. Arrows indicate the six mitochondrial profiles encountered at this level. Note the proximity of mitochondrial profiles to the chloroplast. ×8,400

Fig. 3. A view of the anterior region of the reconstructed cell with one of the four chloroplast lobes removed. The positions of the nucleus (*N*) and mitochondrial branches (*M*) traversing the cell in the basal body region are shown. ×7,700

Fig. 4. Lateral view of the reconstructed chloroplast with associated mitochondrial branches (somewhat displaced from the chloroplast because of reconstruction difficulties). A transverse section representing *A A'* is shown in Fig. 2. ×6,700

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