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### **Title**

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GRANA-LIKE STRUCTURES OF SYNECHOCOCCUS CEDORUM

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Berkeley, California

GRANA-LIKE STRUCTURES OF SYNECHOCOCCUS CEDORUM

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During photosynthetic studies in this laboratory, the desire for an organism possessing a simple internal structure prompted us to investigate the blue-green algae. These organisms have occasionally been reported to contain grana or chloroplasts, but the most widely accepted opinion is that all of their pigments are uniformly distributed throughout the cytoplasm.<sup>1</sup>

A pure culture of a unicellular blue-green alga, Synechococcus cedorum, was grown in an inorganic medium and one-day old cells were used to investigate the pigment distribution.

After the cells were harvested by centrifuging and washed with water, they were broken by grinding with alumina.<sup>2</sup> The mixture was diluted with water and centrifuged at 2,000 g. for 10 minutes to remove unbroken cells, cell debris and alumina. A blue-green supernatant having a strong Tyndall effect and slight fluorescence was obtained. This supernatant was then centrifuged in a refrigerated Spinco ultracentrifuge for 30 minutes at 36,000 g. A clear blue supernatant above a very minute green sediment was obtained. The absorption spectra of the intact cells and of the several

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fractions were determined from 3,500 Å to 7,500 Å with a Cary Recording Spectrophotometer. The curves of the aqueous solutions or suspensions are shown in Figure 1.

The suspension of the green sediment (Figure 1, Curve D) gives an absorption curve for chlorophyll and carotenoids and the clear supernatant (Figure 1, Curve C) shows the absorption spectrum for phycocyanin with no chlorophyll. Centrifugation at 36,000 g. for 20 minutes gives a complete separation of the two groups of pigments, chlorophyll and carotenoids associated with particles and phycocyanin in solution. Breaking of the cells by either ultrasonics or nitrous oxide treatment<sup>3</sup> gave the same results as obtained by alumina grinding.

Electron micrographs of the whole cells of Synechococcus cedorum and of the green sediment (Figure 1, Curve D) obtained after breaking the cells by ultrasonics and ultracentrifuging are shown in Figure 2. The dark areas of the whole cells are believed to correspond to the particles obtained by ultracentrifuging. The polystyrene standards have a diameter of 2,600 Å and from this the diameter of the particles in the whole cells appeared to be approximately 2,200 Å. Therefore, these particles are similar in size to the grana of chloroplasts.

These results show that all of the chlorophyll and carotenoids of Synechococcus cedorum are associated with particles that will centrifuge out at 36,000 g. and that the phycocyanin is in solution. On the basis of size and chlorophyll content, these particles of Synechococcus cedorum might be considered analogous to the grana of other green plants. There is also experimental evidence to indicate that the pigments of the photosynthetic bacteria, Chlorobium<sup>4</sup> and Rhodospirillum,<sup>5</sup> are also aggregated in particles.

We wish to thank Dr. Marybelle Allen for the pure culture of Synechococcus cedorum and Dr. Robley Williams for taking the electron micrographs.

#### References

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Captions to Figures

Figure 1 -- Absorption spectra of Synechococcus cedorum

Curve A - Whole cells.

Curve B - Supernatant (2,000 g.) from alumina-treated cells.

Curve C - Supernatant (36,000 g.) after ultracentrifugation.

Curve D - Resuspended particles obtained from ultracentrifugation.

Figure 2 -- Electron micrographs of Synechococcus cedorum

(a) Whole cells (the polystyrene standard is shown in the lower right corner).

(b) Particles from broken cells sedimenting at 36,000 g. Corresponding to Curve D of Figure 1. (The completely opaque black circles (2) are the polystyrene standards - diameter 2,600  $\mu$ .)

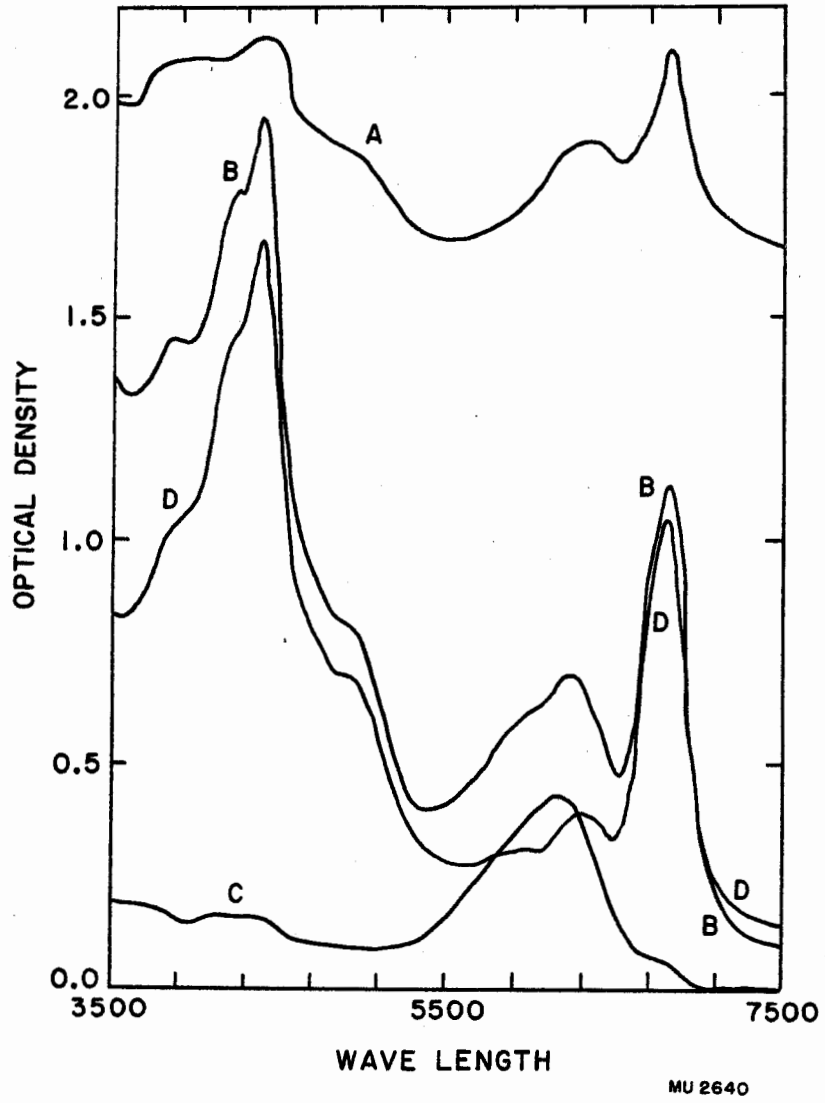


Fig. 1.



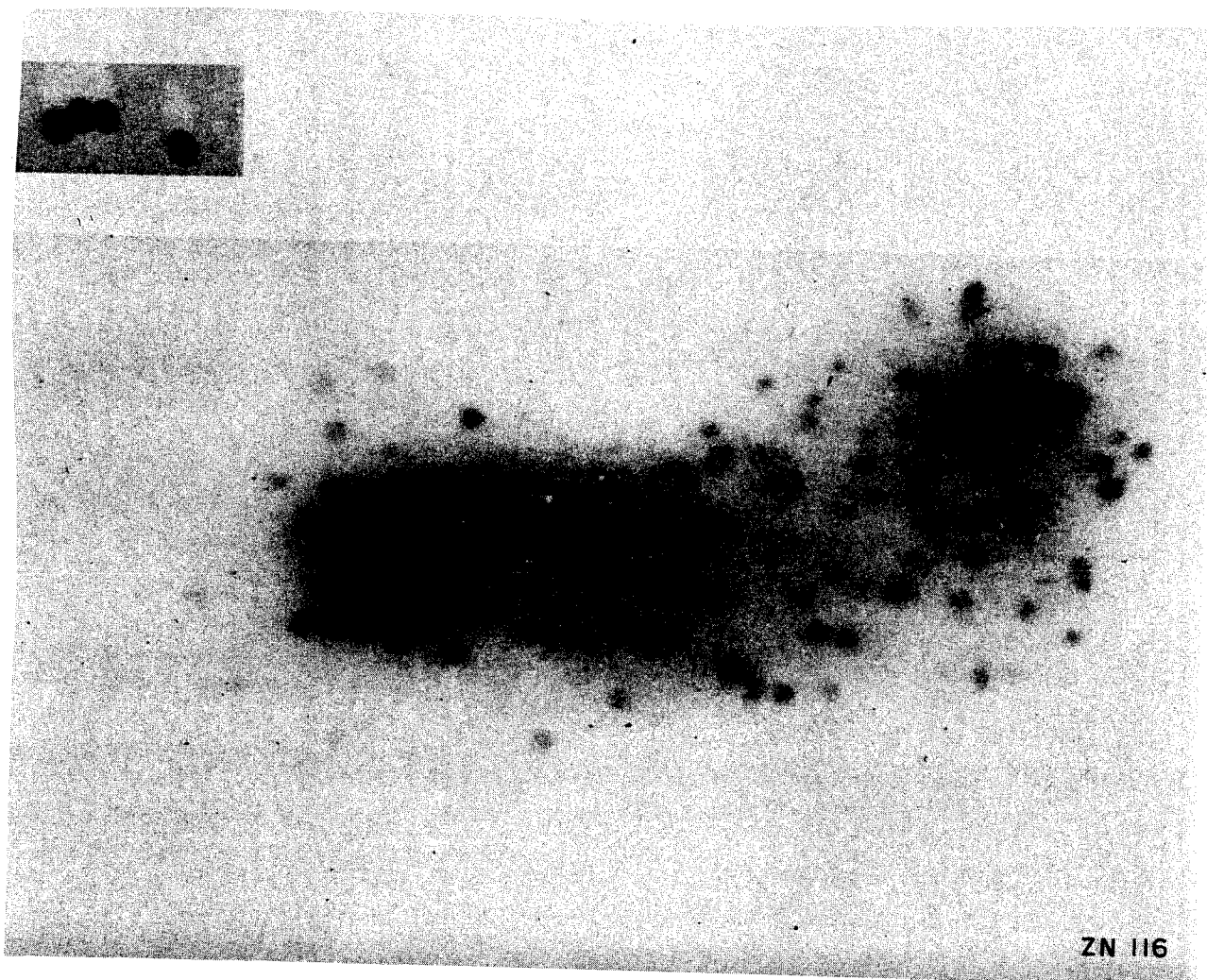


Fig. 2 (a)

