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Three-Dimensional Arrangement of Thylakoid Membranes in *Cyanothece* sp. ATCC 51142, a Unicellular Cyanobacterium

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In cyanobacteria and the chloroplasts of plants and algae, thylakoids are the complex internal membrane system where the light reactions of oxygenic photosynthesis occur. An early cyanobacterium, or cyanobacterial ancestor, was almost certainly the evolutionary precursor of chloroplasts via an endosymbiotic event. Despite this common ancestry, over evolutionary time the thylakoid membrane systems in photosynthetic organisms have diverged into architecturally dissimilar forms, so that a link between thylakoid membrane organization in chloroplasts and that of the cyanobacterial ancestor has been absent. In particular, conspicuously lacking has been evidence of a 3-dimensional organization that could be considered ancestral or intermediate to the present-day arrangement of grana and stromal thylakoids in chloroplasts.

In higher plant chloroplasts, thylakoid membranes have a distinct architecture, forming an intricate network of stacks of flattened or appressed lamellae, the grana, connected by membranes that traverse the stroma matrix, the stroma thylakoids. In the helical model describing this system, multiple right-handed helixes of stroma thylakoids wind around the grana, so that together these membranes enclose a single lumenal space [1]. In contrast, the thylakoid membranes of cyanobacteria typically do not form grana stacks connected by stroma thylakoids, but rather are arranged as sheets of lamellae recently shown to be connected by channels or bridging membranes [2, 3].

We have combined an analysis in 3-D of thylakoid membrane organization in plant chloroplasts with examination of the thylakoid membranes of a cyanobacterium. We prepared samples for electron microscopy by high-pressure freezing followed by freeze substitution, and used electron tomography as a tool for viewing the three-dimensional membrane arrangement of both chloroplasts and cyanobacteria. We found that the thylakoid membranes in a unicellular cyanobacterium, *Cyanothece* sp. ATCC 51142, display a rudimentary helical architecture (Figure 1). Furthermore, large-scale reconstructions of *Cyanothece* cells show that the thylakoid membrane system in this cyanobacterium encloses a single lumen, similar to what has been shown in chloroplasts. Our data suggest that the beginnings of the granal and stromal thylakoid arrangement in chloroplasts were present in the cyanobacterial ancestor, and that helical membrane organization, a defining feature of chloroplast thylakoid membranes, apparently originated from the cyanobacterial ancestor and has been conserved in at least one modern-day cyanobacterial species.

References

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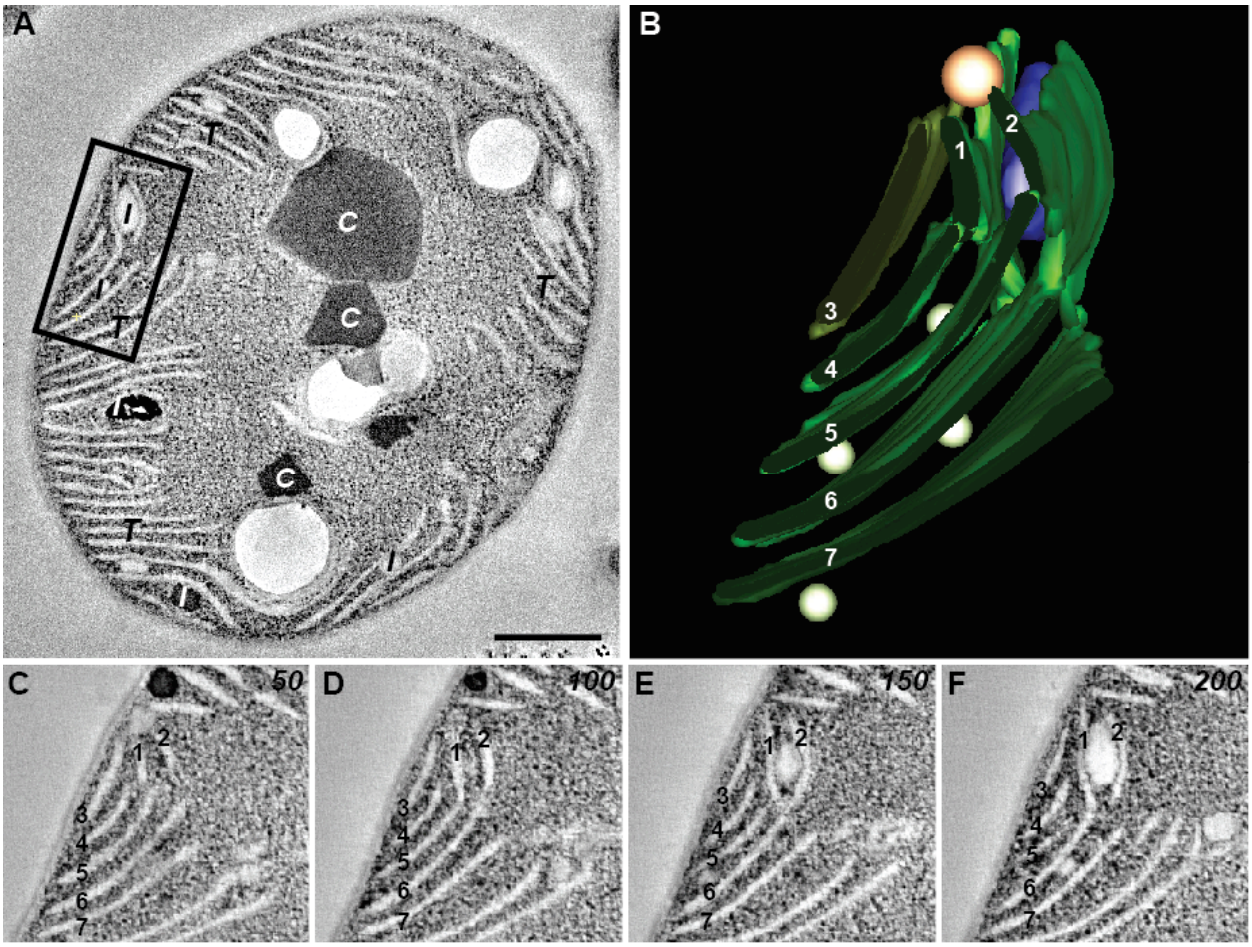

Figure 1. Thylakoid membrane arrangement in *Cyanothece* 51142. (A) Tomographic slice image (constructed from 5 superimposed serial 2 nm slices) of a *Cyanothece* cell. Thylakoid membranes (labeled *T*) radiate from the cell periphery into the cell interior. Some of the inclusion bodies regularly accumulated are labeled *I*, and carboxysomes are labeled *C*. Bar = 200 nm. (B) Model corresponding to the boxed area in (A), showing membrane arrangement and inclusion bodies. Regions of the thylakoid membrane system are labeled *1-7*. Membranes colored the same shade of green in the model enclose a single lumenal space, with the leftmost membrane (labeled *3*) not contiguous in the ~250 nm tomographic volume shown. (C-F) Individual 5 nm-thick slices through the tomogram at 50, 100, 150, and 200 slice intervals. The helical arrangement can be seen in the shift of the numbered membrane regions during the progression through the tomogram.

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