

# Transmission electron microscopic observations of *Chlorella*-free *Paramecium bursaria* infected with yeast cells

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## SUMMARY

Aposymbiotic (*Chlorella*-free) *Paramecium bursaria* was infected with yeast cells (*Pichia capsulata* and *Rhodotorula rubra*) for establishing artificial systems of intracellular symbiosis. Yeast-bearing *Chlorella*-free *P. bursaria* was observed with a transmission electron microscope using freeze-substitution techniques and comparing the results with conventional chemical fixation. Both *P. capsulata* and *R. rubra* were found to be enclosed within the perisymbiont vacuole (PV) membranes in the host cells, and a large amount of microfibrillar materials were observed to be connecting the fungal cell wall and the inner surface of the PV membrane. The distance between the fungal cell wall and the PV membrane was 100–300 nm for *R. rubra* and 100–200 nm for *P. capsulata*, irrespective of the fixation method used, although the cell wall of the symbiotic *Chlorella* was closely apposed to the PV membrane with a distance of only 20–50 nm.