

# Photochemical Properties of Rhodopsins from Algal Bloom-Forming Dinoflagellates and Coral Symbiotic Reveal Unexpected Photochemical Diversity

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## Abstract

Rhodopsins are widely distributed photoreceptive proteins serving essential roles in all domains of life. Unlike animal rhodopsins (Type II), which primarily mediate visual and other light-dependent signaling processes, microbial rhodopsins (Type I) are found in microorganisms, where they exhibit diverse functions, including light-driven ion transport and photosensory signaling. Dinoflagellate rhodopsins were first discovered through environmental metatranscriptomics, and homologs have since been identified in the harmful algal bloom-forming species *Prorocentrum shikokuense*, and *Alexandrium catenella* [1, 2]. Most dinoflagellate rhodopsins share highly conserved residues with bacterial xanthorhodopsin (XRs) involved in retinal binding, spectral tuning, and proton transfer, suggesting potential functions as light-driven proton pumps. For example, the rhodopsin of *P. shikokuense* may enhance survival under low-light or nutrient-limited conditions during red tide formation [2, 3]. We recently discovered that the coral endosymbiotic dinoflagellate *Breviolum minutum* possesses the XRs-like rhodopsin as a potential second engine to capture photoenergy and compensate for impaired photosynthesis under phosphorus limitation [4]. However, the photochemical diversity and ecological significance of microbial rhodopsins in coral symbiotic and harmful algal bloom-forming dinoflagellates remain unclear. Revealing these properties will advance our understanding of alternative light-energy acquisition strategies in dinoflagellates and may reveal naturally optimized photoreceptors with unique photochemical characteristics.

In this study, we set out to characterize the absorption spectra and photochemical properties of rhodopsins from two harmful algal bloom-forming dinoflagellates, *Prorocentrum shikokuense* (PsR) and *Alexandrium catenella* (AcR), two coral symbionts, *Breviolum minutum* (BmR), a well-established experimental model symbiont, and *Cladocopium goreaui* (CgR), the most widespread and ecologically important symbiont in reef-building corals. Rhodopsins were heterologously reconstituted and purified as light-sensitive complexes in HEK293T cells in the presence of all-*trans* retinal following the method of Liénard et al. [5]. One previous study reported absorption wavelength ( $\lambda_{\text{max}}$ ) of approximately 530 nm for PsR expressed in whole *Escherichia coli* cells [2] whereas our results of purified PsR–retinal complexes showed a  $\lambda_{\text{max}}$  of 507 nm. The purified AcR–retinal complex exhibited a  $\lambda_{\text{max}}$  of 515 nm, indicating that both harmful algal bloom-forming dinoflagellate rhodopsins (PsR and AcR) absorb in the green light spectrum. In contrast, in coral symbionts, BmR exhibited a  $\lambda_{\text{max}}$  of 543 nm, indicating sensitivity to yellow-green light while CgR absorbed predominantly blue-green light with a  $\lambda_{\text{max}}$  of 492 nm. Lastly, photochemical assays further evidenced that CgR is the first bistable microbial rhodopsin discovered in dinoflagellates, which can be reversibly photoswitched between two thermally stable UV and green absorbing states.

## References

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