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Structural effects of high laser power densities on an early bacteriorhodopsin photocycle intermediate

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Time-resolved serial crystallography at X-ray Free Electron Lasers offers the opportunity to observe ultrafast photochemical reactions at the atomic level. The technique has yielded exciting molecular insights into various biological processes including light sensing and photochemical energy conversion. However, to achieve sufficient levels of activation within an optically dense crystal, high laser power densities are often used, which has led to an ongoing debate about the extent to which photodamage may compromise the interpretation of the results. Here we compare time-resolved serial crystallographic data of the bacteriorhodopsin K-intermediate collected at laser power densities ranging from 0.04 to 2493 GW/cm² and follow energy dissipation of the absorbed photons logarithmically from picoseconds to milliseconds. Although the effects of high laser power densities on the overall structure are small, in the upper excitation range we observe significant changes in retinal conformation and increased heating of the functionally critical counterion cluster. We compare light-activation within crystals to that in solution and discuss the impact of the observed changes on bacteriorhodopsin biology.

References

[1] Bertrand, Q. et al. Structural effects of high laser power densities on an early bacteriorhodopsin photocycle intermediate. *Nature Communications* 15, 10278 (2024).

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Session Classification: Pump-Laser Excitation Conditions in Time-Resolved Serial Femtosecond Crystallography