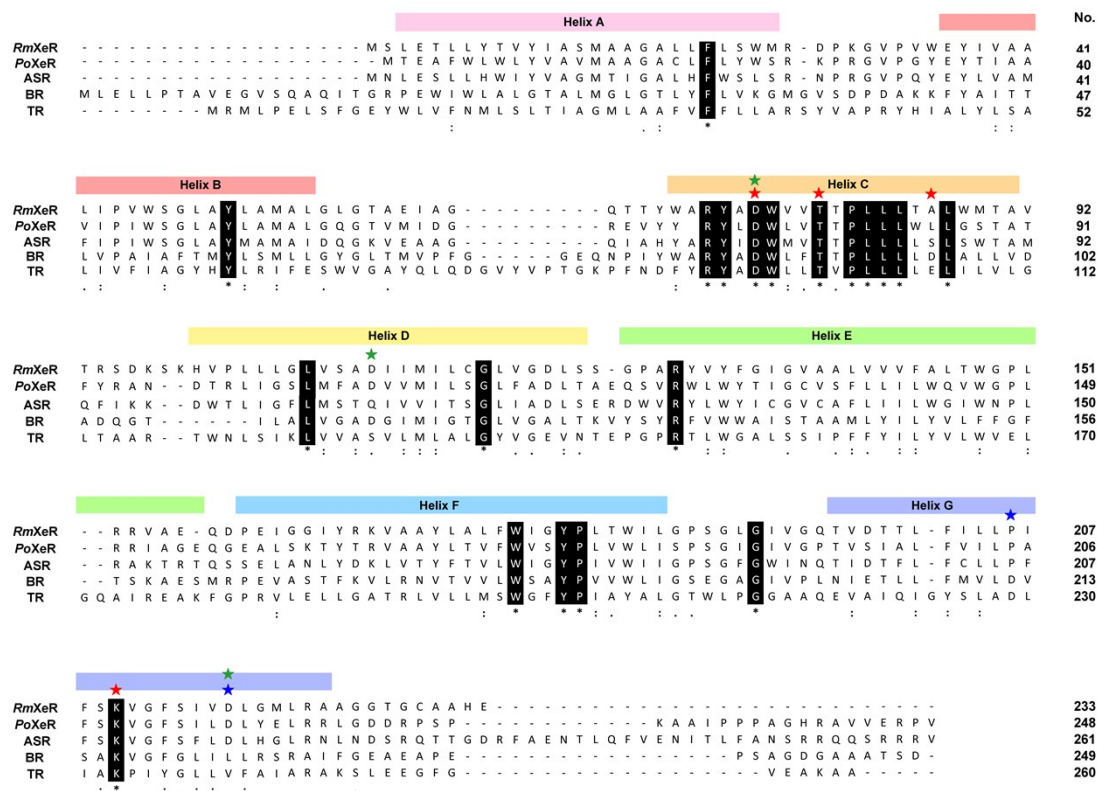
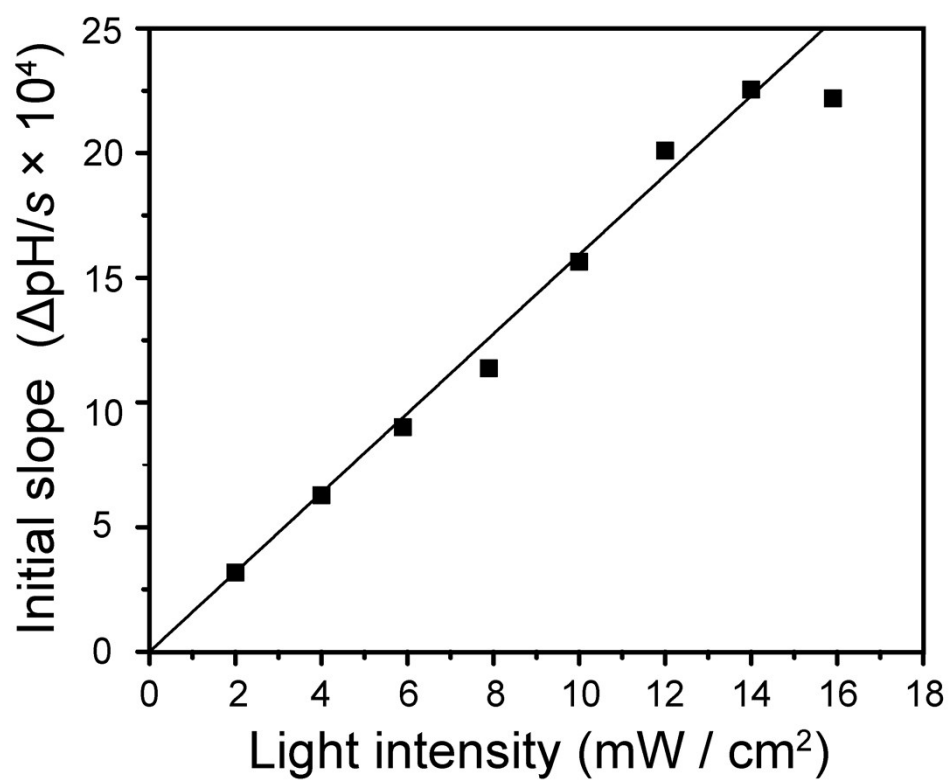


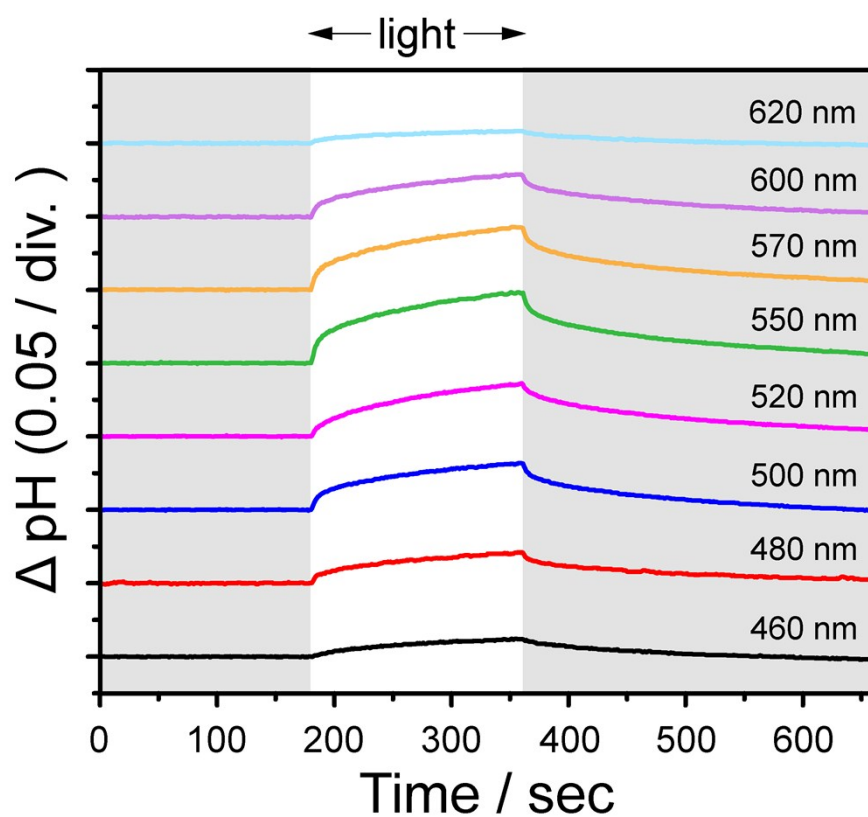
## Supplementary Figures



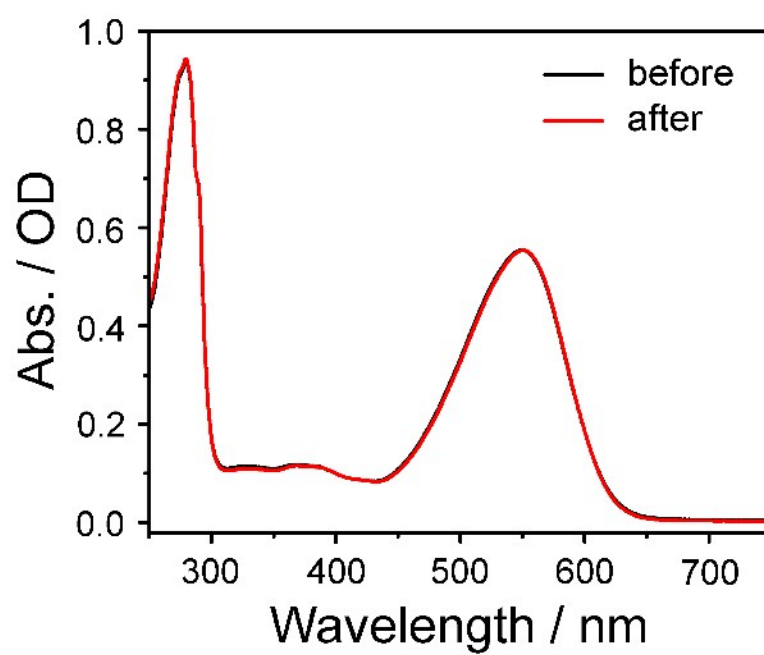
**Figure S1:** Multiple amino acid sequence alignment of microbial rhodopsins. *RmXeR*, *PoXeR*, *ASR*, *BR* and *TR* represent *Rubricoccus marinus* xenorhodopsin, *Parvularcula oceani* xenorhodopsin, *Anabaena* sensory rhodopsin, bacteriorhodopsin and thermophilic rhodopsin, respectively. Red stars show motif sequences (e.g., DTA for *RmXeR*) and Lys involved in binding with retinal as a Schiff base (e.g., Lys210 for *RmXeR*). Blue stars show residues characteristic for XeRs (e.g., Pro206 and Asp217 for *RmXeR*). Green stars show the mutated acidic residues in this study.



**Figure S2:** Correlation between light intensities and initial slope amplitudes of light-induced pH changes of *E. coli* cells expressing *RmXeR*.



**Figure S3:** Light-induced pH changes at various wavelengths of light. The *E. coli* cell suspension was illuminated at approx. 8 mW/cm<sup>2</sup> for 3 min in a solution containing 150 mM NaCl. The temperature was kept at 25°C using a thermostat. The initial slope amplitudes of the ion transport activities ( $\Delta \text{pH/s}$ ) from 0 to 10 sec upon illumination were obtained.



**Figure S4:** Absorption spectra of *RmXeR* before and after laser flash-photolysis experiments.