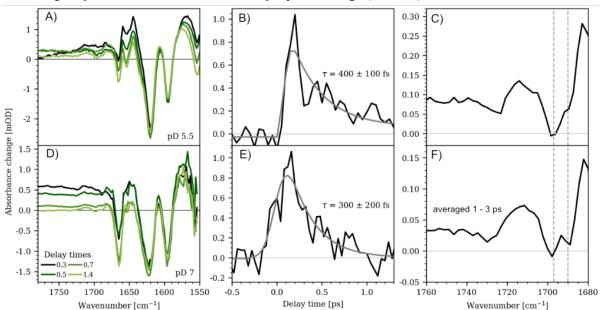
Extended Data Figure 5: Continuum band measurements

All presented datasets show isotropic polarized *Hs*BR dynamics in D₂O solution. Transients of the spectrally averaged CB together with mono-exponential simulations are depicted in B and E. Absorbance difference spectra averaged from 1 to 3 ps reflecting photoproduct formation are shown in C and F in the carbonyl spectral range from 1760 to 1680 cm⁻¹.

In the first graph absorbance difference spectra for selected delay times are presented at pD 5.5 (A, B, and C) and pD 7 (D, E, and F). CB transients are spectrally averaged from 1700 to 1780 cm⁻¹ at pD 5.5 in C and from 1700 to 1835 cm⁻¹ at pD 7.0 in E. Absorbance difference spectra averaged from 1 to 3 ps indicating very similar features in the carbonyl spectral range (C and F).



Measurements on the *Hs*BR Glu194Gln/Glu204Gln variant were also performed (D, E, and F) and are compared to WT *Hs*BR in the next graph: A, B, and C depict are the same as in the graph above. D shows isotropic absorbance difference spectra of the variant, E displays the transient of the CB averaged spectrally from 1700 to 1810 cm⁻¹ and a simulation with a decay constant of 0.3 (1) ps. F shows the averaged absorbance difference spectrum from 1 to 3 ps. Here, we see that the negative dip at 1698 cm⁻¹ is missing or very strongly reduced in the Glu194Gln/Glu204Gln variant compared to WT. The CB shows nearly identical features and decay times in WT and the variant.

