Electronic Supplementary Material (ESI) for Physical Chemistry Chemical Physics.

Supporting Information
The Molecular Mechanism of Light-induced Bond Formation and Breakage in the Cyanobacteriochrome TePixJ

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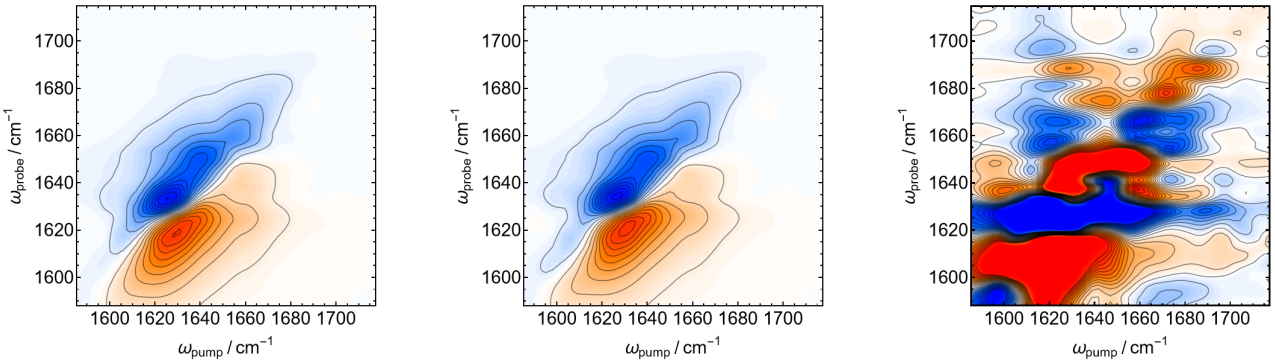

FIG. 1. Absolute 2D-IR spectra of TePixJ in the Pb state (left panel) and in the Pg state (middle panel). Right panel: "Pb-minus-Pg"-difference 2D-IR spectrum reproduced from the main manuscript.

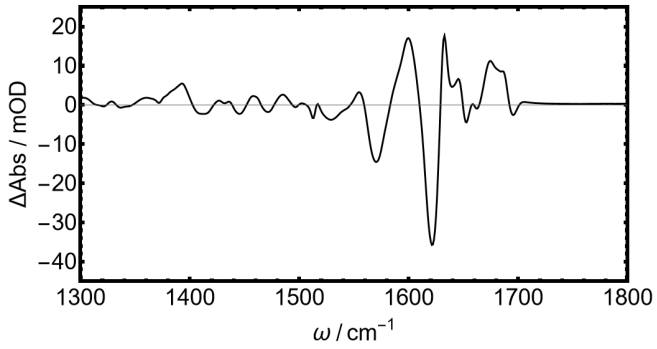

FIG. 2. The "Pg-minus-Pb" FTIR difference spectrum was recorded in a commercial FTIR spectrometer (Tensor 27, Bruker, Ettlingen, Germany). The sample was prepared in the Pb state and a single-channel spectrum was recorded. Then the sample was photoswitched inside the spectrometer by illumination with a green LED array (Thorlabs, 520 nm), and a second single channel spectrum was recorded. The "Pg-minus-Pr" difference spectrum was calculated as the negative logarithm of the ratio of the two single channel spectra.

Supplementary note 1: Lifetime analysis with maximum entropy regularisation. Both TRIR data sets were analyzed with two methods: lifetime analysis and global multiexponential fitting. The fundamental assumption behind both methods is that the data can be described by interconverting discrete states with time-invariant spectra. That is, the data matrix $d(\omega_i, t_j)$ can be written as superposition of the n different components:

$$d(\omega_i, t_j) = \sum_{k=1}^{n} C_k(t_j) A_k(\omega_i), \tag{1}$$

where $C_k(t_j)$ is the concentration profile of component k as a function of time t_j, and $A_k(\omega_i)$ its spectrum at probe frequency ω_i. The data were fit to multiexponential functions[1–3]:

$$f(\omega_i, t_j) = a_0(\omega_i) - \sum_{k=1}^{n} a(\omega_i, \tau_k) e^{-t_j/\tau_k}, \tag{2}$$

where the index k refers to a kinetic component with time constant τ_k. We will abbreviate $a_{i,k} \equiv a(\omega_i, \tau_k)$.

For lifetime analysis, 10 time constants τ_k per decade were fixed and distributed equidistantly on a logarithmic scale, while only the amplitudes $a_{i,k}$ were the free fitting parameters for each kinetic trace. A penalty function that maximizes the generalized absolute Shannon–Jones entropy s_i of the amplitudes $a_{i,k}$ for each frequency i was introduced to regularize the fit and avoid overfitting[3,4]:

$$s_i = \sum_k \left(\sqrt{a_{i,k}^2 + 4m_i^2} - a_{i,k} \ln \frac{\sqrt{a_{i,k}^2 + 4m_i^2} + a_{i,k}}{2m_i} - 2m_i \right) \tag{3}$$

Here, m_i are the so-called *a priori* solutions, which are a measure for the overall amplitude of the data at ω_i[3,4]. The entropy is subtracted from the root mean square deviation χ^2 of the fit, weighted by a regularisation parameter λ:

$$E_i = \frac{\chi_i^2}{\lambda} - s_i, \tag{4}$$

with

$$\chi^2 = \sum_{i,j} \left(d(\omega_i, t_j) - f(\omega_i, t_j) \right)^2 . \tag{5}$$

The metric E_i is minimized with respect to the amplitudes $a_{i,k}$. The sum over the squared amplitudes at all probe frequencies at one time τ_k is termed the "dynamical content" $D(\tau_k)$[5]:

$$D(\tau_k) = \sqrt{\sum_i a_{i,k}^2}. \tag{6}$$

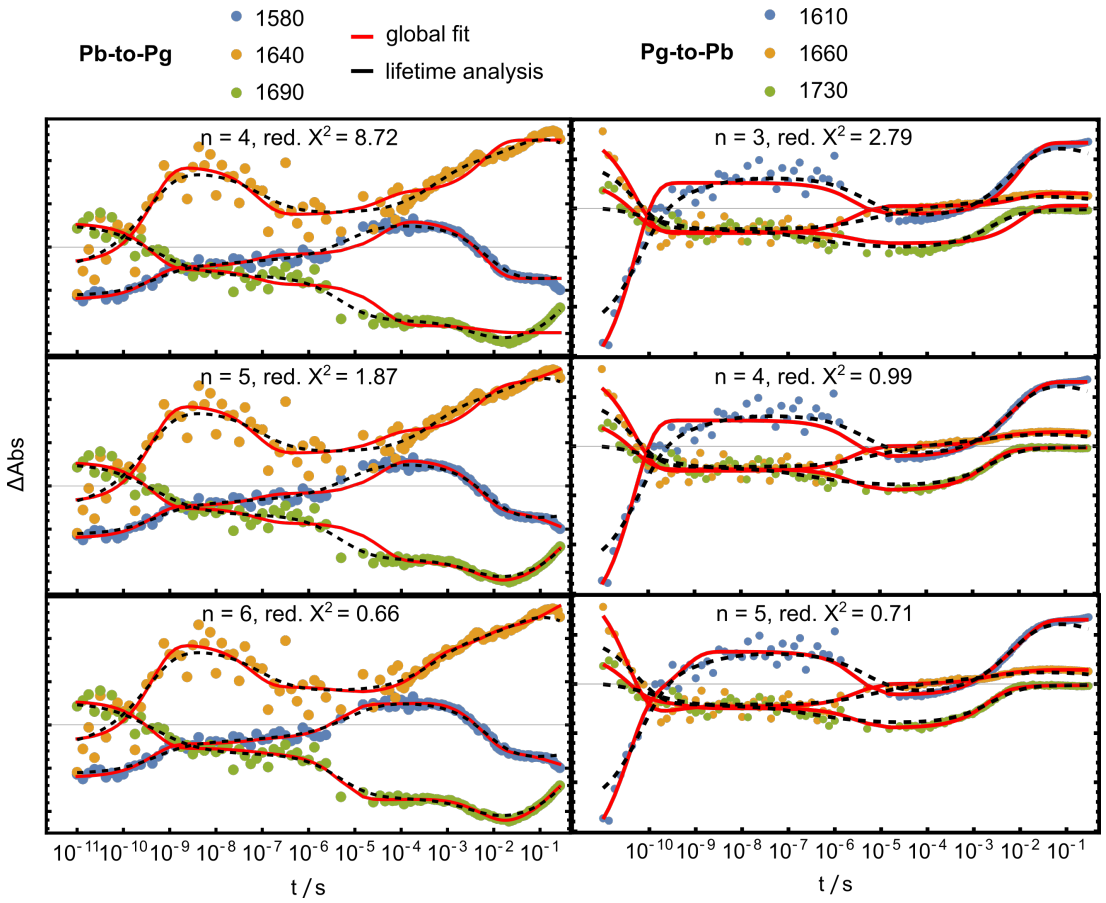

FIG. 3. Lifetime analysis (black dashed lines) and global fits (solid red lines) with different number of components shown for selected kinetic traces at three frequencies (blue, orange and green dots). Left side: Pb-to-Pg reaction, with n = 4, 5 and 6 components (top to bottom). Right side, Pb-to-Pg reaction, with n = 3, 4 and 5 components. Reduced χ^2 statistics were used as a goodness-of-fit criterion and, and calculated values for χ_ν^2 are given in the respective panels.

REFERENCES

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