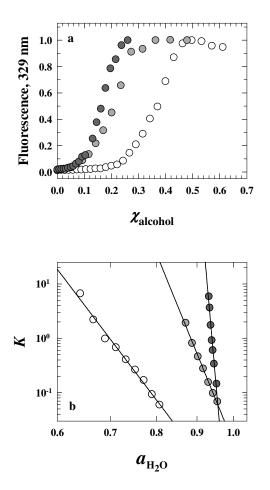
## **Supplemental Data for**

## Unfolding Action of Alcohols on a Highly Negatively Charged State of Cytochrome c

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Unfolding of cytochrome c at pH 7, 50 mM HEPES, 24°C, by methanol ( $\odot$ ), ethanol ( $\odot$ ), and propanol ( $\odot$ ). (a) Normalized fluorescence intensities are plotted as a function of mole fraction of alcohol. The unfolding equilibrium at pH 7 appears to be two-state. (b) Plots used to quantify the preferential hydration parameter across the folding-unfolding equilibrium. The water exclusion values are listed in Table 1 of the main text.