ANGEWANDTE CHEMIE
A Journal of the Gesellschaft Deutscher Chemiker

Supporting Information

for

Angew. Chem. Int. Ed. Z19274

69451 Weinheim, Germany

Naked-Eye Detection of Phosphate in Water of Physiological pH: A Remarkably Selective and Easy to Assemble Colorimetric Phosphate Sensing Probe**

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Supporting Information

Determination of the thermodynamic parameters and association constants for the binding of the indicator as well as the analyte to $[Zn_2(H\text{-}bpmp)]^{3+}$.

Isothermal titration calorimetry (ITC) measures the heat change on formation of a complex at constant temperature, thus to determine the ΔH for the binding interactions. The ITC plots for the titration of $[Zn_2(H\text{-}bpmp)]^{3+}$ with pyrocatechol violet and phosphate are depicted in Figure 5a and Figure 5b, respectively. The integration of the heat pulses obtained in each titration (top figures) gives a titration curve (bottom figures), from which thermodynamic parameters and binding constants for the bindings were calculated.

An Aqueous solution (1.5 mL, pH 7.0) of $[Zn_2(H\text{-}bpmp)]^{3+}$ (0.18 mM) was added to the calorimeter cell. To this solution was injected a 7 μL portion of aqueous HPO_4^{2-} solution (3 mM) 20 times. The mixture was continuously stirred and was kept at the operating temperature of 30 °C. The data were analyzed and fitted using the software Origin provided with the instrument.

An Aqueous solution (1.4 mL, pH 7.0) of $[Zn_2(H\text{-}bpmp)]^{3+}$ (0.2 mM) was added to the calorimeter cell. To this solution was injected a 7 μL portion of aqueous solution of pyrocatechol violet (2 mM) 30 times. The mixture was continuously stirred and was kept at the operating temperature of 30 °C.

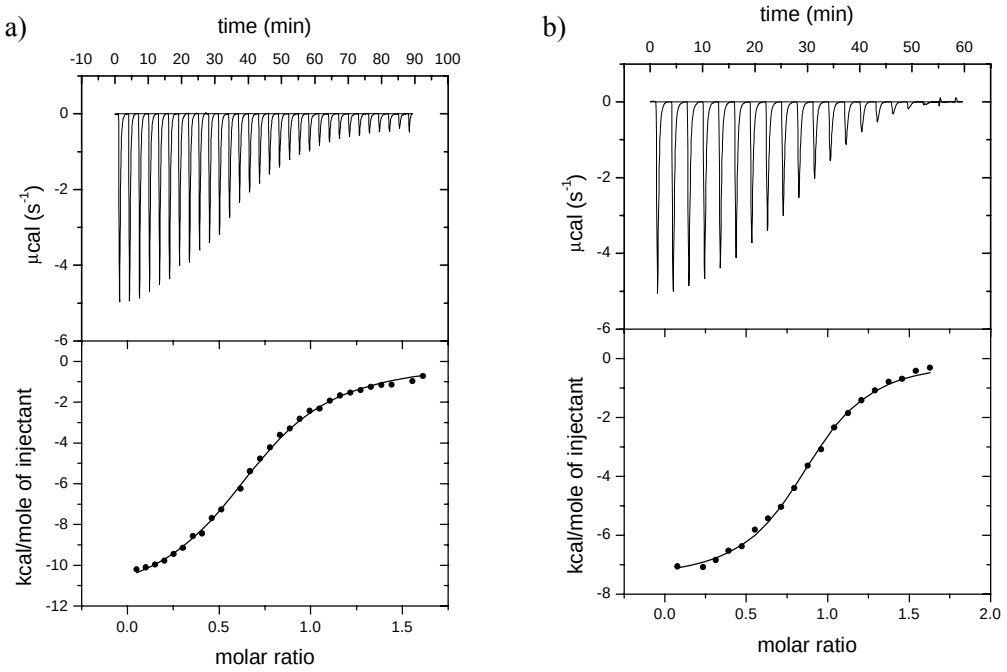
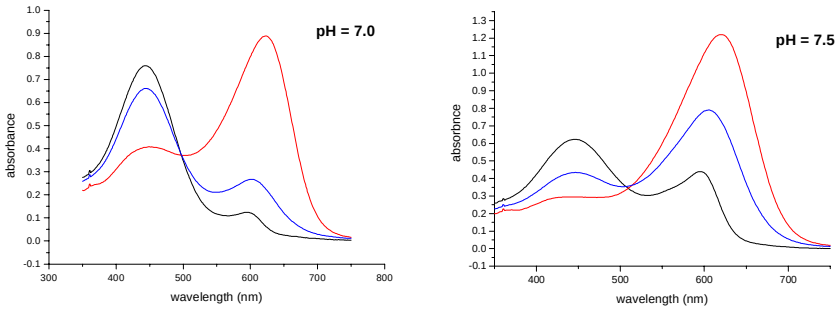


Figure 5. (a) Isothermal calorimetric titration of $[Zn_2(H\text{-}bpmp)]^{3+}$ (0.2 mM) with pyrocatechol violet (2 mM) in buffer solution (pH 7.0, 10 mM HEPES) at 30 °C. (b) Isothermal calorimetric titration of $[Zn_2(H\text{-}bpmp)]^{3+}$ (0.18 mM) with HPO_4^{2-} (3 mM) in buffer solution (pH 7.0, 10 mM HEPES) at 30 °C.

pH Dependence of the Phosphate Sensing



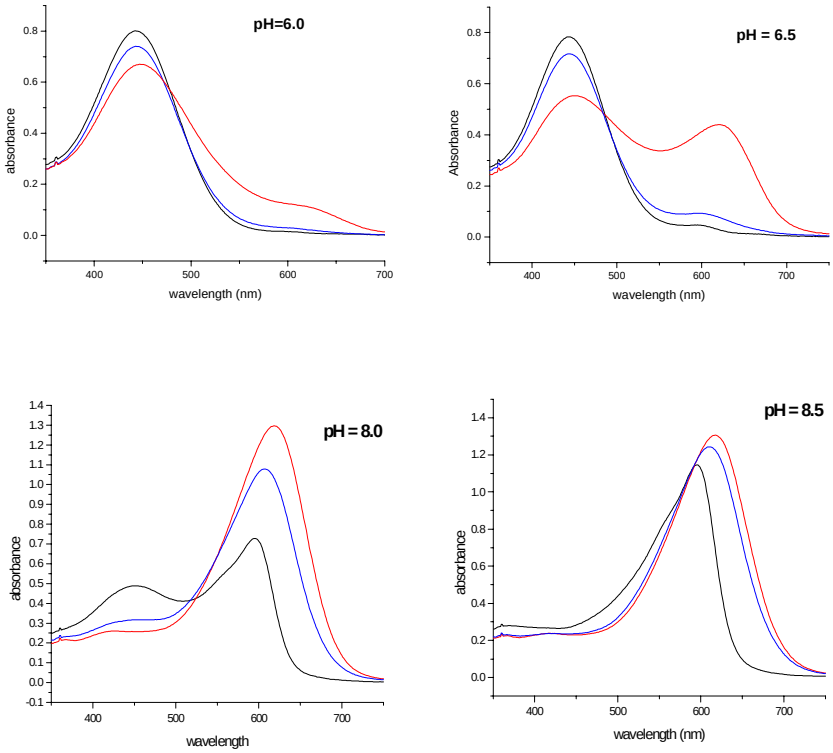


Figure 6. UV-vis absorbance intensities of the aqueous solution of pyrocatechol violet (50 μM, black line), the ensemble (pyrocathechol violet (50 μM) + $[Zn_2(H\text{-bpmp})]^{3+}$ (50 μM), red line), and the ensemble in the presence of Na_2HPO_4 (ensemble (50 μM) + Na_2HPO_4 (250 μM), blue line) at different pH. Compositions of the buffer solutions are as the following : pH 6.0 (MES 10 mM); pH 6.5 (MES 10 mM); pH 7.0 (HEPES 10 mM); pH 7.5 (HEPES, 10 mM); pH 8.0 (HEPES 10 mM); pH 8.5 (CHES 10 mM).

Quantitative Assay for phosphate

The preset ensemble may allow quantitative assay of phosphate in an aqueous solution containing various anions. Thus, no significant reduction within the experimental error in the UV absorbance was observed by the presence of anions and cations that are commonly found in the biological system up to their concentrations being about equal to that of the analyte. Figure 7 is a calibration curve for the quantitative determination of phosphate in the absence and presence of other ions.

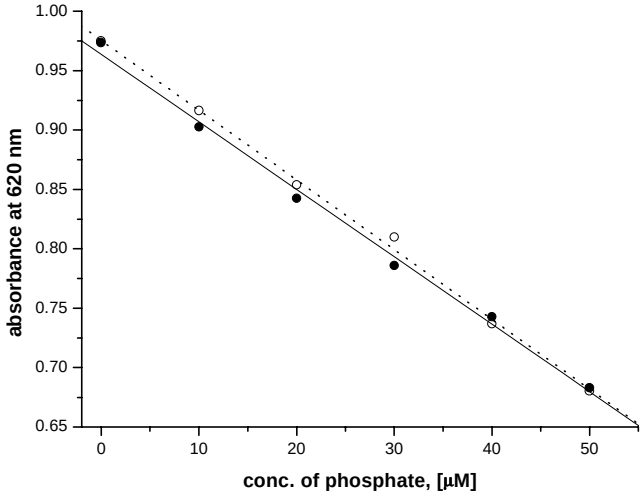


Figure 7. Plot of absorbance at 619 nm against conc. of phosphate in HEPES solution (pH 7.0) in the absence (●, ——) and presence of other ions (○, ·····) (50 μM NaCl, 50 μM NaOAc, 50 μM $NaHCO_3$, and 50 μM Na_2SO_4). The concentration of the sensor was maintained at 50 μM.