Electronic Supplementary Information

A natural cyanobacterial protein C-phycoerythrin as an Hg^{2+} selective fluorescence probe in aqueous systems

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S1. CPE purification and characterization

The growth of the cyanobacterium *Lyngbya* sp. CCNM 2053 was carried out using previously published reports [1]. Briefly, fresh biomass was suspended in 0.1 M potassium phosphate buffer pH 7.0 at a solid liquid ratio of 75 mg ml^{-1} (wet basis) and repeatedly frozen and thawed at -80 and 27 °C respectively. After 3 such cycles, the mixture was centrifuged (10,000 g, 10 °C, 10 minutes) and the supernatant was subjected to 25 % followed by 60 % ammonium sulphate precipitation. The pellet obtained after 60 % ammonium sulphate saturation was dissolved in minimal quantity of extraction buffer and extensively dialyzed against it. The partially purified CPE was further purified by ion-exchange (DEAE Sepharose, Sigma Aldrich, USA) followed by size exclusion chromatography (Sephadex S300 HR, Sigma Aldrich, USA). The pink fractions were pooled together and brought to 60 % ammonium sulphate saturation and left overnight. After centrifugation, the pellet was re-dissolved in minimum quantity of the extraction buffer and dialyzed against it overnight. CPE quantification was carried out using the following equations [2]:

$$\text{CPC (mg ml - 1)} = [A615 - 0.474A652]/5.34 \quad (1)$$

$$\text{APC (mg ml - 1)} = [A652 - 0.208A615]/5.09 \quad (2)$$

$$\text{CPE (mg ml - 1)} = [A562 - 2.41(CPC) - 0.849(APC)]/9.62 \quad (3)$$

The purity ratio of CPE was calculated using the A_{562}/A_{280} ratio while its characteristics were verified using UV-visible and fluorescence spectroscopy.

S2. Loss of CPE color with 7 µM Hg^{2+}

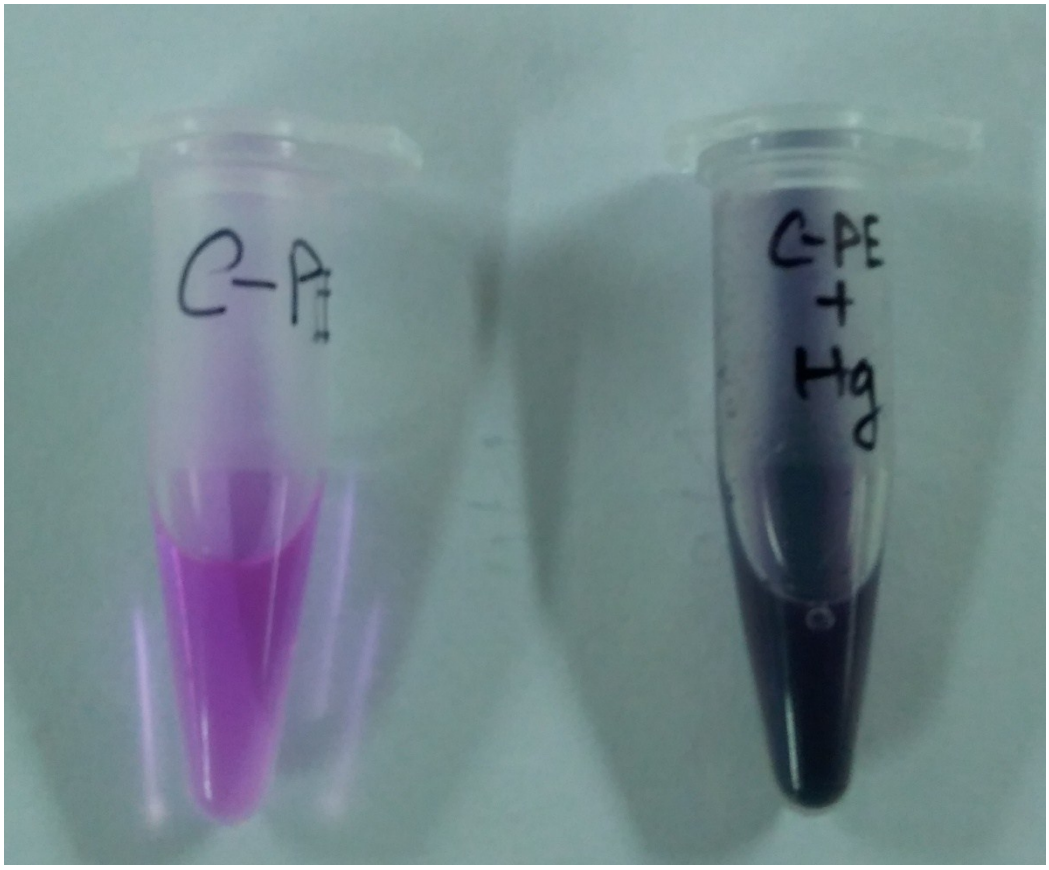

Fig. S1. CPE (left) before and (right) after interaction with 7 µM Hg^{2+}.

S3. Decrease in fluorescence intensity w.r.t [Hg^{2+}]

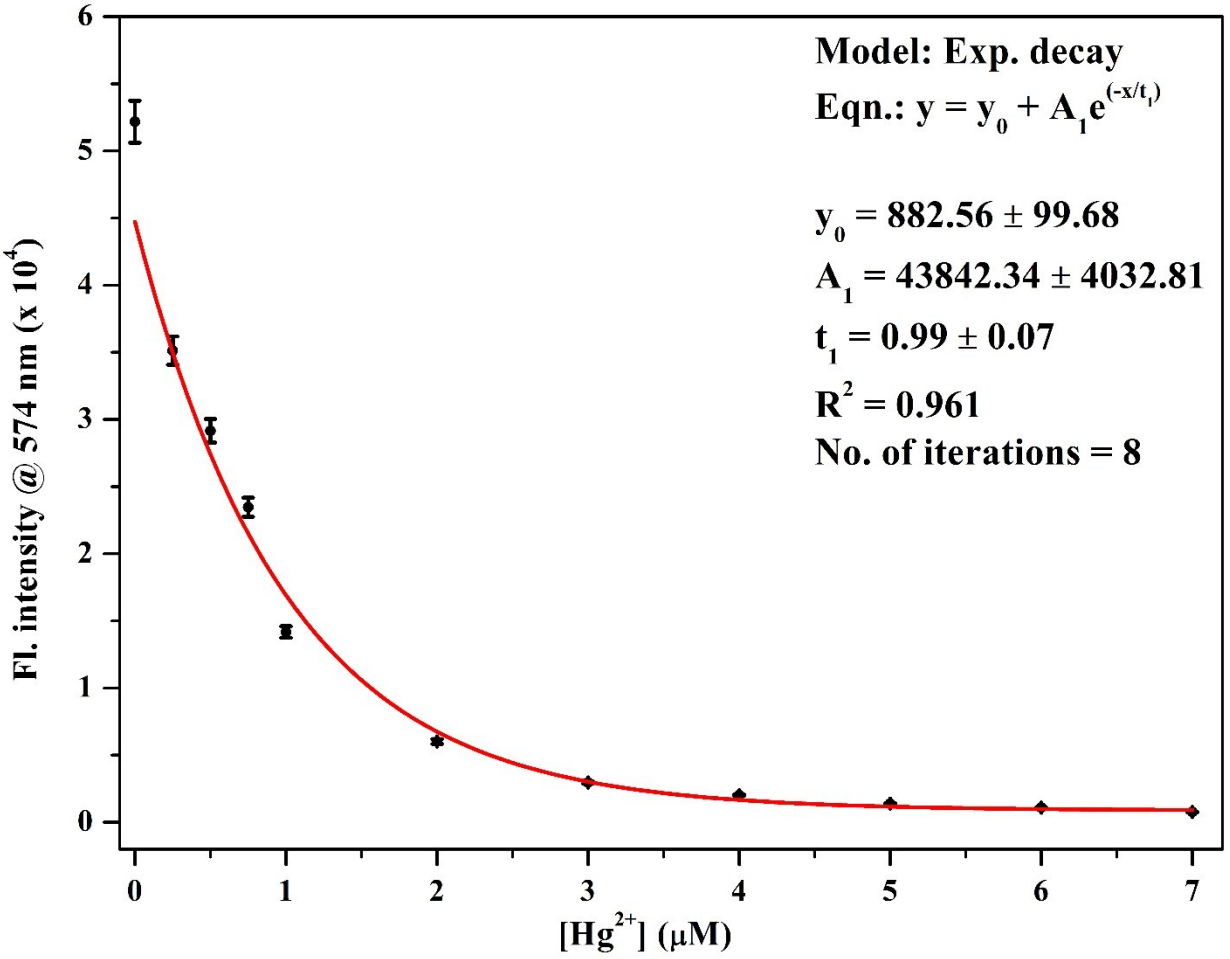

Fig. S2. Decrease in fluorescence emission of CPE under increasing [Hg^{2+}].

S4. Stern Volmer curve for CPE – Hg^{2+} interaction

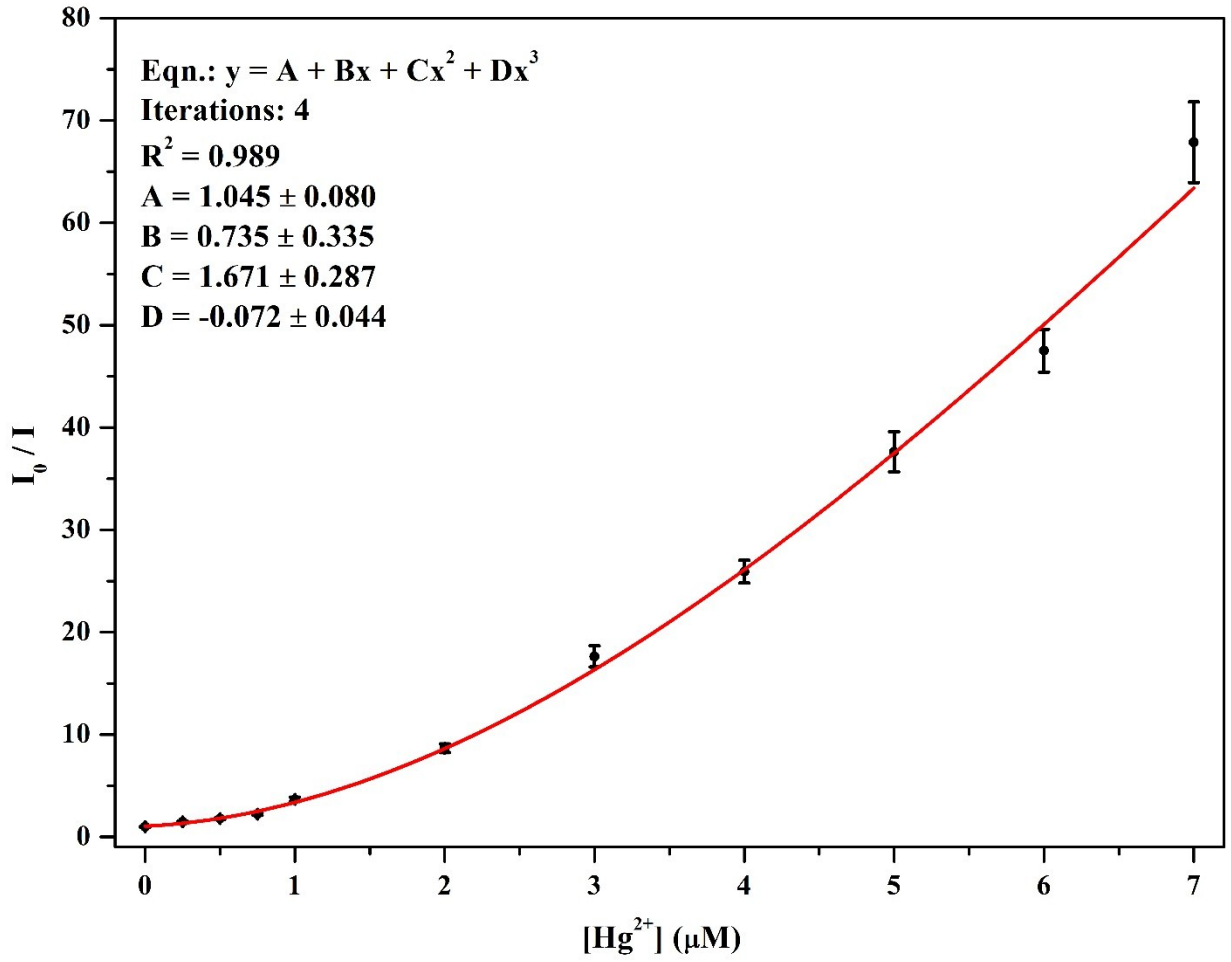

Fig. S3: Stern Volmer curve for CPE - Hg^{2+} interaction

S5. Fluorescence life time analysis

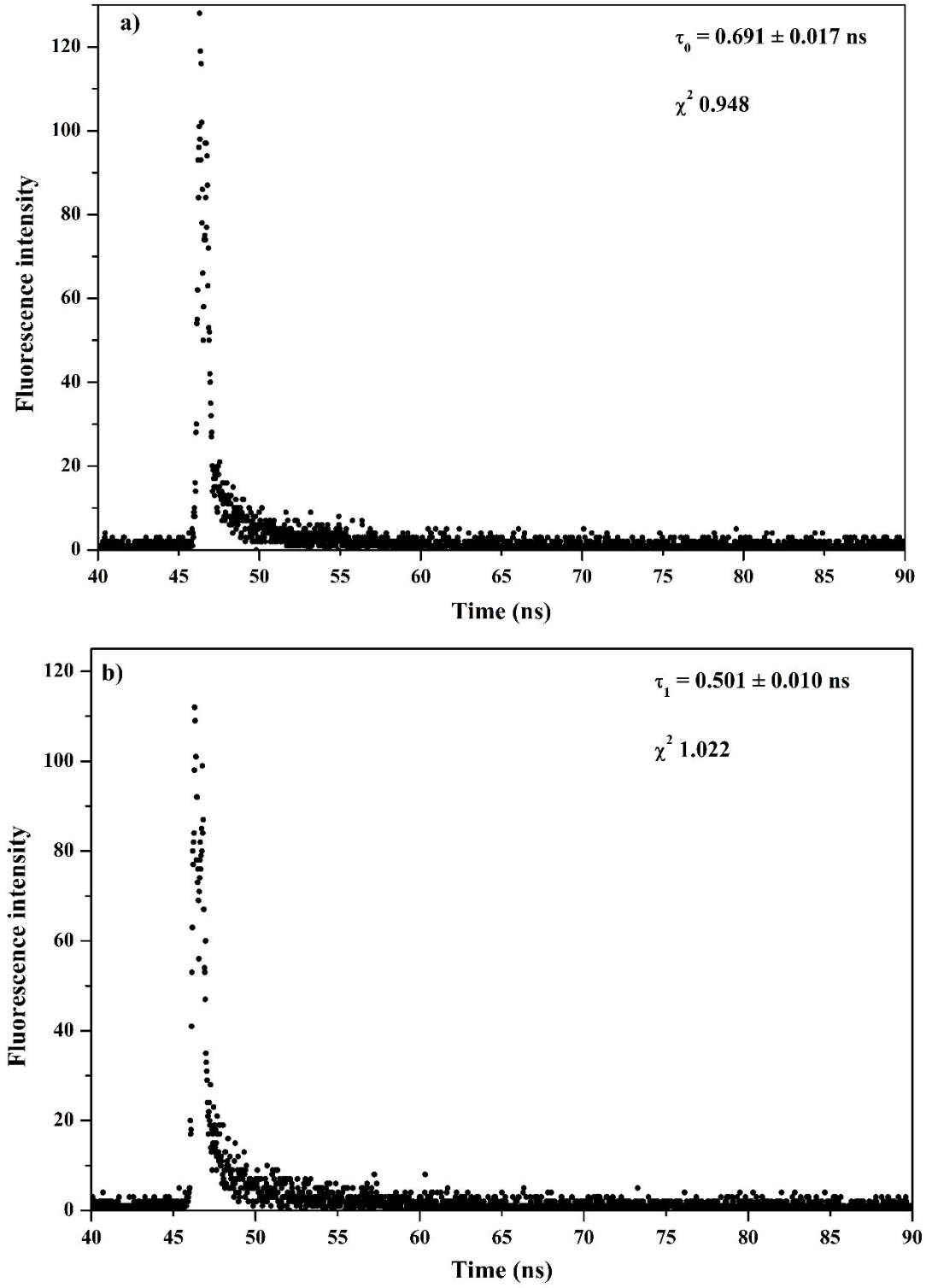

Fig. S4. Fluorescence lifetime of CPE in **a)** absence of Hg^{2+} and, **b)** in presence of 3 μM Hg^{2+}

References

1 T. Ghosh, K. Bhayani, C. Paliwal, R. Maurya, K. Chokshi, I. Pancha and S. Mishra, *Front. Mar. Sci.*, 2016, **3**, 146.

2 A. Bennett and L. Bogorad, *J. Cell Biol.*, 1973, **58**, 419–435.