Electronic Supporting Information

Detection of Hg$^{2+}$ by Cyanobacteria in Aqueous media.
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1. **Absorption spectrum of C-Phycocyanin.**

![Absorption spectrum of C-Phycocyanin](image)

**Figure S1** : Absorption spectrum of C-Phycocyanin ($2.57 \times 10^{-8}$ M) with phosphate buffer at pH 7.2.
2. Emission Spectrum of C-Phycocyanin.

Figure S2: Fluorescence emission spectrum of C-Phycocyanin (2.57 x 10^{-8} M) with phosphate buffer at pH 7.2. Excitation at 580 nm.
3. Emission spectra with different metal ions.

Figure S3: Changes in emission spectra of C-PC upon addition of different metal ions in phosphate buffer at pH 7.2. Excitation wavelength that was used for studies is 580 nm.
4. Absorption spectrum of Phycocyanobilin in phosphate buffer:

**Figure S4**: Absorption spectrum of phycocyanobilin (1 x 10^{-4}M) in phosphate buffer at pH 7.2.
5. Absorption spectrum of Phycocyanobilin in water:

Figure S5: Absorption spectrum of phycocyanobilin (1.0 x 10^{-4}M) in water
6. Binding constant for C-PC with various cations in phosphate buffer (pH = 7.2):

<table>
<thead>
<tr>
<th>Metal ions</th>
<th>Binding constant (K_b)$^a$</th>
</tr>
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<tbody>
<tr>
<td>Na$^+$</td>
<td>$-$</td>
</tr>
<tr>
<td>K$^+$</td>
<td>$-$</td>
</tr>
<tr>
<td>Cs$^+$</td>
<td>$-$</td>
</tr>
<tr>
<td>Ca$^{2+}$</td>
<td>$(2.5\pm0.10)\times10^3\text{M}^{-1}$</td>
</tr>
<tr>
<td>Mg$^{2+}$</td>
<td>$-$</td>
</tr>
<tr>
<td>Sr$^{2+}$</td>
<td>$-$</td>
</tr>
<tr>
<td>Ba$^{2+}$</td>
<td>$-$</td>
</tr>
<tr>
<td>Cr$^{3+}$</td>
<td>$-$</td>
</tr>
<tr>
<td>Fe$^{3+}$</td>
<td>$-$</td>
</tr>
<tr>
<td>Ni$^{2+}$</td>
<td>$-$</td>
</tr>
<tr>
<td>Cu$^{2+}$</td>
<td>$(3.0\pm0.15)\times10^3\text{M}^{-1}$</td>
</tr>
<tr>
<td>Hg$^{2+}$</td>
<td>$(2.7\pm0.2)\times10^4\text{M}^{-1}$</td>
</tr>
<tr>
<td>Cd$^{2+}$</td>
<td>$(3.1\pm0.2)\times10^3\text{M}^{-1}$</td>
</tr>
<tr>
<td>Co$^{2+}$</td>
<td>$-$</td>
</tr>
<tr>
<td>Zn$^{2+}$</td>
<td>$-$</td>
</tr>
</tbody>
</table>

$^a$Binding constant for each metal ions is an average of five independent experiments. Values were evaluated based on the fluorescence titration at room temperature. Spectral changes for Na$^+$, K$^+$, Cs$^+$, Mg$^{2+}$, Sr$^{2+}$, Ba$^{2+}$, Cr$^{3+}$, Fe$^{3+}$, Ni$^{2+}$, Co$^{2+}$ and Zn$^{2+}$ were not significant to enable us to evaluate respective binding constant values.
7. Mass spectra of Phycocyanobilin:

Exact mass:
Phycocyanobilin + H⁺ m/z = 587.2869

**Fig S6:** ESI-mass spectrum of phycocyanobilin extracted from *C-Phycocyanin* using Micromass Q-Tof micro™, equipped with ESI source and Q-Tof analyzer.
8. Mass spectra of Phycocyanobilin with Hg\(^{2+}\):

m/z = 811 Corresponds to Phycocyanobilin + Hg\(^{2+}\) + 23.
m/z for Na\(^+\) ion = 23

**Fig S7:** ESI-mass spectrum of ESI-mass spectrum of phycocyanobilin with Hg\(^{2+}\) using Micromass Q-Tof micro\(^\text{TM}\), equipped with ESI source and Q-Tof analyzer.
9. IR spectra of Phycocynobilin:
10. IR spectra of Phycocynobilin with Hg$^{2+}$:
11. Confocal images at different time intervals:

Fig S8. Confocal images of *spirulina platensis* with 10 μM Hg$^{2+}$ in 1:1 water / ethanol when exposed to 20 μM L$_1$ in a different time interval.
**12. Fluorescence competitive metal ion study of C-PC with Hg^{2+}:**

![Graph showing emission spectra of C-PC and C-PC+Hg^{2+} with various other metal ions](image)

**Fig S9:** Change in the emission spectra of C-PC (4.0 x 10^{-8} M) in the presence of Hg^{2+} (8.0 x 10^{-4} M) with various other metal ions (Co^{2+}, Ni^{2+}, Cu^{2+}, Ca^{2+}, Cd^{2+}, Sr^{2+}, Mg^{2+}, Zn^{2+}, Na^{+}, K^{+}, Li^{+}, Fe^{3+}) (8.0 x 10^{-3} M) in phosphate buffer at pH 7.2. Excitation at 580 nm.
13. HPLC study:

Purity of phycocyanobilin sample is further demonstrated by HPLC studies. A Waters HPLC system 2695 separation module (Alliance) coupled with Waters 2696 photodiode array UV-vis detector (PDA) was used for the HPLC separations on analytical C₈ column, 25cm x 4.6mm, 5μm. The column is equilibrated with mobile combination of 95% water and 5% methanol (0.1% acetic acid). The flow rate was 0.5 ml/min. The PDA detector was set to monitor the absorbance of eluent at 590 nm. Two fractions of phycocyanobilin chromophore were found in HPLC chromatogram at tᵣ = 1.8 min, (69.37%) and tᵣ = 2.0 min (30.63%) which corresponds to the existence of cis-trans isomers of phycocyanobilin.

Fig S10: HPLC profile of phycocyanobilin using analytical C₈ column (25cm x 4.6mm) and water/CH₃OH (95:5, v/v). Figure (a) shows the profile with 10 min retention time, while (b) shows retention time upto 50 minutes.
14. $^1$H NMR spectra for phycocyanobilin in CDCl$_3$:

![NMR Spectra](image)

**Fig S11**: $^1$H NMR spectra for phycocyanobilin in CDCl$_3$