Supporting Information

Contrasting guest binding interaction of cucurbit[7-8]urils with neutral red dye: Controlled exchange of multiple guests

Mhejabeen Shaikh, Sharmistha Dutta Choudhury, Jyotirmayee Mohanty,* Achikanath C. Bhasikuttan,* Haridas Pal

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Fig. S1: Relative fluorescence intensity changes recorded for NRH⁻:CB8 (a) and NRH⁻:CB7 (b) interaction. (c) and (d) represent the recovery of fluorescence signal obtained when Tryptophan or BSA was added to the (NRH⁻)₆:CB8 system, respectively. Inset shows the emission wavelength changes during NRH⁻:CB8 titration (e) and during the titration of (NRH⁻)₆:CB8 with Tryptophan (f) or BSA (g).

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Fig. S2: Absorption spectra of an aqueous solution of NRH⁺ without CB8 (a), with CB8 (b). Spectrum (c) represents the absorption spectrum recorded from a concentrated solution of NRH⁺ using a 1mm path length, indicating the presence of a dimeric species.

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Fig. S3: Computationally optimized geometries of various stoichiometric arrangements for neutral red-CB8 interaction (a-d). Structure (e) represents the optimized geometry for the NRH⁺:CB8:Trp Complex.

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1:1 NR:CB8 Complex
NR+CB8 → NR.CB8
ΔHf = -1.23 kcal/mole

1:1 NRH⁺:CB8 Complex
NRH⁺+CB8 → NRH⁺.CB8
ΔHf = -39.9 kcal/mole

2:1 NR:CB8 Complex
2NR+CB8 → (NR)₂:CB8
ΔHf = +21.64 kcal/mole

2:1 NRH⁺:CB8 Complex
2NRH⁺+CB8 → (NRH⁺)₂:CB8
ΔHf = -9.5 kcal/mole

1:1:1 NRH⁺:CB8:Trp Complex
NRH⁺+CB8+ Trp → NRH⁺.CB8.Trp
ΔHf = -7.15 kcal/mole

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Fig. S4: The fluorescence spectra recorded for the neutral red-CB8 system at different pHs, maintain the excitation parameters the same

Note: 1

The binding curve obtained for NRH⁺-CB8-BSA system (Fig.7, inset) were analyzed by the equations according to the following 1:1 interactions

\[
CB8\cdot(NRH^\cdot)_2 + BSA \leftrightarrow NRH^\cdot\cdot\cdotCB8\cdotBSA + NRH^\cdot
\]

\[\text{S1}\]

\[
NRH^\cdot + BSA \leftrightarrow NRH^\cdot\cdot\cdotBSA
\]

\[\text{S2}\]

Taking \([\text{Dye}]_0\) and \([\text{BSA}]_0\) as the total concentrations of dye and BSA, respectively, eq. S3 applies for the concentration of free (uncomplexed) dye in equilibrium:

\[
[Dye]_{eq} = [K_{eq}[Dye]_0] - K_{eq}[BSA]_0 - 1 + \sqrt{[K_{eq}[Dye]_0] + K_{eq}[BSA]_0 + 1} - 4K_{eq}^2[Dye]_0[BSA]_0 / 2K_{eq}
\]

\[\text{S3}\]

Where \([\text{Dye}]\) represents the concentration of CB8\cdot(NRH^\cdot)_2 complex for Eqn S1 and NRH^\cdot for Eqn S2.

The fluorescence intensity can therefore be understood as a composite of the fluorescence intensity contributions from the complexed and uncomplexed forms according to eq. S4:

\[
I_f = I_{\text{Dye}}^0 + \frac{[\text{Dye}]_{eq}}{[\text{Dye}]_0} + I_{\text{Dye}\cdot\cdot\cdot\text{BSA}}^\infty \frac{[\text{Dye} \cdot \text{BSA}]_{eq}}{[\text{Dye}]_0}
\]

\[\text{S4}\]

where \(I_{\text{Dye}}^0\) is the initial fluorescence intensity in the absence of CB8 and \(I_{\text{Dye}\cdot\cdot\cdot\text{BSA}}^\infty\) corresponds to the fluorescence intensity if all the dye molecules in the solution were complexed by BSA. The change in fluorescence intensity (\(\Delta I_f\)) can be obtained by rearrangement (eq.S5):

\[
\Delta I_f = (1 - \frac{[\text{Dye}]_{eq}}{[\text{Dye}]_0}) (I_{\text{Dye}\cdot\cdot\cdot\text{BSA}}^\infty - I_{\text{Dye}}^0)
\]

\[\text{S5}\]

In the fluorescence titrations, we employed the fluorescence intensity as experimental measure. The concentration of (Dye) was kept constant at a particular pH 5 and the concentration of BSA was varied. The binding constants (K) obtained from the nonlinear fittings of the experimental data using Eqn. S5 were 2.5x10⁵ M⁻¹ for the CB8\cdot(NRH^\cdot)_2 complex with BSA and 3x10³ M⁻¹ for the NRH^\cdot with BSA.