Supplementary Information for

Characterizing Chelation at Surfaces by Charge Tunneling

Li Yuan,^{1,2†} Samuel E. Root,^{1†} Lee Belding,¹ Junwoo Park,¹ Jeff Rawson,¹ Hyo Jae Yoon,³ Mostafa Baghbanzadeh,¹ Philipp Rothemund,^{1,4} and George M. Whitesides^{1*}

¹Department of Chemistry and Chemical Biology, Harvard University, 12 Oxford Street,

Cambridge, MA 02138

²Department of Chemistry, Tsinghua University, Beijing, 100084, China

³Department of Chemistry, Korea University, Seoul, 02841, Korea

⁴John A. Paulson School of Engineering and Applied Sciences, Harvard University, 29

Oxford Street, Cambridge, MA 02138, USA.

[†] Authors contributed equally

*Corresponding author: <u>gwhitesides@gmwgroup.harvard.edu</u>

S1. Materials
S2. Histograms of tunneling currents from the titration of Cu²⁺
S3. bpy-Ag titration
S4. X-ray photoelectron spectroscopy
S5. Binding model

S1. Materials

All reagents were used as supplied unless otherwise specified. All organic solvents in analytical degree were purchased from Sigma-Aldrich. The 1-decanethiol and 1-octadecanethiol were purchased from Sigma-Aldrich (>98% purity). High purity eutectic gallium-indium (EGaIn; 99.99%) was obtained from Sigma-Aldrich and used as supplied. 2,2'-Bipyridyl-terminated 1-undecanethiol (bpy(CH₂)₁₁SH) was synthesized following the procedures previously reported by us.¹ Figure S1shows the ¹H NMR spectrum of bpy(CH₂)₁₁SH.



Figure S1. The NMR spectra of bpy(CH₂)₁₁SH.



S2. Histograms of tunneling Currents from the titration of Cu²⁺

Figure S2. Histograms of log(J) at -1.0 V for the tunneling titration of CuCl₂. The solid lines are the Gaussian fits. The one standard deviation, number of J(V) traces and concentration of CuCl₂ in ethanol in mol/L are given in each panel.

S3. bpy-Ag titration

Figure S3a shows the comparison of J(V) between junctions of bpy-Ag (at $[Ag^+] = 1 \text{ mM}$) and bpy-terminated SAM. The J(V) curve of bpy-Ag is statistically indistinguishable from that of bpy-Cu. Figure S3b and S3c show the determination of K_{off} and K_{on} with the SAM of bpy-Ag, which yields a $K_d \sim 1.0 \times 10^{-6}$ mol/L. Figure S4 shows the results of titration with Ag⁺. The observed behavior is qualitatively analogous that of Cu²⁺. The dissociation constant inferred from these experiments was $K_d^{Ag^+} = 7.6 \times 10^{-6}$ mol/L, and this value is comparable with the K_d obtained by $K_{\text{off}}/K_{\text{on}}$. This value is several orders of magnitude lower than the value reported by Scrocco and coworkers of $K_d^{Ag^+} = 1.9 \times 10^{-4}$ mol/L.



Figure S3. (a) A comparison between the *J-V* curves for an uncomplexed and fully complexed junction contained Ag^+ . (b) A plot of current density at -1.0 V as a function of time that the sample was rinsed with ethanol. (c) A plot of current density at -1.0 V as a function of time that the sample was incubated with an ethanolic solution of AgClO₄ of concentration $5x10^{-4}$ mol/L.



Figure S4. Tunneling titration plots of bpy-Ag junctions with different concentration of $[Ag^+]$ in solution.

S4. X-ray photoelectron spectroscopy (XPS)

Oxidation states of metal cations. We performed the XPS measurement using a Thermo Scientifc Nexsa XPS system with a base pressure of 10^{-7} mbar in the Center for Nanoscale Systems at Harvard University. The energy of the incident X-ray beam was at 1486.6 eV. The SAMs were electrical in contact with the sample stage. We recorded the high-resolution XPS spectra of C 1*s*, O 1*s*, S 2*p*, N 1*s*, Ag 3*d*, Cu 2*p*, Ni 2*p*. Voigt functions (a linear combination of Lorentzian (30%) and Gaussian (70%)) were used to fit the XPS spectra with XPSpeak software. As shown in Figure S2, we observed the chemical states of Ag⁺ and Ni²⁺ are the same as they are in solution, but Cu peak shows there are ~14% Cu are in Cu(I) state which is caused by the photoelectron reduction (see details in the following section).



Figure S5. XPS spectra of metal cations, Cu (a), Ag (b), Ni (c) on the surface of composed of HS(CH₂)₁₁bpy.

The reduction of Cu cations by photoelectron emitted from Au bottom-electrodes.

Figure S3a-c shows that the apparent composition of Cu (as estimated from the XPS spectrum) depends on the acquisition time, and from \sim 3 mins to \sim 30 mins the spectra can be deconstructed into two peaks corresponding to 14% Cu⁺ and 86% Cu²⁺, 51% Cu⁺ and 49% Cu²⁺, and 87% Cu⁺ and 13% Cu²⁺. The 3 mins acquisition time is the shortest time we need to record a reasonable high resolution XPS spectra. It was well-documented that the Cu²⁺ can be reduced by the photoelectrons generated by the X-ray irradiation. The reduction kinetics can be expressed as a first order reaction:

$$\frac{d[Cu^{2+}]}{dt} = -k[Cu^{2+}]$$
 eq. S1

$$\ln([Cu^{2+}]) = \ln([Cu^{2+}]_0) - kt \qquad \text{eq. S2}$$

where $[Cu^{2+}]_0$ is the original concentration of bpy- Cu^{2+} complex and k is the reduction constant with unit of S⁻¹. We calculated the mole fraction of Cu^{2+} by integrating the peak area of Cu(II) (fit in red) and dividing by the entire peak area of Cu $2p_{3/2}$. A plot of the decay of the mole fraction of Cu^{2+} as a function of acquisition time (fit to **eq. S2**) is shown in **Figure S6d**. The mole fraction of $[Cu^{2+}]_0$ calculated from the intercept of the fit at t=0 is $100.8\pm1.2\%$.



Figure S6. XPS spectra of Cu with different acquisition time 3 mins (a), 15 mins (b) and 30 mins (c). d) The first-order kinetic plot of Cu(II) reduction by photoelectrons.

Characterization of Counter Ion. The XPS spectra also provided useful information regarding the nature and location of the chloride counter ion. For the bpy-Ag⁺ complex, as expected, we observed a 1:1 ratio of Ag to chloride. For the bpy-Cu²⁺ complex, we found that the magnitude of chloride peak depends on the way in which the chip was rinsed with EtOH (**Figure S4**). We found that if the chip was rinsed very gently with the nozzle very close to the chip (i.e. a 'soft rinse'), a 1:2 ratio of Cu to chloride was observed. Alternatively, if the chip was rinsed with an impinging stream of EtOH (i.e. a 'hard rinse') a 1:1 ratio of Cu to Chloride was observed. This observation suggests strongly that the chloride counter ion was exchanged for a hydroxide ion during the hard rinse.



Figure S7. XPS characterization of the fully complexed surface. (a) XPS spectra showing that a hard rinse vs. soft rinse does not affect the presence of the Cu ions. (b) XPS spectra showing that a hard rinse results in a significant reduction of the chloride signal. A "soft" wash indicates washing with a very gentle laminar flow of ethanol, while a "hard" wash indicates washing with an impinging jet from the squirt bottle.

S5. Binding model

Titration of one metal ion into SAMs terminated in bpy. The equilibrium equations for a metal ion (M) binding to a chelating group (bpy) are given by equations S3 and S4:

$$bpy + M \rightleftharpoons bpy \cdot M$$
 (eq.S3)

$$K_d^{bpy \cdot M} = \frac{[bpy][M]}{[bpy \cdot M]}$$
(eq.S4)

where [bpy] (mol/cm²) is the density of free bpy groups on the surface, $[bpy \cdot M]$ (mol/cm²) is the density of bound *M* on the surface, [M] (mol/L) is the concentration of metal ions in solution, and $K_d^{bpy \cdot M}$ is the dissociation constant. Conservation of mass requires that

$$[bpy]_0 = [bpy] + [bpy \cdot M]$$
(eq.S5)

where $[bpy]_0 \pmod{2}$ is the total molar concentration of bpy groups on the surface. We combine equations S3 and S4 to obtain the fraction of sites of *bpy* that are bound to M, $\left(\frac{[bpy\cdot M]}{[bpy]_0}\right)$, and the fraction of bpy that remained unbound $\left(\frac{[bpy]}{[bpy]_0}\right)$ (equations S6 and S7).

$$\frac{[bpy \cdot M]}{[bpy]_0} = \frac{\frac{[M]}{\kappa_d^{bpy \cdot M}}}{\left(1 + \frac{[M]}{\kappa_d^{bpy \cdot M}}\right)}$$
(eq.S6)

$$\frac{[bpy]}{[bpy]_0} = \frac{1}{\left(1 + \frac{[M]}{K_d^{bpy \cdot M}}\right)}$$
(eq.S7)

We assume that the current density $J(A/cm^2)$ can be separated into a parallel flow of charges with current density $J^{bpy \cdot M}(A/cm^2)$ through molecules of the SAM that contain metal ions and with current density $J^{bpy}(A/cm^2)$ through molecules that do not.

$$J = J^{bpy \cdot M} \frac{[bpy \cdot M]}{[bpy]_0} + J^{bpy} \frac{[bpy]}{[bpy]_0}$$
(eq.S8)

To relate the current to the dissociation constant and the concentration of metal ions in solution, we apply the definition of $[bpy]_0$ (eq.S5) and combine equation S8 with equations S6 and S7, to produce following relation:

$$J = \frac{\frac{J^{bpy \cdot M}[M]}{\kappa_d^{bpy \cdot M}} + J^{bpy}}{1 + \frac{[M]}{\kappa_d^{bpy \cdot M}}}$$
(eq.S9)

To modify this equation into a more useful form, in which the dependent variables are separated from the independent variables, we add and subtract J^{bpy·M}:

$$J = \frac{\frac{J^{bpy \cdot M}[M]}{\kappa_d^{bpy \cdot M}} + J^{bpy \cdot M} + J^{bpy} - J^{bpy \cdot M}}{1 + \frac{[M]}{\kappa_d^{bpy \cdot M}}} = \frac{J^{bpy \cdot M} \left(\frac{[M]}{\kappa_d^{bpy \cdot M}} + 1\right) + J^{bpy} - J^{bpy \cdot M}}{1 + \frac{[M]}{\kappa_d^{bpy \cdot M}}}$$
(eq.S10)

This trick allows for the algebraic rearrangement such that eq. S9 written in the following form:

$$\frac{J - J^{bpy \cdot M}}{J^{bpy} - J^{bpy \cdot M}} = \frac{1}{1 + \frac{[M]}{\kappa_d^{bpy \cdot M}}}$$
(eq.S11)

Next, both sides of eq. S11 can be inverted to obtain:

$$\frac{J^{bpy} - J^{bpy \cdot M}}{J - J^{bpy \cdot M}} = 1 + \frac{[M]}{\kappa_d^{bpy \cdot M}}$$
(eq.S12)

Subtracting of 1 from both sides of eq. S12 produces:

$$\frac{J^{bpy} - J}{J - J^{bpy} \cdot M} = \frac{[M]}{\kappa_d^{bpy \cdot M}}$$
(eq.S13)

Finally, both sides of eq. S13 are inverted and a logarithm is taken to yield the most useful functional form, which allows for fitting the experimental data with a linear regression:

$$\log\left(\frac{J-J^{bpy\cdot M}}{J^{bpy}-J}\right) = \log(K_d^{bpy\cdot M}) - \log([M])$$
(eq.S14)

Kinetics of Association. The kinetics of association for the reversible reaction by which the metal ions to bind to the surface sites from solution is described by eq. 2, which is reproduced here:

$$d[bpy \cdot M]/dt = k_{on}[bpy][M] - k_{off}[bpy \cdot M], \qquad (eq.2)$$

For the experiments to determine the 'on' rate constant, k_{on} , we consider the following conditions. Initially, the surface coverage, $\theta = 0$. Furthermore, we take [M] = 0.5 mol/L to be a constant due to an excess of metal ions in solution and an assumption that the surface binding is reaction limited. Dividing eq. 2 by $[bpy]_0$, simplifies the notation to use surface coverage as the integration variable for the following differential equation:

$$d\theta/dt = k_{on}[M](1-\theta) - k_{off}\theta.$$
 (eq.S15)

This ordinary differential equation and initial condition can be solved by separating the variables and integrating both sides of the equation to yield the following dependence on time:

$$\theta = \frac{-k_{on}[M]}{(k_{off} + k_{on}[M])} e^{-k_{on}[M]t - k_{off}t} + \frac{k_{on}[M]}{(k_{off} + k_{on}[M])}$$
(eq. S16)

Plugging this relation into eq. 4 and simplifying algebraically yields the final form for the dependence of current density on the incubation time, shown in the main text.

$$J = \left(\langle J^{bpy \cdot M} \rangle - \langle J^{bpy} \rangle \right) \frac{k_{on}[M]}{k_{on}[M] + k_{off}} \left(1 - e^{-k_{on}[M]t - k_{off}t} \right) + \langle J^{bpy} \rangle \qquad (\text{eq. 8})$$

Competitive Binding Titration. There might be instances in which competitive binding of metal ions or other molecular ligands to receptors on surfaces is of interest. Here, we introduce the equations to describe tunneling titration experiments of two metal ions (M1 and M2) competing to bind to the same surface receptor site (bpy). The equilibrium equations are:

$$bpy + M1 \rightleftharpoons bpy \cdot M1$$
 (eq.S15)

$$bpy + M2 \rightleftharpoons bpy \cdot M2$$
 (eq.S16)

$$K_d^{bpy \cdot M1} = \frac{[bpy][M1]}{[bpy \cdot M1]}$$
(eq.S17)

$$K_d^{bpy \cdot M2} = \frac{[bpy][M2]}{[bpy \cdot M2]}$$
(eq.S18)

where $[bpy \cdot M1]$ (mol/cm²) and $[bpy \cdot M2]$ (mol/cm²) are the densities of bound *M1*, and *M2* on the surface, [M1] and [M2] (mol/L) are the concentration of ligands in solution, and $K_d^{bpy \cdot M1}$ (mol/L) and $K_d^{bpy \cdot M2}$ (mol/L) are the dissociation constants of the metal ions on the surface. Conservation of mass requires that

$$[bpy]_0 = [bpy] + [bpy \cdot M1] + [bpy \cdot M2]$$
 (eq.S19)

We combine equations S17, S18, and S19 to obtain the fraction of sites of bpy are bound to M1,

$$\frac{[bpy\cdot M1]}{[bpy]_{0}}, \text{ bound to } M2, \left(\frac{[BIPY\cdot M2]}{[BIPY]_{0}}\right), \text{ or are unbound, } \left(\frac{[bpy]}{[bpy]_{0}}\right):$$

$$\frac{[bpy\cdot M1]}{[bpy]_{0}} = \frac{[M1]}{\left(\frac{K_{d}^{bpy\cdot M1} + [M1] + [M2]\frac{K_{d}^{bpy\cdot M1}}{K_{d}^{bpy\cdot M2}}\right)}$$
(eq.S20)

$$\frac{[bpy \cdot M2]}{[bpy]_0} = \frac{[M2]}{\left(\kappa_d^{bpy \cdot M2} + [M2] + [M1] \frac{\kappa_d^{bpy \cdot M2}}{\kappa_d^{bpy \cdot M1}}\right)}$$
(eq.S21)

$$\frac{[bpy]}{[bpy]_0} = \frac{1}{\left(1 + \frac{[M_1]}{\kappa_d^{bpy\cdot M_1}} + \frac{[M_2]}{\kappa_d^{bpy\cdot M_2}}\right)}$$
(eq.S22)

Analogous to the model for a single metal ion, we assume that the total current density across the junction $(J, A/cm^2)$ is composed of the flow of charges through molecules binding M1 with current density J^{bpy·M1} (A/cm²), molecules binding M2 with current density J^{bpy·M2} (A/cm²), and bpy molecules that have no bound metal ions, with current density J^{bpy} (A/cm²).

$$J = J^{bpy \cdot M1} \frac{[bpy \cdot M1]}{[bpy]_0} + J^{bpy \cdot M2} \frac{[bipy \cdot M2]}{[bpy]_0} + J^{bpy} \frac{[bpy]}{[bpy]_0}$$
(eq.S23)

We obtain a formula for the current density by combining equations S13-S17.

$$J = J^{bpy \cdot M1} \frac{[M1]}{\left(\kappa_{d}^{bpy \cdot M1} + [M1] + [M2] \frac{\kappa_{d}^{bpy \cdot M1}}{\kappa_{d}^{bpy \cdot M2}}\right)} + J^{bpy \cdot M2} \frac{[M2]}{\left(\kappa_{d}^{bpy \cdot M2} + [M2] + [M1] \frac{\kappa_{d}^{bpy \cdot M2}}{\kappa_{d}^{bpy \cdot M1}}\right)} + J^{bpy} \frac{1}{\left(1 + \frac{[M1]}{\kappa_{d}^{bpy \cdot M1}} + \frac{[M2]}{\kappa_{d}^{bpy \cdot M2}}\right)}$$
(eq.S24)

In the limit of [M1] or $[M2] \rightarrow 0$, it can be seen that eq. S24 reduces to eq. S9, the analogous relation for a single metal ion. To express this relation in another form, we introduced $[C_t]$ (mol/m³) for the total concentration of ions in solution ($[C_t] = [MI] + [M2]$), and used the mole fraction of $MI\left(\chi_{M1} = \frac{[M1]}{[C_t]}\right)$ and $M2\left(\chi_{M2} = \frac{[M2]}{[C_t]}\right)$ in solution.

$$J = \frac{J^{bpy} K_{d}^{bpy \cdot M1} + J^{bpy \cdot M1}[C_{t}]}{K_{d}^{bpy \cdot M1} + [C_{t}]} + \left(\frac{J^{bpy} K_{d}^{bpy \cdot M2} + J^{bpy \cdot M2}[C_{t}]}{K_{d}^{bpy \cdot M2} + [C_{t}]} - \frac{J^{bpy} K_{d}^{bpy \cdot M1} + J^{bpy \cdot M1}[C_{t}]}{K_{d}^{bpy \cdot M1} + [C_{t}]}\right) \frac{1}{1 + 10^{\log\left(\frac{K_{d}^{bpy \cdot M2} K_{d}^{bpy \cdot M1} + [C_{t}]}{K_{d}^{bpy \cdot M2} + [C_{t}]}\right) - \log\left(\frac{\chi_{M2}}{\chi_{M1}}\right)}$$
(eq.S25)

General Stoichiometry. Although bpy-terminated SAMs bind to metal ions with 1:1

stoichiometry, other complexes of interest might have a different binding stoichiometry. Here, we present and discuss the equilibrium equations for the general case of a ligand, *L*, binding to a receptor, *R*, on a surface, with arbitrary stoichiometric coefficients, α , β .

$$\alpha R + \beta L \rightleftharpoons R_{\alpha} \cdot L_{\beta} \tag{eq. S26}$$

$$K_d^{R_{\alpha} \cdot L_{\beta}} = \frac{[R]^{\alpha} [L]^{\beta}}{[R_{\alpha} \cdot L_{\beta}]}$$
(eq. S27)

Here $[R] \pmod{cm^2}$ is the density of free receptor groups on the surface, $[R_{\alpha} \cdot L_{\beta}] \pmod{cm^2}$ is the density of complexes on the surface, $[L] \pmod{L}$ is the concentration of ligands in solution which can be assumed to be constant (experiments were designed to ensure that the total moles of *L* in solution was at least 100x greater than the number of sites on the surface), and $K_d^{R_{\alpha} \cdot L_{\beta}}$ is the dissociation constant of the complex. Conservation of mass requires that

$$[R]_0 = [R] + \alpha [R_\alpha \cdot L_\beta]$$
 (eq. S28)

where $[R]_0 \pmod{2}$ is the total molar concentration of receptors on the surface. We combine equations S27 and S28 to obtain the fraction of sites of *R* that are bound to L, $\theta = \frac{\alpha [R_{\alpha} \cdot L_{\beta}]}{[R]_0}$.

$$\theta = \frac{\alpha[R_{\alpha} \cdot L_{\beta}]}{[R]_{0}} = \frac{\frac{\alpha[R]^{\alpha-1}[L]^{\beta}}{R_{\alpha} \cdot L_{\beta}}}{\left(1 + \frac{\alpha[R]^{\alpha-1}[L]^{\beta}}{R_{\alpha} \cdot L_{\beta}}\right)}$$
(eq. S29)

The important aspect to note about this equation is that [*R*] only cancels out for the case of $\alpha =$ 1, leading to an algebraic difficulty in relating the current density directly to $K_d^{R_{\alpha} \cdot L_{\beta}}$ and [*L*]. Following the same line of reasoning for the 1:1 stoichiometry case, we can write the current density as:

$$J = \frac{\frac{J^{R_{\alpha} \cdot L_{\beta}} \alpha[R]^{\alpha - 1}[L]^{\beta}}{K_{d}^{R_{\alpha} \cdot L_{\beta}}}}{1 + \frac{\alpha[R]^{\alpha - 1}[L]^{\beta}}{K_{d}^{R_{\alpha} \cdot L_{\beta}}}} + \frac{J^{R}}{1 + \frac{\alpha[R]^{\alpha - 1}[L]^{\beta}}{K_{d}^{R_{\alpha} \cdot L_{\beta}}}}$$
(eq. S30)

Which can be rearranged to yield the following form:

$$\log\left(\frac{J-J^{R_{\alpha}\cdot L_{\beta}}}{J^{R}-J}\right) = \log\left(K_{d}^{R_{\alpha}\cdot L_{\beta}}\right) - \beta\log([L]) - (\alpha - 1)\log([R]) - \log(\alpha)$$
(eq. S31)

For $\alpha \neq 1$, [*R*] must be eliminated from the equation, as it is an unknown variable. For the case of $\alpha = 2$, this elimination can be achieved by combining equation eq. S27 and eq. S28 to produce the following quadratic equation:

$$[R]^{2} + \frac{K_{d}^{R_{2} \cdot L_{\beta}}[R]}{2[L]^{\beta}} - \frac{K_{d}^{R_{2} \cdot L_{\beta}}[R]_{0}}{2[L]^{\beta}} = 0$$
 (eq. S32)

Applying the quadratic formula, produces the following roots:

$$[R] = -\frac{K_d^{R_2 \cdot L_\beta}}{2[L]^\beta} \pm \sqrt{\left(\frac{K_d^{R_2 \cdot L_\beta}}{2[L]^\beta}\right)^2 - \frac{2K_d^{R_2 \cdot L_\beta}[R]_0}{[L]^\beta}} \qquad (eq. S33)$$

Taking the positive root of eq. S32, eq. S31 can be rewritten as:

$$\log\left(\frac{J-J^{R_2 \cdot L_\beta}}{J^{R}-J}\right) = \log\left(K_d^{R_2 \cdot L_\beta}\right) - \beta \log([L]) - \log\left(-\frac{K_d^{R_2 \cdot L_\beta}}{2[L]^\beta} + \sqrt{\left(\frac{K_d^{R_2 \cdot L_\beta}}{2[L]^\beta}\right)^2 - \frac{2K_d^{R_2 \cdot L_\beta}[R]_0}{[L]^\beta}}\right) - \log(2)$$
(eq. S34)

This equation cannot be written in a linear form but can be fit to the experimental data with a nonlinear curve fitting algorithm.

References

Yoon, H. J.; Liao, K. C.; Lockett, M. R.; Kwok, S. W.; Baghbanzadeh, M.; Whitesides, G.
 M. Rectification in Tunneling Junctions: 2,2'-Bipyridyl-Terminated n -Alkanethiolates. J.

Am. Chem. Soc. 2014, 136 (49), 17155–17162.