Supporting Information

Molecular Basis of Coupled Protein and Electron Transfer Dynamics of Cytochrome *c* in Biomimetic Complexes

Damián Alvarez-Paggi,^a Diego F. Martín,^a Pablo M. De Biase,^a Peter Hildebrandt,^b

Marcelo A. Martí^{a*} and Daniel H. Murgida^{a*}

^aDepartamento de Química Inorgánica, Analítica y Química Física / INQUIMAE-
CONICET, Facultad de Ciencias Exactas y Naturales, Universidad de Buenos Aires.
Ciudad Universitaria, Pab. 2, piso 1, C1428EHA-Buenos Aires, Argentina.

^bTechnische Universität Berlin, Institut für Chemie, Str. des 17. Juni 135, Sekr. PC14, D-
10623-Berlin, Germany.

*Corresponding authors. Fax.: +54 11 4576-3341.

E-mail: marcelo@qi.fcen.uba.ar, dhmurgida@qi.fcen.uba.ar;

Figure S1. Work versus distance plot for the SMDs of Cyt^{+3} (top) and Cyt^{+2} (bottom). The distance is measured from the Fe atom to the plane defined by the Au atoms. As described in *Computational Methods*, SMDs were performed for 26 different starting conformations for each redox state. Left: Work plots corresponding to unfavorable binding orientations. Right: Work plots corresponding to favourable binding orientations, ie. where minima can be seen.

Figure S2. α versus ϕ plots for all MD simulations in explicit water. Each point represents a snapshot from the corresponding MD. Snapshots were taken every 10 ps. The color code is similar to Figure 1A.

Table S3. Binding energy (Kcal.mol⁻¹) contributions for Cyt/SAM complexes obtained with MM/PBSA calculations

Orientation	ΔE_{ELEC}^a	ΔE_{VDW}^b	ΔE_{GAS}^c	ΔG_{SV}^d	ΔG_{B}^e
o1	-2413.06	-58.39	-2471.45	2425	-45.95
o2	-2473.69	-82.03	-2555.72	2478.49	-77.23
o3	-2217.94	-79.97	-2297.90	2255.04	-56.84
o4	-2179.13	-66.57	-2245.70	2207.53	-49.10
o5	-2144.75	-39.20	-2183.95	2145.71	-47.03
o6	-2163.00	-63.61	-2226.61	2180.56	-57.35
o7	-1931.05	-84.80	-2015.85	1956.89	-58.96
r1	-2170.66	-71.79	-2242.45	2207.05	-47.53
r2	-2303.70	-84.34	-2388.03	2337.46	-65.10
r3	-2428.81	-84.27	-2513.08	2461.40	-66.57
r4	-2080.26	-51.49	-2131.76	2098.36	-33.40
r5	-1583.74	-44.70	-1628.44	1614.04	-22.77

^a ΔE_{ELEC} : Electrostatic contribution to the interaction energy in gas phase

^b ΔE_{VDW} : van der Waals contribution to the interaction energy in gas phase

^c ΔE_{GAS} : Interaction energy in gas phase, estimated as $\Delta E_{\text{ELEC}} + \Delta E_{\text{VDW}}$

^d ΔG_{SV} : Free energy of solvation estimated using the generalized Born implicit solvation method

^e ΔG_{B} : Binding energy estimated as $\Delta E_{\text{GAS}} + \Delta G_{\text{SV}}$

Figure S4. Superimposed average structures for Cyt⁺² (red) and Cyt⁺³ (blue) from the different MDs. The results show that the protein does not suffer major structural changes during the simulations.

Table S5. RMSD values for Cyt during the MDs. The values were computed both against the average structure of the simulations and the X-Ray structure. The results show that for Cyt⁺³ the mobility of the α A β 1 loop is similar to the whole protein and remains close to the starting structure. For Cyt⁺² the loop and whole protein mobilities are also similar but the establishment of new contacts in the loop induces larger structural changes.

Redox state	Reference structure			
	Average		X-ray	
	Whole Protein	α A β 1 Loop	Whole Protein	α A β 1 Loop
Cyt ⁺³	0.9	0.8	1.0	1.0
Cyt ⁺²	1.53	0.9	2.5	2.8

Figure S6. Orientation of the protein dipole moment plotted as the angle between the dipole moment vector and the Z axis (perpendicular to the SAM) versus simulation time for selected MDs. Data shown from top to bottom correspond to MDs labeled as o6, r3 and r1, respectively (see Figure 1).

Figure S7. Total protein dipole moment computed with respect to the center of charge of the protein for selected MDs. Data shown from top to bottom correspond to MDs labeled as o6, r3 and r1, respectively (see Figure 1).

Figure S8. Boltzmann analysis of the probability of orientational distributions for Cyt^{+2} (top) and Cyt^{+3} (bottom). The color code indicates increasing probabilities from $< 10^{-6}$ (black) to $> 10^{-2}$ (brown).

The distribution of probabilities in Figure S8 was estimated based on the calculated binding energies and working with the canonical ensemble assuming a Boltzmann distribution:

$$\frac{N_j}{N} = \frac{e^{-E_j/(RT)}}{\sum_i e^{-E_i/(RT)}}$$

Summation is done over all binding energy values for every pair of (α, ϕ) explored by each redox state.