Supplementary Material

The key role played by charge in the interaction of cytochrome c with cardiolipin

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Figure S1: Comparison of the UV-Vis electronic absorption spectra of the Lys73Arg (red), Lys73Ala (blue), Lys73Asn (green) mutants compared with wt cyt c (black). The 470–650nm and 650–800 nm regions are expanded 5-fold and 20-fold, respectively. The spectra have been shifted along the ordinate axis to allow better visualization. The experimental conditions were 20 mM Hepes + 0.1 mM EDTA, pH 7.0, and 25°C.
Figure S2: Near-UV CD spectra of the Lys72Arg and Lys73Arg (-- ... ), Lys72Ala and Lys73Ala (---- ... ), and Lys72Asn and Lys73Asn (--- ... ) mutants. For each type of mutation, the Lys72 and Lys73 variants show almost identical spectra. The concentration for all mutants was 10 μM. The CD spectrum of wt cyt c (-----) is reported for comparative purposes. The experimental conditions were 20 mM Hepes + 0.1 mM EDTA, pH 7.0, and 25°C.
Figure S3: RR spectra in the high (A) and low (B) frequency regions of the Lys73Arg (red), Lys73Ala (blue) and Lys73Asn (green) mutants compared with wt cyt c (black). Experimental conditions: 20 mM Hepes + 0.1 mM EDTA, pH 7.0, 25°C, excitation wavelength 406.7 nm; WT: laser power at the sample 5 mW, average of 5 spectra with 25 min integration time (A) and average of 4 spectra with 20 min integration time (B); K73R: average of 6 spectra with 30 min integration time; K73A: average of 6 spectra with 30 min integration time (A) and average of 8 spectra with 40 min integration time (B); K73N: average of 4 spectra with 20 min integration time (A) and average of 3 spectra with 15 min integration time (B). The intensities are normalized to that of the ν4 band. The spectra have been shifted along the ordinate axis to allow better visualization.
Figure S4: Unfolding profiles of the Lys72Arg and Lys73Arg mutants (●) and wild type cyt c (▼). The two variants showed almost identical profiles. Points refer to CD measurements taken at 222 nm. The experimental conditions were 20 mM Hepes + 0.1 mM EDTA, pH 7.0, and 25°C.