

Supplementary Information

Investigations were carried out to probe the reversibility of the binding process between cyt. *c* and CL. We carried out experiments on the cyt. *c*/CL complex to demonstrate that CL can be removed to yield authentic cyt. *c*. The results of these experiments are given in Figure 1 below in which we show the spectra of ferrous cyt. *c* during a cycle that takes it from the native form to the CL bound nitrosyl complex and back again.

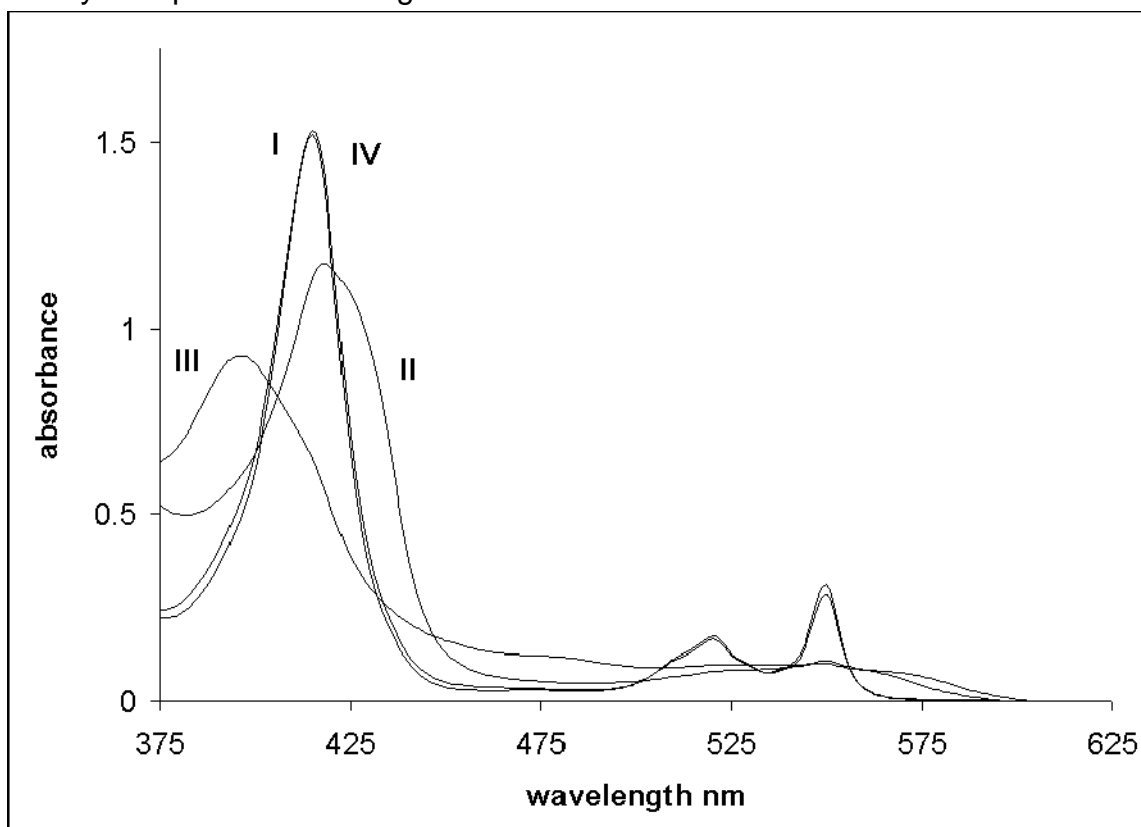


Figure 1. Spectrum I is that of native ferrous cyt. *c*, spectrum II is that of the CL complex of ferrous cyt. *c*, and spectrum III is of the ferrous NO complex. CL was removed from the NO cyt. *c*/CL adduct by incubation (~5 mins) with 250 mM NaCl and 10 mM Triton X-100, and the NO was removed by photolysis in the presence of an NO scavenger (carboxy-PTIO). The protein was then “freed” from CL and Triton etc by elution from a column containing an ion exchange resin (CM 52 cation exchange resin). The spectrum of the resulting protein, diluted to a concentration comparable to that of the original sample, is given in spectrum IV. It is clear that this spectrum is essentially identical to spectrum I showing that the transition brought about by CL is fully reversible. Conditions: [cyt. *c*]=~12 μ M, [CL]=360 μ M, [NO]=100 μ M, temperature=22^oC, buffer used was 20 mM Hepes pH 7.4.