

Spin Label EPR Studies of *E. coli* Cytochrome *bo*₃

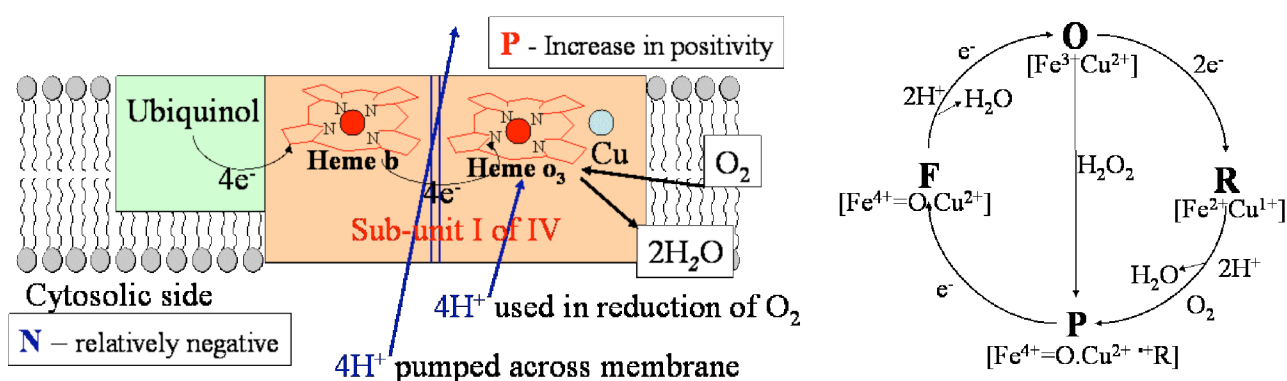
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Site-directed spin labelling (SDSL) has a wide range of applications in the analysis of protein structure, function and dynamics and can be complementary to x-ray crystallography and NMR.¹⁻³ The technique has been successfully applied to studies of high molecular weight proteins and membrane systems where preparation of crystals is difficult and NMR approaches are generally not applicable due to lack of spectral resolution.⁴⁻⁶ Cell respiration in *E. coli* is catalysed by the membrane-bound, Cytochrome *bo*₃ ubiquinol oxidase.⁷ During its catalytic cycle O₂ is reduced to H₂O, coupled with the translocation of 4 protons across the bacterial membrane. Cytochrome *bo*₃ is a 160 kDa, 4 unit, heme-copper oxidase where all the redox centres are located in sub-unit I. Unlike Cytochrome *c* oxidases, sub-unit II has no Cu_A centre, instead heme b receives electrons directly from a ubiquinol molecule. Spin label EPR is being used to investigate interactions between key parts of Cytochrome *bo*₃ and to detect any conformational changes that occur during different stages of its catalytic cycle.



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