

P007 Clustering and Dynamics of the *Escherichia coli* cytochrome *bd* terminal quinol oxidase *in vivo*.

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Oxidative phosphorylation (OXPHOS) is a multi-step process requiring the co-ordinated activity of multiple enzymes. Much is known about the genetics, biochemistry and structures of individual OXPHOS enzymes but direct observations relating to the spatial and temporal interactions of these enzymes are relatively scarce. The isolation and imaging of OXPHOS super-complexes provides strong *in vitro* evidence of organisation of OXPHOS enzymes on a nano-scale and dynamic localisation of fluorescent protein fusions of the ATP synthase and succinate dehydrogenase, expressed under the control of inducible promoters, in *Bacillus subtilis in vivo* has been reported. Here we have engineered an *Escherichia coli* strain, YTL01, where the native *cydB* allele is exchanged for a chimeric *cydB-gfp* gene and therefore expresses functional, GFP-tagged cytochrome *bd-I* terminal quinol oxidase complexes under the control of native promoters. Video-rate fluorescence imaging of YTL01 cells using a custom built epifluorescence/TIRF (Total Internal Reflection Fluorescence) microscope reveals that GFP-tagged cytochrome *bd-I* complexes form mobile clusters in the *E. coli* plasma membrane. Single particle tracking and step-wise photobleaching of clusters show that they contain 10s – 100s of cytochrome *bd-I* complexes and show both restricted and free diffusion in the membrane plane. We therefore hypothesise that in *E. coli*, OXPHOS is localised to patches of membrane (“respiration zones”) where OXPHOS enzymes congregate, and now test this hypothesis by similarly visualising other OXPHOS enzymes.