



Figure 1S. Treatment of Arabidopsis leaves with 5 mM DTT in the dark leads to an increase in the redox-activation state of mitochondrial alternative oxidase (AOX) within 1h.

To investigate whether DTT treatment of leaf discs affects the *in-vivo* thiol status of mitochondrial proteins, we analysed the dimerisation state of the mitochondrial AOX. This protein has been found previously to be activated by reduction of an intermolecular disulfide bond formed between the Cys 126 residues of the AOX homodimer (Umbach and Siedow 1993). Leaf discs were rapidly extracted using trichloroacetic acid, proteins separated using non-reducing SDS-PAGE, and AOX protein detected via immunoblot. Reduction of the inter-molecular disulfide-bridge leads to an increased mobility of the alternative oxidase protein during gel-electrophoresis since the protein is running as a dimer in the oxidised and as a monomer in the reduced form. The monomer/dimer ratio increased in discs treated with 5 mM DTT. Results are the mean \pm SD ($n = 2$ independent incubations).