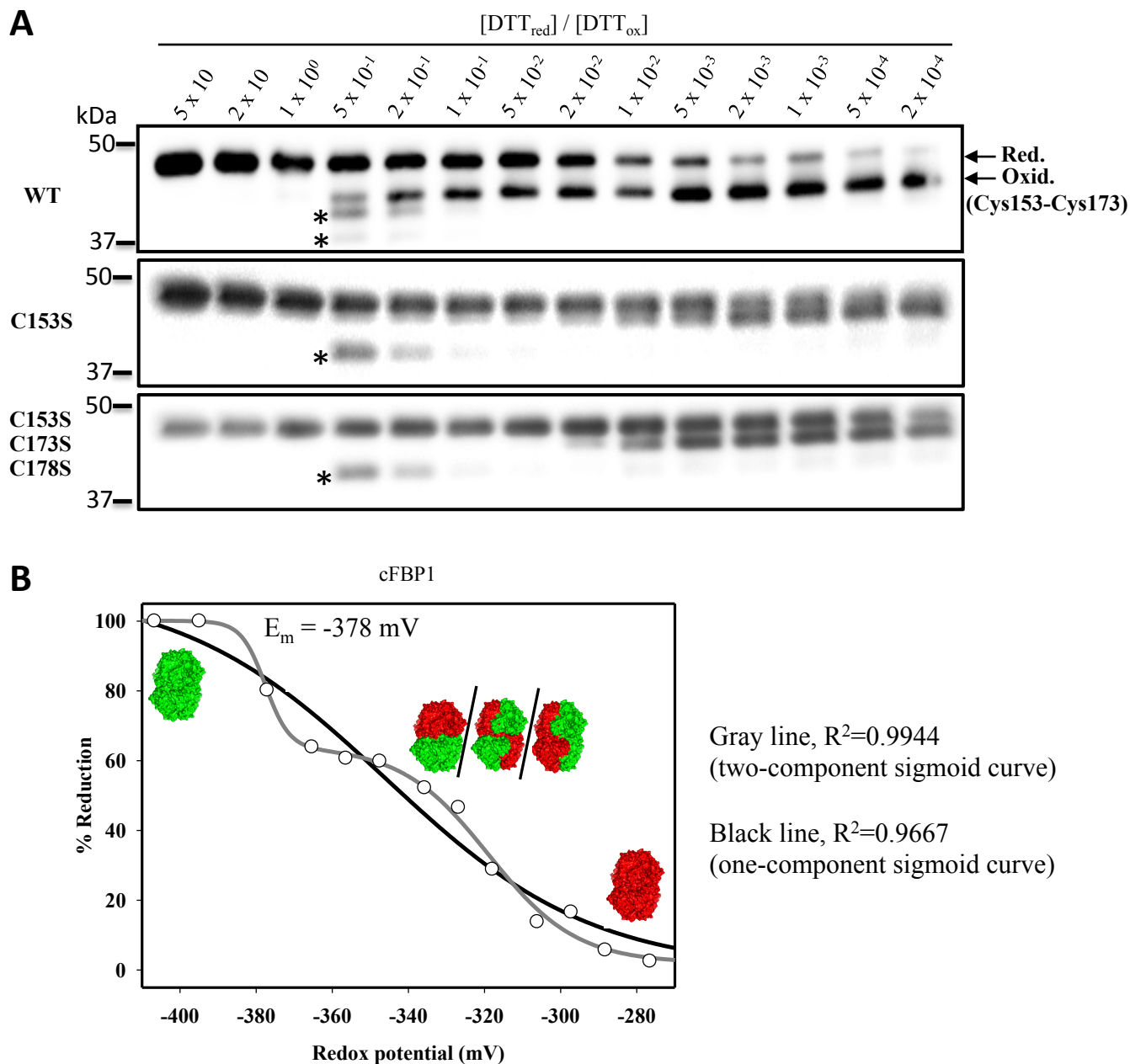


Supplemental figure 2



Redox potential of the regulatory disulphide Cys153-Cys173. A, western blot for the redox titration of the cFBP1 regulatory disulfide and redox pattern of the mutant C153S and triple mutant C153S C173S C178S. FBPase, at a final concentration of 100 $\mu\text{g/ml}$, was incubated in 50 μl of 100 mM Tris-HCl buffer (pH 8.0) that contained different DTT_{red}/DTT_{ox} ratios. Concentration of oxidized DTT was fixed at 50 mM and reduced DTT was adjusted to obtain the ratios displayed in the figure. After a 3 h incubation at room temperature, free cysteines were blocked as shown in supplemental figure 1. B, Titration curve representing the midpoint redox potentials of the disulphide bridge Cys153-Cys173. Values were obtained by quantification of the oxidized and reduced bands of the WT FBPase. For the calculation of the redox potentials the value of n in the Nerst equation was set at 2. Curves were fitted using SigmaPlot software.