Electronic Supporting Information (ESI)

PEGylation as an efficient tool to enhance cytochrome c thermostability: a kinetic and thermodynamic study

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Table S1. Thermodynamic parameters of Cyt-c catalysed reaction and reversible unfolding of native Cyt-c, Cyt-c-PEG-4 and Cyt-c-PEG-8 estimated according to Arrhenius. The concentration of non-PEGylated and PEGylated Cyt-c was 10 µM in 0.01 M potassium phosphate buffer (0.14 M of NaCl, pH 7.4). The catalytic oxidation of ABTS was performed in the temperature range of 30–100 °C (ΔT = 10 °C).

Protein	***E**** **(kJ.mol^{-1})**	**ΔH^{o}_{U} (kJ.mol^{-1})**
Cyt-c	10.22±0.40	33.82±4.92
Cyt-c-PEG-4	7.51±0.06	109.4±13.1
Cyt-c-PEG-8	8.87±0.29	58.43±3.11

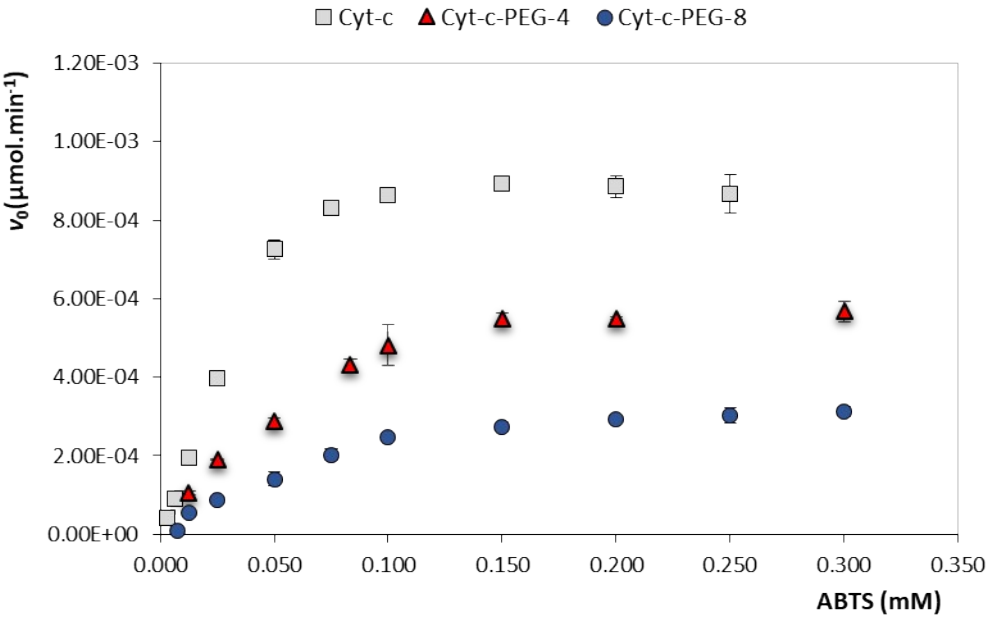

Figure S1. Kinetics of ABTS (0.025-0.300 mM) by 25 mM H_2O_2 catalyzed by Cyt-c, Cyt-c-PEG-4 or Cyt-c-PEG-8. Enzyme concentration: 0.6 mg.mL^{-1} Cyt-c.

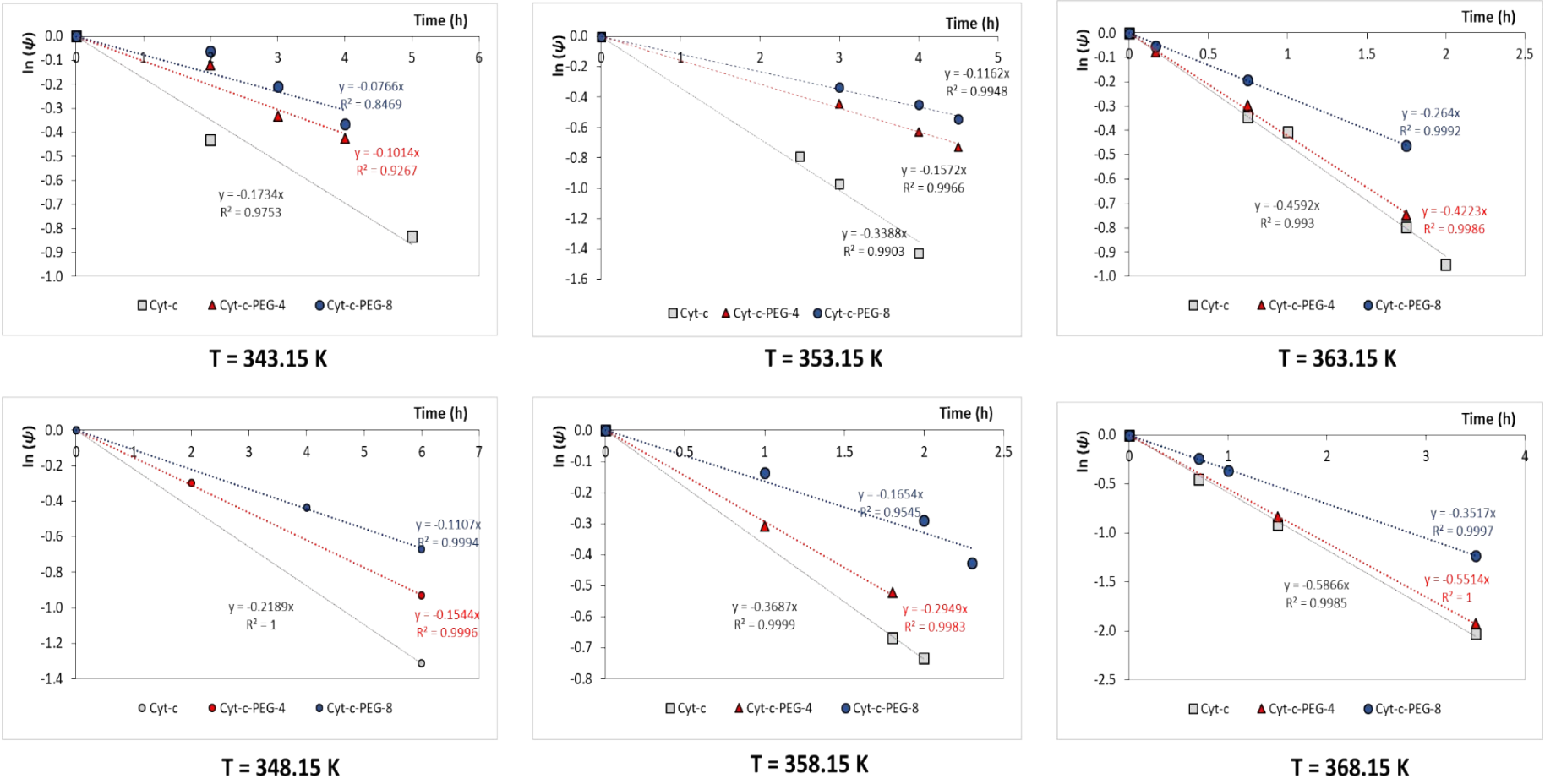

Figure S2. Semi-log plots of irreversible denaturation of Cyt-c, Cyt-c-PEG-4 and Cyt-c-PEG-8, at different temperatures. Experimental conditions: 0.24 mL of Cyt-c ($1.2x10^{-3}$ µM), 0.03 mL of ABTS (25 mM) and 0.03 mL of H_2O_2 (25 mM).