Supplementary Information

Real-time electrochemical monitoring of isothermal helicase-dependent amplification of nucleic acids

Francine Kivlehan,[a] François Mavré,[a] Luc Talini,[b] Benoît Limoges,*[a] and Damien Marchal*[a]

[a]*Laboratoire d'Electrochimie Moléculaire, UMR 7591 CNRS, Univ Paris Diderot, Sorbonne Paris Cité, 15 rue Jean-Antoine de Baïf, F-75205 Paris Cedex 13, France. E-mails: marchal@univ-paris-diderot.fr, limoges@univ-paris-diderot.fr.*

[b]*Easy Life Science, 68, Boulevard de Port-Royal, 75005 Paris Cedex 05, France.*

Real-time fluorescent-based PCR

Figure S1 shows the real-time PCR amplification curves for serial dilutions (10-fold) of the 89-bp target DNA sequence of *E. Coli*. The experimental conditions are described in the Experimental Section. Graph A in Figure S1 corresponds to the baseline corrected relative fluorescence intensity (ΔRn) of EvaGreen® dye versus amplification time, while graph B is the corresponding semi-logarithmic representation. The horizontal dashed line in graph B was used to determine the threshold cycle numbers that were next used to establish the calibration plot in C. A linear dynamic range between 2×10^3 to 5×10^9 initial target copies was determined from graph C, with a detection limit of ~10^3 initial target copies, while a PCR efficiency of 1.8 can be calculated from the slope, which is closed to the theoretical value of 2.

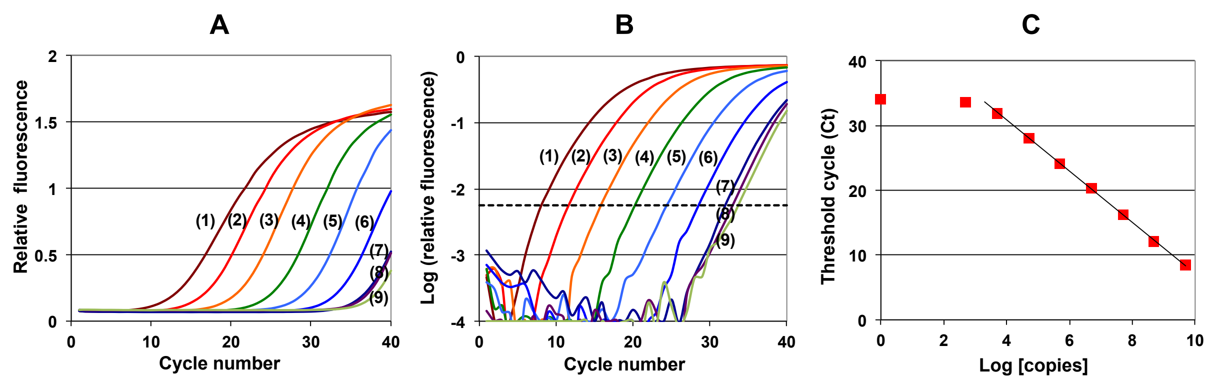

Figure S1. (A, B) EvaGreen®-based Real-time PCR amplification curves of the 89-bp DNA target. The concentration of primers in each reaction solution was 75 nM and the initial number of target DNA copies per assay was: (1) 5×10^9; (2) 5×10^8; (3) 5×10^7; (4) 5×10^6; (5) 5×10^5; (6) 5×10^4; (7) 5×10^3; (8) 5×10^2 and (9) 0 (NTC). (B) Semi-logarithmic representation of the relative fluorescence intensity as a function of PCR cycle number. (C) Calibration plot (slope: -3,96, intercept: 46.7, r = 0.9995).