

Supporting Information

Low cost platform for multiplexed electrochemical melting curve analysis

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Table S-1. Oligonucleotides used for the melting curve analysis in this study: capture probes, 21-mer targets, 124-mer targets, and the primers used to generate the ferrocene-targets by asymmetric-PCR. (The regions of the target sequences complementary to the capture probes are underlined)

Oligonucleotides	Sequences (5' to 3')	
Approach 1: 21-mer targets		
Wild type (Full complementary)	5'-Fc-CGAAGTGTGAACTAGTCCCAC-3'	
SNP at the top	5'-Fc- <u>AGA</u> AGTGTGAACTAGTCCCAC-3'	
SNP at the middle	5'-Fc-CGAAGTGTGAA <u>AT</u> AGTCCCAC-3'	
SNP at the bottom	5'-Fc-CGAAGTGTGAACTAGTCCC <u>AA</u> -3'	
Approach 2 (4 Fc-targets / 1 capture probe) and Approach 3 (1 Fc-target / 4 capture probes): 124-mer targets		
No SNP (Full complementary)	5'- AGCTCCAGAAGATAAATTACAGGCGAAGTGTGAACTAGTCCCACCACCTTAATTTCACTGTG TGTTAACTTGTAAGAAGCTGCATAATGTGTATCTTACAAGTACTAGGATACTATGACCCC- 3'	
SNP at the top	5'- AGCTCCAGAAGATAAATTACAGGCGAAGTGTGAACTAGTCCC <u>CA</u> ACCTTAATTTCACTGT GTGTTAACTTGTAAGAAGCTGCATAATGTGTATCTTACAAGTACTAGGATACTATGACCC C-3'	
SNP at the middle	5'- AGCTCCAGAAGATAAATTACAGGCGAAGTGTGAA <u>AT</u> AGTCCCACCACCTTAATTTCACTGT GTGTTAACTTGTAAGAAGCTGCATAATGTGTATCTTACAAGTACTAGGATACTATGACCC C-3'	
SNP at the bottom	5'- AGCTCCAGAAGATAAATTACAGG <u>AGA</u> AGTGTGAACTAGTCCCACCACCTTAATTTCACTGT GTGTTAACTTGTAAGAAGCTGCATAATGTGTATCTTACAAGTACTAGGATACTATGACCC C-3'	
PCR primers for Ferrocene incorporation	Fc-Forward primer	5'-Fc-AGCTCCAGAAGATAAATTACAGG-3'
	Reverse primer	5'-pho-GGGGTCATAGTATCCTAGTTG-3'
Capture probes for all approaches		
Full complementary to the target (thiol 3'-)	5'- <u>GTGGGACTAGTTCACACTTCGTTT</u> -3'-Thiocticacid	
SNP at the top	5'- <u>ATGGGACTAGTTCACACTTCGTTT</u> -3'-Thiocticacid	
SNP at the middle	5'- <u>GTGGGACTAATTCACACTTCGTTT</u> -3'-Thiocticacid	
SNP at the bottom	5'- <u>GTGGGACTAGTTCACACTTCATTT</u> -3'-Thiocticacid	
Surface control		
Fc-DNA-thiol	5'-Fc-GTGGGACTAGTTCACACTTCGTTT-3'-C6-Thiol	

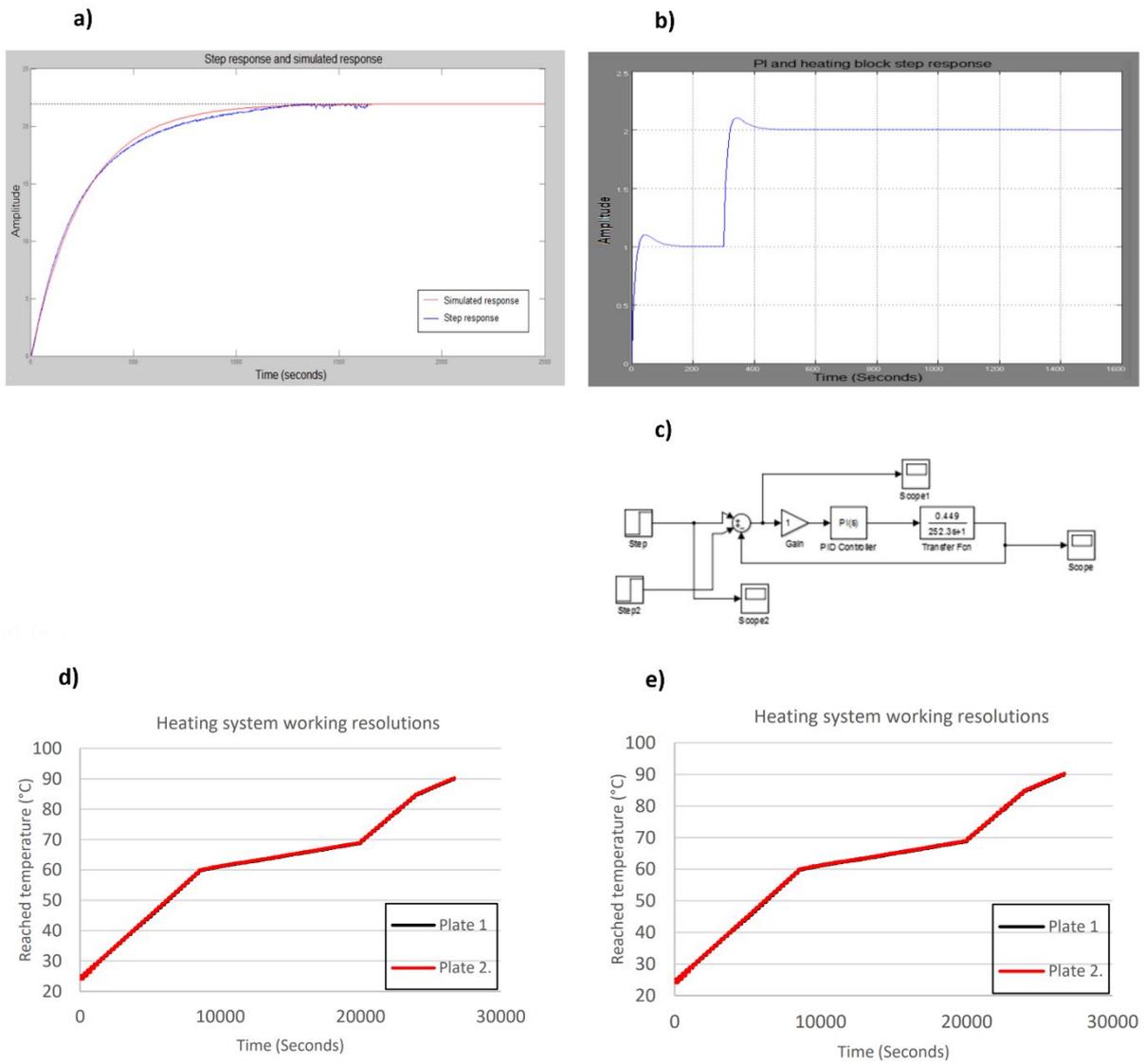


Figure S-1 (a) Step response and simulated transfer function of the heating plate 1 obtained with Matlab; (b) Modelled closed loop response of the PID controller in series with the heating block; (c) Modelled system of the PID controller in series with the transfer function plate that simulates the behaviour of the heating plate 1; (d) Example of the system's behaviour for a heating test starting at 25°C and finishing at 90°C. Both heating plates produced a stable output; (e) The heating ramp of the complete system with a glass slide – PMMA template in the middle of the heating plates.

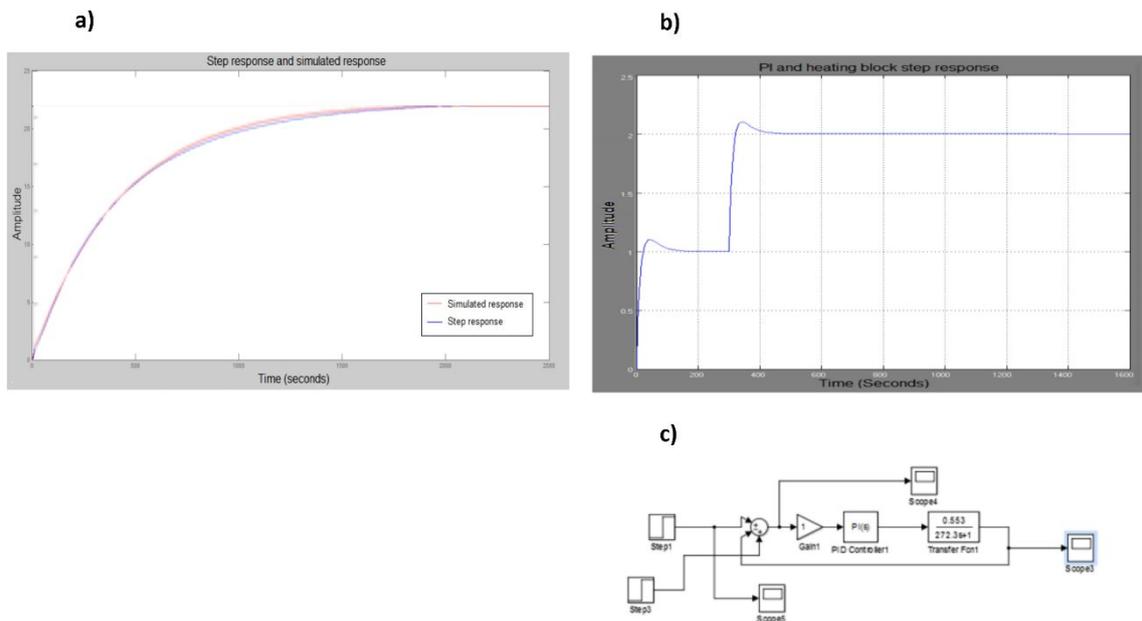


Figure S-2 (a) Step response and simulated transfer function of the heating plate 2 obtained with Matlab; (b) Modelled closed loop response of the PID controller in series with the heating block; (c) Modelled system of the PID controller in series with the transfer function plate that simulates the behaviour of the heating plate 2.

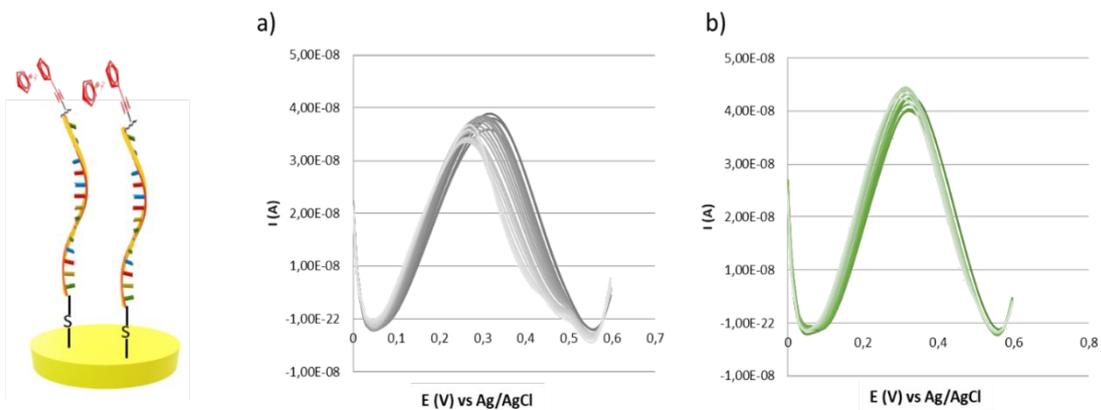


Figure S-3 (a) Repetitive SWVs of double functionalized Fc-DNA-SH on gold surface with temperature ramping; (b) Repetitive SWVs of double functionalized Fc-DNA-SH on gold surface during 1 h at 25°C.

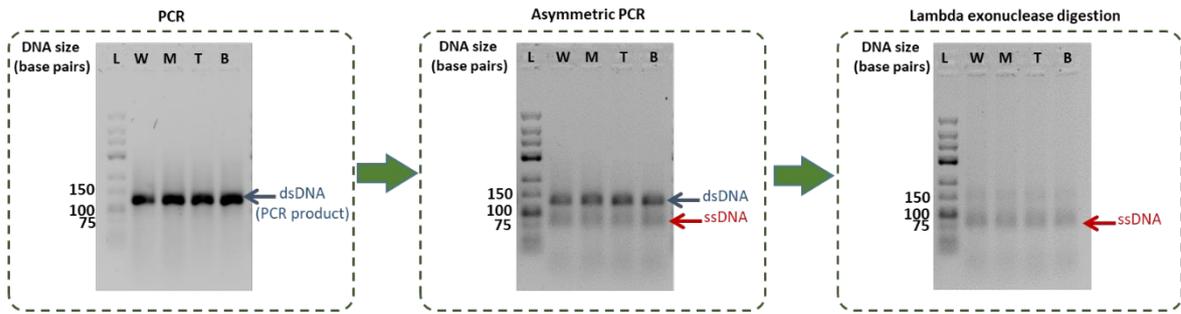


Figure S-4 Agarose gel electrophoresis after each step of the single-stranded redox labelled PCR amplicon generation based on a combination of asymmetric PCR and Lambda exonuclease digestion.