

Capture and direct amplification of DNA on chitosan microparticles in a single PCR-optimal solution.

Kunal R. Pandit*, Imaly A. Nanayakkara†, Weidong Cao,†† Srinivasa R. Raghavan*,†, Ian M. White†.

*Department of Chemical & Biomolecular Engineering, University of Maryland, College Park, MD

†Fischell Department of Bioengineering, University of Maryland, College Park, MD

††Canon US Life Sciences, Inc., Rockville MD.

Supporting Information

The supporting information includes product melt peaks and gels for amplicon validation, as well as PCR results investigating reasons for the shift in cycle threshold for direct PCR.

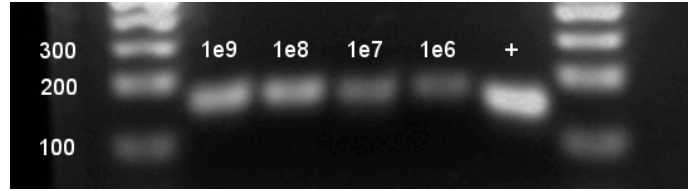


Figure S1. Direct PCR products run on a 1% agarose gel and stained with SYBR Green I. Copies of pUC19 captured by beads in reactions from Fig. 6 are listed above each lane; the positive control lane (+) contains product from solution-based PCR, and DNA ladders are in the first and last lanes.

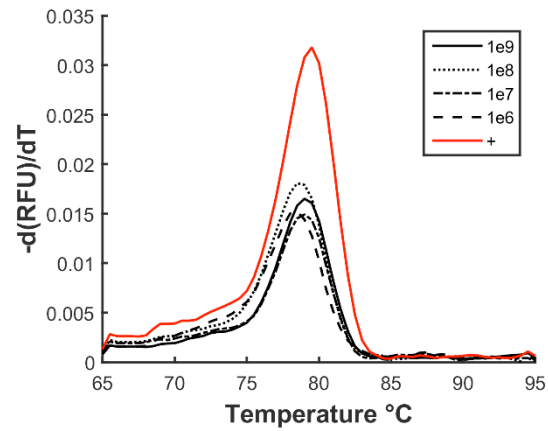


Figure S2. Melt curves from one calibration curve of PCR products from Fig. 6. The melt temperatures, 79.0 ± 0.5 °C, of the amplicons with beads in solution ($10^9 - 10^6$) match the melt temperature, 79.5 °C, of amplicons generated without beads (+).

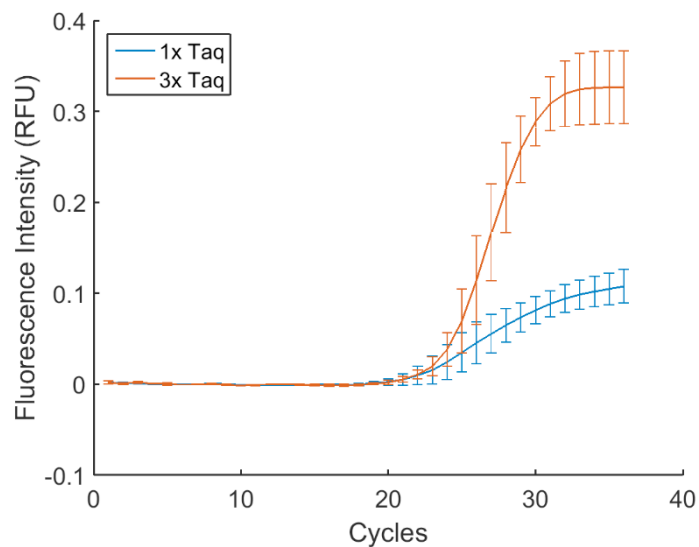


Figure S3. Amplification curves of 10^6 copies of pUC19 on chitosan beads with 1X or 3X Taq polymerase. Cycle thresholds determined for 1X and 3X polymerase were 24.06 ± 1.95 and 23.24 ± 0.82 , respectively. Error bars represent the standard deviation for $n = 3$.

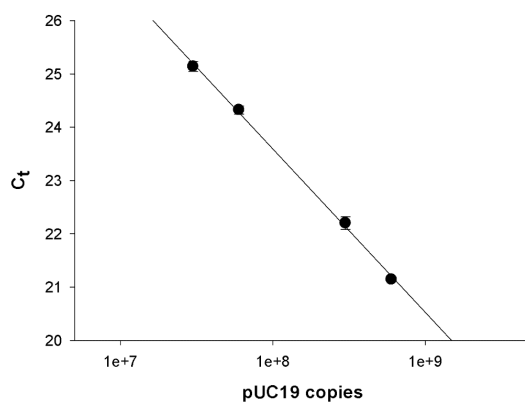


Figure S4. PCR calibration curve constructed from primers that had been first aspirated with chitosan microparticles before being added to the solution-based PCR reaction. The efficiency of the reaction was 112%, suggesting that minimal primers were absorbed onto the chitosan microparticles.