

# Smartphone-Integrated Point-of-Care Sensor for Rapid and Sensitive Malaria Detection in Resource-Limited Settings

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## Supplementary Information

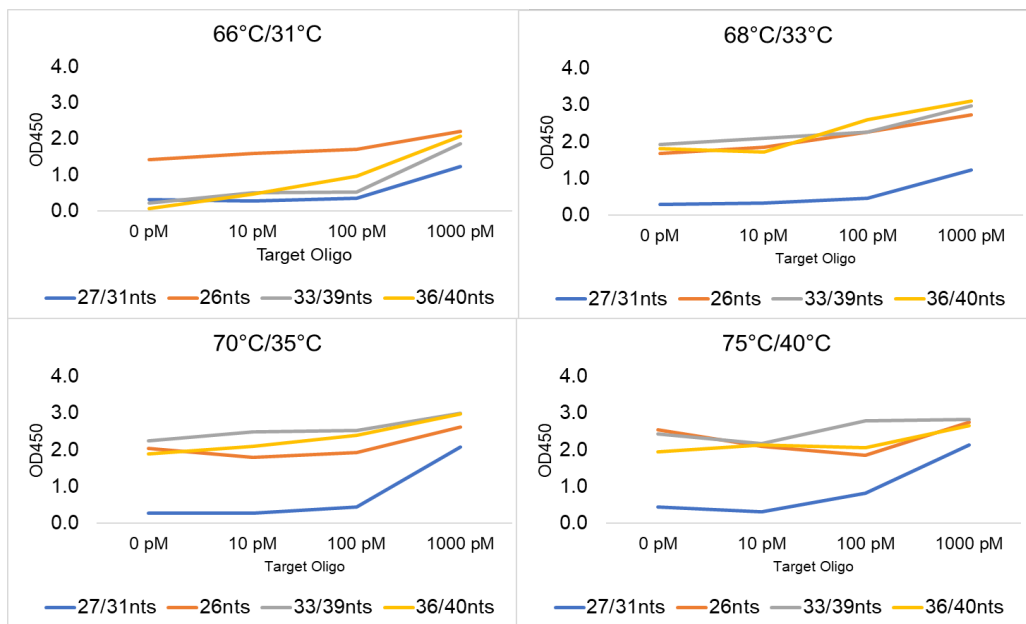
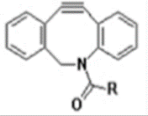
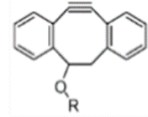
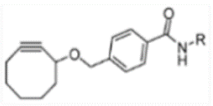
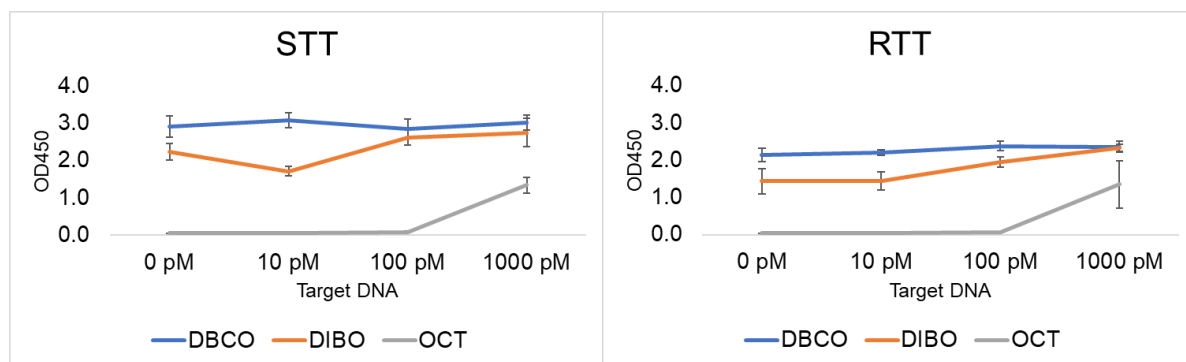


Figure S1. Primer Temperature Screen. Each set of primers was tested with four different thermocycling conditions. Primers were reacted in 50 mM KCl, 5 mM ammonium sulfate, 5% DMSO and 0.1% tween20 for 20 cycles with 30 seconds at the high (denaturing temperature) followed by 60 seconds at the low (annealing temperature).

"Click" suitable alkyne	Chem. Structure	2 <sup>nd</sup> order rate constant (M <sup>-1</sup> s <sup>-1</sup> )
Dibenzocyclooctyne (DBCO)		0.31
Dibenzocyclooctyne (DIBO)		0.057
Cyclooctyne (OCT)		0.0024

**Figure S2. SPAAC-Ligation Reaction Groups.** Dibenzocyclooctyne (DBCO/DIBO) and Cyclooctyne (OCT) reactive groups react with azide to form ligated nucleic acid. Groups are listed in order from fastest to slowest reaction rate.



**Figure S3. Comparison of SPAAC Reaction Groups.** 27/31nts primer sets labeled with either DBCO, DIBO, or OCT reactive groups were tested in samples containing specific concentrations of target DNA and 25% bovine serum, 50 mM KCl, 20 mM ammonium sulfate, 5% DMSO, and 0.1% tween-20. Samples were incubated at 95°C for 10 minutes followed by 50 cycles of denaturing temperature (66°C) for 30 seconds followed by annealing temperature (31°C) for 60 seconds (Standard Thermocycling Time (STT), left) or denaturing for 20 seconds followed by annealing for 45 seconds (Reduced Thermocycling Time (RTT), right). All samples were tested in duplicate.

**Table S1. Comparison of Chronoamperometric Results Generated by PSTrace v.5.8 or the Giner Smartphone Application.**

<b>Target (pM)</b>	<b>GC/pL</b>	<b>-<math>\mu</math>A/s</b>	<b>App Result</b>	<b>App Concentration</b>	<b>Relative Error</b>
1500	841	0.407	Negative	BLoQ	NA
700	392	0.447	Negative	BLoQ	NA
100	56.0	0.318	Negative	BLoQ	NA
25	14.0	0.215	Positive	21.7 GC/pL	-55.0%
5	2.80	0.0559	Positive	0.8 GC/pL	71.4%
1	0.560	0.0172	Positive	0.5 GC/pL	10.7%
0	0	0.0588	Positive	ALoQ	NA

*Samples were spiked with target at concentrations ranging from 1500 pM to 1 pM with unspiked sample serving as a negative control. Each sample was first measured using a potentiostat connected to a laptop running PSTrace v.5.8 software (PalmSens). Samples were then tested using the Giner smartphone application to determine the accuracy of the reported values. Only 3/6 positive samples returned calculated sample concentrations. Highly positive samples were reported as negative while the negative sample was reported as positive.*