Supplementary Information for

Smartphone dongle for simultaneous measurement of hemoglobin concentration and

detection of HIV antibodies

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Materials and Methods

Red blood cell lysis. We tested the following blood lysis agents, as blood lysis is for reproducible hemoglobin important measurements: 3-[(3-Cholamidopropyl)dimethylammonio]-1-propanesulfonate (CHAPS, Sigma), Triton X-100 (Sigma), and sodium deoxycholate (Sigma). Each lysis agent was dissolved in 1x phosphate buffered saline (PBS) or deionized water at a range of concentrations, 8 µL of the solution was placed in a microcentrifuge tube, and solvent was removed using a centrifugal evaporator. CHAPS was tested at 12, 24, 48 mM; Triton X-100 was tested at 1, 2, 4, and 10 mM; and sodium deoxycholate was tested at 1.25, 2.5, 5% (w/v). Concentrations were chosen based on previous publications^{13, 18}. To lyse blood, 8 µL of whole blood (collected within the past 3 days) was gently mixed in the microcentrifuge tube with the lysis agent. The sample was drawn into the microfluidic cassette by negative-pressure driven flow (-34 kPa). In this study, all fluid flow was generated by connecting a 60 mL-syringe to the cassette outlet, and pulling the plunger from 20 mL to 30 mL, held in place by a metal spacer between the plunger and the barrel. Each sample was observed in the microfluidic channels under 10x magnification on a bright-field microscope. Blood was considered completely lysed if there were no visible red blood cells.



Figure S1. Calibration curve for a second device showing correlation between optical density

measurements compared with Hemocue readings. Data are averages \pm SD (n = 3).

Hemocue Reading (g/dL)		OD (530 nm)		Average	Standard deviation	Coefficient of variation
0	-0.004	-0.002	-0.002	-0.003	0.001	-0.354*
0.9	0.007	0.009	0.010	0.009	0.001	0.144
1.7	0.019	0.021	0.020	0.020	0.001	0.041
3.2	0.037	0.034	0.034	0.035	0.001	0.040
3	0.034	0.034	0.035	0.034	0.000	0.014
5.2	0.058	0.059	0.057	0.058	0.001	0.014
6.3	0.068	0.068	0.071	0.069	0.001	0.020
6.8	0.077	0.078	0.078	0.078	0.000	0.006
8.5	0.096	0.098	0.100	0.098	0.002	0.017
9.3	0.105	0.104	0.103	0.104	0.001	0.008
10.7	0.115	0.113	0.117	0.115	0.002	0.014
10.4	0.112	0.114	0.119	0.115	0.003	0.026
12.9	0.141	0.139	0.141	0.140	0.001	0.007
13.7	0.145	0.149	0.150	0.148	0.002	0.015
14.9	0.149	0.156	0.159	0.155	0.004	0.027
16.4	0.170	0.173	0.166	0.170	0.003	0.017
16.9	0.178	0.179	0.180	0.179	0.001	0.005
18	0.180	0.182	0.185	0.182	0.002	0.011
18.2	0.184	0.189	0.186	0.186	0.002	0.011
19.9	0.204	0.198	0.198	0.200	0.003	0.014
21.2	0.210	0.208	0.216	0.211	0.003	0.016

Table S1. Table showing data used to create calibration curve in Figure 1E. OD measurements from 3 trials, average OD, standard deviation, and coefficient of variation for each concentration are displayed. *Was not included to calculate average coefficient of variation.

Hemocue Reading (g/dL)		OD (530 nm)		Average	Standard deviation	Coefficient of variation
0	0.000	0.001	-0.001	0.000	0.001	#DIV/0!*
2.5	0.033	0.032	0.031	0.032	0.001	0.026
4.85	0.059	0.060	0.061	0.060	0.001	0.014
7.4	0.095	0.090	0.086	0.090	0.004	0.041
8.3	0.099	0.098	0.098	0.098	0.000	0.005
9.75	0.112	0.114	0.111	0.112	0.001	0.011
10.75	0.128	0.126	0.125	0.126	0.001	0.010
11.55	0.133	0.136	0.132	0.134	0.002	0.013
13.05	0.146	0.142	0.143	0.144	0.002	0.012
13.5	0.161	0.162	0.149	0.157	0.006	0.038
15.15	0.165	0.163	0.165	0.164	0.001	0.006
16.6	0.184	0.182	0.179	0.182	0.002	0.011
18.55	0.193	0.198	0.194	0.195	0.002	0.011
20.4	0.206	0.210	0.209	0.208	0.002	0.008
22.2	0.224	0.225	0.221	0.223	0.002	0.008

Table S2. Table showing data used to create calibration curve in Figure S2. OD measurements from 3 trials, average OD, standard deviation, and coefficient of variation for each concentration are displayed. *Was not included to calculate average coefficient of variation.

	Dongle Results		Commercial Test		
Sample #	HIV (signal to cutoff)	Estimated Hemoglobin (g/dL)	HIV Immunoassay (Abbott ARCHITECT)	Hemoglobin (g/dL) (Hemocue)	
1	0.90	21.2	Pos	21.7	
2	1 20	20.9	Pos	21.8	
3	1.46	18.1	Pos	18.7	
4	1.61	18.1	Pos	18.3	
5	1.47	19.1	Pos	18.3	
6	1.65	21.0	Pos	21.3	
7	1.33	18.8	Pos	18.8	
8	1.78	16.4	Pos	16.2	
9	1.81	17.9	Pos	17.8	
10	1.33	17.3	Pos	16.6	
11	1.09	19.7	Pos	20.0	
12	1.69	19.1	Pos	19.1	
13	1.39	19.3	Pos	19.7	
14	0.61	12.9	Neg	12.0	
15	0.73	16.0	Neg	15.9	
16	0.52	14.9	Neg	14.5	
17	0.30	14.2	Neg	13.8	
18	0.38	17.3	Neg	17.5	
19	1.77	17.5	Pos	18.2	
20	1.75	15.3	Pos	14.9	
21	1.29	17.8	Pos	18.3	
22	1.56	17.2	Pos	17.2	
23	1.47	16.8	Pos	17.2	
24	1.67	13.3	Pos	12.9	
25	0.71	15.6	Neg	14.3	
26	0.74	16.6	Neg	15.5	
27	0.91	18.6	Neg	18.7	
28	0.52	15.5	Neg	15.6	
29	0.39	17.0	Neg	16.6	
30	0.51	17.3	Neg	17.9	
31	0.45	16.0	Neg	16.0	
32	0.11	14.7	Neg	14.4	
33	0.43	13.8	Neg	13.3	
34	0.50	17.0	Neg	18.1	
35	0.42	17.9	Neg	18.3	
36	0.25	15.7	Neg	16.2	
37	0.57	15.5	Neg	15.5	
38	1.28	17.1	Neg	15.6	

Table S3. Table showing HIV immunoassay results obtained from the dongle, estimatedhemoglobin concentration obtained from the dongle, HIV status, and hemoglobin concentrationfor 38 clinical samples.