

Electronic Supplementary Information

Paper Microchip with Graphene-modified Silver Nano-composite Electrode for Electrical Sensing of Microbial Pathogens

Mohammadali Safavieh^a, Vivasvat Kaul^{a,†}, Sultan Khetani^{a,†}, Anupriya Singh^a, Karan Dhingra^a, Manoj Kumar Kanakasabapathy^a, Mohamed Shehata Draz^{a,b}, Adnan Memic^c, Daniel R. Kuritzkes^{d,e}, Hadi Shafiee^{a,e*}

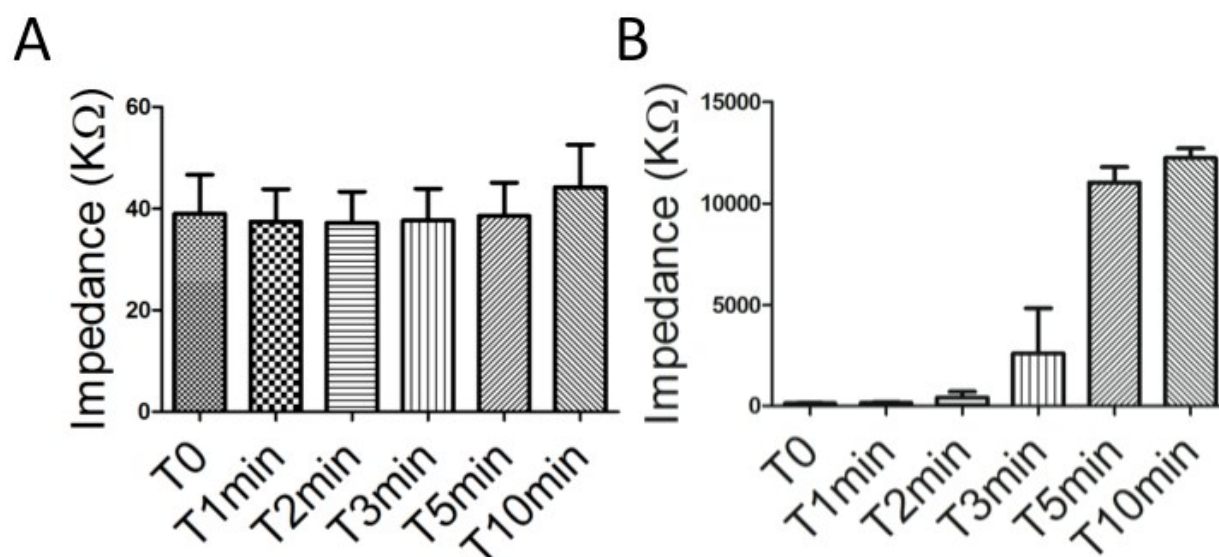


Figure S1. Chip stability evaluation through monitoring impedance magnitude of DI water over time. (A) Impedance magnitude of DI water on a cellulose paper pad at 10 KHz over 10 min incubation time and at 0, 1 min, 2 min, 3 min, 5 min, and 10 min. (B) Impedance magnitude of DI water on a Whatman chromatography over 10 min incubation time and at 0, 1 min, 2 min, 3 min, 5 min, and 10 min. The impedance magnitude of the DI samples on the Whatman chromatography paper changed significantly over 10 min incubation time, which may be due to sample evaporation.

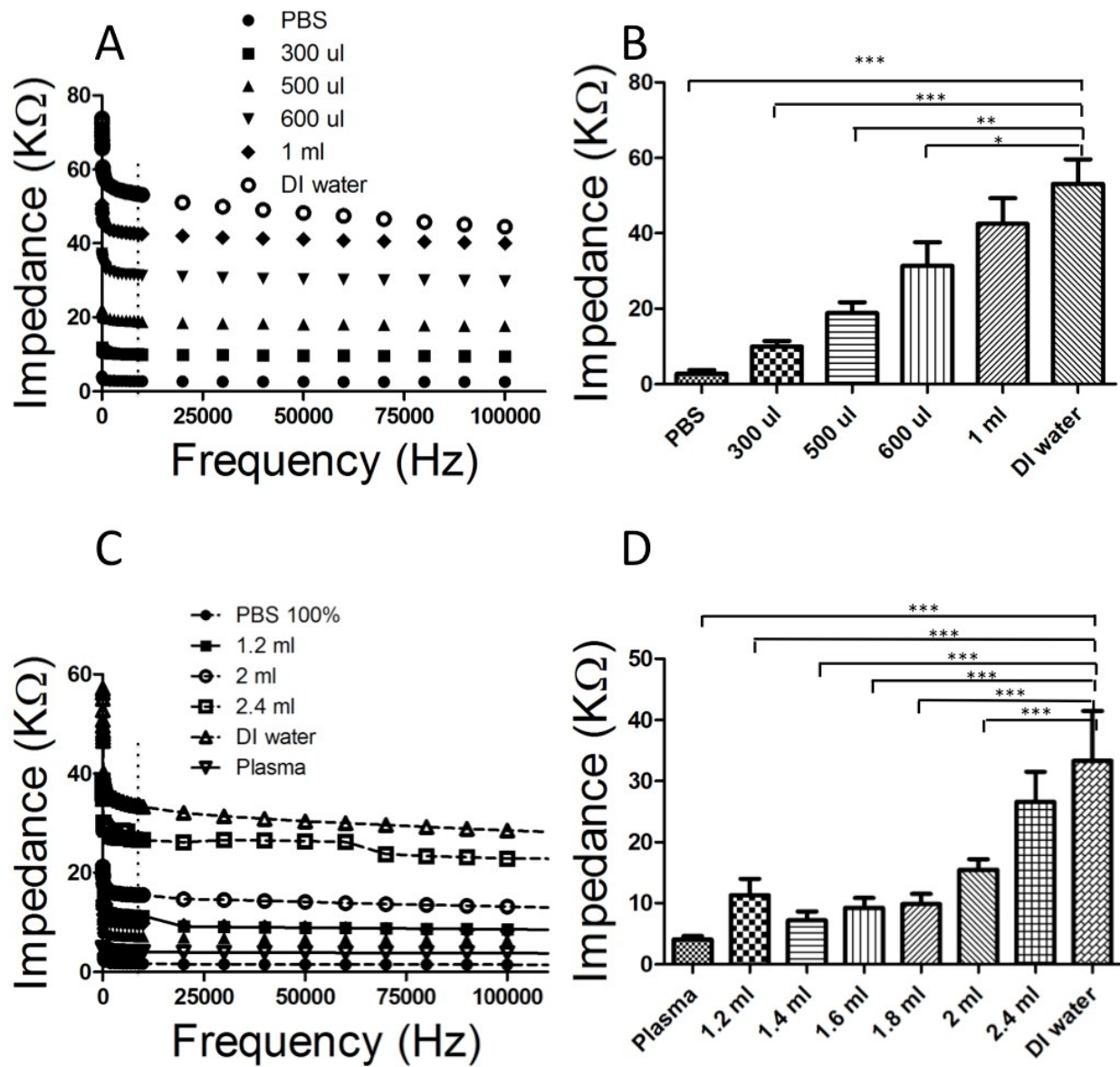


Figure S2. Washing step optimization. (A) Impedance spectroscopy of the cellulose paper chip after each washing step when 100% PBS was used as an initial sample. (B) Impedance magnitude of the solution on cellulose chips at 10 KHz. 1 ml DI water was required to completely remove the cellulose paper chip when 100% PBS was used as an initial sample. (C) Impedance spectroscopy of solution on cellulose paper chip when plasma was used as the initial sample. (D) Impedance magnitude of the chips with different wash volumes (1.2 ml, 1.4 ml, 1.6 ml, 1.8 ml, 2 ml, and 2.4 ml) at 10 KHz. 2.4 ml of DI water was required to completely remove the 100% PBS from the cellulose chip. All error bars are SEM based on repeating the experiments three times (n=3).

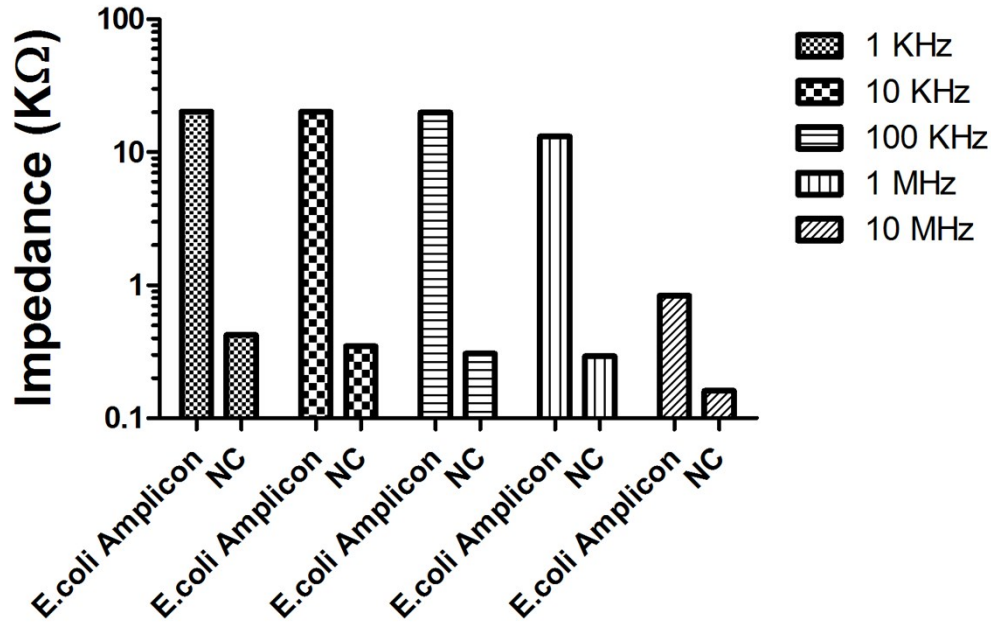


Figure S3. LAMP electrical signal optimization and comparison of impedance magnitude between *E.coli* amplicon and negative control at different frequencies.

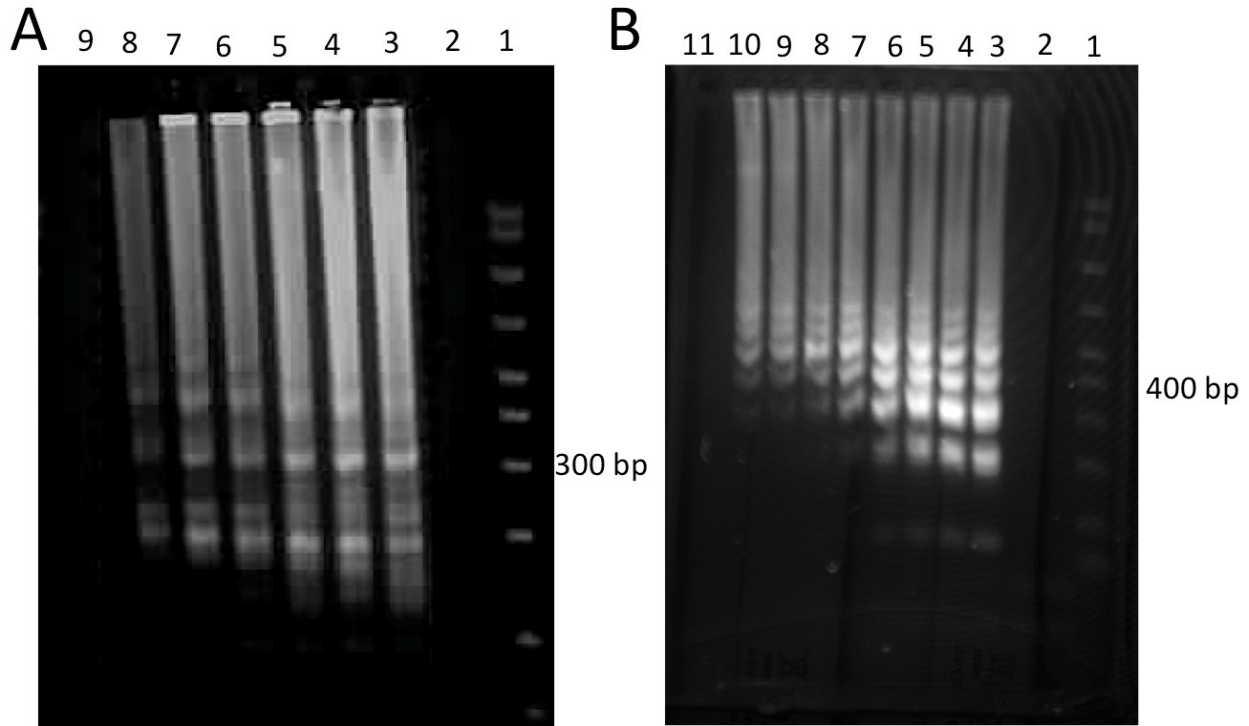


Figure S4. Gel electrophoresis image of *p24 HIV* and *E.coli Tuf* gene after 40 minutes amplification using RT-LAMP and LAMP. (A) Gel image of post processing RT-LAMP amplicon consisting of (1) 2000 bp molecular ladder, (2) DI water and negative control, (3) 1 ng/ μ l *RNA HIV-1*, (4) 100 pg/ μ l *RNA HIV-1*, (5) 10 pg/ μ l *RNA HIV-1*, (6) 1 pg/ μ l *RNA HIV-1*, (7) 100 fg/ μ l *RNA HIV-1*, (8) 10 fg/ μ l *RNA HIV-1*, (9) 10 fg/ μ l *RNA HIV-1*. (B) Gel image of LAMP amplicon of *E.coli Tuf* gene consisting of (1) (1) 2000 bp molecular ladder, (2) DI water and negative control, (3) 10 ng/ μ l *E.coli* DNA, (4) 1 ng/ μ l *E.coli* DNA, (5) 100 pg/ μ l *E.coli* DNA, (6) 10 pg/ μ l *E.coli* DNA, (7) 1 pg/ μ l *E.coli* DNA, (8) 100 fg/ μ l *E.coli* DNA, (9) 10 fg/ μ l *E.coli* DNA, (10) 1 fg/ μ l *E.coli* DNA and (11) 0.1 fg/ μ l *E.coli* DNA.

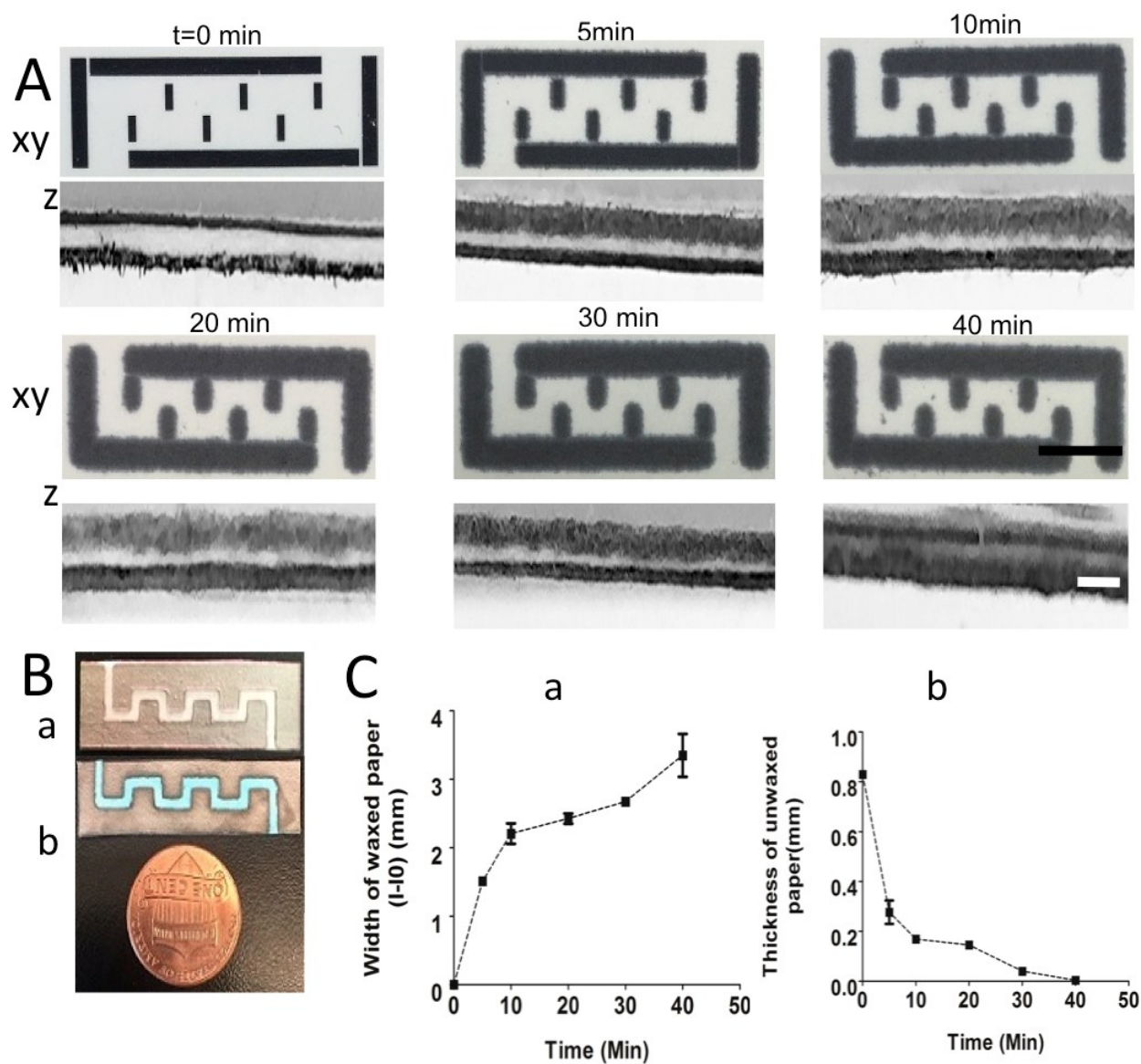


Figure S5. Wax printing optimization on cellulose paper pads. (A) Printed wax spreads on the surface and through the thickness of the paper chips over heating time (90°C). Paper chips were placed in the oven for 20 min and then flipped and incubated for another 20 min. The scale for x is 1 cm. The scale for z direction is 8 mm. (B) Image of (a) wax printed cellulose paper chip with printed graphene-modified silver electrodes. (b) a tested paper chip with DI water. (C) (a) Expanded width of printed wax on the x direction. (b) Width of non-waxed region on cellulose paper during an incubation of 40 min.

Table S1. LAMP and RT-LAMP Primer sequences for amplification of *Tuf* gene of *E.coli* and P24 in HIV-1.

Primers name	Primers Sequences	Size (bp)
<i>E.coli Tuf</i> gene		
F3	CTG CTG GGT CGT CAG GTA	18
B3	GGA TTT TCG CTT CCC ACT CT	20
LF	GGA TTT TCG CTT CCC ACT CT	20
LB	CGA CGA CAC TCC GAT CGT T	19
FIP	AGC AGC TCT TCG TCA TCA ACC AGG CGT TCC GTA CAT CAT CG	41
BIP	TGT CTC AGT ACG ACT TCC CGG GCG CTT TCA GAG CAG AAC CAC	42
<i>HIV-1 P24</i>		
F3	ATT ATC AGA AGG AGC CAC C	19

B3	CAT CCT ATT TGT TCC TGA AGG	21
LF	TTT AAC ATT TGC ATG GCT GCT TGA T	25
LB	GAG ATC CAA GGG GAA GTG A	19
FIP	CAG CTT CCT CAT TGA TGG TTT CTT TTT AAC ACC ATG CTA AAC ACA GT	47
BIP	TGT TGC ACC AGG CCA GAT AAT TTT GTA CTG GTA GTT CCT GCT ATG	45

Table S2. Material cost analysis for the viral immunoassay cellulose paper microchip using electrical sensing.

Material	Cost (cents)
Cellulose paper	6
Anti-gp-120	160
Silver/graphene nano-composite	4
DSA	6
Total	176

Table S3. Material cost analysis for the nucleic acid test for HIV-1 cellulose paper microchip using electrical sensing.

Material	Cost (cents)
Cellulose paper	6
Primers	52
LAMP reagent	168
Silver/graphene nano-composite	4
DSA	6
Total	226