

Supplementary material for

Novel pH sensing semiconductor for point-of-care detection of HIV-1 viremia

Gurrala, R¹, Lang, Z², Shepherd, L², Davidson, D², Harrison, E², McClure, M¹, Kaye, S¹, Toumazou, C^{2,3}, Cooke, GS¹

1 Division of Infectious Diseases, Imperial College, London

2 DNA Electronics Ltd, Wood Lane, London

3 Department of Bioengineering, Imperial College, London

Supplementary data

Supplementary table 1- Primer sequences used in pH-LAMP reactions generating a primary amplicon of 317bp

Primer designation	Sequence	Location ¹
F3	5'- CCC TAC AAT CCC CAA AG – 3'	4199-4215
F2	5'- ATG GCA GTA TTC ATY CAC AAT TTT AA– 3'	4307-4332
FLP	5'- CCA ATC CCC CCT TTT– 3'	4336-4350
F1C	5'- TAT YCT I ² TC CCC TGC ACT GTA CCC C– 3'	4351-4370
R1C	5'- CGG GTT TAT TAC A– 3'	4445-4457
RLP	5'- GAC AGC AGA GAI CC– 3'	4460-4473
R2	5'- AGY TTT GCT GGT CCT TTC CAA– 3'	4477-4497
R3	5'- CCT TCA CCT TTC CA– 3'	4502-4515
FIP	5'-TAT YCT 6TC CCC TGC ACT GTA CCC <u>CTT TT</u> ³ A TGG CAG TAT TCA TYC ACA ATT TTA A – 3'	
RIP	5'-CGG GTT TAT TAC <u>ATT TTA</u> GYT TTG CTG GTC CTT TCC AA– 3'	

¹ Location in GenBank K03455 ² inosine ³ thymidine spacer

Supplementary table 2- Clade distribution of clinical samples used for validation

Clade	Samples analysed in “tube assay”	Samples analysed in “chip assay”
A	52 (5.2%)	14 (8.5%)
AE	18 (1.8%)	5 (3%)
AG	58 (5.8%)	16 (9.7%)
B	581 (58.6%)	19 (11.5%)
C	135 (13.6%)	83 (50.6%)
D	34 (3.4%)	8 (4.8%)
F	6 (0.6%)	0
G	21 (2.1%)	4 (2.4%)
H	2 (0.2%)	0
J	4 (0.4%)	0
K	1 (0.1%)	0
mixed	79 (7.9%)	10 (6%)
Unknown	0	5 (3%)
Total	991	164

Supplementary table 3- Comparison of clade distribution of samples included in the assays with failed pH RT-lamp tube assay reactions **(a)** and on-chip assay reactions **(b)** performed on clinical samples. Only the samples with viral load > 1000 copies per ml of plasma are represented above.

a

Clade	Proportion of total	Proportion of failures
A	5.2%	0%
AG	5.8%	14.3%
B	58.6%	58.9%
C	13.6%	12.5%
D	3.4%	5.3%
Other	13.6%	8.9%

b

Clade	Proportion of total	Proportion of failures
A	6.25%	0%
AG	10.4%	10%
AE	2.0%	50%
B	9.3%	0%
C	57.2%	8.1%
D	6.25%	33.33%
G	2.0%	100%
Other	6.25%	16.6%

Supplementary figure 1a-d- Optimisation of pH RT-LAMP reaction conditions for dNTP (a) Betaine (b), MgSO₄ (c) and KCl (d). 1000 copies of RNA standard was used as template for the reactions. The concentration that produced lowest time to detection in minutes was selected for the final protocol.



