

## SUPPLEMENTAL INFORMATION

### Optimization of spotting conditions

Spotting conditions were optimized for certain proteins/peptides to fit the spot size, shape and adherence to the glass surface required for accurate measurements. To reduce the spot size and obtain round-shaped compact spots the printing was performed at lower protein/peptide concentrations or using printing buffers with increased salt content (i.e. 300 mM NaCl in 2 x PBS): CA, NEF, VIF, Phlp 5a, gp41, NC and TAT (0.33 mg/ml, phosphate buffer pH 8.4); PR, HSA, Phl p2, Bos d6, Bos d8, Can f2, Der p1, Der p2, Jug r2, Ses i1 (0.25 mg/ml, phosphate buffer pH 8.4); 120/08, 11, 12, 14, 41/02,03,15, VIF/06 (0.5 mg/ml, PBS pH 7.2); 41/06, MA, VP1 89 (0.33 mg/ml, PBS pH 7.2); 41/04, 41/05, VPU/03, REV/04 (0.25 mg/ml, PBS pH 7.2); 120/17, 41/08, REV/01 (0.125 mg/ml, PBS pH 7.2); VPU/02 (0.125 mg/ml, 2 x PBS pH 7.2); gp120 (0.25 mg/ml, MOPS pH 8.0); 41/07 (0.065 mg/ml, MOPS pH 6.5).

To increase adherence to the glass surface IN was spotted in MOPS PH 8.0, 1mM DTT, 0.01% SDS, 0.33 mg/ml and RR was immobilized through spotting in MOPS pH 8.0, 0.2 mM EDTA, 0.01% SDS, 1 mM spermidin at 0.33 mg/ml.

**Table S1.** Comparison of IgG antibody levels measured on the HIV chip for HIV-positive and HIV-negative serum samples.

HIV component	HIV positive ISU Median (min-max)	HIV negative ISU Median (min-max)	Mann Whitney U P value	Area under ROC
120/01	2.98 (0.31-15.82)	2.18 (0.32-20.81)	ns	0.641
120/02	0.00 (0.00-0.77)	0.17 (0.00-0.96)	<0.0001	0.818
120/03	0.04 (0.00-0.97)	0.27 (0.03-1.96)	0.0002	0.804
120/04	0.00 (0.00-5.00)	1.06 (0.00-21.13)	<0.0001	0.886
120/05	0.16 (0.00-1.76)	0.43 (0.00-12.39)	0.0315	0.679
120/06	0.16 (0.00-1.48)	1.22 (0.00-6.77)	<0.0001	0.887
120/07	0.05 (0.00-1.34)	0.26 (0.00-19.76)	0.0053	0.729
120/08	0.54 (0.09-2.97)	1.00 (0.12-30.18)	ns	0.662
120/09	0.22 (0.00-0.41)	1.29 (0.23-9.09)	<0.0001	0.985
120/10	0.59 (0.08-7.53)	1.72 (0.27-31.91)	<0.0001	0.816
120/11	0.24 (0.05-3.01)	1.33 (0.23-5.19)	<0.0001	0.876
120/12	0.12 (0.00-0.68)	0.58 (0.07-11.97)	<0.0001	0.851
120/13	0.00 (0.00-1.72)	0.28 (0.00-12.16)	<0.0001	0.867
120/14	0.38 (0.09-3.29)	8.91 (0.50-39.06)	<0.0001	0.975
120/15	0.29 (0.02-1.90)	123.5 (0.37-131.89)	<0.0001	0.994
120/16	0.42 (0.12-0.97)	37.13 (1.89-100.24)	<0.0001	1.000
120/17	0.60 (0.00-0.54)	6.71 (0.07-25.39)	<0.0001	0.989
120/18	0.08 (0.00-96.02)	7.01 (0.22-43.12)	<0.0001	0.917
120/19	0.11 (0.00-0.63)	5.05 (0.09-25.65)	<0.0001	0.968
120/20	0.72 (0.14-2.02)	2.59 (0.29-19.33)	<0.0001	0.853
120/21	0.2 (0.01-0.48)	3.17 (0.09-22.57)	<0.0001	0.959
120/22	0.00 (0.00-1.46)	0.38 (0.00-6.70)	<0.0001	0.897
120/23	0.04 (0.00-3.64)	1.40 (0.05-7.78)	<0.0001	0.890
120/24	0.11 (0.00-1.67)	25.46 (0.33-132.27)	<0.0001	0.981
41/01	0.07 (0.00-0.21)	0.70 (0.00-9.81)	<0.0001	0.942
41/02	0.28 (0.05-1.32)	3.78 (0.09-32.30)	<0.0001	0.970
41/03	0.23 (0.03-8.36)	2.06 (0.17-31.49)	<0.0001	0.850
41/04	0.54 (0.06-1.85)	85.50 (1.35-127.81)	<0.0001	0.998
41/05	0.27 (0.09-0.88)	77.16 (0.52-113.44)	<0.0001	0.995
41/06	0.45 (0.06-1.06)	17.97 (0.75-91.95)	<0.0001	0.997
41/07	0.4 (0.06-1.12)	7.81 (0.15-118.43)	<0.0001	0.983
41/08	0.12 (0.00-1.10)	3.40 (0.25-61.87)	<0.0001	0.953
41/09	0.14 (0.00-0.82)	2.02 (0.13-24.34)	<0.0001	0.972
41/10	0.85 (0.15-5.18)	2.65 (0.77-10.24)	<0.0001	0.836
41/11	0.00 (0.00-3.21)	1.22 (0.00-131.62)	<0.0001	0.912
41/12	0.10 (0.00-0.20)	0.87 (0.01-9.39)	<0.0001	0.976
41/13	0.09 (0.01-0.57)	1.33 (0.10-18.51)	<0.0001	0.967
41/14	0.1 (0.02-0.76)	1.16 (0.04-74.77)	<0.0001	0.938
41/15	0.18 (0.02-1.16)	0.56 (0.08-61.77)	0.0003	0.792
41/16	0.04 (0.00-0.22)	1.10 (0.04-22.51)	<0.0001	0.988
41/17	0.00 (0.00-0.07)	0.78 (0.00-99.44)	<0.0001	0.902
gp120	0.14 (0.00-0.57)	19.55 (0.13-53.02)	<0.0001	0.991
gp41	0.17 (0.00-1.96)	4.08 (1.24-65.50)	<0.0001	0.972
MA	0.23 (0.00-4.86)	45.86 (0.06-131.63)	<0.0001	0.984
CA	0.99 (0.15-7.31)	123.00 (1.05-132.75)	<0.0001	0.993
NC	0.45 (0.00-3.95)	17.97 (0.00-18.58)	<0.0001	0.945
PR	0.55 (0.18-8.11)	14.49 (0.47-71.57)	<0.0001	0.923
RR	0.07 (0.00-7.06)	0.46 (0.00-6.56)	0.0007	0.774
IN	0.21 (0.05-0.94)	6.10 (0.20-45.92)	<0.0001	0.990
NEF	2.24 (0.16-10.11)	5.45 (1.40-53.81)	0.0002	0.802
TAT	1.36 (0.00-8.81)	2.16 (0.32-129.85)	0.0443	0.668
VIF	0.20 (0.02-2.91)	0.57 (0.06-4.35)	0.0085	0.717
VIF/01	0.10 (0.05-0.52)	0.48 (0.08-3.30)	<0.0001	0.920
VIF/02	0.08 (0.00-0.47)	0.29 (0.00-1.30)	<0.0001	0.831
VIF/03	0.11 (0.01-0.50)	0.41 (0.07-4.09)	<0.0001	0.886
VIF/04	0.03 (0.00-16.22)	0.19 (0.00-2.57)	<0.0001	0.890
VIF/05	0.06 (0.00-20.33)	0.40 (0.00-23.81)	0.0166	0.698
VIF/06	0.15 (0.00-0.82)	0.61 (0.00-5.06)	<0.0001	0.883
VIF/07	0.37 (0.00-17.57)	0.82 (0.00-18.54)	0.0495	0.664
VIF/08	0.07 (0.01-1.32)	0.21 (0.00-3.14)	ns	0.651
VPR/01	0.03 (0.00-0.68)	0.195 (0.00-1.65)	<0.0001	0.836
VPR/02	0.09 (0.00-0.75)	0.40 (0.00-4.03)	<0.0001	0.830
VPR/03	0.02 (0.00-0.61)	0.16 (0.00-0.84)	0.0008	0.772
VPR/04	0.10 (0.04-0.71)	0.24 (0.05-1.56)	0.0067	0.723
VPU/01	0.02 (0.00-0.16)	0.54 (0.00-29.67)	<0.0001	0.911
VPU/02	0.07 (0.00-0.65)	1.57 (0.02-53.26)	<0.0001	0.905
VPU/03	0.13 (0.02-0.42)	0.46 (0.10-2.65)	<0.0001	0.895
REV/01	0.06 (0.00-0.38)	0.43 (0.07-68.71)	<0.0001	0.941
REV/02	0.15 (0.00-3.31)	0.53 (0.08-24.07)	<0.0001	0.820
REV/03	0.16 (0.00-4.14)	0.75 (0.02-32.88)	0.0005	0.783
REV/04	0.12 (0.00-2.85)	0.81 (0.10-25.25)	<0.0001	0.886
REV/05	0.05 (0.00-0.80)	0.20 (0.01-0.95)	0.0186	0.695

**Fig. S1.** Microarray-based mapping of HIV-specific IgG responses in African and European HIV-infected individuals and controls. IgG levels (y-axes, ISAC standardized units, ISU) to micro-arrayed peptides from HIV accessory proteins (a) as well as to VP1 and allergens (b) are shown for African HIV-positive patients (n=15), European HIV-positive patients (n=15) and controls (n=15). Median IgG levels are indicated for each peptide/protein (horizontal lines).

**Fig. S2.** IgG reactivity profiles in sequential serum samples from HIV-infected individuals (e.g., 1a, 1b, 1c etc.). (a) IgG levels towards HIV-derived peptides and proteins determined by microarray, expressed in ISAC standardized units. (b) IgG levels towards selected HIV-derived peptides and proteins measured by ELISA, expressed in optical density. For both assays, IgG levels are colour-coded with increasing intensities from white to red and the reactivity of negative controls is expressed as mean IgG reactivity + 3 standard deviations (SD) on the lowest row.

**Fig. S1**



