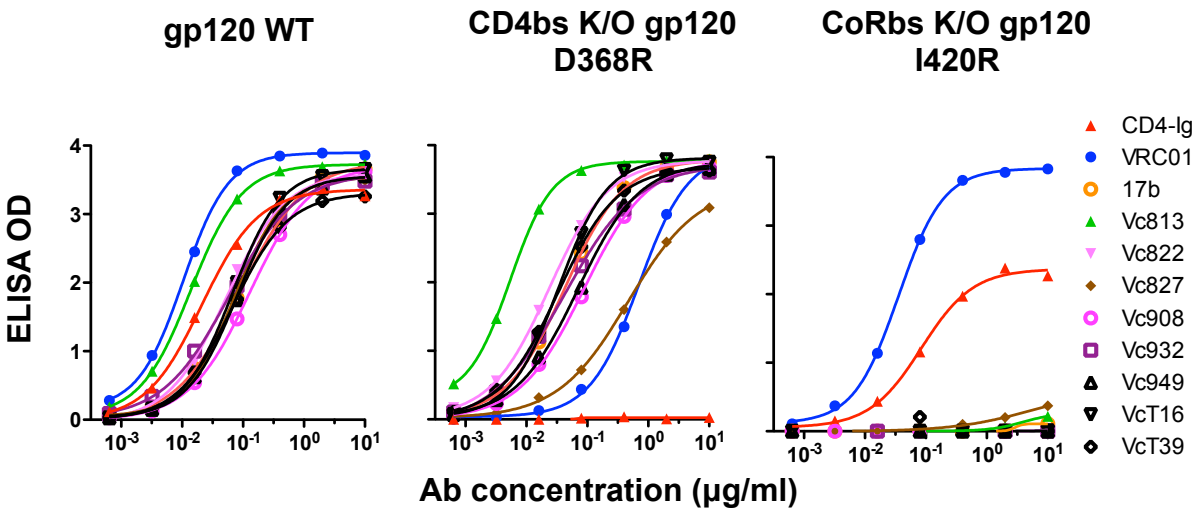
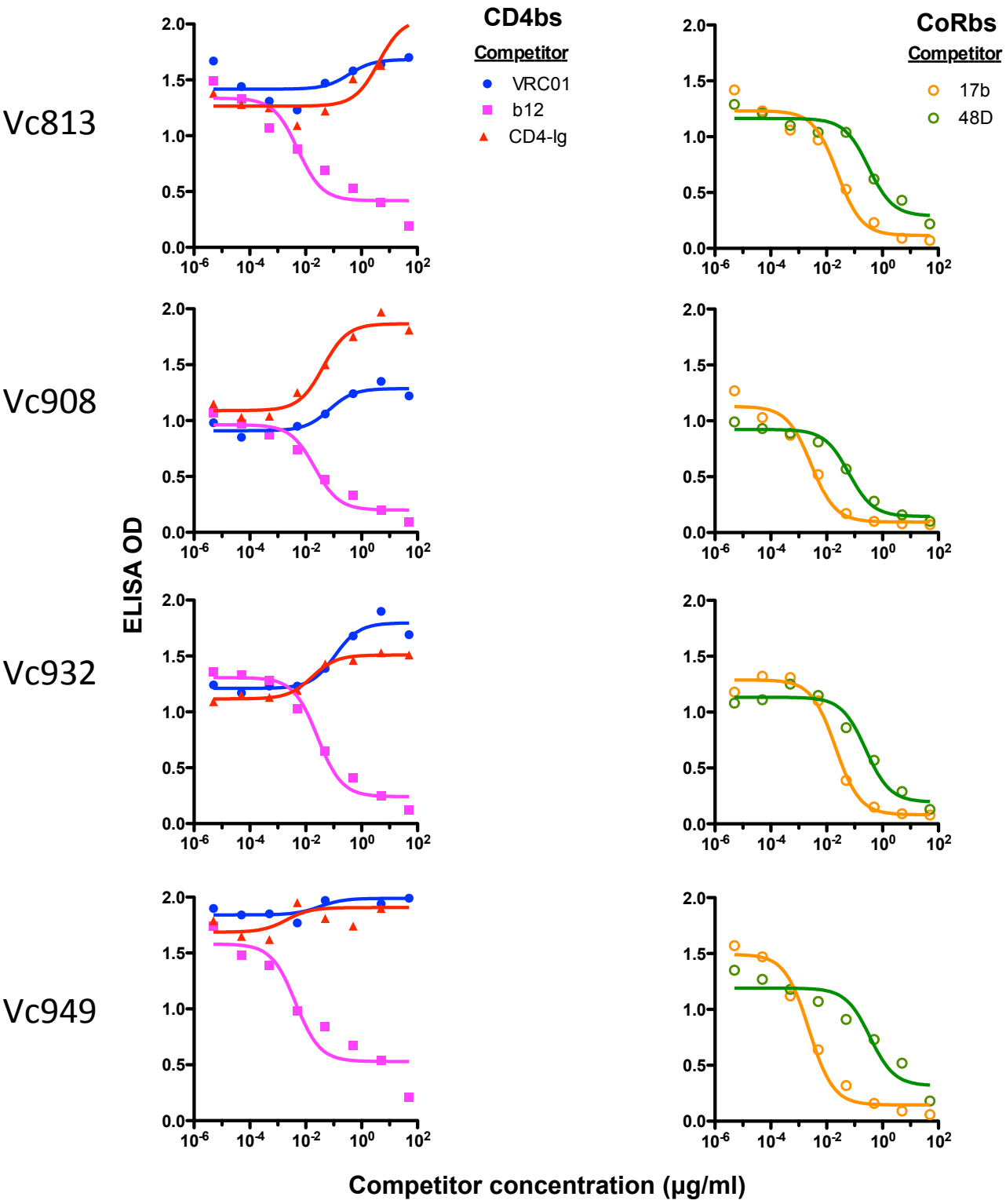
Supplementary Fig.S1

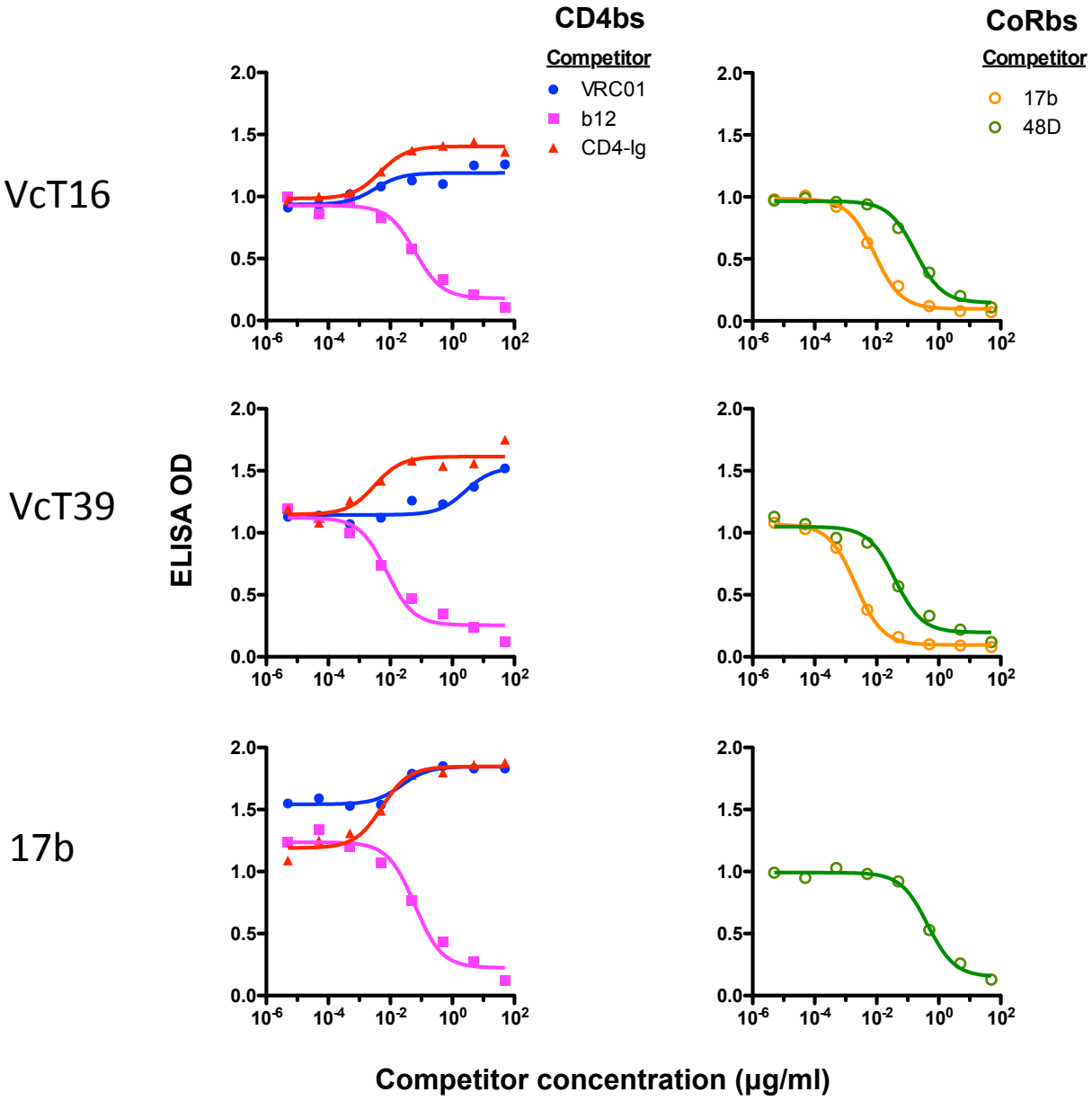


Supplementary Fig.S1. Binding specificity of other CoRbs mAbs to gp120. YU2gp120 CD4bs knockout mutant D368R and CoRbs knockout mutant, I420R were used to define binding specificity. Note that these CoRbs mAbs have the same binding pattern as conventional CoRbs mAb such as 17b, of which binding to YU2gp120 is sensitive to gp120 mutations in CoRbs (I420R), but not in CD4bs (D368R). Control ligands include CD4bs ligands, CD4-Ig and VRC01; and CoRbs mAb, 17b.

Supplementary Fig.S2



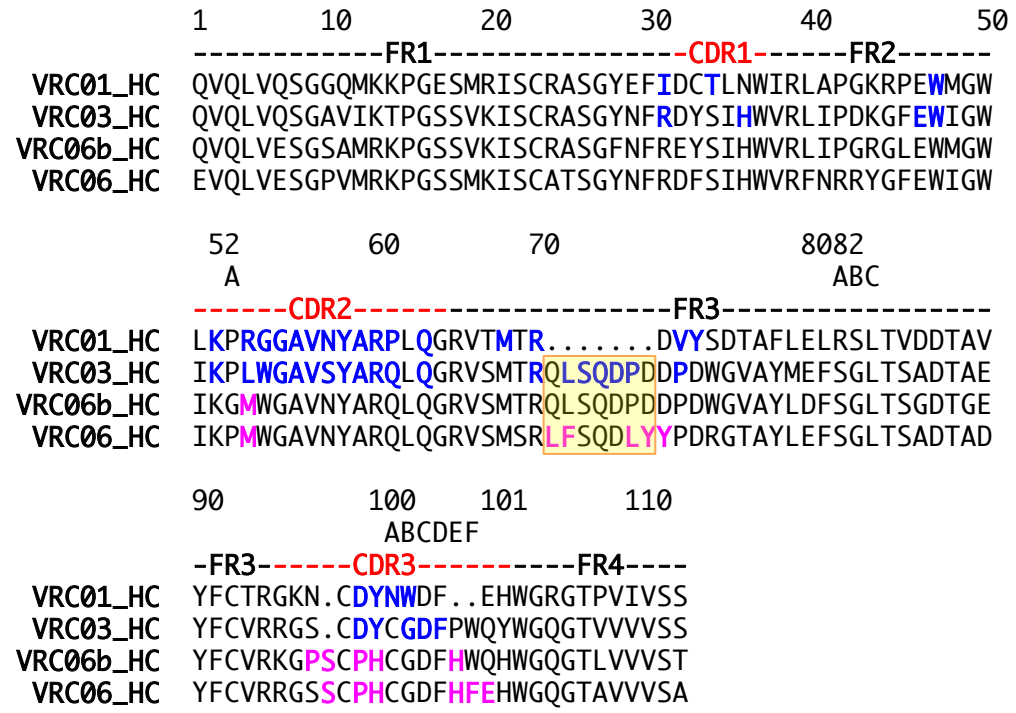
Supplementary Fig.S2 (continued)



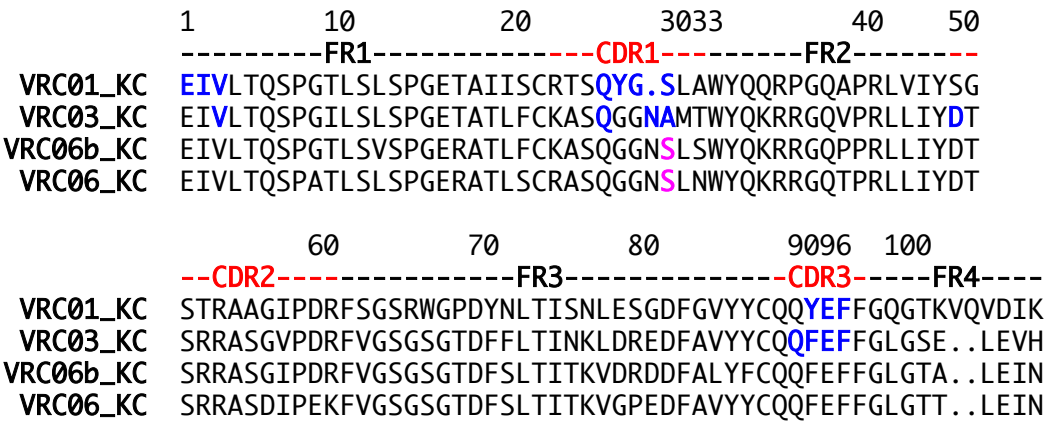
Supplementary Fig.S2. Binding specificity of other CoRbs mAbs to gp120 defined by competition ELISA. Biotin-labeled CoRbs mAbs were incubate with Yu2 gp120 in the presence of CD4bs or CoRbs ligands as competitors. Note that these CoRbs mAbs have the same competition pattern as conventional CoRbs mAb such as 17b, of which binding to YU2 gp120 compete with CD4bs mAb b12 and CoRbs mAbs, 17b and 48D. However, their binding to gp120 was enhanced by CD4-Ig and VRC01 as previously observed. CoRbs mAb, 17b was used as control.

Supplementary Fig.S3.

Heavy chain



Kappa chain



Supplementary Fig.S3. Heavy chain and kappa chain amino acid sequence alignment of VRC06 and VRC06b with genetically related mAbs, VRC03 and VRC01, which were previously isolated, following Kabat numbering system. The residues involved in contacting gp120 core of VRC03 and VRC01 defined by previous crystallography study were highlighted in blue, and the according divergent residues of VRC06 and VRC06b were highlighted in magenta. The 7 amino acid insertions, resulted from 21 bp insertion in the heavy chain frame work 3 (FR3) of VRC03, 06, and 06b, were highlighted with yellow box..

Table S1: Antibody neutralization data against 27 HIV-1 clade A Env-pseudoviruses

			Neutralization IC_{50}* (µg/ml)			Neutralization IC_{80}* (µg/ml)		
Virus ID	**Clade**	**Origin**	**VRC03^**	**VRC06**	**VRC06b**	**VRC03^**	**VRC06**	**VRC06b**
0260.v5.c36	A	Tanzania	0.872	>50	0.804	3.6	>50	3.1
0330.v4.c3	A	Tanzania	>50	>50	>50	>50	>50	>50
0439.v5.c1	A	Tanzania	0.224	1.8	>50	0.916	8.3	>50
3365.v2.c20	A	Tanzania	2.3	>50	2.1	21.5	>50	14.7
3415.v1.c1	A	Tanzania	0.033	0.978	0.026	0.106	3.8	0.099
3718.v3.c11	A	Tanzania	>50	>50	>50	>50	>50	>50
398-F1_F6_20	A	Tanzania	0.171	>50	>50	1.1	>50	>50
BB201.B42	A	Kenya	24.6	>50	2.4	>50	>50	37.1
BB539.2B13	A	Kenya	10.4	1.9	1.0	>50	10.4	10.1
BI369.9A	A	Kenya	>50	>50	>50	>50	>50	>50
BS208.B1	A	Kenya	0.263	3.8	0.099	2.3	>50	0.432
KER2008.12	A	Kenya	0.435	>50	28.1	1.7	>50	>50
KER2018.11	A	Kenya	0.415	8.9	0.937	1.4	49.9	7.4
KNH1209.18	A	Kenya	20.7	>50	39.9	>50	>50	>50
MB201.A1	A	Kenya	>50	>50	19.5	>50	>50	>50
MB539.2B7	A	Kenya	>50	>50	>50	>50	>50	>50
MI369.A5	A	Kenya	>50	>50	>50	>50	>50	>50
MS208.A1	A	Kenya	>50	>50	>50	>50	>50	>50
Q168.a2	A	Kenya	3.1	49.0	>50	>50	>50	>50
Q23.17	A	Kenya	0.041	1.7	0.052	0.199	12.2	0.318
Q259.17	A	Kenya	0.021	0.155	0.019	0.073	0.540	0.069
Q461.e2	A	Kenya	>50	>50	>50	>50	>50	>50
Q769.h5	A	Kenya	0.025	0.223	0.015	0.092	1.0	0.076
Q842.d12	A	Kenya	>50	>50	>50	>50	>50	>50
QH209.14M.A2	A	Kenya	>50	>50	40.5	>50	>50	>50
RW020.2	A	Rwanda	>50	0.515	1.5	>50	19.3	29.0
UG037.8	A	Uganda	>50	>50	>50	>50	>50	>50
Breadth	**N=27**	**Titer < 50**	**56%**	**37%**	**56%**	**41%**	**30%**	**41%**
		Titer < 1	**37%**	**15%**	**26%**	**19%**	**4%**	**19%**
Potency		**GMT#**	**0.510**	**1.7**	**0.899**	**0.775**	**6.2**	**1.7**

*Values < 1 µg/ml are highlighted in red, and values 1 – 50 µg/ml are in green.
^VRC03 was published previously (1). However, the data used in this paper are from a completely independent experimental run, and are consistent with those generated from the previous study.
#Geometric mean titers (GMT) were calculated for neutralization sensitive viruses with an IC_{50} or IC_{80} value < 50 µg/ml.

Table S2: Antibody neutralization data against 38 HIV-1 clade B Env-pseudoviruses

			Neutralization IC_{50}* (µg/ml)			Neutralization IC_{80}* (µg/ml)		
Virus ID	**Clade**	**Origin**	**VRC03^**	**VRC06**	**VRC06b**	**VRC03^**	**VRC06**	**VRC06b**
3988.25	B	USA	3.9	>50	>50	22.7	>50	>50
5768.04	B	USA	0.172	>50	0.315	0.878	>50	2.6
6101.10	B	USA	0.036	1.9	>50	0.114	6.4	>50
6535.3	B	USA	1.3	11.2	12.9	3.5	>50	35.2
7165.18	B	USA	>50	>50	>50	>50	>50	>50
89.6	B	USA	0.165	>50	0.245	0.541	>50	1.2
AC10.29	B	USA	>50	>50	>50	>50	>50	>50
ADA	B	USA	0.097	0.716	0.083	0.347	3.2	0.366
BaL.01	B	USA	>50	>50	>50	>50	>50	>50
BG1168.01	B	USA	>50	>50	22.8	>50	>50	>50
BL01	B	USA	>50	>50	>50	>50	>50	>50
BR07	B	USA	3.2	>50	18.3	10.3	>50	>50
BX08.16	B	USA	0.089	0.385	0.089	0.269	1.0	0.249
CAAN.A2	B	USA	8.2	>50	3.7	30.4	>50	10.0
HO86.8	B	Peru	>50	>50	>50	>50	>50	>50
HT593.1	B	Haiti	0.185	>50	0.061	0.711	>50	0.367
HXB2	B	USA	0.045	>50	0.462	0.121	>50	1.2
JR-CSF	B	USA	0.148	>50	3.9	0.718	>50	>50
JR-FL	B	USA	0.007	0.710	0.007	0.027	4.7	0.025
MN.3	B	USA	0.024	>50	0.072	0.059	>50	0.548
PVO.04	B	Italy	0.713	>50	0.093	2.1	>50	0.341
QH0515.01	B	Trinidad	0.120	>50	0.222	0.573	>50	4.3
QH0692.42	B	Trinidad	0.471	9.1	0.222	0.921	>50	1.2
REJO.67	B	USA	0.036	1.3	0.218	0.084	>50	1.2
RHPA.7	B	USA	12.1	34.3	0.037	>50	>50	0.210
SC422.8	B	Trinidad	0.023	3.1	0.048	0.064	9.1	0.088
SF162	B	USA	0.023	0.152	0.094	0.058	0.632	0.280
SS1196.01	B	USA	0.023	0.210	0.023	0.046	0.527	0.048
THRO.18	B	USA	>50	>50	>50	>50	>50	>50
TRJO.58	B	USA	0.040	0.247	0.023	0.127	0.772	0.149
TRO.11	B	Italy	0.271	>50	3.8	1.6	>50	49.3
WITO.33	B	USA	>50	>50	>50	>50	>50	>50
YU2	B	USA	0.049	0.135	0.031	0.140	0.470	0.086
CNE10	B'	China	0.515	20.7	0.814	1.6	>50	3.6
CNE12	B'	China	0.175	>50	>50	0.489	>50	>50
CNE14	B'	China	0.322	>50	2.0	1.4	>50	15.0
CNE4	B'	China	0.450	3.4	0.493	1.4	11.2	1.7
CNE57	B'	China	0.040	>50	0.174	0.275	>50	0.546
Breadth	**N=38**	**Titer < 50**	**79%**	**39%**	**74%**	**71%**	**26%**	**66%**
		Titer < 1	**66%**	**18%**	**55%**	**53%**	**11%**	**34%**
Potency		**GMT#**	**0.173**	**1.5**	**0.290**	**0.504**	**2.0**	**0.831**

*Values < 1 µg/ml are highlighted in red, and values 1 – 50 µg/ml are in green.
^VRC03 was published previously (1). However, the data used in this paper are from a completely independent experimental run, and are consistent with those generated from the previous study.
#Geometric mean titers (GMT) were calculated for neutralization sensitive viruses with an IC_{50} or IC_{80} value < 50 µg/ml.

Table S3: Antibody neutralization data against 54 HIV-1 clade C Env-pseudoviruses

			Neutralization IC_{50}* (µg/ml)			Neutralization IC_{80}* (µg/ml)		
Virus ID	**Clade**	**Origin**	**VRC03^**	**VRC06**	**VRC06b**	**VRC03^**	**VRC06**	**VRC06b**
286.36	C	Ethiopia	12.6	>50	3.5	>50	>50	29.0
288.38	C	Ethiopia	0.291	>50	0.130	1.6	>50	0.507
0013095-2.11	C	India	0.849	30.6	8.3	4.9	>50	>50
001428-2.42	C	India	0.023	0.166	0.023	0.023	2.0	0.023
0077_V1_C16	C	India	>50	>50	>50	>50	>50	>50
00836-2.5	C	India	0.052	0.669	0.039	0.118	>50	0.095
0921.v2.c14	C	India	0.633	>50	>50	2.5	>50	>50
16055-2.3	C	India	0.417	>50	0.035	>50	>50	0.336
16845-2.22	C	India	>50	>50	>50	>50	>50	>50
16936-2.21	C	India	0.083	>50	1.8	0.615	>50	>50
25710-2.43	C	India	0.180	23.1	0.107	0.570	>50	0.297
25711-2.4	C	India	1.3	>50	>50	3.4	>50	>50
25925-2.22	C	India	0.213	>50	0.553	0.993	>50	3.4
26191-2.48	C	India	>50	>50	43.6	>50	>50	>50
3168_V4_C10	C	India	0.061	1.3	0.087	0.166	3.8	0.204
3637_V5_C3	C	India	>50	>50	>50	>50	>50	>50
3873_V1_C24	C	India	>50	35.1	>50	>50	>50	>50
6322_V4_C1	C	India	>50	>50	>50	>50	>50	>50
6471_V1_C16	C	India	>50	>50	>50	>50	>50	>50
6631_V3_C10	C	India	>50	>50	>50	>50	>50	>50
6644_V2_C33	C	India	0.267	>50	2.7	0.901	>50	46.2
6785_V5_C14	C	India	0.138	>50	0.253	0.302	>50	0.574
6838_V1_C35	C	India	1.3	3.0	1.1	8.0	16.0	9.0
96ZM651_02	C	Zambia	>50	>50	30.1	>50	>50	>50
BR025.9	C	Brazil	>50	>50	>50	>50	>50	>50
CAP210.E8	C	South Africa	>50	>50	>50	>50	>50	>50
CAP244.D3	C	South Africa	>50	>50	>50	>50	>50	>50
CAP45.G3	C	South Africa	>50	>50	>50	>50	>50	>50
CNE30	C	China	>50	>50	6.8	>50	>50	>50
CNE31	C	China	0.629	3.3	0.825	1.5	8.5	2.3
CNE53	C	China	>50	0.395	0.180	>50	4.8	0.667
CNE58	C	China	16.0	>50	0.153	>50	>50	2.8
DU123.06	C	South Africa	>50	>50	>50	>50	>50	>50
DU151.02	C	South Africa	>50	>50	>50	>50	>50	>50
DU156.12	C	South Africa	>50	>50	>50	>50	>50	>50
DU172.17	C	South Africa	>50	>50	>50	>50	>50	>50
DU422.01	C	South Africa	>50	>50	>50	>50	>50	>50
MW965.26	C	Malawi	4.1	>50	>50	30.3	>50	>50
SO18.18	C	Malawi	0.154	>50	0.069	0.484	>50	0.271
TV1.29	C	South Africa	>50	>50	>50	>50	>50	>50
TZA125.17	C	Tanzania	>50	>50	>50	>50	>50	>50
TZBD.02	C	Tanzania	2.3	>50	0.085	>50	>50	1.6
ZA012.29	C	South Africa	17.7	>50	>50	>50	>50	>50
ZM106.9	C	Zambia	0.180	>50	10.6	1.1	>50	>50
ZM109.4	C	Zambia	>50	>50	>50	>50	>50	>50
ZM135.10a	C	Zambia	>50	>50	>50	>50	>50	>50
ZM176.66	C	Zambia	0.029	>50	>50	0.147	>50	>50
ZM197.7	C	Zambia	2.3	>50	16.5	11.7	>50	31.8
ZM214.15	C	Zambia	>50	>50	>50	>50	>50	>50
ZM215.8	C	Zambia	>50	>50	>50	>50	>50	>50
ZM233.6	C	Zambia	>50	>50	>50	>50	>50	>50
ZM249.1	C	Zambia	13.3	>50	>50	>50	>50	>50
ZM53.12	C	Zambia	28.9	7.9	14.1	>50	30.8	>50
ZM55.28a	C	Zambia	>50	>50	>50	>50	>50	>50
Breadth	**N=54**	**Titer < 50**	**48%**	**19%**	**44%**	**35%**	**11%**	**31%**
		Titer < 1	**30%**	**6%**	**24%**	**19%**	**0%**	**17%**
Potency		**GMT[#]**	**0.652**	**3.2**	**0.778**	**0.992**	**7.3**	**1.2**

*Values < 1 µg/ml are highlighted in red, and values 1 – 50 µg/ml are in green.
^VRC03 was published previously (1). However, the data used in this paper are from a completely independent experimental run, and are consistent with those generated from the previous study.
[#]Geometric mean titers (GMT) were calculated for neutralization sensitive viruses with an IC_{50} or IC_{80} value < 50 µg/ml.

Table S4: Antibody neutralization data against 9 HIV-1 clade D Env-pseudoviruses

			Neutralization IC_{50}* (µg/ml)			Neutralization IC_{80}* (µg/ml)		
Virus ID	**Clade**	**Origin**	**VRC03^**	**VRC06**	**VRC06b**	**VRC03^**	**VRC06**	**VRC06b**
231965.c1	D	Uganda	39.5	>50	>50	>50	>50	>50
247-23	D	Cameroon	>50	>50	>50	>50	>50	>50
3016.v5.c45	D	Tanzania	>50	>50	>50	>50	>50	>50
57128.vrc15	D	Uganda	>50	>50	>50	>50	>50	>50
6405.v4.c34	D	Tanzania	>50	>50	>50	>50	>50	>50
A03349M1.vrc4a	D	Uganda	>50	>50	>50	>50	>50	>50
NKU3006.ec1	D	Kenya	0.093	>50	0.055	0.286	>50	0.191
UG021.16	D	Uganda	>50	36.9	>50	>50	>50	>50
UG024.2	D	Uganda	>50	>50	>50	>50	>50	>50
Breadth	**N=9**	**Titer < 50**	**22%**	**11%**	**11%**	**11%**	**0%**	**11%**
		Titer < 1	**11%**	**0%**	**11%**	**11%**	**0%**	**11%**
Potency		**GMT#**	**1.9**	**36.9**	**0.055**	**0.286**	**>50**	**0.191**

*Values < 1 µg/ml are highlighted in red, and values 1 – 50 µg/ml are in green.
^VRC03 was published previously (1). However, the data used in this paper are from a completely independent experimental run, and are consistent with those generated from the previous study.
#Geometric mean titers (GMT) were calculated for neutralization sensitive viruses with an IC_{50} or IC_{80} value < 50 µg/ml.

Table S5: Antibody neutralization data against 16 HIV-1 CRF01_AE Env-pseudoviruses

			Neutralization IC_{50}* (µg/ml)			Neutralization IC_{80}* (µg/ml)		
Virus ID	**Clade**	**Origin**	**VRC03^**	**VRC06**	**VRC06b**	**VRC03^**	**VRC06**	**VRC06b**
620345.c1	AE	Thailand	>50	>50	>50	>50	>50	>50
C1080.c3	AE	Thailand	>50	>50	>50	>50	>50	>50
C2101.c1	AE	Thailand	>50	>50	>50	>50	>50	>50
C3347.c11	AE	Thailand	0.677	0.232	0.064	10.8	1.0	0.930
C4118.09	AE	Thailand	>50	>50	>50	>50	>50	>50
CNE3	AE	China	>50	>50	>50	>50	>50	>50
CNE5	AE	China	6.6	>50	>50	25.2	>50	>50
CNE55	AE	China	1.5	>50	>50	8.2	>50	>50
CNE56	AE	China	>50	>50	>50	>50	>50	>50
CNE59	AE	China	>50	>50	>50	>50	>50	>50
M02138	AE	Thailand	>50	>50	>50	>50	>50	>50
R1166.c1	AE	Thailand	>50	>50	1.3	>50	>50	7.2
R2184.c4	AE	Thailand	0.054	>50	>50	0.197	>50	>50
R3265.c6	AE	Thailand	>50	>50	>50	>50	>50	>50
TH966.8	AE	Thailand	>50	>50	>50	>50	>50	>50
TH976.17	AE	Thailand	>50	>50	>50	>50	>50	>50
Breadth	**N=16**	**Titer < 50**	**25%**	**6%**	**13%**	**25%**	**6%**	**13%**
		Titer < 1	**13%**	**6%**	**6%**	**6%**	**0%**	**6%**
Potency		**GMT#**	**0.774**	**0.232**	**0.291**	**4.6**	**1.0**	**2.6**

*Values < 1 µg/ml are highlighted in red, and values 1 – 50 µg/ml are in green.
^VRC03 was published previously (1). However, the data used in this paper are from a completely independent experimental run, and are consistent with those generated from the previous study.
#Geometric mean titers (GMT) were calculated for neutralization sensitive viruses with an IC_{50} or IC_{80} value < 50 µg/ml.

Table S6: Antibody neutralization against 16 CRF02_AG Env-pseudoviruses

			Neutralization IC_{50}* (µg/ml)			Neutralization IC_{80}* (µg/ml)		
Virus ID	**Clade**	**Origin**	**VRC03^**	**VRC06**	**VRC06b**	**VRC03^**	**VRC06**	**VRC06b**
235-47	AG	Cameroon	0.659	>50	0.073	10.9	>50	0.532
242-14	AG	Cameroon	>50	>50	>50	>50	>50	>50
T250-4	AG	Cameroon	>50	>50	>50	>50	>50	>50
T251-18	AG	Cameroon	>50	>50	>50	>50	>50	>50
T255-34	AG	Cameroon	>50	>50	>50	>50	>50	>50
T257-31	AG	Cameroon	>50	>50	>50	>50	>50	>50
263-8	AG	Cameroon	0.044	0.511	0.023	0.258	1.5	0.176
T266-60	AG	Cameroon	>50	>50	>50	>50	>50	>50
269-12	AG	Cameroon	>50	>50	2.6	>50	>50	5.6
271-11	AG	Cameroon	>50	>50	>50	>50	>50	>50
T278-50	AG	Cameroon	>50	>50	>50	>50	>50	>50
T280-5	AG	Cameroon	0.035	0.783	0.023	0.310	8.7	0.097
928-28	AG	Cote d'Ivoire	>50	>50	>50	>50	>50	>50
DJ263.8	AG	Kenya	>50	>50	>50	>50	>50	>50
T253-11	AG	Cameroon	>50	>50	>50	>50	>50	>50
T33-7	AG	Cameroon	>50	>50	>50	>50	>50	>50
Breadth	**N=16**	**Titer < 50**	**19%**	**13%**	**25%**	**19%**	**13%**	**25%**
		Titer < 1	**19%**	**13%**	**19%**	**13%**	**0%**	**19%**
Potency		**GMT#**	**0.100**	**0.633**	**0.100**	**0.955**	**3.6**	**0.474**

*Values < 1 µg/ml are highlighted in red, and values 1 – 50 µg/ml are in green.
^VRC03 was published previously (1). However, the data used in this paper are from a completely independent experimental run, and are consistent with those generated from the previous study.
#Geometric mean titers (GMT) were calculated for neutralization sensitive viruses with an IC_{50} or IC_{80} value < 50 µg/ml.

Table S7: Antibody neutralization against 17 HIV-1 recombinant and 1 clade G Env-pseudoviruses

			Neutralization IC_{50}* (µg/ml)			Neutralization IC_{80}* (µg/ml)		
Virus ID	**Clade**	**Origin**	**VRC03^**	**VRC06**	**VRC06b**	**VRC03^**	**VRC06**	**VRC06b**
3301_V1_C24	AC	Tanzania	0.098	>50	>50	0.304	>50	>50
3589_V1_C4	AC	Tanzania	>50	>50	>50	>50	>50	>50
6540.v4.c1	AC	Tanzania	>50	>50	>50	>50	>50	>50
6545.v3.c13	AC	Tanzania	>50	>50	>50	>50	>50	>50
0815_V3_C3	ACD	Tanzania	0.043	0.305	0.023	0.075	2.4	0.053
6095_V1_C10	ACD	Tanzania	>50	>50	>50	>50	>50	>50
3468_V1_C12	AD	Tanzania	>50	>50	>50	>50	>50	>50
CH038.12	BC	China	>50	>50	>50	>50	>50	>50
CH070.1	BC	China	>50	>50	>50	>50	>50	>50
CH117.4	BC	China	0.117	>50	0.617	0.689	>50	16.1
CH181.12	BC	China	8.1	>50	1.7	>50	>50	20.8
CNE15	BC	China	0.422	>50	>50	3.9	>50	>50
CNE7	BC	China	0.419	>50	0.240	2.6	>50	0.926
CNE40	BC	China	0.421	>50	0.096	1.3	>50	0.330
3326_V4_C3	CD	Tanzania	>50	>50	>50	>50	>50	>50
3337_V2_C6	CD	Tanzania	>50	>50	>50	>50	>50	>50
3817.v2.c59	CD	Tanzania	>50	>50	>50	>50	>50	>50
X2088_c9	G	Ghana	>50	>50	>50	>50	>50	>50
Breadth	**N=18**	**Titer < 50**	**39%**	**6%**	**28%**	**33%**	**6%**	**28%**
		Titer < 1	**33%**	**6%**	**22%**	**17%**	**0%**	**17%**
Potency		**GMT#**	**0.313**	**0.305**	**0.224**	**0.770**	**2.4**	**1.4**

*Values < 1 µg/ml are highlighted in red, and values 1 – 50 µg/ml are in green.
^VRC03 was published previously (1). However, the data used in this paper is from a completely independent experimental run, which are consistent with that generated from the previous study.
#Geometric mean titers (GMT) were calculated for neutralization sensitive viruses with an IC_{50} or IC_{80} value < 50 µg/ml.

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