Supplementary Information

Broad and potent HIV-1 neutralization by a human antibody that binds the gp41-120 interface

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Supplementary Tables (PDF format)

Supplementary Table 1. 35O22 neutralizing breadth and potency.

a. Neutralization by 35O22 and its variants against an 8-isolate Env-pseudovirus mini-panel.

	Antibody variants							
Virus ID	**35O22**	**7B9**	**4O20**	**10J4**	**7K3**	**13I10**	**2N5**	**10M6**
Q168.a2	0.009	3.7	>20	>20	3.28	>20	>20	>20
RW020.2	0.010	0.853	0.041	>20	>20	>20	>20	>20
Q769.h55	0.004	0.593	>20	>20	>20	>20	>20	>20
JRFL.JB	0.013	1.36	0.073	3.93	>20	>20	0.098	>20
THRO.18	0.908	>20	>20	>20	>20	>20	>20	>20
CAAN.A2	0.003	0.212	0.007	0.006	0.004	0.005	>20	0.003
ZAO12.29	>20	>20	>20	>20	>20	13.8	>20	>20
DU156.12	>20	>20	>20	>20	>20	>20	>20	>20

[a] $IC_{50}<1\mu g/ml$ is highlighted in red, $1<IC_{50}<10$ μg/ml in yellow, and $10<IC_{50}<50$ μg/ml in green.

b. Neutralization profile of patient N152 serum and monoclonal antibodies.

		Serum[a]	Monoclonal antibody[b]				
Clade	**Virus ID**	**N152**	**35O22**	**10E8**	**VRC01**	**PG9**	**PGT121**
A	KER2018	397	0.004	2.71	0.070	0.0009	>50
	RWO20.2	1341	0.010	1.30	0.303	0.103	0.002
	Q168.a2	509	0.009	0.768	0.140	0.106	>50
	Q769.d22	1834	0.002	1.11	0.015	0.007	>50
	Q769.h5	836	0.004	2.74	0.014	0.002	>50
B	JRFL.JB	860	0.013	0.247	0.033	>50	0.017
	BaL.01	1590	0.003	0.715	0.102	0.052	0.011
	YU2.DG	279	0.287	0.895	0.055	3.69	0.068
	PVO.04	359	0.011	1.78	0.386	6.24	0.132
	TRO.11	2773	>50	0.026	0.343	41.5	0.006
	CAAN.A2	1364	0.003	1.37	1.06	13.0	0.005
	TRJO.58	1231	0.002	0.881	0.079	0.246	4.31
	THRO.18	351	0.908	0.048	4.42	15.0	>50
	BG1168.1	167	>50	0.136	0.449	>50	>50
	6101.1	1372	>50	0.009	0.104	>50	0.002
C	ZA012.29	39	>50	0.827	0.250	27.0	0.005
	DU156.12	2021	11.7	0.007	0.082	0.023	0.005
	DU422.01	325	0.021	1.06	>50	0.303	0.164
	ZM106.9	256	0.372	>50	0.248	0.639	0.005
	ZM55.28a	85	1.93	2.31	0.144	0.571	0.070
Control	SIVmac251.SG3	5	>50	>50	>50	>50	>50
Geometric Mean IC_{50} ($\mu g\ ml^{-1}$)			0.031	0.417	0.149	0.355	0.020
Median IC_{50} ($\mu g\ ml^{-1}$)			0.010	0.881	0.140	0.303	0.009

[a] The data for N152 shows the ID_{50} of serum against each virus. $ID_{50}>1000$ is highlighted in red, $500<ID_{50}<1000$ is in orange, and $100< ID_{50}<500$ is in yellow. [b] The data for the monoclonal antibodies shows the IC_{50}. $IC_{50}<0.1\mu g\ ml^{-1}$ is in red, $0.1<IC_{50}<1\mu g\ ml^{-1}$ is in orange, $1<IC_{50}<10$ $\mu g\ ml^{-1}$ $\mu g\ ml^{-1}$ is in yellow, and $10<IC_{50}<50$ $\mu g\ ml^{-1}$ is in green.

Supplementary Table 2. Amino acid sequences of the variable regions of 35O22 and its variants.

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               1       10        20        30          40         5052A   57
               |        |         |         |           |          | ||    |
Heavy Chain    ___________FR1________________ _CDR1 _____FR2______ ___CDR2__
IGHV1-18*02    QVQLVQSGAEVKKPGASVKVSCKASGYTFT SYGIS WVRQAPGQGLEWMG WISAYNGNT
       10J4    QGQLVQSGGELKKPGASVKISCKTSGYRFS FYHIN WIRQLVGRGPEWMG WISPYNGGT
       10M6    QGQLVQSGGELKRPGASVKISCKTSGYRFS SYHIN WIRQVIGRGPEWMG WISPYSGGT
      13I10    QGQLVQSGGELKKPGASVKISCKTSGYRFS FYHIN WIRQVVGRGPEWMG WISPYNGGT
        7K3    QGQLVQSGGELKKPGASVKISCKTSGYRFS FYHIN WIRQVSGRGPEWMG WISPYSGDT
        2N5    QGQLVQSGAELKKPGASVKISCKTSGYKFS FFHIN WIRQTAGRGPEWLG WISPYSGDK
       4O20    QGQLVQSGAELKKPGGSVKISCKTSGYRFN FYHIN WIRQTAGRGPEWMG WISPYSGDK
        7B9    QGQLVQSGAELKKPGDSVKISCKTSGYRFN FYHIN WIRQTAGRGPEWMG WISPYSGDK
      35O22    QGQLVQSGAELKKPGASVKISCKTSGYRFN FYHIN WIRQTAGRGPEWMG WISPYSGDK

                60         7072ABCDEFGH      8082ABC      90        100ABCDEF 103    110
                 |          | |||||||||       | ||||       |          |||||||   |      |
Heavy Chain    __CDR2__ ____________FR3_________________________ _____CDR3_____ _FR4(HJ4*02)
IGHV1-18*02    NYAQKLQG RVTMTTD--------TSTSTAYMELRSLRSDDTAVYYCAR    RDGYNYYFDY  WGQGTLVTVSS
       10J4    NLAPELRG RLVLTTEREVVDTMTLSTGTAHMELRNLRSDDTGIYFCAK GLLRDGSSTWLPHL WGQGTLLTVSS
       10M6    NLAPEFRG RLVLTTEREVVDTMTLSTGTAHMELRNLKSDDTGIYFCAK GLLRDGSSTWLPHL WGQGTLLTVSS
      13I10    NLAPEFRG RLVLTTEREVVDTMTMSTGTAHMELRNLKSDDTGLYFCAK GLLRDGPSTWLPHL WGQGTLLTVSS
        7K3    NLAPDFRG RVVLTTDREVVDTMTMSTGTAHMELRNLKSDDTGLYFCAK GLLRDGSSTWLPHL WGQGTLLTVSS
        2N5    NYAPAFQD RVIMTTDKEVPVTSFTSTGTAYLEIRSLKPDDTGIYFCAR GLLRDGSSTWLPYL WGQGTLLTVSS
       4O20    NLAPAFQD RVIMTTDKEVPVTAFTSTGTAYMEIRNLKFDDTGIYFCAK GLLRDGSSTWLPYL WGQGTLLTVSS
        7B9    NLAPAFQD RVIMTTDNEVPVTAFTSTGTAYMEIRNLRFDDTGIYFCAK GLLRDGSSTWLPYL WGQGTLLTVSS
      35O22    NLAPAFQD RVIMTTDTEVPVTSFTSTGAAYMEIRNLKFDDTGTYFCAK GLLRDGSSTWLPYL WGQGTLLTVSS
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               1       10        20      27ABC 30         40         50
               |        |         |       ||||  |          |          |
Light Chain    ___________FR1_________ _____CDR1_____ ______FR2______ _CD
IGLV2-14*02    QSALTQPAS.VSGSPGQSITISC TGTSSDVGSYNLVS WYQQHPGKAPKLMIY EGS
       10J4    QSALTQPAS.VSGSLGQSVTISC TGPSSVCCSHKSIS WYQWPPGRAPTLLIF EDS
       10M6    QSALTQPAS.VSGSLGQSVTISC TGPSSVCCSHKSIS WYQWPPGRAPTLLIF EDS
      13I10    QSALTQPAS.VSGSLGQSVTISC TGPSSVCCSHKSIS WYQWPPGRAPTLLIF EDS
        7K3    QSALTQPAS.VSGSLGQSVTISC TGPNSVCCSHKSIS WYQWPPGRAPTLLIF EDS
        2N5    QPVLTQPAS.VSGSLGQSVTISC TGPSSVCCSHKSIS WYRWPPGRAPTLIIY EDN
       4O20    QSALTQPAS.VSGSLGQSVTISC TGPNSVCCSHKSIS WYQWPPGRAPTLIIY EDN
        7B9    QSALTQPAS.VSGSLGQSVTISC TGPNSACCSHKSIS WHQWPPGRAPTLIIY EDN
      35O22    QSVLTQSAS.VSGSLGQSVTISC TGPNSVCCSHKSIS WYQWPPGRAPTLIIY EDN

                      60        70        80         90   95A   100   106A
                       |         |         |          |    ||     |     ||
Light Chain    R2__ ____________FR3_________________ ___CDR3___ FR4(LJ1*01)
IGLV2-14*02    KRPS GVSNRFSGSKSGNTASLTISGLQAEDEADYYC SSYTSSSTLV FGTGTKVTVL
       10J4    ERSW GISDRFSGYKSYWSASLTISNLRPEDETTYYC CSYTHNSGCV FGTGTNVSVL
       10M6    ERSW GISDRFSGYKSYWSASLTISNLRPEDETTYFC CSYTHNSGCV FGTGTKVSVL
      13I10    ERSW GISDRFSGYKSYWSASLTISNLRPEDETTYYC CSYTHNSGCV FGTGTKVSVL
        7K3    ERSW GISDRFSGYKSYWSASLTISNLRPEDETTYYC CSYTHNSGCV FGTGTKVSVL
        2N5    KRFS EISPRFSGYKSYWSAYLTISDLRPEDETTYYC CSYTHNSGCV FATGTKVSVL
       4O20    EKAP GISHRFSGYKSYWSAYLTISDLRPEDETTYYC CSYTHNSGCV FGTGTKVSVL
        7B9    EKAP GISHRFSGYKSYWSAYLTISDLRPEDETTYYC CSYTHNSGCV FGTGTKVSVL
      35O22    ERAP GISPRFSGYKSYWSAYLTISDLRPEDETTYYC CSYTHNSGCV FGTGTKVSVL
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Residues in red represent substitutions from the germline sequence. The dot symbol denotes the residue deletion. Kabat numbering is used to identify specific residues in the 35O22 heavy and light chains.

Supplementary Table 3. Antibody neutralization data against 181 HIV-1 Env-pseudoviruses.

		ID50[a]	IC50 (μg ml^{-1})[b]									
Virus ID	Clade	N152	35O22	10E8	4E10	VRC01	NIH 45-46	3BNC 117	PG9	PG16	PGT121	PGT128
0260.v5.c36	A	3906	0.393	8.96	31.7	0.529	0.397	0.200	2.18	2.10	0.039	0.062
0330.v4.c3	A	9263	0.736	1.27	6.86	0.064	0.049	0.013	0.018	0.006	0.041	1.09
0439.v5.c1	A	4111	0.118	0.924	8.24	0.052	0.185	0.215	>50	>50	>50	6.41
3365.v2.c20	A	9810	0.780	2.45	1.38		0.038	0.011			0.059	>50
3415.v1.c1	A	12934	6.99	6.61	11.5	0.092	0.082	0.094	0.149	0.036	>50	0.018
3718.v3.c11	A	2167	>50	1.63	8.55	0.218	0.871	>50	0.050	0.019	1.40	>50
398-F1_F6_20	A	11290	>50	0.942	0.784	0.058	0.157	0.071	>50	>50	0.002	0.002
BB201.B42	A	470	1.73	0.231	2.19	0.343	0.303	3.35	0.014	0.003	0.003	0.004
BB539.2B13	A	1947	3.91	1.00	0.712	0.094	0.022	0.033	0.106	0.012	>50	>50
BI369.9A	A	6422	0.003	0.135	1.36	0.149	0.043	0.020	0.029	0.007	0.008	0.030
BS208.B1	A	9293	>50	0.127	0.618	0.029	0.006	0.002	0.031	0.004	>50	>50
KER2008.12	A	8198	0.0008	>50	1.28	0.563	0.567	0.248	0.017	0.006	2.22	14.7
KER2018.11	A	397	0.004	2.71	8.46	0.070	0.828	0.417	0.0009	0.0006	>50	>50
KNH1209.18	A	2457	0.103	0.422	1.63	0.087	0.246	0.040	0.367	0.678	0.002	0.006
MB201.A1	A	4508	7.85	0.230	2.21	0.237	0.165	0.464	0.024	0.001	0.005	0.016
MB539.2B7	A	726	0.257	8.03	21.7	0.544	0.402	0.087	0.058	0.025	>50	3.57
MI369.A5	A	2259	0.005	0.617	2.88	0.162	0.074	0.033	0.058	0.011	0.022	0.020
MS208.A1	A	14167	>50	0.254	1.56	0.147	0.090	0.019	0.071	0.047	>50	>50
Q23.17	A	5898	0.002	0.350	0.132	0.086	0.106	0.017	0.007	0.002	0.004	0.013
Q259.17	A	953	1.92	3.22	10.2	0.051	0.046	0.017	0.045	0.028	>50	>50
Q461.e2	A	4282	0.001	1.62	5.53	0.410	0.212	0.069	3.01	4.11		
Q769.d22	A	1834	0.002	1.11	1.73	0.015	0.013	0.007	0.007	0.010	>50	>50
Q769.h5	A	836	0.004	2.74	1.17	0.014	0.019	0.006	0.002	0.002	>50	>50
Q842.d12	A	7407	0.003	4.53	8.99	0.006	0.015	0.002	0.005	0.001	0.016	0.026
QH209.14M.A2	A	1490	1.13	1.45	5.93	0.024	0.011	0.008	>50	>50	>50	>50
RW020.2	A	1341	0.010	1.30	8.21	0.303	0.144	0.020	0.103	0.070	0.002	0.008
UG037.8	A	2490	0.026	0.039	0.504	0.035	0.056	0.020	0.021	0.0008	0.065	0.018
3301.V1.C24	AC	9904	0.348	2.81	8.64	0.084	0.055	0.046	0.281	0.023	0.009	0.174
3589.V1.C4	AC	143	0.010		1.94	0.073	0.351	0.061	0.025	0.728	>50	0.028
6540.v4.c1	AC	3626	7.73	2.62	14.3	>50	>50	>50	0.035	0.017	>50	3.13
6545.V3.C13	AC	5118	0.722	1.61	6.07		>50	>50				
6545.V4.C1	AC	3797	4.92	1.54	12.5	>50	>50	>50	0.095	0.068	>50	>50
0815.V3.C3	ACD	465	0.005	0.190	1.66	0.036	0.055	0.018	>50	>50	0.020	0.036
6095.V1.C10	ACD	7918	>50	0.0006	0.004	0.464	0.601	0.096	0.242	0.023	37.3	>50
3468.V1.C12	AD	12304	0.004	0.366	0.964	0.040	0.104	0.073	2.09	2.38	0.042	>50
Q168.a2	AD	509	0.009	0.768	18.7	0.140	0.138	0.050	0.106	0.031	>50	>50
620345.c1	AE	187	>50	0.748	0.469	>50	>50	>50	0.393	>50	>50	>50
C1080.c3	AE	8632	0.0009	0.038	0.598	1.50	0.539	0.096	0.004	0.001	>50	0.117
C2101.c1	AE	12603	0.002	0.671	2.45	0.097	2.38	0.064	0.026	0.009	>50	0.010
C3347.c11	AE	32066	0.022	0.016	0.161	0.037	0.055	0.029	0.038	0.006	>50	0.004
C4118.09	AE	25357	0.0006	0.498	5.26	0.110	0.035	0.019	0.037	0.021	>50	>50
CNE3	AE	70.5	0.939	1.36	2.23	3.56	1.99	0.125	0.079	0.173	>50	>50
CNE5	AE	38.3	>50	1.13	2.99	0.228	0.329	0.386	0.023	0.023	>50	0.096
CNE55	AE	22242	0.195	0.044	0.901	0.292	0.294	0.147	0.146	1.37	>50	>50
CNE56	AE	1145	>50	0.024	0.207	0.442	0.483	0.075	>50	>50	>50	25.0
CNE59	AE	3223	>50	0.001	0.042	0.516	0.102	0.043	0.091	0.113	>50	>50

		ID50	IC50 (μg ml^{-1})									
Virus ID	**Clade**	**N152**	**35O22**	**10E8**	**4E10**	**VRC01**	**NIH 45-46**	**3BNC 117**	**PG9**	**PG16**	**PGT121**	**PGT128**
M02138	AE	15153	0.217	0.002	0.151	0.742	0.270	0.154	0.122	0.022	>50	>50
R1166.c1	AE	6893	2.29	0.550	2.69	1.77	1.73	0.230	1.55	0.587	>50	>50
R2184.c4	AE	1474	0.013	0.679	3.79	0.052	0.065	0.035	0.204	0.280	>50	4.84
R3265.c6	AE	35.5	>50	5.10	15.6	0.731	0.019	0.020	1.30	0.036	>50	>50
TH966.8	AE	2803	>50	0.012	0.079	0.331	0.094	0.056	0.042	0.008	>50	0.006
TH976.17	AE	3259	0.325	0.339	0.213	0.066	0.116	0.025	>50	>50	>50	>50
235-47	AG	457	>50	0.077	0.802	0.049	0.250	0.022	0.322	0.246	0.110	>50
242-14	AG	6609	>50	1.35	>50	>50	>50	>50	0.025	0.023	>50	>50
263-8	AG	3500	>50	0.029	0.319	0.119	0.196	0.047	0.353	1.31	1.23	0.488
269-12	AG	95876	>50	0.049	0.133	0.163	9.79	0.151	1.52	0.281	0.164	0.019
271-11	AG	2733	0.711	0.623	3.11	0.052	0.215	0.007	0.097	0.102	11.7	>50
928-28	AG	5652	>50	0.022	0.009	0.378	0.164	0.155	0.060	0.023	31.0	>50
DJ263.8	AG	5021	0.021	0.010	0.081	0.072	0.018	0.025	0.100	0.048	0.064	20.0
T250-4	AG	24732	>50	0.658	0.361	>50	>50	>50	0.0006	0.0006	0.001	0.003
T251-18	AG	6192	0.007	0.132	9.42	3.58	1.36	0.203	>50	10.5	10.8	>50
T253-11	AG	243	>50	0.863	0.755	0.265	0.268	0.116	0.127	4.44	>50	18.9
T255-34	AG	687	0.021	0.153	0.023	0.252	0.779	0.051	0.015	0.005	>50	>50
T257-31	AG	89.7	>50	0.224	2.00	1.68	0.579	0.181	0.020	0.003	>50	>50
T266-60	AG	14457	0.795	>50	4.74	0.353	0.219	0.032	24.0	>50	0.160	0.014
T278-50	AG	2342	3.00	0.362	3.17	>50	>50	>50	0.913	1.13	>50	0.050
T280-5	AG	2740	3.18	2.88	0.726	0.017	0.036	0.019	0.379	0.233	0.002	0.005
T33-7	AG	1731	0.632	0.715	2.89	0.023	0.074	0.007	0.023	0.023	>50	>50
3988.25	B	1332	0.119	0.053	1.37	2.10	0.358	>50	0.010	0.002	0.002	0.007
5768.04	B	140	0.192	2.01	15.0	0.099	0.074	0.201	0.073	0.020	0.039	4.50
6101.1	B	1372	>50	0.0009	0.381	0.104	0.016	0.022	>50	>50	0.002	0.004
6535.3	B	7906	0.114	0.054	0.458	2.16	0.067	0.262	0.465	>50	0.003	0.015
7165.18	B	9575	>50	0.036	0.895	>50	>50	6.54	>50	>50	0.019	0.013
89.6.DG	B	1943	0.459	0.040	1.02	0.460	0.274	0.109	>50	>50	0.016	0.010
AC10.29	B	13018	0.833	0.048	0.408	1.43	0.364	6.05	0.078	0.023	0.028	0.009
ADA.DG	B	15877	>50	0.016	0.656	0.424	0.112	0.086	0.342	0.023	0.002	0.017
Bal.01	B	1590	0.003	0.715	6.74	0.102	0.017	0.012	0.052	8.00	0.011	0.300
BaL.26	B	5407	0.0006	0.264	2.70	0.047	0.013	0.006	0.034	0.136	0.010	0.021
BG1168.01	B	167	>50	0.136	3.34	0.449	1.32	0.179	>50	>50	>50	>50
BL01.DG	B	1044	0.006	0.136	4.68	>50	>50	>50	>50	>50	>50	>50
BR07.DG	B	4626	0.0009	0.031	2.01	1.67	0.362	0.135	>50	>50	0.064	2.28
BX08.16	B	74924	0.019	0.099	0.469	0.281	0.366	0.102	0.024	0.062	0.002	0.006
CAAN.A2	B	1364	0.003	1.37	8.49	1.06	0.205	0.673	13.0	7.43	0.005	0.102
CNE10	B	8422	>50	0.013	0.307	0.776	0.173	0.050	0.243	9.17	0.005	0.008
CNE12	B	2475	0.027	0.117	1.11	0.785	0.516	0.085	>50	>50	0.002	0.017
CNE14	B	7920	0.005	0.080	2.52	0.389	0.091	0.024	>50	>50	0.002	0.014
CNE4	B	13863	0.0009	0.015	0.056	0.871	0.238	0.113	>50	>50	11.5	3.15
CNE57	B	1687	10.2	0.010	0.156	0.535	0.182	0.066	>50	>50	0.008	0.009
HO86.8	B	1372	0.051	0.104	0.344	>50	>50	>50	0.014	0.001	>50	>50
HT593.1	B	7037	>50	0.016	0.397	0.438	0.099	0.229	0.271	0.153	>50	>50
HXB2.DG	B	6443	0.013	0.002	0.043	0.040	0.009	0.037	0.553	>50	>50	11.0
JRCSF.JB	B	4641	0.002	0.065	4.12	0.234	0.060	0.028	0.003	0.002	0.061	0.012

		ID50	**IC50 (μg ml^{-1})**									
Virus ID	**Clade**	**N152**	**35O22**	**10E8**	**4E10**	**VRC01**	**NIH 45-46**	**3BNC 117**	**PG9**	**PG16**	**PGT121**	**PGT128**
JRFL.JB	B	860	0.013	0.247	5.37	0.033	0.009	<0.003	>50	>50	0.017	0.011
MN.3	B	48721	0.007	0.0006	0.016	0.033	0.004	>50	>50	>50	>50	>50
PVO.04	B	359	0.011	1.78	1.99	0.386	0.148	0.074	6.24	19.8	0.132	0.010
QH0515.01	B	7984	0.033	1.78	2.00	0.523	0.689	0.175	>50	>50	8.70	>50
QH0692.42	B	6528	0.021	0.358	1.60	1.16	0.991	0.275	>50	>50	0.940	0.046
REJO.67	B	9758	0.0001	0.093	0.167	0.045	0.011	0.039	0.005	0.005	8.87	>50
RHPA.7	B	1939	>50	0.640	13.3	0.047	0.022	0.019	>50	1.32	0.014	0.029
SC422.8	B	3070	25.5	0.196	1.49	0.132	0.031	0.049	0.535	1.20	0.098	2.41
SF162.LS	B	17317	>50	0.107	0.412	0.237	0.025	0.019	>50	>50	0.004	0.004
SS1196.01	B		0.062	0.193	0.686	0.276	0.047	0.038	0.293	0.069	0.002	0.011
THRO.18	B	351	0.908	0.048	1.77	4.42	1.88	2.80	15.0	0.975	>50	>50
TRJO.58	B	1231	0.002	0.881	8.75	0.079	0.029	0.062	0.246	0.393	4.31	0.014
TRO.11	B	2773	>50	0.026	0.599	0.343	1.04	0.033	41.5	4.86	0.006	0.017
WITO.33	B	133	>50	0.247	1.20	0.112	0.037	0.030	0.023	0.023	0.787	>50
YU2.DG	B	279	0.287	0.895	16.0	0.055	0.031	0.029	3.69	0.041	0.068	0.058
CH038.12	BC	4565	1.10	0.543	4.57	0.379	0.183	>50	0.500	49.0	0.004	0.004
CH070.1	BC	3504	>50	1.13	21.4	18.7	>50	7.89	0.006	0.002	0.003	0.027
CH117.4	BC	8831	0.0006	0.146	0.343	0.059	0.022	0.663	0.008	0.005	>50	>50
CH181.12	BC	4947	>50	0.287	5.45	0.540	0.189	0.124	0.008	0.002	0.007	0.018
CNE15	BC	29955	>50	0.802	3.36	0.080	0.052	>50	0.023	0.023	19.0	>50
CNE40	BC	43426	0.002	0.0006	0.003	0.425	0.320	0.116	1.16	49.0	0.224	>50
CNE7	BC	82869	0.004	0.034	0.128	0.540	0.147	>50	1.66	0.393	0.032	0.058
286.36	C		0.001	1.05	0.988	0.103	0.953	0.067	0.071	0.005	0.002	0.014
288.38	C	1949	>50	0.580	0.456	1.52	0.128	0.063	3.14	0.186	0.006	0.019
0013095-2.11	C	1930	>50	0.003	0.078	0.142	0.019	0.208	0.023	0.023	>50	>50
001428-2.42	C	2688	>50	0.750	9.10	0.023	0.009	0.010	0.023	0.023	0.023	0.039
0077_V1.C16	C	1658	0.003	1.19	0.886	1.04	0.308	>50	0.091	0.023	>50	>50
00836-2.5	C	265	>50	0.492	1.43	0.128	0.015	>50	49.0	>50	31.8	>50
0921.V2.C14	C	46	>50	0.773	5.63		0.181	0.243			>50	>50
16055-2.3	C	7672	>50	0.571	4.12	0.105	0.068	3.24	0.014	0.005	1.02	>50
16845-2.22	C	11099	>50	0.001	0.459	2.41	3.19	29.6	2.38	27.8	9.41	0.233
16936-2.21	C	8448	0.001	0.166	1.79	0.109	0.023	0.059	>50	>50	0.003	0.030
25710-2.43	C	9868	>50	0.013	0.376	0.545	0.202	0.100	0.038	0.023	0.014	0.021
25711-2.4	C	4093	>50	0.448	6.24	0.712	0.718	>50	1.50	0.037	0.010	0.019
25925-2.22	C	3217	>50	0.287	2.23	0.559	0.141	0.136	0.023	0.023	0.024	0.014
26191-2.48	C	35.2	>50	1.07	4.20	0.195	0.115	0.043	0.142	1.95	0.150	0.025
3168.V4.C10	C	7844	0.038	1.23	2.63	0.131	0.178	0.110	0.162	0.037	0.485	>50
3637.V5.C3	C	777	47.9	1.66	3.51	4.09	1.77	>50	>50	>50	>50	>50
3873.V1.C24	C	1353	>50	3.81	3.05	0.954	0.141	6.97	>50	12.2	0.015	0.015
6322.V4.C1	C	8201	3.19	1.80	7.95	>50	>50	>50	>50	>50	>50	>50
6471.V1.C16	C	3099	>50	4.62	23.1	>50	>50	>50	>50	>50	>50	>50
6631.V3.C10	C	49.4	>50	0.643	>50	>50	>50	>50	>50	>50	>50	1.99
6644.V2.C33	C	2654	0.0008	0.002	0.781	0.164	0.071	0.033	0.033	35.3	0.018	0.100
6785.V5.C14	C	17458	>50	0.435	1.96	0.332	0.311	0.195	0.023	0.023	0.019	0.019
6838.V1.C35	C	719	>50	0.207	1.66		1.52	0.281			0.119	>50
96ZM651.02	C	7423	>50	0.004	0.113	0.525	0.416	0.443	>50	>50	0.009	0.131

		ID50	IC50 (μg ml^{-1})									
Virus ID	Clade	N152	35O22	10E8	4E10	VRC01	NIH 45-46	3BNC 117	PG9	PG16	PGT121	PGT128
BR025.9	C	7102	0.006	0.143	3.73	0.271	0.067	>50	0.044	0.009	0.002	0.004
CAP210.E8	C	33972	0.042	0.267	4.00	>50	>50	8.16	0.087	0.023	>50	>50
CAP244.D3	C	2859	20.9	0.178	1.98	0.857	0.277	0.073	0.088	0.023	>50	>50
CAP45.G3	C	1909	0.010	0.527	2.36	9.47	>50	0.589	0.023	0.023	2.08	>50
CNE30	C	3150	>50	0.424	1.73	0.927	0.624	0.291	>50	>50	0.061	0.853
CNE31	C	88.2	>50	1.14	3.12	0.962	0.253	9.95	13.5	2.51	0.789	14.9
CNE53	C	14560	>50	0.185	0.032	0.108	0.035	0.051	0.147	>50	0.022	0.034
CNE58	C	368	>50	0.346	0.870	0.124	1.04	0.389	0.023	0.023	>50	7.84
DU123.06	C	12736	6.33	0.089	0.343	13.6	>50	0.183	0.091	0.023	0.033	0.072
DU151.02	C	4533	0.094	0.353	2.97	7.70	>50	>50	0.023	0.023	0.005	0.012
DU156.12	C	2021	11.7	0.007	0.023	0.082	0.029	0.035	0.023	0.023	0.005	0.024
DU172.17	C	2564	>50	0.008	0.023	>50	>50	0.289	0.262	0.030	0.104	0.044
DU422.01	C	325	0.021	1.06	1.65	>50	>50	>50	0.303	0.023	0.164	0.175
MW965.26	C	42805	5.60	0.001	0.025	0.038	0.024	0.005	1.99	0.961	0.011	0.315
SO18.18	C	2603	>50	0.422	8.79	0.071	0.028	0.032	0.061	0.023	0.002	0.056
TV1.29	C	241	>50	0.353	1.95	>50	>50	>50	0.008	0.002	0.118	0.077
TZA125.17	C	11592	>50	0.405	0.431	>50	>50	>50	0.231	0.024	9.96	1.20
TZBD.02	C	409	>50	1.67	2.32	0.072	0.011	45.9	0.266	0.025	0.005	>50
ZA012.29	C	39	>50	0.827	5.80	0.250	0.098	0.063	27.0	0.631	0.005	0.028
ZM106.9	C	256	0.372	>50	16.1	0.248	0.058	0.082	0.639	1.10	0.005	0.024
ZM109.4	C	1856	>50	0.273	0.708	0.134	0.046	0.041	0.106	4.93	13.7	>50
ZM135.10a	C		>50	0.338	0.034	1.28	0.280	0.067	>50	>50	1.50	30.0
ZM176.66	C	551	>50	0.184	0.669	0.038	0.096	>50	0.007	0.002	13.8	0.021
ZM197.7	C	784	>50	0.046	0.425	0.624	0.314	0.398	0.414	0.650	>50	>50
ZM214.15	C	6130	0.034	1.69	1.45	0.881	0.132	0.088	>50	>50	0.682	1.44
ZM215.8	C	2557	0.690	0.017	0.641	0.276	0.015	0.010	0.023	>50	0.014	0.039
ZM233.6	C	955	0.007	0.103	2.06	4.25	5.44	0.202	0.023	0.023		
ZM249.1	C	896	0.001	0.787	2.75	0.082	0.038	0.039	0.033	0.073	>50	10.7
ZM53.12	C	7255	>50	2.56	4.52	0.839	0.205	0.212	0.041	0.023	0.002	>50
ZM55.28a	C	133	1.93	2.31	8.79	0.144	0.085	0.040	0.571	>50	0.070	0.029
3326.V4.C3	CD	111	>50	1.18	1.93	0.073	4.27	48.1	0.023	0.023	>50	>50
3337.V2.C6	CD	22394	0.002	1.23	1.91	0.063	0.034	0.008	>50	>50	21.1	0.006
3817.v2.c59	CD		>50	0.549	4.78	>50	>50	0.216	0.007	0.006	>50	0.022
231965.c1	D	7990	>50	8.03	21.7	0.487	0.060	0.035	1.51	4.72	>50	>50
247-23	D	9426	0.006	0.368	2.37	24.2	47.8	0.028	0.195	>50	>50	>50
3016.v5.c45	D	6403	9.85	0.312	4.80	0.111	>50	0.648	0.286	>50	>50	>50
57128.vrc15	D	11880	0.167	0.203	1.93	>50	>50	0.432	0.104	0.162	2.16	0.069
6405.v4.c34	D	19040	0.115	0.325	3.92	2.63	0.731	0.171	>50	>50	0.019	6.86
A03349M1.vrc4a	D	15192	0.122	0.292	3.12	4.66	>50	0.512	>50	>50	0.013	0.018
NKU3006.ec1	D	12585	0.008	0.588	5.83	0.506	0.615	0.089	>50	>50	>50	45.0
UG021.16	D	7703	>50	0.022	0.196	0.266	1.48	0.410	>50	>50		
UG024.2	D	11754	>50	0.013	0.091	0.106	>50	0.057	3.94	>50	>50	>50
X2088.c9	G	8786	3.62	>50	>50	>50	>50	>50	>50	>50	0.003	>50
SIVmac251.30.SC	NA	<5	>50	>50	>50	>50	>50	>50	>50	>50	>50	>50
SVA.MLV	NA	<5	>50	>50	>50	>50	>50	>50	>50	>50	>50	>50

[a] The data for N152 shows the ID_{50} of serum against each virus. $ID_{50}>1000$ is highlighted in red, $500<ID_{50}<1000$ is in orange, and $100<ID_{50}<500$ is in yellow. [b] The data for the monoclonal antibodies shows the IC_{50}. $IC_{50}<0.1$μg ml-1 is in red, $0.1<IC_{50}<1$μg ml^{-1} is in orange, $1<IC_{50}<10$ μg ml^{-1} μg ml-1 is in yellow, and $10<IC_{50}<50$ μg ml^{-1} is in green.

Supplementary Table 4. 35O22 neutralization of pseudotyped $\text{HIV}_{\text{JRCSF}}$ alanine mutants.

	Domain	Virus ID	IC_{50} ($\mu\text{g ml}^{-1}$)^{a}	Ratio
		WT	0.0106	NA
gp120	C1	E83A	0.0210	1.98
		V84A	0.0606	5.71
		V85A	0.0436	4.12
		L86A	0.1562	14.74
		E87A	0.0107	1.01
		N88A	>20	>1886
		V89A	>20	>1886
		T90A	>20	>1886
		E91A	0.0136	1.29
		D92A	0.0126	1.19
		F93A	No infection	NA
	V1	N134A	0.0073	0.69
		N141A	0.0049	0.46
		N156A	0.0046	0.44
	V2	N160K	0.0365	3.44
		N186A	0.0071	0.67
	C2	N197A	0.0018	0.17
		L226A	0.0143	1.35
		K227A	>20	>1886
		C228A	0.0115	1.08
		N229A	0.0127	1.20
		N230A	>20	>1886
		K231A	0.1303	12.29
		T232A	0.3346	31.57
		F233A	0.0176	1.66
		N234A	0.0138	1.30
		G235A	0.0078	0.74
		K236A	0.0239	2.25
		K237A	0.0139	1.31
		Q238A	0.0407	3.84
		C239A	No infection	NA
		K240A	0.0025	0.24
		N241A	>20	>1886
		V242A	0.0224	2.11
		S243A	>20	>1886
		T244A	0.0275	2.60
		V245A	0.0155	1.46
		Q246A	0.0121	1.14
		N262A	0.0068	0.64
		N276A	0.0088	0.83
		N280A	0.0027	0.26
		N289A	0.0152	1.44
		N295A	0.0229	2.16
	V3	N301A	0.0140	1.32

		N302A	0.0153	1.44
	C3	N332A	0.0065	0.61
		N339A	0.0347	3.27
		N355A	0.0969	9.14
	V4	N392A	0.0089	0.84
		N396A	0.0106	1.00
		N411A	0.0117	1.11
	C4	N425A	0.0063	0.60
		N448A	0.0185	1.75
		N461A	0.0238	2.24
		N478A	0.0189	1.78
gp41	**NHR**	N553A	0.0128	1.21
		N554A	0.0079	0.75
		N611A	0.0158	1.49
		N616A	0.0144	1.36
	CHR	D620A	0.0076	0.72
		S621A	0.0242	2.29
		I622A	0.0050	0.47
		W623A	0.0844	7.96
		N624A	0.0568	5.36
		N625A	>20	>1886
		M626A	0.0154	1.45
		T627A	0.0034	0.32
		W628A	0.0145	1.37
		M629A	0.1015	9.58
		E630A	0.0366	3.45
		N637A	0.0147	1.39
		N656A	0.0237	2.24
	TR	N671A	0.0390	3.68

Double/Triple Mutants

Virus ID	**IC$_{50}$ (μg ml^{-1})^{a}**	**Ratio**
N230A N241A	>20	>1886
N241A N625A	>20	>1886
N230A N625A	>20	>1886
K227A N230A	>20	>1886
K227A N241A	>20	>1886
K227A S243A	>20	>1886
K227A N625A	>20	>1886
N230A S243A	>20	>1886
N241A S243A	>20	>1886
S243A N625A	>20	>1886
N230A N241A N625A	>20	>1886

[a] IC$_{50}$ values >30-fold compared to HIV wild-type virus are highlighted in yellow.

Supplementary Table 5. 35O22 neutralization of HIV_{LAI} alanine mutants.

	Domain	Virus ID	IC_{50} (μg ml^{-1})[a]	Ratio
		WT	0.0141	NA
gp120	C1	Y40F	0.0143	1.02
		L52V	0.0074	0.52
		F53V	0.0035	0.25
		N88A	>1	>91
		V89A	0.4041	28.69
	C2	T232Y	0.1901	13.50
		N234A	0.0197	1.40
		N241A	0.1480	10.51
		T244V	0.3240	23.00
	C5	V488A	0.0053	0.38
		V489A	0.3527	25.04
		K490T	0.0203	1.44
		K490F	0.0142	1.01
		T499A	0.0730	5.19
		K500A	0.0315	2.23
		A501T	0.0075	0.53
		K502T	0.0359	2.55
		R504T	0.0533	3.78
		V505A	0.0091	0.65
		V505K	0.0200	1.42
		V506A	0.0191	1.35
		V506K	0.0370	2.63
gp41	NHR	K588A	0.0091	0.65
		K588N	0.0064	0.46
		K588F	0.0067	0.47
		K601Q	0.0099	0.70
		K601A	0.0893	6.34
		A607N	0.0130	0.92
		N611D	0.0080	0.57
		N611A	0.0018	0.13
		N616Q	0.0046	0.32
		N624Q	0.5010	35.57
	CHR	N637Q	0.0048	0.34
		N637A	0.0019	0.14
		E647A	0.0017	0.12

[a] IC_{50} values >10-fold compared to HIV wild-type virus are highlighted in yellow.

Supplementary Table 6. Data collection and refinement statistics for 35O22 Fab.

	35O22 Fab
Data collection	
Space group	$P4_12_12$
Cell dimensions	
a, *b*, *c* (Å)	57.2, 57.2, 267.7
a, b, g (°)	90.0, 90.0, 90.0
Resolution (Å)	50-1.55 (1.58-1.55)*
R_{sym} or R_{merge}	11.1 (48.6)
I / s*I*	20.3 (2.9)
Completeness (%)	98.0 (92.8)
Redundancy	11.2 (5.0)
Molecules/ASU	1
Refinement	
Resolution (Å)	39.1-1.55
No. reflections	64,474
R_{work} / R_{free}	0.1665/0.1822
No. atoms	6,866
Protein	
Ligand/ion	0
Water	449
B-factors	
Protein	32.75
Ligand/ion	0
Water	38.59
R.m.s. deviations	
Bond lengths (Å)	0.005
Bond angles (°)	1.035
PDB ID	4TOY

*Values in parentheses are for highest-resolution shell.
The dataset was collected from a single crystal.

1. Supplementary Table 7. 35O22 neutralizing activity with reversion of an 8 amino acid FR3 insertion to germline

Clade	Virus ID	Monoclonal antibody[a]		Fold change[b]
		35O22	35O22 d8aa	
A	KER2018	0.004	>20	>4933
	RWO20.2	0.010	1.110	>114
	Q168.a2	0.009	>20	>2317
	Q769.d22	0.002	>20	>8029
	Q769.h5	0.004	>20	>4765
B	JRFL.JB	0.013	0.060	4.7
	BaL.01	0.003	0.023	6.8
	YU2.DG	0.287	>20	>70
	PVO.04	0.011	0.400	36.5
	TRO.11	>50	>20	NA
	CAAN.A2	0.003	0.482	142.1
	TRJO.58	0.002	0.005	2.2
	THRO.18	0.908	>20	>22
	BG1168.1	>50	>20	NA
	6101.1	>50	>20	NA
C	ZA012.29	>50	>20	NA
	DU156.12	11.700	>20	>2
	DU422.01	0.021	>20	>971
	ZM106.9	0.372	>20	>54
	ZM55.28a	1.930	>20	>10
Control	SIVmac251.SG3	>50	>50	>50
Geometric Mean IC$_{50}$ (µg ml^{-1})		0.031	0.107	3.5
Median IC$_{50}$ (µg ml^{-1})		0.010	0.230	22.0

[a] Concentration is µg ml^{-1}. IC$_{50}$<0.1µg ml^{-1} is in red, 0.1<IC$_{50}$<1µg ml^{-1} is in orange, 1<IC$_{50}$<10 µg ml^{-1} is in yellow, and 10<IC$_{50}$<50 µg ml^{-1} is in green.

[b] Fold change=IC$_{50}$ of 35O22 d8aa / IC$_{50}$ of 35O22. Fold changes >10 are highlighted in yellow.

Supplementary Table 8. Conservation of N-linked glycosylation sequons in HIV-1 Env.

Residue number		% Glycan Conservation	Residue number		% Glycan Conservation	Residue number		% Glycan Conservation
Alignment	HXB2		Alignment	HXB2		Alignment	HXB2	
44	29	3.8	272	-	6.75	586	395	1.48
78	49	9.85	273	-	3.82	587	396	25.21
125	88	98.59	274	-	1.55	588	397	9.94
170	130	60.56	311	-	1.24	589	398	37.91
172	132	1.13	312	-	2.42	590	399	8.32
173	133	16.69	313	-	4.97	591	400	7.34
174	134	3.45	314	-	7.2	592	401	8.25
175	135	26.35	315	-	11.04	593	402	20.49
176	136	15.92	316	-	13.6	594	403	11.37
177	137	24.76	317	187	32.45	595	404	12.4
178	138	16.3	318	188	24.45	596	405	8.68
179	139	15.45	327	197	97.84	597	406	5.72
180	140	18.5	366	229	1.69	598	407	5.16
181	141	15.83	367	230	32.68	599	408	4.2
182	142	13.79	369	232	3.02	600	-	1.62
183	-	9.33	371	234	79.7	613	-	1.66
184	-	5.63	380	241	97	614	409	3.05
185	-	4.38	404	262	99.41	615	410	9.99
186	-	3.12	411	268	1.17	616	411	45.96
187	-	1.97	424	276	94.7	617	412	1.24
188	-	1.29	440	289	69.36	618	413	24.01
189	-	1.08	446	293	5.84	667	442	26.89
215	-	1.27	450	295	60.12	669	444	6.57
216	-	1.9	456	301	93.13	675	446	7.22
217	-	3.17	498	332	71.61	677	448	86.4
218	-	4.1	500	334	22.06	693	460	14.82
219	-	7.62	510	337	4.31	694	461	26.68
220	143	11.44	512	339	65.79	695	462	31.82
221	144	10.88	517	344	2.91	696	463	20.28
222	145	12.8	530	354	9.05	697	-	3.87
223	146	5.39	539	355	4.78	698	-	1.62
224	147	9.8	540	356	73.62	733	-	1.83
225	148	4.45	542	358	4.99	734	-	28.14
226	149	12.71	544	360	3.42	736	465	27.15
227	150	1.81	546	362	35.19	919	611	98.41
234	156	95.85	553	363	8.3	937	616	91.07
238	160	91.3	577	386	86.96	939	618	4.48
269	185	7.76	583	392	78.8	949	625	95.99
270	186	15.76	584	393	2.06	962	637	96.2
271	-	5.89	585	394	4.64	1003	674	13.22

The conservation (in percent, on a color scale of white to red) of N-linked glycosylation sequons at HIV-1 gp140 residues is based on analysis of 4265 HIV-1 sequences (http://www.hiv.lanl.gov/content/sequence/NEWALIGN/align.html). Shown are gp140 residue positions where an N-linked glycosylation sequon is present in more than 1% of the sequences.