LETTER

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A single injection of anti-HIV-1 antibodies protects against repeated SHIV challenges

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Despite the success of potent anti-retroviral drugs in controlling human immunodeficiency virus type 1 (HIV-1) infection, little progress has been made in generating an effective HIV-1 vaccine. Although passive transfer of anti-HIV-1 broadly neutralizing antibodies can protect mice or macaques against a single high-dose challenge with HIV or simian/human (SIV/HIV) chimaeric viruses (SHIVs) respectively[1–8], the long-term efficacy of a passive antibody transfer approach for HIV-1 has not been examined. Here we show, on the basis of the relatively long-term protection conferred by hepatitis A immune globulin, the efficacy of a single injection (20 mg kg^{-1}) of four anti-HIV-1-neutralizing monoclonal antibodies (VRC01, VRC01-LS, 3BNC117, and 10-1074 (refs 9–12)) in blocking repeated weekly low-dose virus challenges of the clade B SHIV$_{AD8}$. Compared with control animals, which required two to six challenges (median = 3) for infection, a single broadly neutralizing antibody infusion prevented virus acquisition for up to 23 weekly challenges. This effect depended on antibody potency and half-life. The highest levels of plasma-neutralizing activity and, correspondingly, the longest protection were found in monkeys administered the more potent antibodies 3BNC117 and 10-1074 (median = 13 and 12.5 weeks, respectively). VRC01, which showed lower plasma-neutralizing activity, protected for a shorter time (median = 8 weeks). The introduction of a mutation that extends antibody half-life into the crystallizable fragment (Fc) domain of VRC01 increased median protection from 8 to 14.5 weeks. If administered to populations at high risk of HIV-1 transmission, such an immunoprophylaxis regimen could have a major impact on virus transmission.

It is now recognized that, unlike most other prophylactic vaccines for human viral pathogens, an effective vaccine against HIV-1 will probably need to completely block the establishment of a productive infection within a very short time frame (1–3 days of transmission). Such protection has, in fact, been achieved by administering polyclonal and monoclonal anti-HIV-1-neutralizing antibodies (NAbs) to humanized mice or macaques before challenge with SIV/HIV chimaeric viruses (SHIVs)[1–8].

During the past 7 years, monoclonal antibodies (MAbs) have been isolated from selected HIV-1 infected individuals, who generate anti-viral NAbs (bNAbs) with broad and potent activity against isolates of diverse genetic and geographical origin[13]. Several of these bNAbs have been used to suppress ongoing viral infections in humanized mice, macaques, and humans[14–18]. Pre-exposure immunoprophylaxis with bNAbs has also been evaluated in macaque models. In most of these experiments, a single dose of antibody, typically infused 24–48 h before a single high-dose virus challenge, was sufficient to block infection by a virus challenge, capable of establishing an infection in all untreated animals[4,19–21]. Humans, however, are usually exposed to much lower doses of virus on several occasions before becoming infected with HIV-1 (ref. 22).

It is worth noting that before the development of an effective hepatitis A virus vaccine, pre-exposure immunoprophylaxis with hepatitis A immune globulin was common practice for travellers to endemic regions of the world; protective effects lasted 3–5 months[23]. Prophylactic administration of antibodies against other microbial pathogens has also been used to prevent disease[24]. On the basis of this idea, we explored the possibility that a single administration of a potent neutralizing anti-HIV MAb, in the setting of repeated low-dose SHIV challenges, might protect for extended periods of time, thereby providing a proof of concept for periodic administration of MAb as an alternative to HIV-1 vaccination.

We initially selected three MAbs for the repeated low-dose SHIV challenge experiment on the basis of their previously described activity in blocking virus acquisition in a cohort of 60 macaques after a single high-dose SHIV challenge[21]. Two of these antibodies (VRC01 (ref. 12) and 3BNC117 (ref. 11)) target the gp120 CD4bs and one (10-1074 (ref. 10)) is dependent on the presence of HIV-1 gp120 N332 glycan, located immediately downstream of the V3 loop. The challenge virus selected for the present study was SHIV$_{AD8\text{-}EO}$ (ref. 25), an R5-tropic molecular-cloned derivative of the clade B SHIV$_{AD8}$ (ref. 26), which possesses multiple properties typical of pathogenic HIV-1 isolates[27].

When tested against large HIV-1 pseudovirus panels including multiple clades, 3BNC117 and VRC01 neutralize more than 80% of the viral isolates and 10-1074 neutralizes between 60% and 70%. Against sensitive viruses, 10-1074 is the most potent, followed by 3BNC117 and VRC01 (ref. 28). Consistent with this trend, the 50% inhibitory concentration (IC_{50}) values for VRC01, 3BNC117, and 10-1074 against SHIV$_{AD8\text{-}EO}$ were 0.67, 0.06, and 0.08 μg ml^{-1}, respectively, and the 80% (IC_{80}) values were 2.04, 0.19, and 0.18 μg ml^{-1}, respectively (Extended Data Fig. 1a). Neutralization sensitivities were also measured using the SHIV challenge stock in a single round of infection assay in TZM-bl cells, using replication-competent SHIV$_{AD8\text{-}EO}$. The IC_{50} and IC_{80} values for VRC01, 3BNC117, and 10-1074 in this assay system were 2.06, 0.12, and 0.05, and 7.14, 0.32, and 0.14 μg ml^{-1}, respectively (Extended Data Fig. 1b).

In an initial experiment designed to simulate low-dose mucosal transmission in humans, a cohort of nine monkeys was challenged weekly by the intrarectal route with ten 50% tissue culture infectious doses ($TCID_{50}$) of SHIV$_{AD8\text{-}EO}$, in the absence of antibody treatment.

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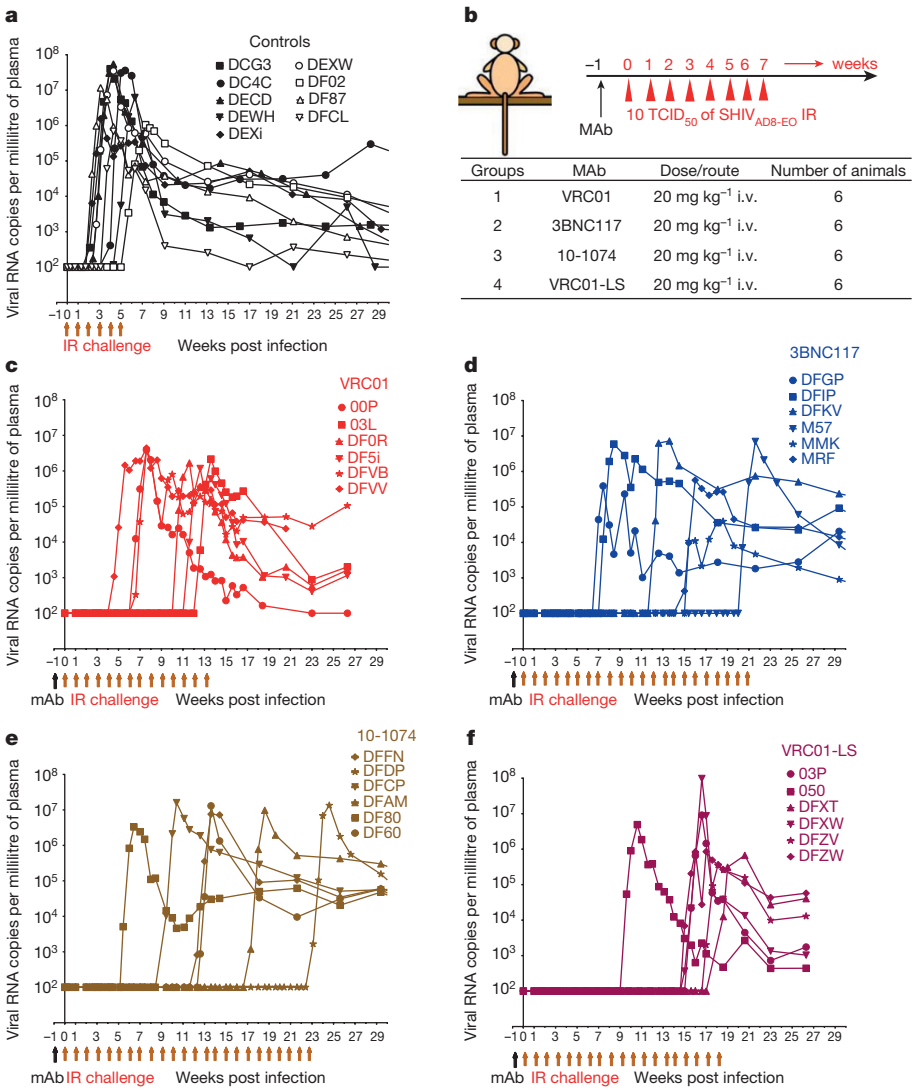


Figure 1 | HIV MAbs delay virus acquisition after repeated low-dose intrarectal $SHIV_{AD8\text{-}EO}$ challenges. a, Plasma viral loads in macaques receiving no MAb (controls, $n = 9$). IR, intrarectal. **b**, Representation of the regimen used to assess the protective efficacy of MAbs. Macaques were intravenously (i.v.) administered the indicated MAbs at a dose of 20 mg kg^{-1} and challenged 1 week later and every week thereafter. **c**–**f**, Plasma viral loads in macaques administered VRC01, 3BNC117, 10-1074, and VRC01-LS bNAbs, respectively.

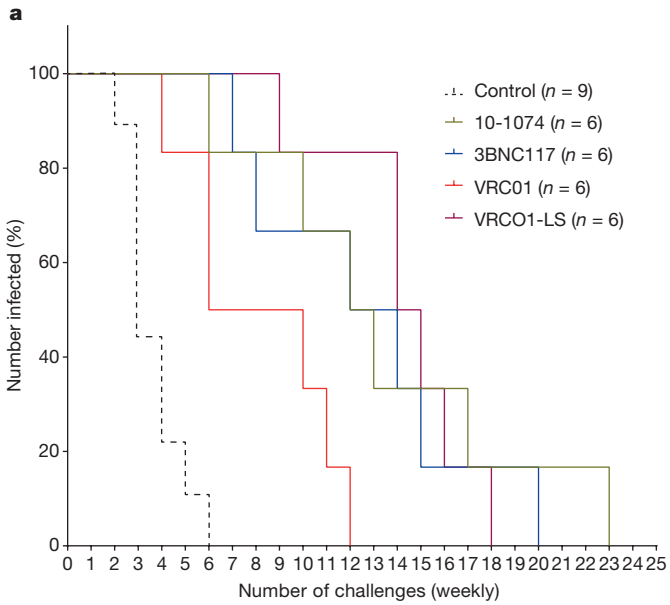
As shown in Fig. 1a, plasma viraemia became detectable after two to six challenges, with a median of 3.0 weekly virus exposures needed to infect all nine animals. On the basis of these results, the inoculum size administered to each monkey per challenge was estimated to be 0.27 50% animal infectious doses (AID_{50}).

The regimen used to assess the protective efficacy of the three anti-HIV-1 MAbs against a repeated low-dose rectal challenge of $SHIV_{AD8\text{-}EO}$ is shown in Fig. 1b. Individual MAbs (20 mg kg^{-1}) were administered a single time intravenously to three cohorts of six animals. Starting 1 week later, each group was challenged weekly by the intrarectal route with ten $TCID_{50}$ of $SHIV_{AD8\text{-}EO}$. Samples of blood, collected at regular intervals, were monitored for levels of viral RNA, concentrations of MAb, and anti-SHIV-neutralizing titres. The number of virus challenges required to establish a $SHIV_{AD8\text{-}EO}$ infection, indicated by measurable viraemia (>100 viral RNA copies per millilitre of plasma), in the recipients of the anti-HIV-1 MAbs was compared with that needed for virus acquisition in the control group.

In all cases, the administration of MAbs delayed virus acquisition. Animals receiving VRC01 required 4–12 challenges; 3BNC117 required 7–20 challenges; and 10-1074, 6–23 challenges (Fig. 1c–e). The differences in the number of challenges required for infection, and thus the median times to virus acquisition compared with control monkeys, were 8 weeks for VRC01, 13 weeks for 3BNC117, and 12.5 weeks for 10-1074.

The pharmacokinetic profile of VRC01 was altered by introducing two amino-acid mutations (M428L and N434S, referred to as 'LS') into its Fc domain, which increased its half-life in both plasma and tissues[9]. The neutralization activity of this VRC01 derivative, designated VRC01-LS, was first tested in the TZM-bl assay and, as expected, it exhibited IC_{50} and IC_{80} values similar to VRC01 (Extended Data Fig. 1a, b). When administered to six macaques in the previously described repeated low-dose $SHIV_{AD8\text{-}EO}$ challenge system, the VRC01-LS-treated animals required 9–18 challenges (median = 14.5) for all of the monkeys to become infected (Fig. 1f). Thus the modified VRC01-LS Fc domain conferred an estimated 1.8-fold increase in the number of challenges, resulting in successful acquisition compared with the parental VRC01 MAb.

The protective effects of the four anti-HIV-1 MAbs are described by Kaplan–Meier analysis in which the percentage of macaques remaining uninfected is plotted against the number of $SHIV_{AD8EO}$ challenges (Fig. 2a). Significantly increased numbers of challenges were required to establish infections in the recipients of the VRC01, 3BNC117, 10-1074, and VRC01-LS MAbs than in the control animals ($P = 0.007$, 0.002, 0.002, and 0.002, respectively), using the Wilcoxon rank-sum test



b

Groups compared		Wilcoxon *P* value
Control	VRC01	0.007
Control	3BNC117	0.002
Control	10-1074	0.002
Control	VRC01-LS	0.002
VRC01	3BNC117	0.077
VRC01	10-1074	0.106
VRC01	VRC01-LS	0.02
10-1074	3BNC117	0.5

Figure 2 | Kaplan–Meier analysis and magnitude of protection by HIV MAbs in repeated low-dose challenge. a, Kaplan–Meier survival curves for recipients of the four bNAbs and the cohort of control animals. The percentage of macaques remaining uninfected is plotted against the number of ten $TCID_{50}$ $SHIV_{AD8\text{-}EO}$ intrarectal challenges required to establish infections. **b**, Statistical differences are represented as *P* values (Wilcoxon rank-sum test) by comparing the number of challenges resulting in infection between control animals and an individual MAb recipient group or between different MAb-treated groups.

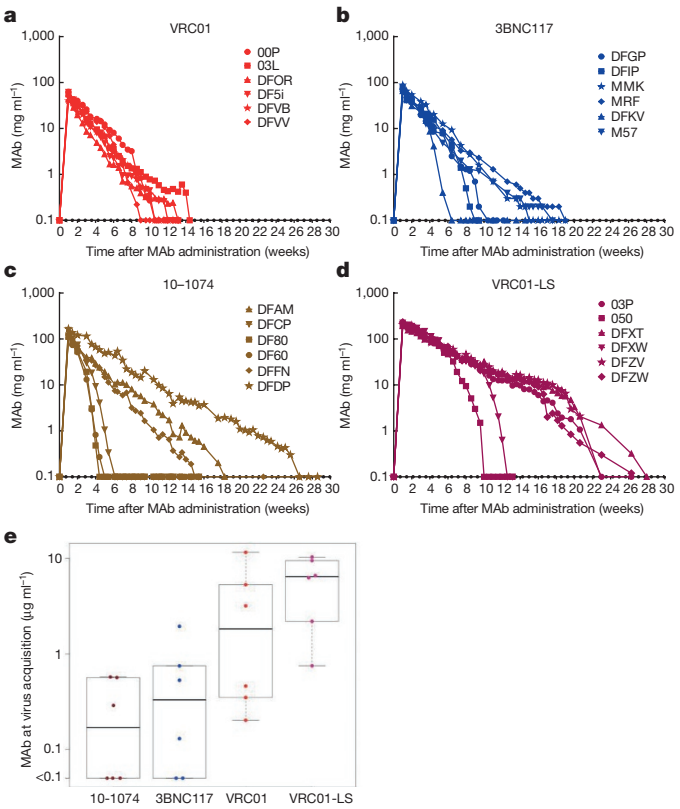


Figure 3 | Plasma concentrations of the infused MAbs in macaques correlate with long-term protection from SHIV infection. a–d, Plasma antibody concentrations in macaques administered VRC01, 3BNC117, 10-1074, and VRC01-LS decay over time. **e**, Median plasma concentrations at the times of virus breakthrough in bNAb recipients were 0.169 (10-1074), 0.330 (3BNC117), 1.825 (VRC01), and 6.446 (VRC01-LS), respectively. Boxes represent the twenty-fifth and seventy-fifth percentiles, and the heavier line represents the median value for each group. The top and bottom horizontal bars outside the boxes represent the maximum and minimum of the data, respectively.

(Fig. 2b). A comparison of the individual pairs of Kaplan–Meier curves revealed that 10-1074, 3BNC117, and VRC01-LS were not significantly different from each other in blocking infection.

Ultrasensitive nested quantitative reverse-transcription PCR (qRT–PCR) and qPCR assays for plasma viral RNA and cell-associated viral RNA and DNA[29] were performed on plasma and peripheral blood mononuclear cell samples, collected from recipients of the different neutralizing MAbs, before $SHIV_{AD8\text{-}EO}$ breakthrough infections, as assessed by plasma viraemia measured in our standard assay. In all cases, the levels of viral RNA and DNA measured with the ultrasensitive assays were below detectable limits (Extended Data Table 1).

The plasma concentrations of the infused MAbs were measured longitudinally in individual animals beginning 1 week after infusion. (Fig. 3 and Extended Data Tables 2 and 3). The median plasma concentrations at the times of virus breakthrough for the 10-1074 and 3BNC117 recipient cohorts were 0.169 and 0.330 $\mu g\,ml^{-1}$, respectively (Fig. 3e). These values are comparable to the IC_{80} values determined *in vitro*, using the TZM-bl assay with replication competent $SHIV_{AD8\text{-}EO}$ (Extended Data Fig. 1b). The median plasma concentrations at the times of virus acquisition for VRC01 and VRC01-LS were 10- to 20-fold higher (1.825 and 6.446 $\mu g\,ml^{-1}$) and were also in the same range as the IC_{80} values determined *in vitro* (Extended Data Fig. 1b). It is worth noting that three of the six recipients of the 10-1074 MAb experienced rapid decay of plasma antibody, which fell to background levels between weeks 4 and 6 after administration (Fig. 3c). A similar pattern occurred for three of the 3BNC117 MAb and VRC01-LS recipients, although the decline of antibody in plasma was delayed in these two groups animals (Fig. 3b). This rapid clearance of plasma MAbs in the subgroups of the 10-1074 and 3BNC117 recipients tracked with the emergence of anti-antibody responses to the infused anti-HIV-1 human MAbs (Extended Data Fig. 2). For monkeys infused with the 10-1074 MAb, the median number of challenges for successful infection in the three-animal subgroup not experiencing the rapid anti-antibody induced decay was 17.0 weeks compared with 12.5 for the entire 10-1074 recipient cohort.

Probit analysis was also used to estimate the probability of infection as a function of the imputed plasma MAb concentration at the time of each challenge. The probability of infection per infection for the control monkeys was 0.27, estimated by pooling all of the $SHIV_{AD8\text{-}EO}$ challenges to this group of animals; this is indicated by the single open circle along the ordinate of Extended Data Fig. 3. Not unexpectedly, the curves relating antibody concentration and virus acquisition for VRC01 and VRC01-LS were superimposed on one another even though VRC01-LS had a longer half-life *in vivo*. In this same analysis, the curves for 10-1074 and 3BNC117, which conferred lower probabilities for infection at each plasma MAb concentration, reflected their greater neutralization potency against the challenge virus, relative to the VRC01 antibodies. At a 10-1074 MAb plasma concentration of 1 $\mu g\,ml^{-1}$, the model predicts a probability of infection, for a single challenge, of 0.044, approximately sixfold less than that estimated for animals receiving no antibodies.

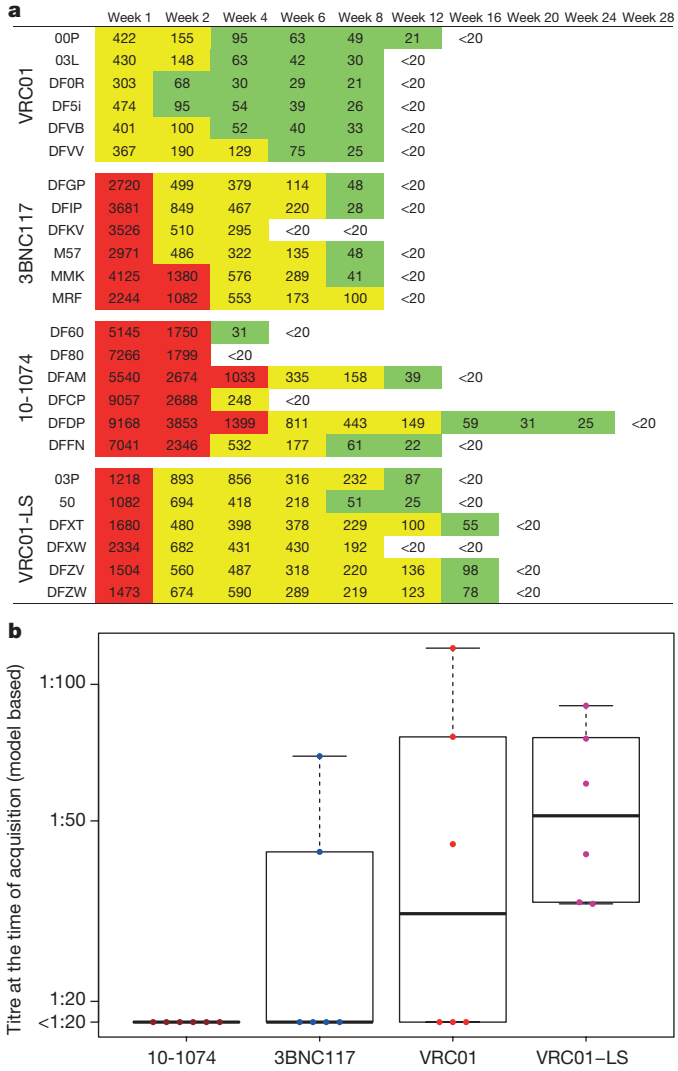


Figure 4 | The decline of neutralizing antibody titres in plasma over time in macaques corresponds to the time of virus acquisition. a, Plasma IC_{50} titres of the indicated MAbs were determined longitudinally using the TZM-bl cell assay. IC_{50} values are colour coded: 21–99 as green; 100–999 as yellow; and ≥1,000 as red. **b**, Plasma-neutralizing titres at the time of virus acquisition for the four groups of MAb recipients. Boxes represent the twenty-fifth and seventy-fifth percentiles, and the heavier line represents the median value for each group. The top and bottom horizontal bars outside of the boxes represent the maximum and minimum of the data, respectively.

The plasma neutralization titre was also determined for each of the MAb recipients at multiple times after infusion (Fig. 4a). The median plasma-neutralizing titres for the four groups of macaques at the time of $SHIV_{AD8\text{-}EO}$ acquisition were low: <1:20 (below the level of detection) for 10-1074 and 3BNC117 recipients; 1:27 for the VRC01 group; and 1:51 for the VRC01-LS cohort (Fig. 4b). As noted earlier for plasma MAb concentrations, the levels of detectable neutralizing activity in members of each cohort inversely correlated with the emergence of anti-antibodies (compare Fig. 4a and Extended Data Fig. 2).

In conclusion a single administration of potent anti-HIV-1-neutralizing MAbs to naive macaques was protective against repeated low-dose SHIV infection for several months. The duration of protection was directly related to antibody potency and half-life. When considered in the context of a potential exposure to HIV-1 in regions of the world where the HIV-1 is endemic, the barrier to infection when antibody concentrations remain above protective levels in infused individuals could have a profound impact on virus transmission. As noted earlier, anti-antibodies directed against some of the administered MAbs emerged quite rapidly in some macaques, and diminished their prophylactic efficacy. However, this is not likely to occur in humans as reported in a recent study of VRC01 (ref. 30). On the basis of the results obtained with VRC01 and VRC01-LS, it is also anticipated that the creation and use of 3BN117 and/or 10-1074 derivatives with the LS mutation should exhibit increased durability *in vivo*, resulting in protection of up to 6 months against $SHIV_{AD8\text{-}EO}$-infected macaques. The administration of a multivalent cocktail of these anti-viral bNAbs could augment their efficacy by increasing overall breadth and their capacity to block the transmission of resistant HIV-1 strains.

Online Content Methods, along with any additional Extended Data display items and Source Data, are available in the online version of the paper; references unique to these sections appear only in the online paper.

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Author Contributions R.G., Y.N., M.A.M., M.C.Nu., and J.R.M. designed experiments; R.G., Y.N., A.P., F.K., A.G., J.G., A.B.W., R.S., K.W., Z.M., and S.D.S. performed experiments; R.G., Y.N., M.C.Na., M.A.M., M.C.Nu., J.R.M., and J.D.L. analysed data; R.G., Y.N., M.A.M., M.C.Nu., J.R.M., and J.D.L. wrote the manuscript.

Author Information Reprints and permissions information is available at www.nature.com/reprints. The authors declare no competing financial interests. Readers are welcome to comment on the online version of the paper. Correspondence and requests for materials should be addressed to M.A.M. (malm@nih.gov).

METHODS

Animal experiments. Thirty-three male and female rhesus macaques (*Macaca mulatta*) of Indian genetic origin from 2 to 4 years of age were housed and cared for in accordance with Guide for Care and Use of Laboratory Animals Report number NIH 82-53 (Department of Health and Human Services, Bethesda, Maryland, 1985) in a biosafety level 2 National Institute of Allergy and Infectious Diseases (NIAID) facility. All animal procedures and experiments were performed according to protocols approved by the Institutional Animal Care and Use Committee of NIAID, NIH. Animals were not randomized and the data collected were not blinded. Phlebotomies, euthanasia, and sample collection were performed as previously described[31]. All of the macaques used in this study were negative for the major histocompatibility complex (MHC) class I *Mamu-A*01*, *Mamu-B*08*, and *Mamu-B*17* alleles. No animals were excluded from the analysis.

Antibodies. The VRC01, 3BNC117, 10-1074, and VRC01-LS anti-HIV-1 monoclonal NAbs were isolated and produced as described elsewhere[9–12]. MAb 10-1074 was produced by transient transfection of IgH and IgL expression plasmids into the human embryonic kidney cells whereas VRC01, VRC01-LS, and 3BNC117 were produced from Chinese hamster ovary cells. All of the MAbs were IgG1. All of the monoclonal antibodies were purified by chromatography and sterile filtration and were endotoxin free. A single dose (20 mg kg^{-1}) of each MAb was administered intravenously to individual animals in four cohorts of monkeys.

Virus challenge. The origin and preparation of the tissue-culture-derived $SHIV_{AD8\text{-}EO}$ stock have been previously described[25]. One week after MAb infusion, animals were challenged intrarectally with ten $TCID_{50}$ of $SHIV_{AD8\text{-}EO}$, and every week thereafter, until a virus infection was established. A paediatric nasal speculum was used to gently open the rectum and a 1 ml suspension of virus was slowly infused into rectal cavity using a plastic tuberculin syringe. An intrarectal challenge $SHIV_{AD8\text{-}EO}$ inoculum size of ten $TCID_{50}$ was chosen for repeated low-dose experiments on the basis of previous results indicating that (1) 1,000 $TCID_{50}$ of $SHIV_{AD8\text{-}EO}$ administered by the intrarectal route resulted in the establishment of infections of 30 of 30 rhesus monkeys and (2) an intrarectal virus titration suggested that 1,000 $TCID_{50}$ of $SHIV_{AD8\text{-}EO}$ was equivalent to approximately ten AID_{50} (ref. 21).

Quantification of viral nucleic acids. Viral RNA levels in plasma were determined by qRT–PCR (ABI Prism 7900HT sequence detection system; Applied Biosystems) as previously described[31]. Ultrasensitive measurement of plasma SIV gag RNA was performed as described, and cell-associated levels of SIV RNA and DNA were determined by a nested, hybrid real-time/digital PCR assay, essentially as reported previously[29].

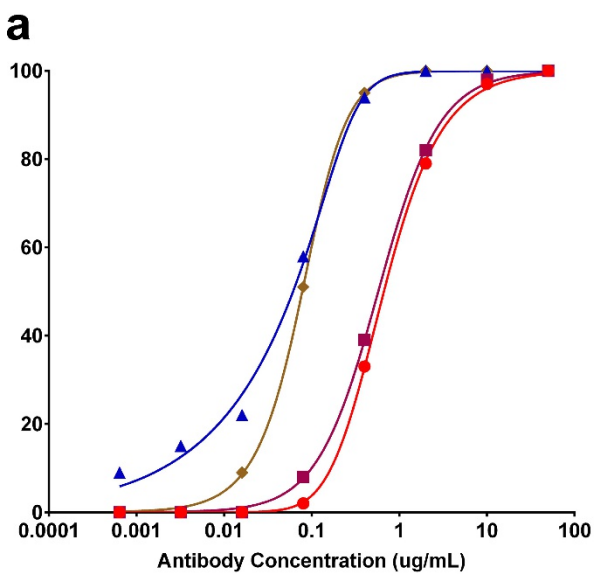
Antibody concentrations in plasma. Plasma antibody levels were quantified by ELISA using purified MAbs as a standard and anti-antibody responses in plasma were also evaluated as reported earlier[9]. These assays were performed twice.

Neutralization assays. The titres of each MAb against $SHIV_{AD8\text{-}EO}$ was assessed by two types of *in vitro* neutralization assay: (1) TZM-bl entry assay with pseudotype challenge virus[25,27] and (2) a single-round TZM-bl infectivity assay with replication competent challenge virus[32]. Antibody concentrations required to inhibit infection by 50% or 80% are reported as IC_{50} or IC_{80}, respectively. TZM-bl cells were obtained through the NIH AIDS Reagent Program, Division of AIDS, NIAID, NIH, from J. C. Kappes, X. Wu and Tranzyme[33].These cells were not authenticated for this study and not tested for mycoplasma contamination. The neutralization activity present in plasma samples collected from rhesus macaques was assessed by TZM-bl entry assay with pseudotype challenge virus. The IC_{50} titre was calculated as the plasma dilution causing 50% reduction in RLUs compared with virus controls. The neutralization assays were repeated twice.

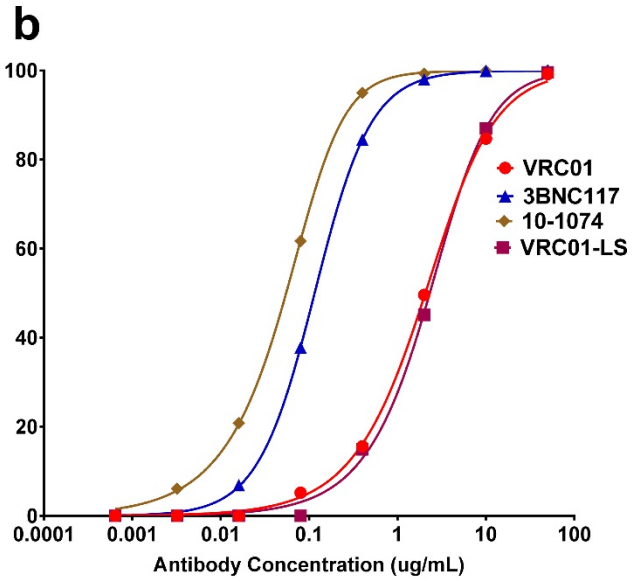
Statistical analyses. No statistical methods were used to predetermine sample size. The experiments were not randomized. The investigators were not blinded to allocation during experiments and outcome assessment.

A Wilcoxon rank-sum test was used to compare number of challenges until infection between each MAb group and control; these comparisons were considered primary and were compared with a Bonferroni-adjusted α of $0.05/4 = 0.0125$ to determine significance. Comparisons between antibodies were considered secondary and not adjusted for multiple comparisons. Finally, probit models were used to model the probability of infection at each challenge as a function of concurrent antibody concentration. Since these values were not always measured at the precise time of challenge, antibody concentrations were modelled separately for each animal over time, and these models were used to impute the concentration at the exact time of each challenge for the probit model.

31. Endo, Y. *et al.* Short- and long-term clinical outcomes in rhesus monkeys inoculated with a highly pathogenic chimeric simian/human immunodeficiency virus. *J. Virol.* **74,** 6935–6945 (2000).
32. Li, M. *et al.* Human immunodeficiency virus type 1 env clones from acute and early subtype B infections for standardized assessments of vaccine-elicited neutralizing antibodies. *J. Virol.* **79,** 10108–10125 (2005).
33. Wei, X. *et al.* Emergence of resistant human immunodeficiency virus type 1 in patients receiving fusion inhibitor (T-20) monotherapy. *Antimicrob. Agents Chemother.* **46,** 1896–1905 (2002).

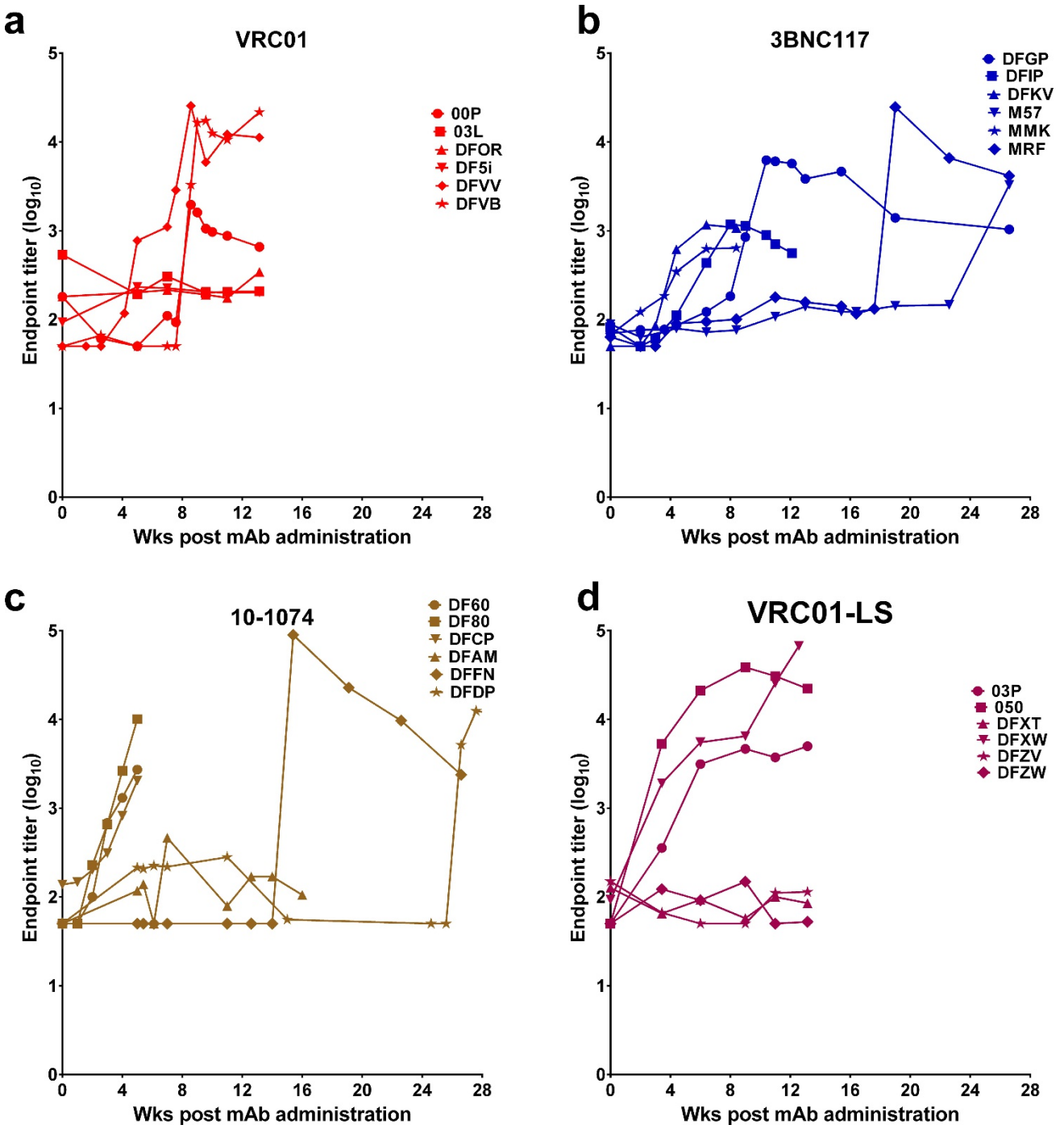


	IC_{50} (ug/mL)	IC_{80} (ug/mL)
VRC01	0.67	2.04
VRC01-LS	0.58	1.76
3BNC117	0.06	0.19
10-1074	0.08	0.18

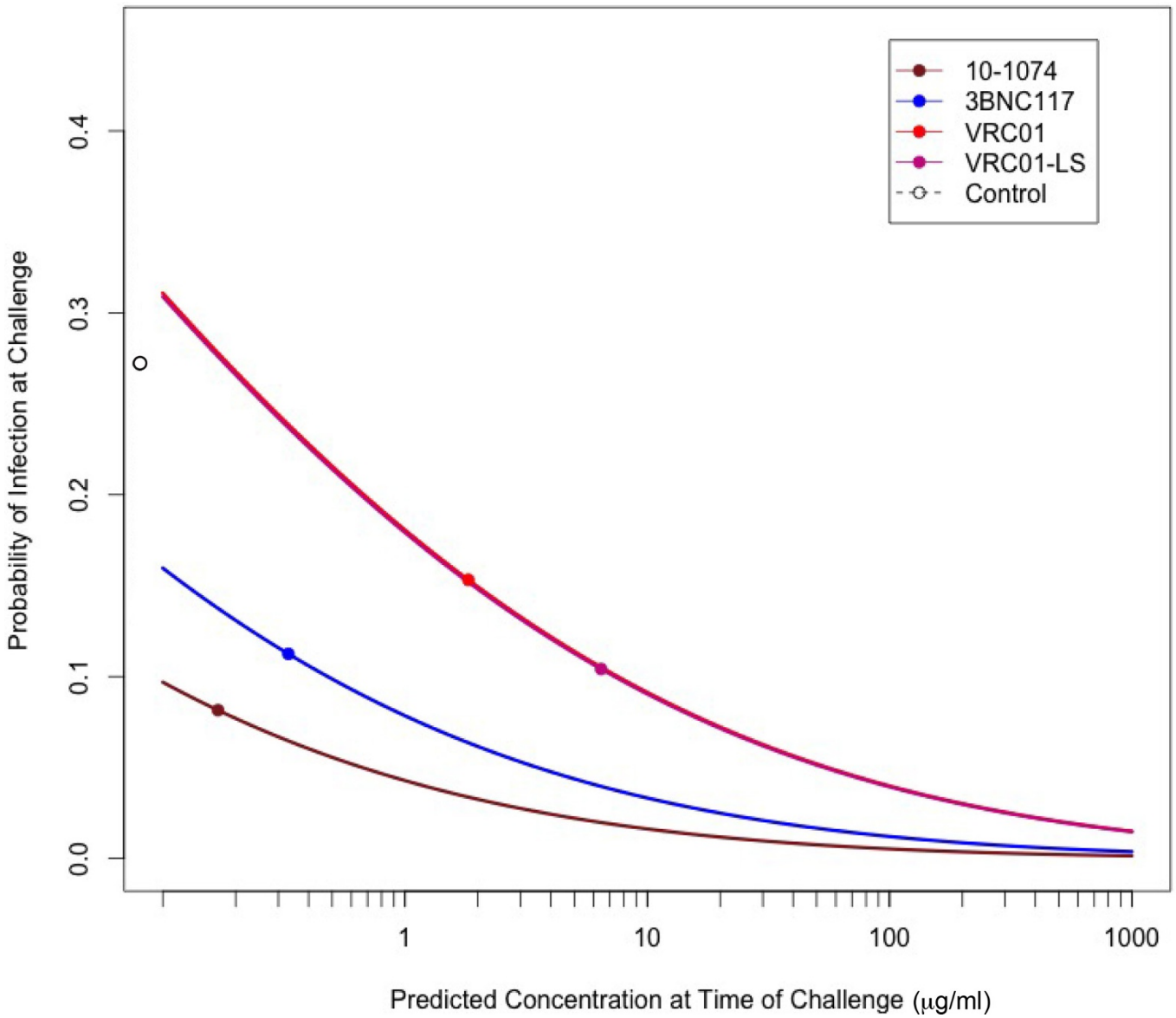


	IC_{50} (ug/mL)	IC_{80} (ug/mL)
VRC01	2.06	7.14
VRC01-LS	2.32	6.83
3BNC117	0.12	0.32
10-1074	0.05	0.14

Extended Data Figure 1 | Neutralization sensitivity of $SHIV_{AD8\text{-}EO}$ to four broadly acting neutralizing anti-HIV-1 MAbs. a, Neutralizing activity of the indicated bNAbs was determined against $SHIV_{AD8\text{-}EO}$ pseudovirions using TZM-bl target cells. The calculated IC_{50} and IC_{80} values are shown at the bottom. **b**, Neutralizing activity of the indicated bNAbs was determined against replication competent $SHIV_{AD8\text{-}EO}$ in a single round TZM-bl infectivity assay. The calculated IC_{50} and IC_{80} values are shown at the bottom. The assay was performed in the presence of indinavir. Both experiments were performed twice.



Extended Data Figure 2 | Development of anti-MAb immune responses in recipients of anti-HIV-1 bNAbs. a–d, Longitudinal analysis of anti-VRC01, anti-3BNC117, anti-10-1074, and anti-VRC01-LS antibody responses, respectively, after a single intravenous infusion of indicated MAbs. This assay was performed twice.



Extended Data Figure 3 | Predicted probability of infection as a function of antibody levels. The per-challenge probability of infection was modelled as a function of antibody concentration at the time of each challenge using a probit regression model. The fitted probabilities from the models are plotted separately for each MAb group, with the estimated probability of infection for the control animals (0.27) indicated by the open circle adjacent to each ordinate. The VRC01 and VRC01-LS curves are superimposed. The points on each curve represent the median concentration at the time of breakthrough infection for each group of monkeys.

Extended Data Table 1 | Plasma viral RNA and cell-associated viral RNA/DNA in rhesus macaques before breakthrough of infection

Animal	Wks post mAb treatment	Plasma Viral RNA (copies/ml)	SIV Gag RNA copies per 10^6 cell eq	SIV Gag DNA copies per 10^6 cell eq
DF60	7.4	<2	<1	<1
	11.4	<2	<1	<1
DF80	3.6	<2	<1	<1
	5.4	<2	<1	<1
DFAM	11.4	<2	<1	<1
	15.4	<2	<1	<1
DFCP	5.4	<2	<1	<1
	7.4	<2	<1	<1
DFDP	13.6	<2	<1	<1
	26.6*	3,600,000	140000	610
DFFN	7.4	<2	<1	<1
	11.4	<2	<1	<1
DFGP	3.6	<2	<1	<1
	5.4	<2	<1	<1
DFIP	5.4	<2	<1	<1
	6.4	<2	<1	<1
DFKV	7.4	<2	<1	<1
	11.4*	10	<1	<1
M57	11.4	<2	<1	<1
	17.6	<2	<1	<1
MMK	11.4	<2	<1	<1
	15.4	<2	<1	<1
MRF	9.4	<2	<1	<1
	13.6	<2	<1	<1
00P	4.6	<2	<1	<1
	6.6*	270	3.2	4.2
03L	8.6	<2	<1	<1
	12.6*	25,000	5100	25
DF0R	6.6	<2	<1	<1
	10.6*	73,000	16000	22
DF5i	6.6	<2	<1	<1
	10.6	<2	<1	<1
DFVB	4.6	<2	<1	<1
	6.6*	1,300	<1	<1
DFVV	2.6	<2	<1	<1
	4.6*	930	<1	<1
03P	10.6	<2	<1	<1
	14.6	<2	<1	<1
O50	4.6	<2	<1	<1
	8.6	<2	<1	<1
DFXT	10.6	<2	<1	<1
	14.6	<2	<1	<1
DFXW	10.6	<2	<1	<1
	14.6	<2	<1	<1
DFZV	10.6	<2	<1	<1
	14.6	<2	<1	<1
DFZW	10.6	<2	<1	<1
	14.6*	10	<1	<1

*Time point collected after breakthrough of infection. Ultrasensitive measurements of plasma SIV RNA or cell-associated SIV RNA and SIV DNA in peripheral blood mononuclear cells were determined by a nested, hybrid real-time/digital PCR assay.

Extended Data Table 2 | VRC01 and 3BNC117 antibody concentrations in the plasma of macaques after a single administration of the indicated MAbs

VRC01 conc (μg/ml)

Wks	00P	03L	DFOR	DF5i	DFVB	DFVV
0.0	0.1	0.1	0.1	0.1	0.1	0.1
1.0	63	55.4	43.1	62.41	40.07	50.86
1.6	43.23	35.25	28.4	42.53	31.03	39.16
2.0	40.16	30.21	19.51	34.13	22.88	32.85
2.6	32.65	22.64	12.98	22.54	19.93	30.92
3.0	25.98	16.11	8.49	14	13.46	23.38
3.4	18.19	12.74	7.17	10.93	11.03	16.62
4.1	16.16	9.53	4.87	8.12	8.41	13.97
4.6	14.43	8.15	3.77	6.47	7.53	14.69
5.0	12.23	6.65	2.75	4.78	7	10.77
5.6	9.28	3.78	1.66	2.5	3.81	6.87
6.0	8.08	3.55	1.61	2.26	3	4.45
6.6	6.03	2.51	1.39	1.52	2.45	2.41
7.0	4.38	1.85	0.7	0.96	2.11	1.61
7.6	3.46	1.53	0.67	0.95	1.55	0.66
8.0	3.21	1.35	0.55	0.92	1.38	
8.6	1.06	1.32	0.5	0.78	0.8	0.22
9.0	0.83	1.12	0.39	0.51	0.52	0.1
9.6	0.33	0.89	0.32	0.48	0.5	0.1
10.0	0.27	0.78	0.31	0.45	0.37	0.1
10.6	0.1	0.73	0.3		0.1	0.1
11.0	0.1	0.58	0.29	0.27	0.1	0.1
11.6	0.1	0.46	0.29	0.26	0.1	0.1
12.0	0.1	0.43	0.23	0.1	0.1	0.1
12.6	0.1	0.48	0.24	0.1	0.1	0.1
13.1	0.1	0.42	0.1	0.1	0.1	0.1
13.6		0.60				
14.0		0.40				
14.4		0.10				

3BNC117 conc (μg/ml)

Wks	DFGP	DFIP	MMK	MRF	DFKV	M57
0.0	0.1	0.1	0.1	0.1	0.1	0.1
1.0	72.6	78.1	88.7	64.3	65.5	63
1.4	57	41.4	65.7	49	44.2	41.9
2.0	47.4	39	54.6	35.1	30.8	37.9
3.0	19.6	29.2	38.8	29.1	23.2	23.4
3.6	14.4	20.9	32.9	17.2	14.7	12.7
4.0	12.2	16.8	20.7	14.7	8.6	11
4.4	8.6	15.5	20.3	11.3	4.1	8.8
5.4	7.8	8	12.6	9.1	0.4	4.9
6.4	2.5	4.4	9.3	5.7	0.1	3.3
7.4	1.5	1.4	4.3	3.3	0.1	2
8.0	1.3	0.5			0.1	
8.4	1.2	0.2	2.7	3	0.1	1.3
9.0	0.7	0.1				
9.4	0.2	0.1	1.6	2.3	0.1	1.2
10.4	0.1					
11.0	0.1		0.7	1.3	0.1	0.7
11.4	0.1					
12.1	0.1					
12.6			0.3	0.7	0.1	0.4
13.6			0.3	0.6	0.1	0.3
14.0			0.2	0.5		0.3
14.6			0.2	0.4	0.1	0.2
15.0			0.1	0.4		0.2
15.4			0.1	0.3		0.2
16.0			0.1	0.3		0.2
16.4			0.1	0.2		0.2
17.0			0.1	0.2		
17.6			0.1	0.2		0.1
18.3			0.1	0.2		
19.0			0.1	0.1		

The plasma concentrations of the infused VRC01 and 3BNC117 were measured longitudinally in the indicated animals.

Extended Data Table 3 | 10-1074 and VRC01-LS antibody concentrations in the plasma of macaques after a single administration of the indicated MAbs

	10-1074 conc (μg/ml)					
Wks	DFAM	DFCP	DF80	DF60	DFFN	DFDP
0.0	0.1	0.1	0.1	0.1	0.1	0.1
1.0	112.2	157.1	118.4	123.2	105	165.2
1.4	85.79	102.9	83.04	73.83	100.2	137.1
2.0	65.71	75.66	54.04	51.46	70.47	121.3
3.0	40.26	30.14	19.31	13.14	34.16	115.3
3.6	38.54	18.54	3.09	2.92	33.9	76.13
4.0	29.05	9.41	0.48	0.82	26.9	66.49
4.4	23.98	4.13	0.1	0.27	23.48	58.02
5.0	19.22	1.07	0.1	0.1	14.98	48.57
5.4	15.36	0.34	0.1	0.1	11.37	50.83
6.1	10.25	0.1	0.1	0.1	7.47	43.68
6.4	11.31	0.1	0.1	0.1	7.15	53.86
7.0	11.17	0.1	0.1	0.1	5.95	34.49
7.4	10.01	0.1	0.1	0.1	6.78	22.26
8.0	8.04	0.1	0.1	0.1	3.19	18.74
8.4	6.29	0.1	0.1	0.1	2.74	16.32
9.0	5.69	0.1	0.1	0.1	2.43	14.52
9.4	4.52	0.1	0.1	0.1	1.65	20.28
10.4	3.78	0.1	0.1	0.1	1.05	13.05
11.0	2.51	0.1	0.1	0.1	0.71	9.75
11.4	2.21	0.1	0.1	0.1	0.84	8.53
12.1	1.71	0.1	0.1	0.1	0.65	6.48
12.6	0.85	0.1	0.1	0.1	0.37	3.94
13.3	0.72	0.1		0.1	0.27	5.44
13.6	1.3		0.1	0.1	0.34	4.5
14.0	0.91			0.1	0.24	4.42
14.6	0.72	0.1	0.1	0.1	0.19	4.19
15.0	0.61				0.1	4.21
15.4	0.58	0.1	0.1	0.1	0.1	3.5
16.0	0.45					3.72
17.6						1.87
18.3						1.95
19.0					0.1	1.8
19.6						1.6
20.0						1.09
20.6						1.03
21.0						0.93
22.0						0.78
22.6					0.1	0.73
23.0						0.62
23.4						0.59
24.1						0.46
24.6						0.41
25.0						0.43
25.6						0.3
26.6					0.1	0.1

	VRC01-LS conc (μg/ml)					
Wks	03P	050	DFXT	DFXW	DFZV	DFZW
0.0	0.1	0.1	0.1	0.1	0.1	0.1
1.0	225.6	192.3	191.8	226.3	206	234
1.6	169.9	181.9	153.7	205.5	185.8	169.8
2.0	147.1	183.8	144.9	192.1	196.5	180.9
2.6	161.1	158.4	134.1	175.7	143.3	160
3.0	139.6	121	114.2	146.9	122.4	126.1
3.4	100.8	123.2	97.5	149.2	99.62	109.9
4.1	85.8	99.2	85.39	115.4	83.86	110.5
4.6	80.75	81.47	84.05	96.66	90.35	89.88
5.0	74.15	70.61	81.56	94	76.03	78.98
5.6	66.4	50.01	62.09	66.09	64.61	57.82
6.0	68.3	46.59	68.85	59.62	64.89	55.32
6.6	48.25	32.37	48.55	49.35	59.46	52.37
7.0	41.47	19.19	42.24	42.01	45.4	43.91
7.6	35.11	12.16	35.04	35.43	37.21	33.66
8.0	35.83	9.45	44.51	32.74	46.14	40.3
8.6	28.34	5.09	33.09	28.2	37.1	34.81
9.0	27.42	3.01	29.52	25.81	31.81	29.42
9.6	20.82	1.5	27.67	20.34	26.1	21.27
10.0	19.85	0.2	32.19	17.53	27.91	27.6
10.6	15.94	0.2	20.39	9.9	24.64	21.42
11.0	13.36	0.2	19.64	5.28	20.15	19.54
11.6	12.62	0.2	17.85	2.06	17.7	16.72
12.0	12.52	0.2	16.9	0.64	18.82	18.1
12.6	12.4	0.2	15.67	0.2	15.06	15.68
13.1	9.66	0.2	14.02	0.2	13.15	13.11
13.6	10.40		15.20		16.80	14.10
14.0	9.70		14.10		16.20	13.60
14.4	8.10		12.70		13.50	11.90
15.0	8.00		12.00		14.40	11.80
15.6	7.00		11.00		12.80	10.00
16.0	6.00		10.40		12.70	9.20
16.6	4.40		9.00		11.10	6.30
17.0	4.60		9.30		10.90	2.00
17.6	4.10		7.50		10.00	2.40
18.1	2.40		6.60		8.20	1.70
18.6	1.95		5.76		7.52	1.29
19.0			5.64		6.57	
19.6	1.80		4.70		2.98	0.84
20.0			3.29			
20.6	1.07		2.06		2.10	0.55
23.0	0.10		1.33		0.10	0.30
26.3	0.10		0.32		0.10	0.12

The plasma concentrations of the infused 10-1074 and VRC01-LS were measured longitudinally in the indicated animals.