Figure S1. Amino acid sequence alignment resulting from the DNA sequencing of CIS43 and CIS43LS heavy chain. The LS point mutations are boxed in red.
Figure S2. Autoreactivity of CIS43LS.
(A) Representative confocal fluorescence microscopy images of human HEp-2 epithelial cell staining following reactivity with CIS43LS or CIS43. Test antibodies and concentrations (25 and 50 µg) used are indicated. Anti-HIV1 antibodies were used as negative (VRC01LS) and positive (4E10, VRC07-523LS and VRC07-G54W) controls. Images were acquired with 20x objective N.A. = 0.7. Reactivity was assigned scores of 0, 1, 2, and 3 in the 25 µg/mL samples, using the no-mAb sample as baseline.
(B) Binding of varying concentrations (100 and 33.3 µg/ml) of mAbs to cardiolipin determined by ELISA. Test and control antibodies are as in (A). Values are expressed as optical density at 450 nm (OD450nm) and IgG phospholipid signal (GPL) units. One unit represents 1 µg control IgG antibody.
Figure S3. Binding kinetics of CIS43-expressed AAV mAb in mouse serum to PfCSP.
CIS43 mAb levels in serum (A) and skin (B) at indicated times after IM administration with 10^{11} GC of CIS43-AAV in C57BL/6 albino mice (n = 5). For (B), differences in CIS43 concentration at indicated timepoints were determined using the Kruskal-Wallis test for multiple comparisons with Dunn’s correction. ns, not significant. Data represent the mean with SD (A and B). (C) Apparent binding affinity of CIS43-, 2A10-, VRC01-AAV in mouse sera to PfCSP, 8 weeks following AAV administration. CIS43 and 2A10 IgG have been used as controls. Antibody binding curves are shown in red (raw data) and black (fitted data). The apparent affinity is displayed as K_D (equilibrium dissociation constant) in nM on each panel. ND, no fit could be determined. mAb serial concentrations (2, 1, 0.5, 0.25, and 0.125 μg/ml) used are displayed.
Figure S4. Mouse PfCSP-specific antibody responses in CIS43-AAV-administered mice following the first malaria challenge. Previously challenged and protected mice administered with CIS43-AAV (Figure 3B) were assessed for mouse PfCSP-specific antibodies (anti-mouse) measured by ELISA at 32 weeks just prior to rechallenge. Human IgG induced by CIS43-AAV (anti-human) are shown for comparison. Naïve indicates serum samples collected prior to AAV administration and prior to any challenge. OD$_{405}$ nm, optical density at 405 nm. Data points represent the mean with SEM. Naïve and anti-human, n = 4 per group; anti-mouse, n = 8.
**Supplemental Table 1. Summary of binding of CIS43 or CIS43LS to rhesus or human neonatal Fc receptor (FcRn).**

| Fc Receptor  | Sample ID | Kd (nM)  | Kd (M)  | Kd Error | Kon (1/Ms) | Kon Error | Koff (1/s) | Koff Error | Fold Kd * |
|--------------|-----------|----------|---------|----------|------------|-----------|-------------|------------|-----------|
| Human FcRn  | CIS43     | 54.2     | 5.4E-08 | 1.5E-09  | 4.8E+05    | 1.2E+04   | 2.6E-02    | 3.5E-04    |           |
| pH 6.0       | CIS43LS   | 6.2      | 6.2E-09 | 6.7E-11  | 2.7E+05    | 2.8E+03   | 1.7E-03    | 3.6E-06    | 9.0       |
| Human FcRn  | CIS43     | ND       | No Fits Obtained |          |            |           |             |             |           |
| pH 7.4       | CIS43LS   | 234.1    | 2.3E-07 | 5.8E-09  | 4.8E+05    | 1.1E+04   | 1.1E-01    | 8.9E-04    |           |
| Rhesus FcRn | CIS43     | 325.8    | 3.3E-07 | 5.4E-09  | 5.9E+04    | 9.0E+02   | 1.9E-02    | 1.2E-04    |           |
| pH 6.0       | CIS43LS   | 25.1     | 3.1E-08 | 3.4E-10  | 3.7E+04    | 2.2E+02   | 1.1E-03    | 1.1E-05    | 13.0      |
| Rhesus FcRn | CIS43     | ND       | No Fits Obtained |          |            |           |             |             |           |
| pH 7.4       | CIS43LS   | 262.1    | 2.6E-07 | 3.8E-09  | 2.35E+05   | 2.9E+03   | 6.15E-02   | 4.4E-04    |           |

* Fold change in Kd versus unmodified CIS43.  
Kd, equilibrium dissociation constant; Kon, association rate constant; Koff, dissociation rate constant.
### Supplemental Table 2. Binding affinities of CIS43 and CIS43LS for human Fc gamma receptors.

| Human Fc Receptor (pH 7.4) | Sample ID | Ko (nM) | Ko (M) | Ko Error | kon (1/Ms) | kon Error | Koff (1/s) | Koff Error | Fold K0 * |
|---------------------------|-----------|---------|--------|----------|------------|-----------|------------|------------|-----------|
| FcyRI                     | CIS43     | 14.84   | 1.48E-08 | 2.15E-10 | 1.87E+05  | 2.17E+03  | 2.78E-03  | 2.41E-05  |           |
|                           | CIS43LS   | 15.84   | 1.58E-08 | 1.96E-10 | 1.94E+05  | 1.99E+03  | 3.08E-03  | 2.14E-05  | 1.0       |
| FcyRIIA H167              | CIS43     | 3835    | 3.84E-06 | 6.85E-07 | 1.76E+05  | 3.11E+04  | 6.76E-01  | 1.91E-02  |           |
|                           | CIS43LS   | 2489    | 2.49E-06 | 2.76E-07 | 2.40E+05  | 2.60E+04  | 5.98E-01  | 1.35E-02  | 1.5       |
| FcyRIIIA R167             | CIS43     | 1479    | 1.48E-06 | 1.16E-07 | 3.48E+05  | 6.85E+04  | 2.17E+03  | 2.78E-03  |           |
|                           | CIS43LS   | 2489    | 2.49E-06 | 2.76E-07 | 2.40E+05  | 2.60E+04  | 5.98E-01  | 1.35E-02  | 1.5       |
| FcyRIIB                   | CIS43     | 3132    | 3.13E-06 | 8.28E-07 | 2.59E+05  | 6.74E+04  | 8.12E-01  | 3.81E-02  |           |
|                           | CIS43LS   | 3803    | 3.80E-06 | 1.15E-06 | 2.30E+05  | 6.84E+04  | 8.74E-01  | 4.11E-02  | 0.8       |
| FcyRIIA V176              | CIS43     | 652     | 6.52E-07 | 1.55E-08 | 1.61E+05  | 3.44E+03  | 1.05E-01  | 1.09E-03  |           |
|                           | CIS43LS   | 550     | 5.50E-07 | 2.22E-08 | 7.87E+04  | 2.50E+03  | 4.33E-02  | 1.08E-03  | 1.2       |
| FcyRIIA F176              | CIS43     | 1736    | 1.74E-06 | 1.21E-07 | 1.57E+05  | 1.06E+04  | 2.73E-01  | 5.11E-03  |           |
|                           | CIS43LS   | 2919    | 2.92E-06 | 2.54E-07 | 4.40E+05  | 3.73E+04  | 1.29E-01  | 2.56E-03  | 0.6       |
| FcyRIIB NA1 Protein       | CIS43     | 1027    | 1.03E-06 | 1.20E-07 | 4.30E+05  | 4.80E+04  | 4.42E-01  | 1.56E-02  |           |
|                           | CIS43LS   | 1187    | 1.19E-06 | 7.75E-08 | 3.00E+05  | 1.88E+04  | 3.56E-01  | 6.68E-03  | 0.9       |
| FcyRIIB NA2 Protein       | CIS43     | 1283    | 1.28E-06 | 1.01E-07 | 3.27E+05  | 2.46E+04  | 4.19E-01  | 9.08E-03  |           |
|                           | CIS43LS   | 853.6   | 8.54E-07 | 5.20E-08 | 2.93E+05  | 1.70E+04  | 2.50E-01  | 4.61E-03  | 1.5       |

* Fold change in Ko versus unmodified CIS43; Ko, equilibrium dissociation constant; kon, association rate constant; Koff, dissociation rate constant. Human polymorphisms: FcyRIIA H167, FcyRIIA H167; FcyRIIA R167; FcyRIIIA V176; FcyRIIIA F176; FcyRIIB NA1; FcyRIIB NA2.
Supplemental Table 3: Pharmacokinetic parameters in sera from rhesus macaques administered with CIS43LS or CIS43 (10 mg/kg) by IV route.

| Animal ID | mAb   | Route | T₁₂ (Day) | Observed Tₘₐₓ (Day) | Observed Cₘₐₓ (µg/ml) | AUC (Day x µg/mL) | Clearance (mL/Day/Kg) | Serum Concentration (µg/mL) |
|-----------|-------|-------|-----------|---------------------|-----------------------|-------------------|----------------------|--------------------------|
| 12M069    | CIS43 | IV    | 46.5      | 0.01                | 261.5                 | 5,467             | 1.83                 | 37.3                     | 18.8                     |
| 14D017    | CIS43 | IV    | 31.0      | 0.01                | 313.2                 | 4,107             | 2.43                 | 28.3                     | 11.5                     |
| AVG       |       |       |           |                     | 287.3                 | 4,787             | 2.13                 | 32.8                     | 15.1                     |
| SE        |       |       |           | 7.8                 | 0.0                   | 25.9              | 0.30                 | 4.5                      | 3.7                      |
| 13C084    | CIS43 | IV    | 24.5      | 0.01                | 289.2                 | 1,887             | 5.30                 | 4.1                      | 1.1                      |
| 15C049    | CIS43 | IV    | 20.1      | 0.01                | 271.7                 | 1,592             | 6.28                 | 1.9                      | 0.5                      |
| AVG       |       |       |           | 22.3                | 0.01                 | 280.4             | 1,739               | 5.79                     | 3.0                      | 0.8                      |
| SE        |       |       |           | 2.2                 | 0.0                  | 8.8               | 148                 | 0.49                     | 1.1                      | 0.3                      |

T₁₂ (elimination half-life), time taken for the plasma concentration of a drug to fall by half its original value; Tₘₐₓ, time taken to attain Cₘₐₓ; Cₘₐₓ, maximum drug concentration in plasma; AUC (Area Under the Curve), measure of exposure of all the body to the drug; Clearance, measurement of plasma volume from which a drug is entirely removed per day per Kg body weight.