Supporting Information

Chennamsetty et al. 10.1073/pnas.0904191106

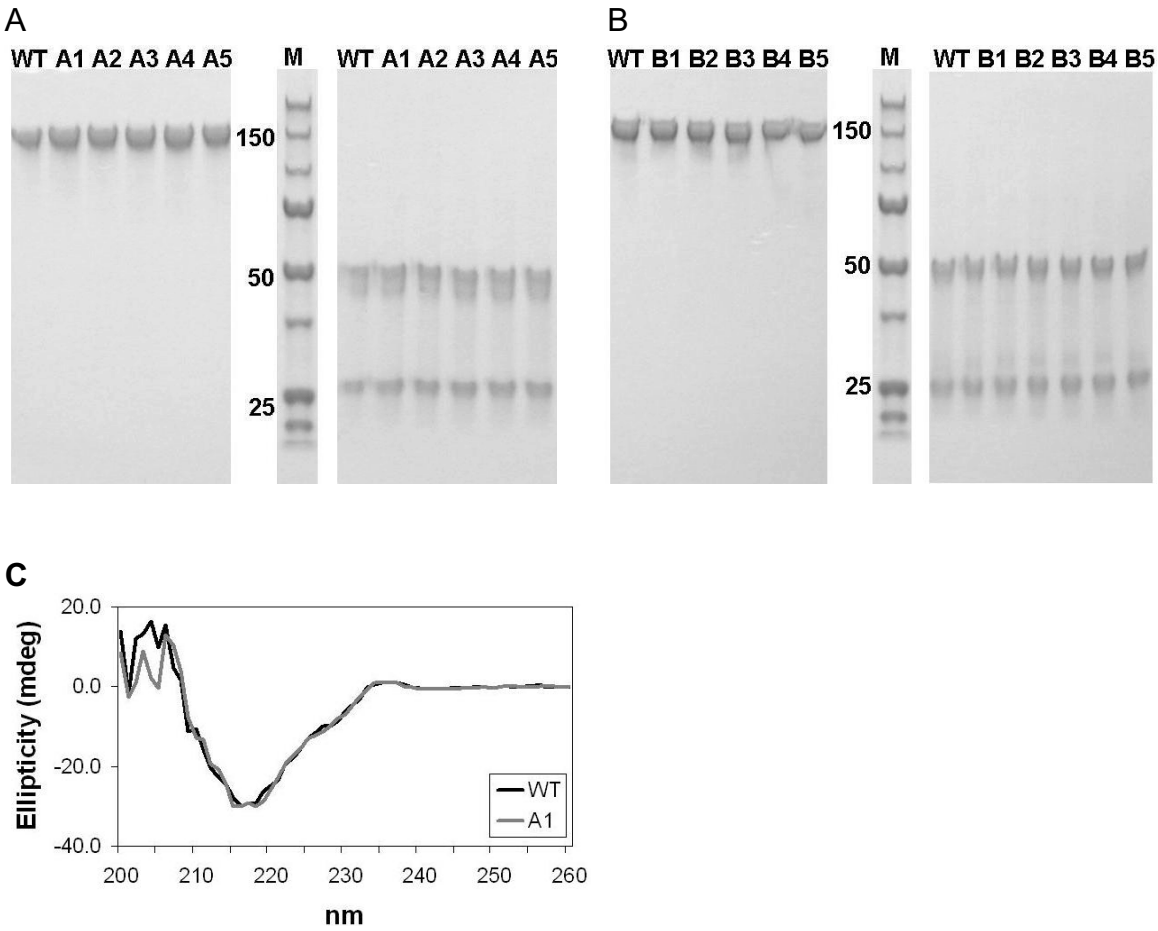

Fig. S1. Quality control of engineered variants. (*A*) Comparison of antibody A Variants A1-A5 to antibody A WT by SDS/PAGE under nonreducing (left) and reducing (right) conditions. The bands in the marker lane M that correspond to 150, 50, and 25 kDa are indicated. (*B*) Comparison of antibody B Variants B1-B5 to antibody B WT by SDS/PAGE under nonreducing (left) and reducing (right) conditions. (*C*) Circular dichroism spectra of antibody A WT and Variant A1. This overlapped pattern shows that the secondary structure is intact upon mutation.

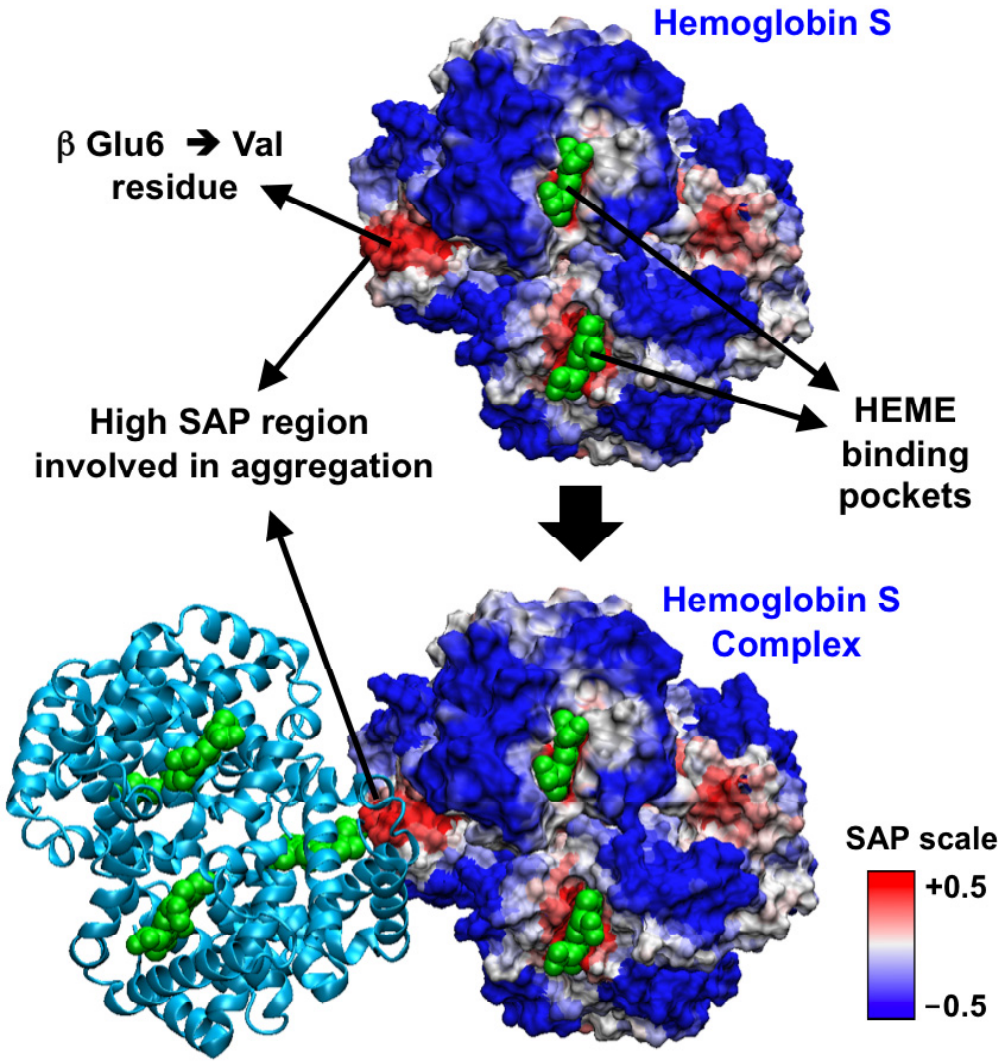

Fig. S2. The SAP analysis at $R = 10$ Å for deoxyhemoglobin S, along with the structure of the aggregated complex (PDB entry 2HBS). The high SAP region that includes the β-Glu-6 to Val mutation is also marked.

Table S1. Antigen binding activity of antibody-A wild type and variants

Sample	Content, mg/mL	Relative potency, %		Mean potency %
		Dilution 1	Dilution 2	
WT1	1	103.6	102.8	103
WT2	1	83.4	84.1	84
A1	1	85.0	75.7	80
A2	1	89.9	87.9	89
A3	1	83.1	82.1	83
A4	1	86.6	86.5	87
A5	1	87.3	86.9	87

The antigen binding activity is measured in a cell-based gene-reporter bioassay. All values of potency are relative to wild type standards. WT1 and WT2 represent two different preparations along with the corresponding variants, independently of the original standards.

Table S2. The antigen binding activity* of antibody-B wild type and variants.

Sample	Relative potency, %
WT1	92
WT2	90
B1	<10
B2	<10
B3	99
B4	<10
B5	<10

See Table S1 for details.