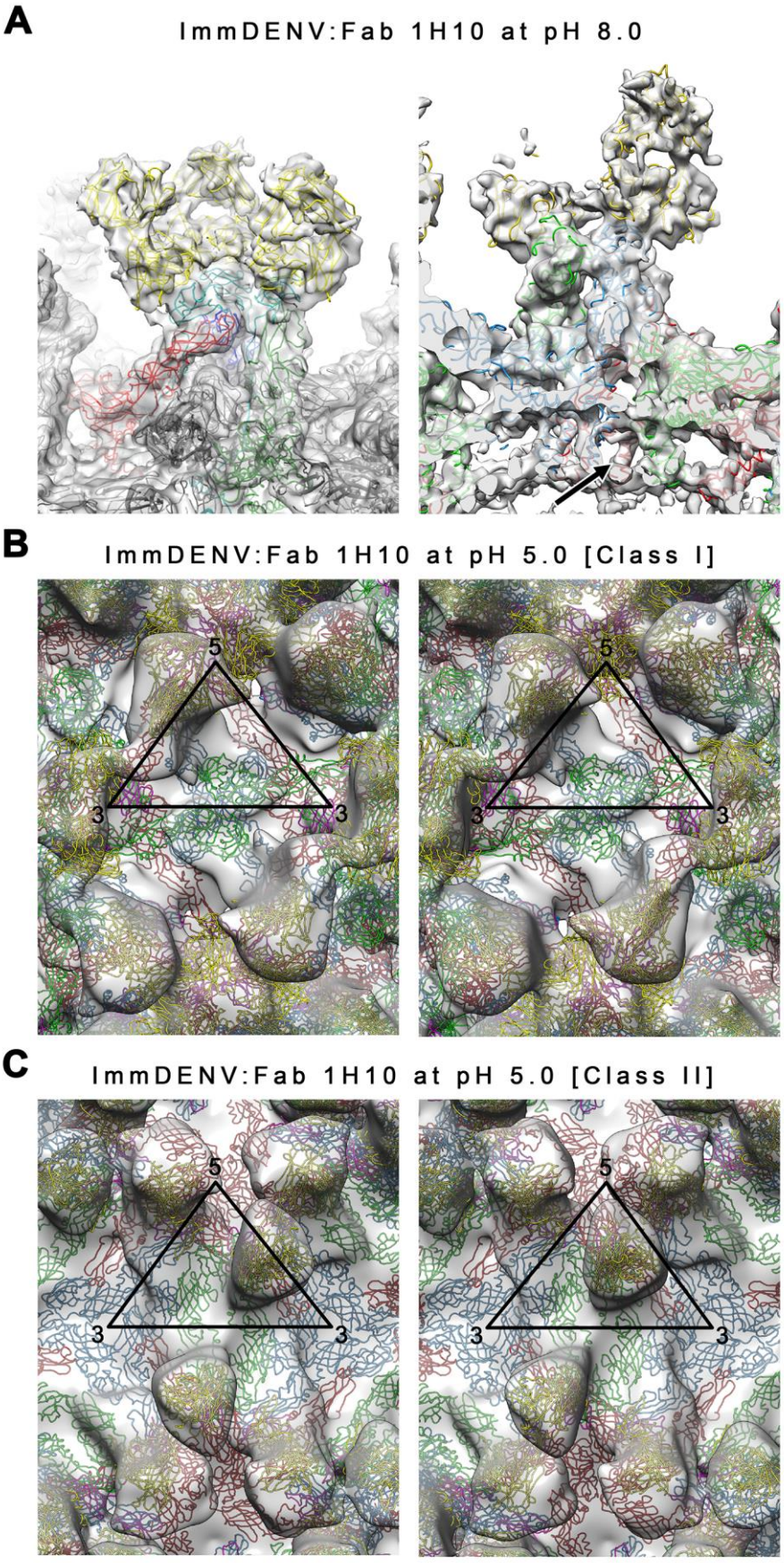

A
ImmDENV:Fab 1H10 at pH 8.0
B
ImmDENV:Fab 1H10 at pH 5.0 [Class I]
5
3
3
5
3
3
C
ImmDENV:Fab 1H10 at pH 5.0 [Class II]
5
3
3
5
3
3

Figure S1. Model fitting of the cryoEM maps of immDENV3:Fab 1H10 at pH 8.0, and class I and class II immDENV3:Fab 1H10 at pH 5.0, Related to Figures 2, 3 and 4.

(A) [Left] The E and prM proteins and Fab 1H10 molecules fit into the cryoEM map of immDENV3:Fab 1H10 at pH 8.0. In one of the trimeric spikes on the cryoEM map, pr molecules and Fab are shown as cyan and yellow wires, respectively, while the E proteins near the 5-, 3- and 2- fold vertices in an asymmetric unit, red, blue and green wires, respectively. The other nearby E protein:prM:Fab 1H10 complexes as grey wires. The density of the cryoEM map is displayed as transparent grey surface. [Right] Another view showing the cross-section of the cryoEM map near one of the trimeric spikes. The 12 Å cryoEM map of immDENV3:Fab 1H10 at pH 8.0 showed resolved helical densities of the trans-membrane regions of E and prM proteins (indicated by black arrow) consistent with the claimed resolution.

(B-C) Stereo-diagram of the E protein:pr:Fab 1H10 complex fitted into the class I and II cryoEM maps of immDENV3:Fab 1H10 complex at pH 5.0.

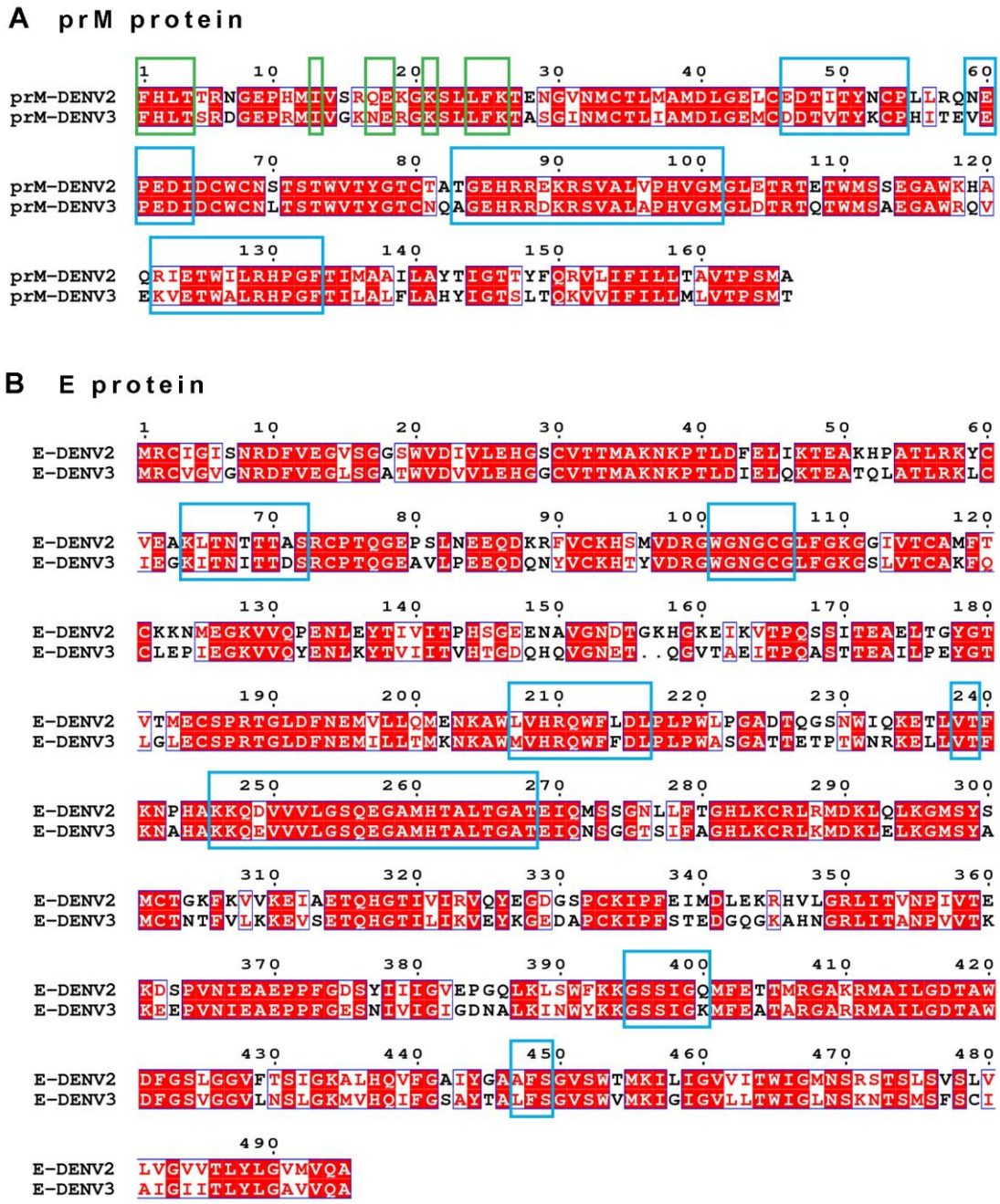


Figure S2. Sequence alignment between DENV2 and DENV3, Related to Figure 1.

(A-B) The prM (A) and E (B) proteins of DENV2 and DENV3 share sequence similarity of 68 and 71%, respectively. The conserved residues are indicated by white letters on red background, whereas residues with high similarity are shown in red letters. The residues involve in prM-E interactions are indicated by cyan box, whereas those of pr-Fab 1H10 in green box.

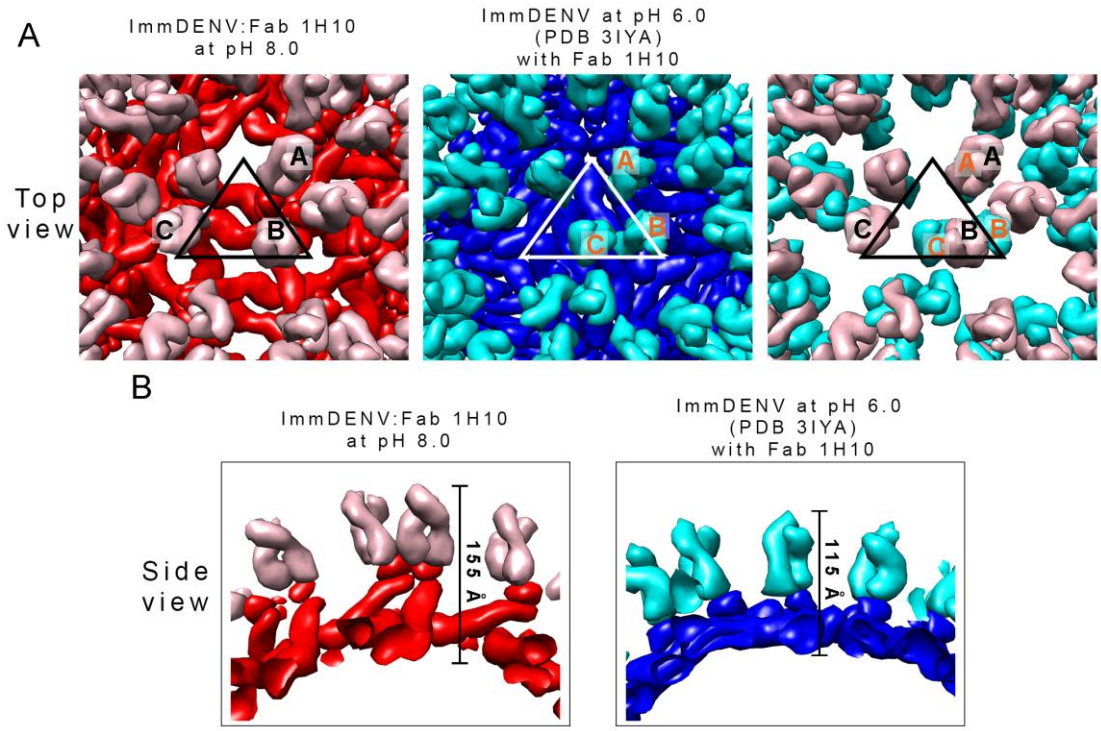
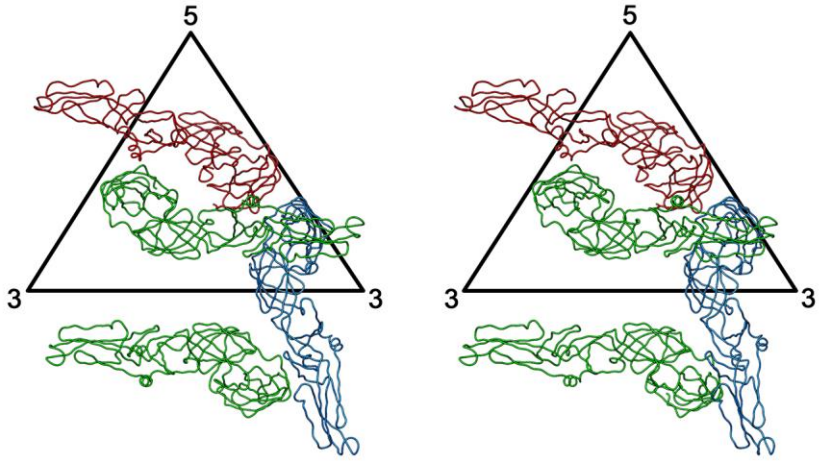


Figure S3. Top and side views of immDENV3:Fab 1H10 at pH 8.0 structure and immDENV2:Fab 1H10 at pH 6.0 model, Related to Figure 4.

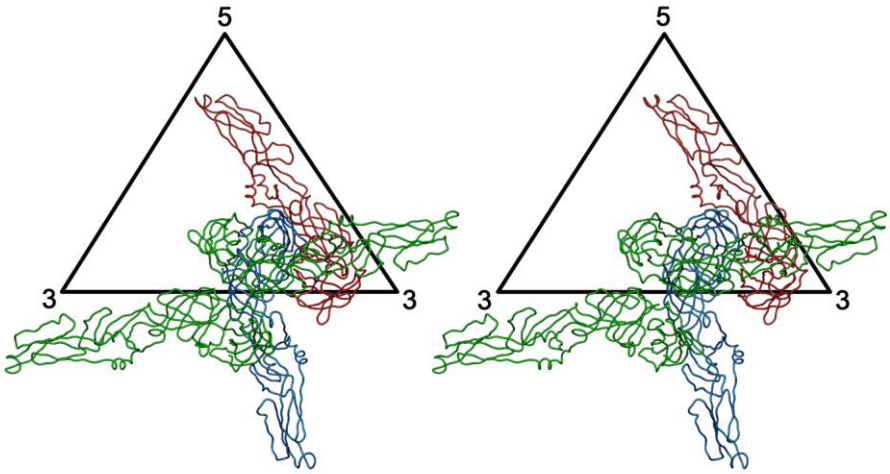
(A) The top view of immDENV3:Fab 1H10 at pH 8.0 structure (left), immDENV2 at pH 6.0 (PDB 3IYA) superimposed with Fab 1H10 model (center), and the display of only the Fab molecules from both models (right). Based on the Fab positions only, although the E protein organization is very different between the two models, the positions of the Fab molecule from the top view are very similar.

(B) The side view of the structures of immDENV3:Fab 1H10 at pH 8.0 and immDENV2:Fab 1H10 model. The structure of the E protein:prM:Fab complex is "standing up" in the immDENV3:Fab 1H10 at pH 8.0, whereas it is lying down in the immDENV2 pH 6.0 superimposed with Fab 1H10 model. There is a 40 Å difference (155 vs 115 Å) on the height of E protein:pr:Fab 1H10 between the two structures. This difference can be used to assign which model is more appropriate to be used in the fitting of class I and II cryoEM maps of immDENV3:Fab 1H10 at pH 5.0.

A ImmDENV:Fab 1H10 at pH 8.0



B ImmDENV:Fab 1H10 at pH 5.0 [Class I]



C ImmDENV:Fab 1H10 at pH 5.0 [Class II]

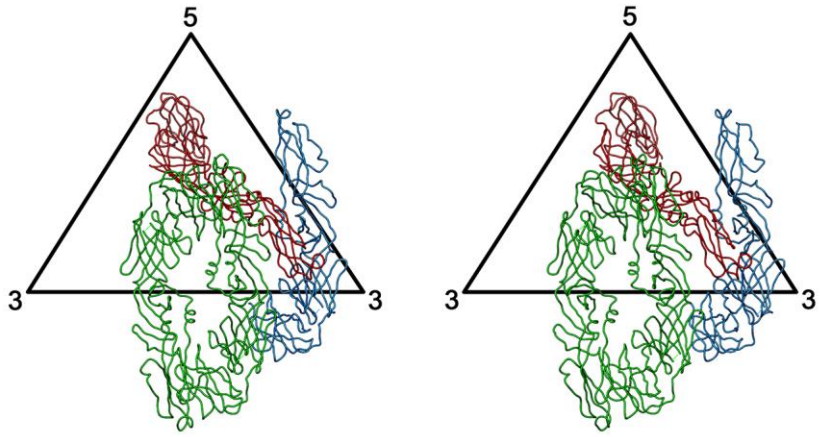


Figure S4. Stereo diagrams of the cryoEM structure of immDENV3:Fab 1H10 at pH 8.0 and class I and II immDENV3:Fab 1H10 at pH 5.0, Related to Figure 5.

(A) The structure of immDENV3:Fab 1H10 at pH 8.0.

(B) The structure of class I immDENV3:Fab 1H10 at pH 5.0.

(C) The structure of class II immDENV3:Fab 1H10 at pH 5.0.

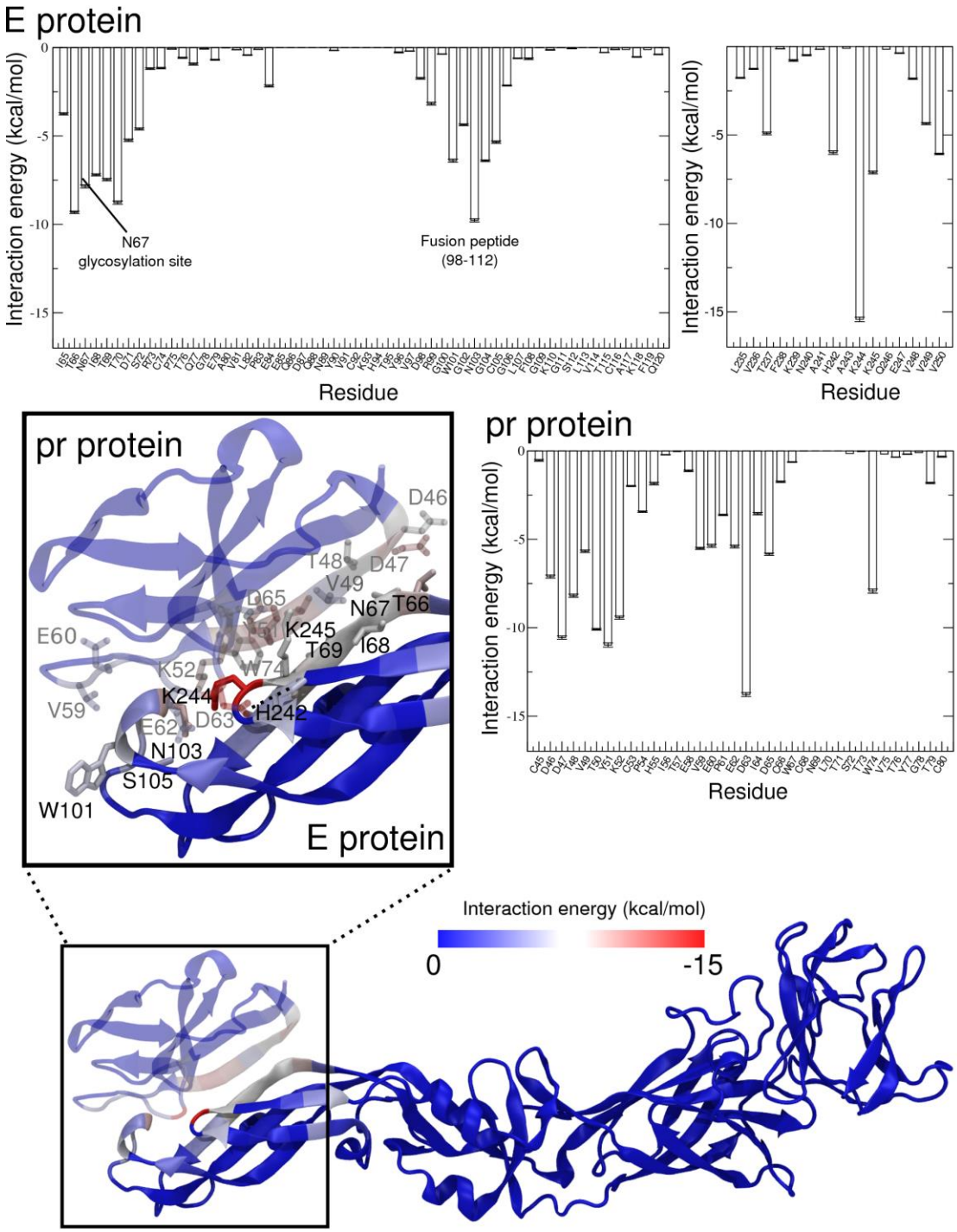


Figure S5. The per-residue interaction energies between E and pr proteins, Related to Figure 6.

Interaction energies were calculated as a sum of short-ranged electrostatic and van der Waals interactions. The protein is shown in cartoon representation and colored according to interaction energies. Residues with interaction energies stronger than -5 kcal mol^{-1} are shown in licorice representation and are denoted inset.

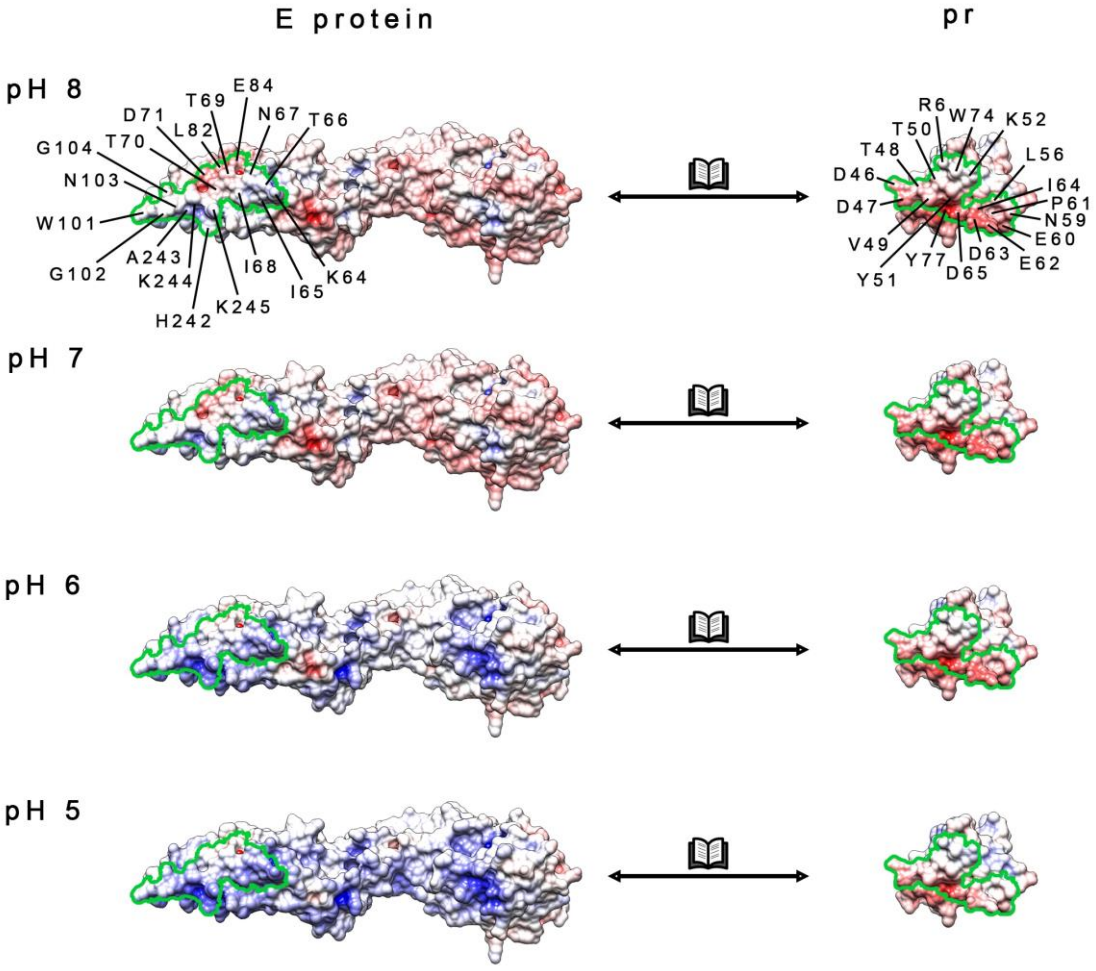


Figure S6. Pr molecule bound to E protein through charge-charge and hydrophobic interactions, Related to Figure 6.

The electrostatic potential surfaces of E and prM proteins at pH 8, 7, 6 and 5 are shown in open book representation. The positive, neutral and negative charges are shown in blue, white, and red colors, respectively. The interacting residues are outlined in green.

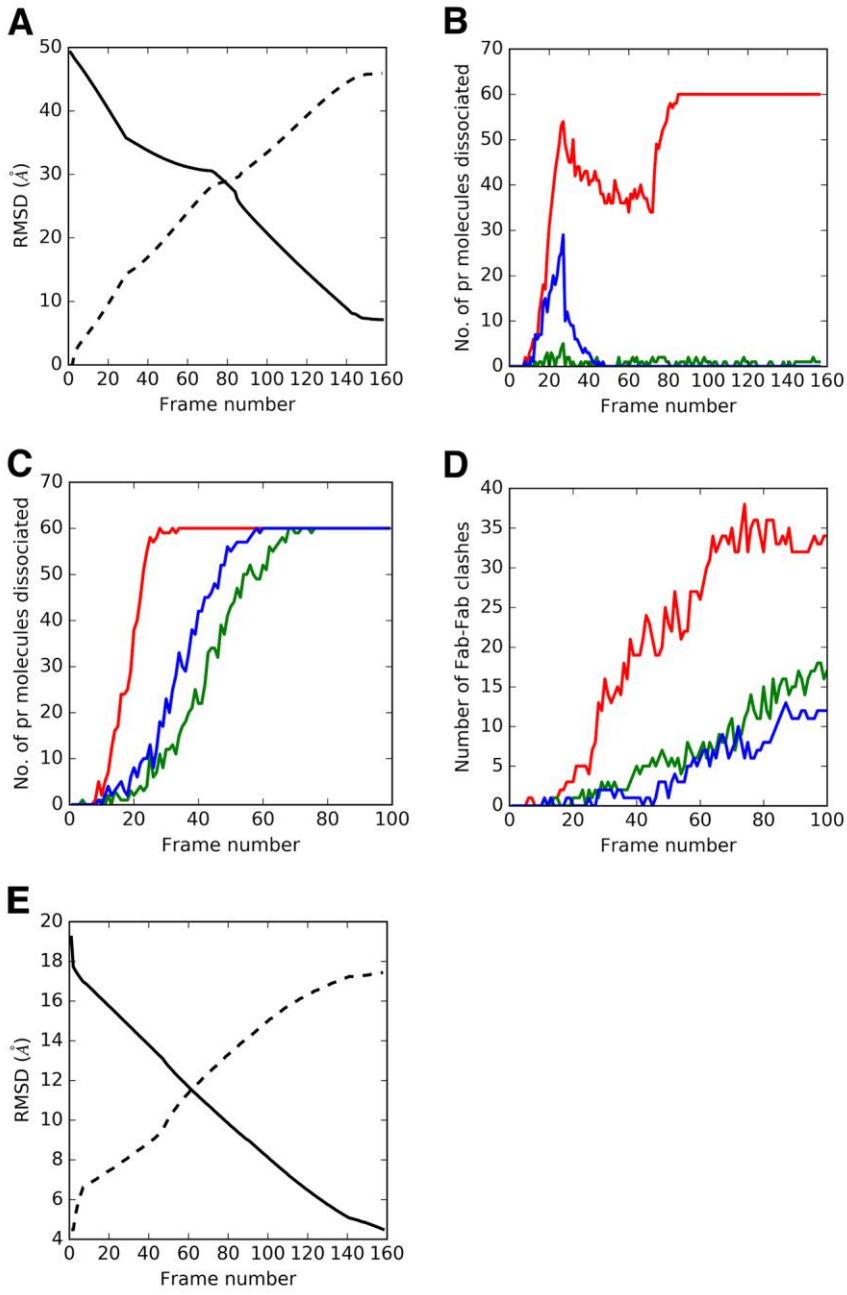


Figure S7. Modeling the E and prM transitions between stages I and II and between stages III and IV, Related to Figures 6 and 7.

(A) Root mean squared deviation (RMSD) for the transition between stages I and II, when all surface proteins were biased towards the final structure. RMSD was calculated for the backbone of all E proteins on virus with respect to initial (dashed line) and final (solid line) structures.

(B) Number of dissociated pr molecules during the simulated transition between stages I and II, when all surface proteins were biased towards the final structure. Pr molecules associated with either red, blue or green E proteins in an asymmetric unit are indicated by their respective color lines.

(C) Number of pr dissociations during the simulated transition between stages I and II over three independent targeted molecular dynamics simulations, where in each experiment a bias is only applied to one of the E proteins (red, or blue or green molecules). Since only one set of E protein molecules is biased per experiment, all molecules eventually will lose their pr molecules, due to steric hindrances with the other neighboring E proteins that were unbiased and hence remained close to their initial positions.

(D) Same experiment as (C), but tracking the number of Fab-Fab clashes after superimposing the Fab-pr on the pr molecules on virus. Results suggest red E protein complexed with pr and Fab likely experienced the most steric hindrance compared to the blue and green E proteins.

(E) Root mean squared deviation (RMSD) for the transition between stages III and IV. RMSD was calculated for the backbone of all E proteins with respect to initial (dashed line) and final (solid line) structures.

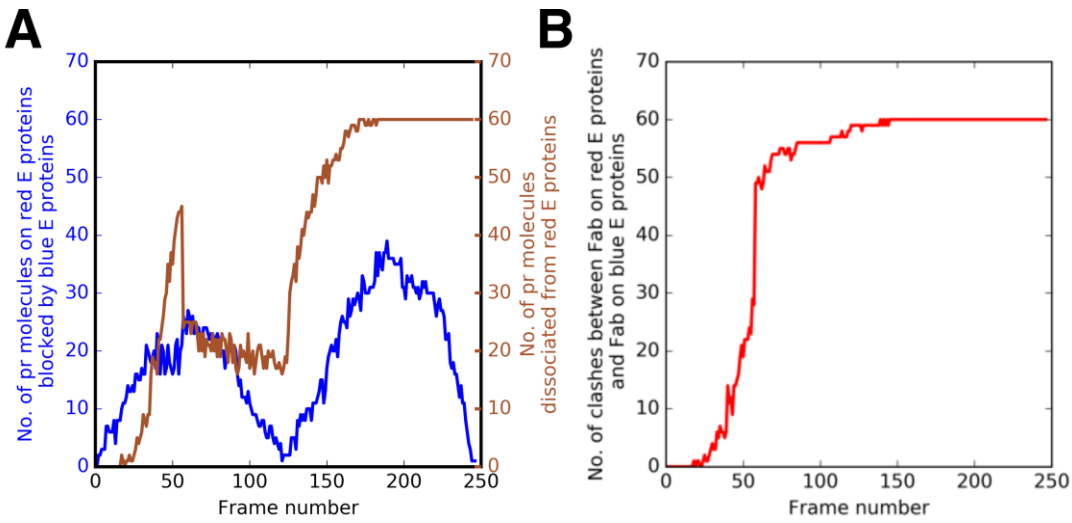


Figure S8. Molecular insights into the stage I-II transition using molecular dynamics simulations with twice slower pulling velocity, Related to Figure 6.

(A) Pr molecules interacting with the red E proteins were blocked by the neighboring blue E protein molecules within a trimeric spike during the structural rearrangement from stage I to II. Plot showing number of pr molecules on the red E proteins that are blocked by neighboring blue E protein molecules during the structural arrangement (blue line) against frame number. The number of dissociated pr proteins from the E proteins (right y-axis) is also shown in brown.

(B) The number of Fab-Fab clashes between the Fab on the pr-blue E protein and that on the pr-red E protein (red line) throughout the simulation. At around frame 50, severe clashes between the Fab:pr:E red molecules and Fab:pr:E blue molecules were observed when Fabs were superimposed onto the simulated pr-E virus molecule.

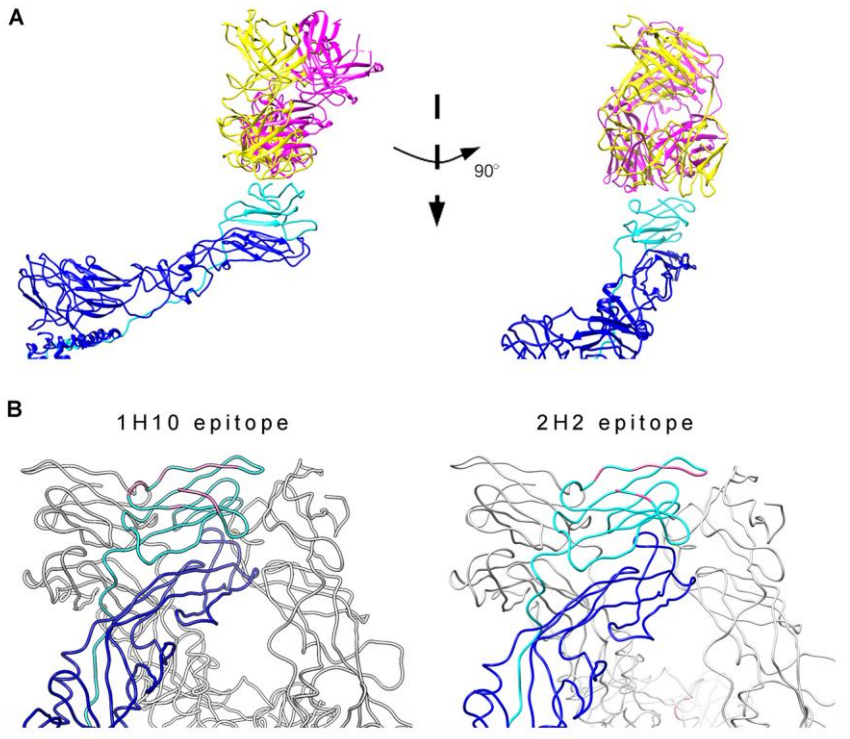


Figure S9. Comparison of the HMAb 1H10 and MAb 2H2 complexed with prM:E structures, Related to Figure 3.

(A) Superposition of immDENV:Fab 2H2 and immDENV:Fab1H10 complex structures showed that the antibodies bind to the pr molecule in a similar manner. Fab 1H10 and 2H2 are colored yellow and magenta, respectively, while prM and E proteins are colored cyan and blue, respectively.

(B) Epitope (pink) of Fab 1H10 and 2H2 on one prM molecule (cyan). The other E proteins and prM molecules in a trimeric spike are colored in grey.