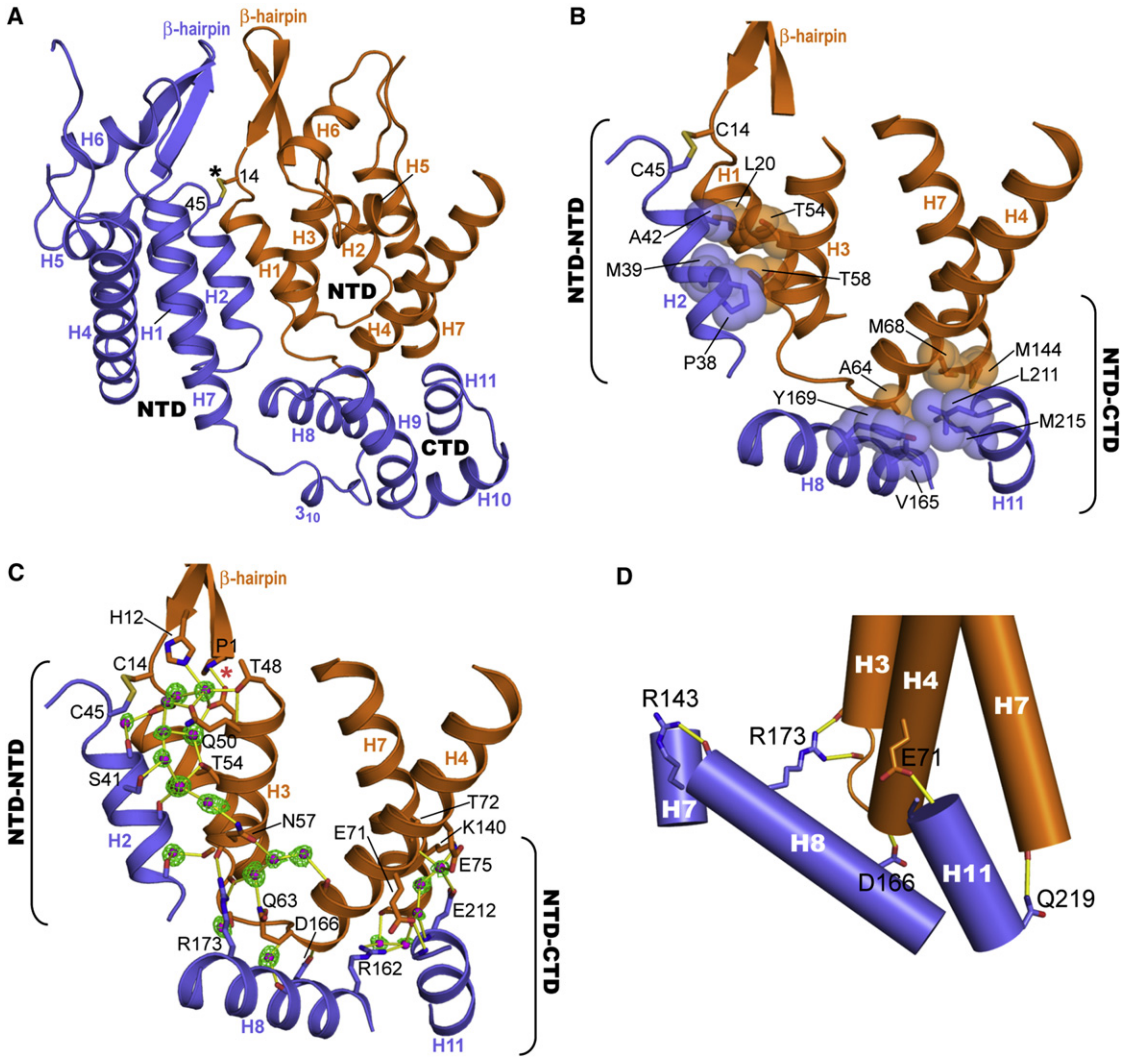

Figure 3. Atomic Details of the Hexamerization Interface

(A) Side view of one representative protomer (colored blue) and its interaction with the adjacent NTD subunit (orange), as seen in the crosslinked hexamers. Secondary structural elements are labeled. The engineered disulfide is marked by the black asterisk.

(B) Hydrophobic contacts. The side chain atoms of hydrophobic interfacial residues are represented in stick and translucent space-filling representations and labeled.

(C) Polar and water-mediated contacts. Selected side chains are shown explicitly and labeled. Green mesh shows unbiased F_O-F_C density contoured at +3σ. These were modeled as water molecules (magenta spheres) in the structure derived from hexagonal crystals. Putative hydrogen bonds are represented by yellow lines. Note that the region around the salt bridge between P1 and D51 (red asterisk), which forms only upon maturation of CA, is particularly water rich. These two residues coordinate water-mediated hydrogen bonds with H12, T48, and Q50. We speculate that the missing E45 side chain (mutated to cysteine in this construct) would participate in this network.

(D) Helix-capping hydrogen bonds at the NTD-CTD interface. Relevant side chains are shown explicitly and labeled. Hydrogen bonds are shown as yellow lines. The most critical of these caps appear to be R173 (to helix 3), since it is located in the middle of the hexamerization interface, and is conserved in 1668/1670 sequences in the Los Alamos HIV database, with the remaining two conservatively substituted with lysine. An intermolecular C cap for helix 7 in the blue protomer is not shown.

1–3 in HIV-1 CA therefore switch from an 18 helix barrel in the hexamer to a 15 helix barrel in the pentamer, with concomitant adjustments in intermolecular contacts. Although details of the pentameric interactions are not yet known, the highly hydrated character of the hexameric interface is compatible with this quasi-equivalent switching mechanism, because water molecules should be particularly adept at repositioning to accommodate altered orientations in hydrogen bonding and side chain packing geometries.

Hexamer-Stabilizing NTD-CTD Contacts

Intermolecular NTD-CTD interactions are made primarily by extended side chains from helix 8 of the CTD, which pack against the C-terminal end of helix 3, the intervening loop, and the