Fig. S1. DSP cross-linking of reconstituted D44A(18–60) and L40A/I42A/L43A triple mutant indicating tetrameric assembly. The reaction was carried out in 2.5 mM DSP and lasted for 60 min.
Fig. S2. $^1$H-$^{15}$N correlation spectra of $^{15}$N-labeled WT(18–60) (Left) and S31N(18–60) mutant (Right) recorded at 30 °C and $^1$H frequency of 500 and 600 MHz, respectively. The NMR samples contain 0.7 mM (monomer) protein, 300 mM DHPC, 40 mM rimantadine, and 30 mM glutamate in a 40 mM sodium phosphate buffer with a pH of 7.5. The labels on the spectrum are sequence specific assignments of resonances.
Fig. S3. Superposition of 10 low energy structures obtained from NOE restraints. The ensemble of structures has a backbone rmsd of 0.87 Å for the transmembrane (TM) helix, and 1.01 Å for the amphipathic helix.
Fig. S4. Possible source of the pore density that was originally proposed to be amantadine. (a) The proposed drug-M2 complex based on 3.5-Å diffraction data positions the hydrophobic adamantane cage in contact with the Ser-31 hydroxyls (3C9J). (b) In a much higher resolution data set (3BKD; 2.05 Å) that allows detergent, ions, water, and polyethylene glycol to be identified, a cluster of water molecules are bound to the Ser-31 hydroxyls.