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Supplemental Information

The Structure of Mouse Cytomegalovirus m04 Protein Obtained from Sparse NMR Data Reveals a Conserved Fold of the m02-m06 Viral Immune Modulator Family

Nikolaos G. Sgourakis, Kannan Natarajan, Jinfa Ying, Beat Vogeli, Lisa F. Boyd, David H. Margulies, and Ad Bax

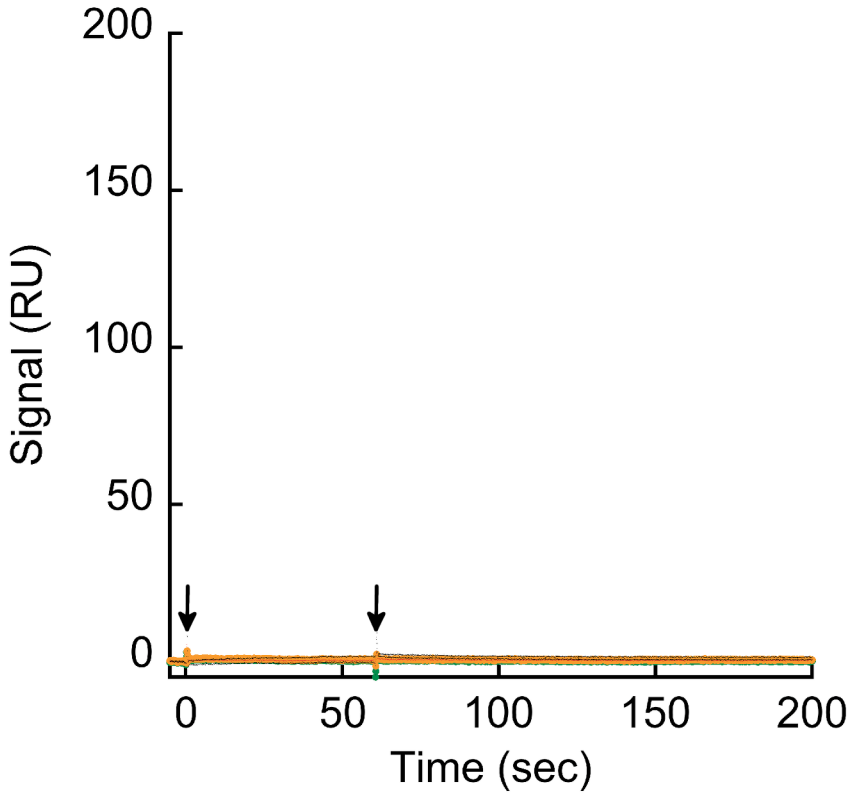

Figure S1, reference to Figure 1. Negative control binding experiments using the MHC-like molecule MULT-1. SPR binding sensograms collected using immobilized m04ED (WT) with increasing concentrations of MULT-1 flow-through, as outlined in Experimental Procedures. The start of the injection (association) and wash out (dissociation) phases are indicated with vertical arrows. Unlike full-length D^d (Figure 1b), this MHC-like molecule, lacking the α_3 domain, bound petide and β_2-microglobulin shows a flat binding profile (after correction of non-specific refracive index chages using a mock surface without m04ED) that is typically of a lack of specific binding to the m04ED surface.

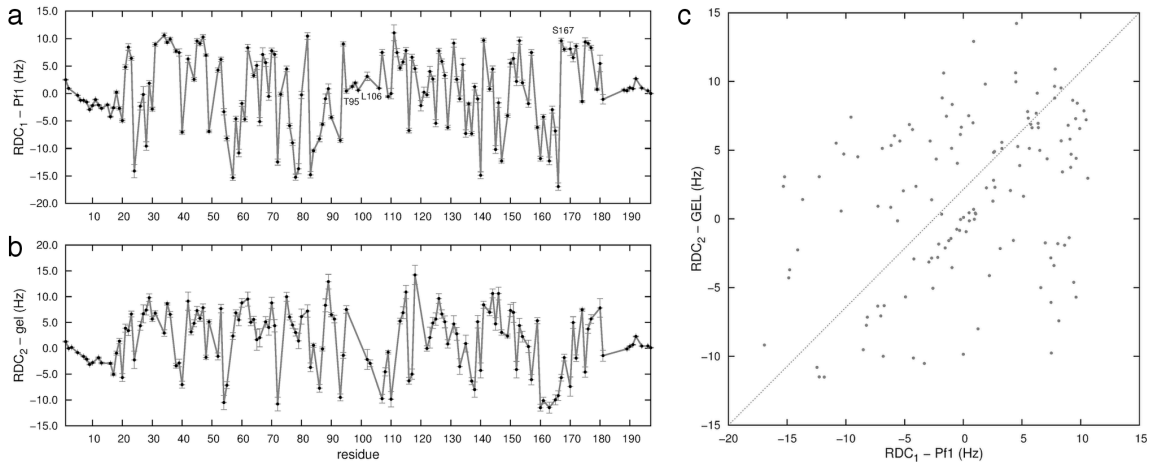

Figure S2, reference to Table 2. ^{15}N-^{1}H backbone RDC values measured in two alignment media for m04ED (Figure 1c). Values measured in **(a)** bacteriophage Pf1 and **(b)** positively charged gels, correspond to approximate alignment tensor magnitudes and Rhombicities of -8.3 Hz/0.19 and -7.0 Hz/0.40 respectively, as estimated using the histogram method (Clore et al., 1998). The RDC splittings were measured for well-resolved peaks in the 2D TROSY-HSQC spectrum using a quantitative ARTSY experiment (Fitzkee and Bax, 2010). Error bars report experimental uncertainties as propagated from the spectral peak-to-noise ratio. Key residues defining the central flexible loop region and kinked C-terminal helix are indicated. **(c)** Correlation diagram between the two RDC datasets, showing a linear correlation coefficient of 0.36 (Tolman and Ruan, 2006), indicating the datasets are nearly independent.

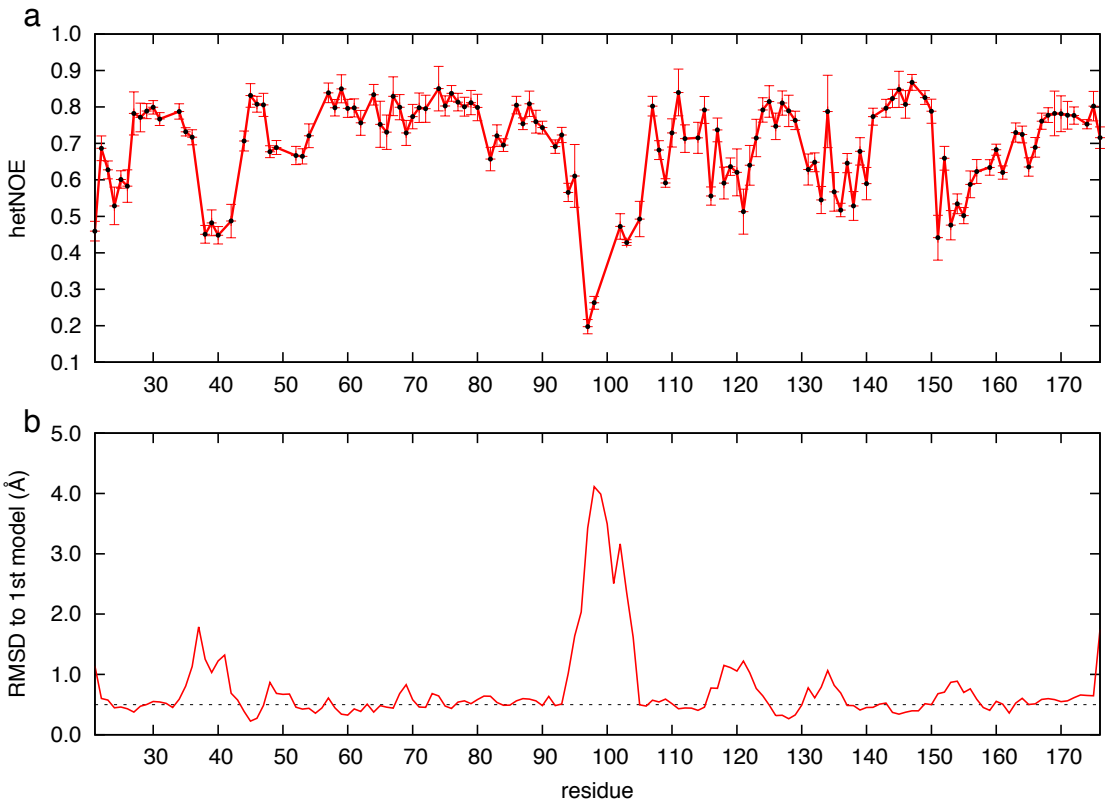

Figure S3, reference to Figure 3. Structural convergence in the final CS-Rosetta ensemble shown in Figure 3c. **(a)** Experimental ^{15}N-{^{1}H} NOE values from Figure 2a plotted only for m04ED core residues 21-176 included in the Rosetta structure calculations. Errors are propagated from the spectral peak-to-noise ratios. **(b)** Average backbone heavy atom RMSD values computed over the 10 lowest-energy models from the final CS-Rosetta calculations (Table 1, #7 and Figure 3c).

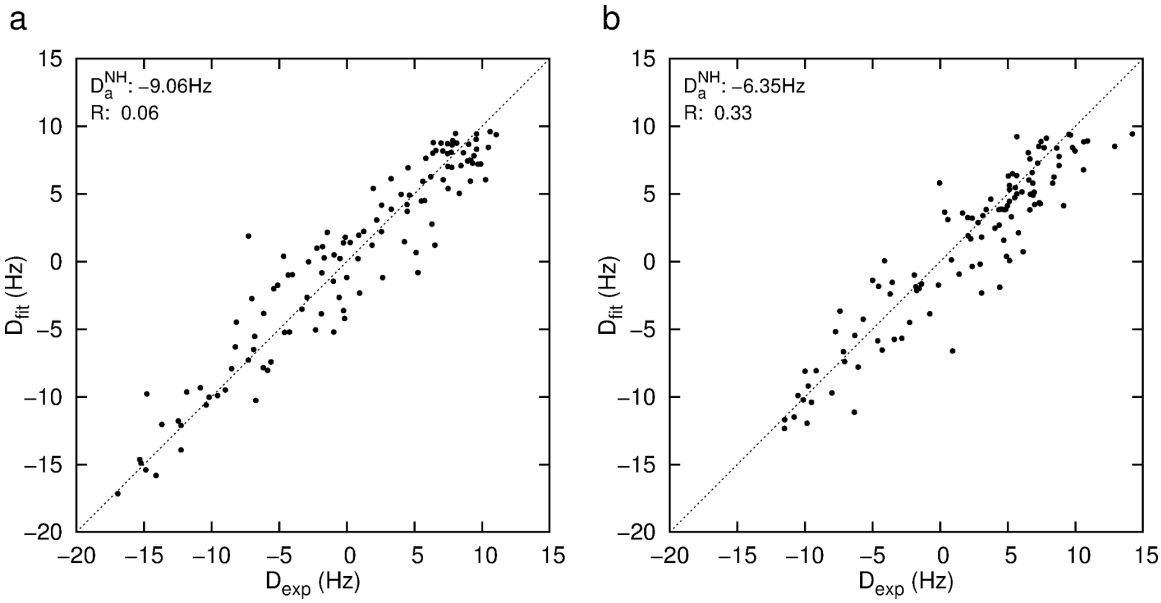

Figure S4, reference to Figure 3, Table 2. Structure fits to the RDC experimental data, shown for the top-ranking model from the final NMR ensemble of Figure 3c. The final RDC quality (Q) factors (Cornilescu et al., 1998) for the Pf1 **(a)** and gel **(b)** datasets are 0.29 and 0.41 respectively (Pearson's linear correlation coefficients (R_P) of 95% and 90%). The structure-based fitted alignment tensor magnitudes and rhombicities are indicated in each plot.

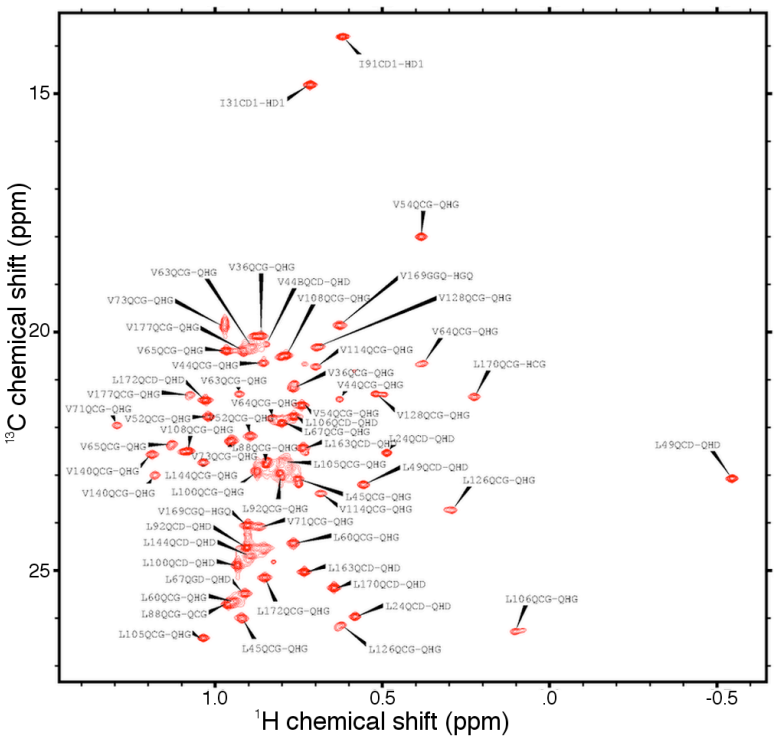

Figure S5, reference to Figure 1. Methyl region of a ^{1}H-^{13}C HMQC spectrum recorded at 900 MHz using an ILV methyl ^{13}C-labeled sample of m04ED (Figure 1c). The complete resonance assignments of 2 Ile $C^{\delta 1}$, 16 Leu $C^{\delta 1}$, $C^{\delta 2}$ and 15 Val $C^{\gamma 1}$, $C^{\gamma 2}$ are indicated.