A Compare-and-Contrast NMR Dynamics Study of Two Related RRMs: U1A and SNF

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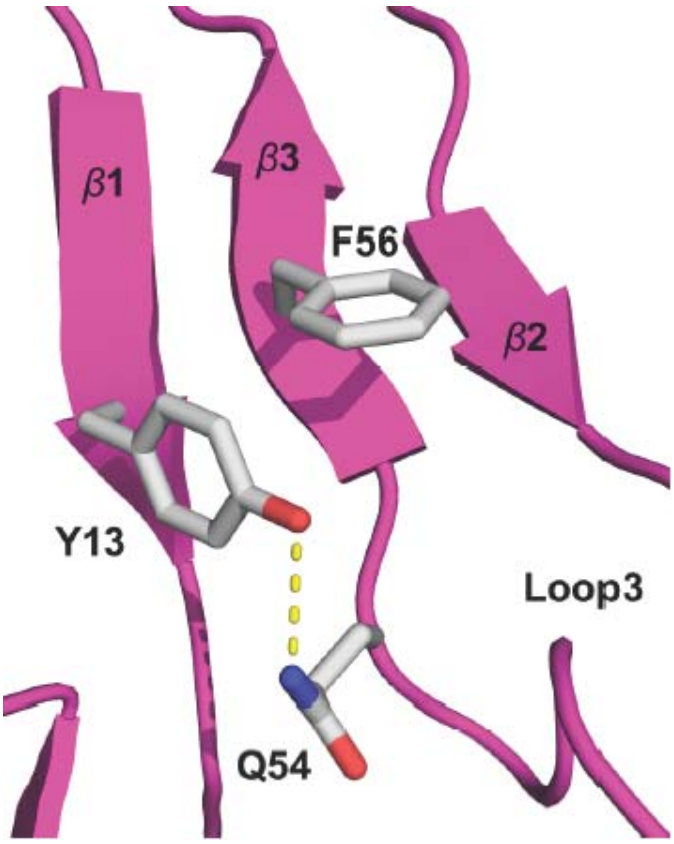

Figure S1. Ribbon of the NMR solution structure of apo U1A β-sheet surface (1fht.pdb, model 1, (11)). Dotted line indicates the distance between Tyr13 ring oxygen to Gln54 sidechain amide nitrogen where the average distance in the 43 structures is 2.9 ± 0.4 Å.

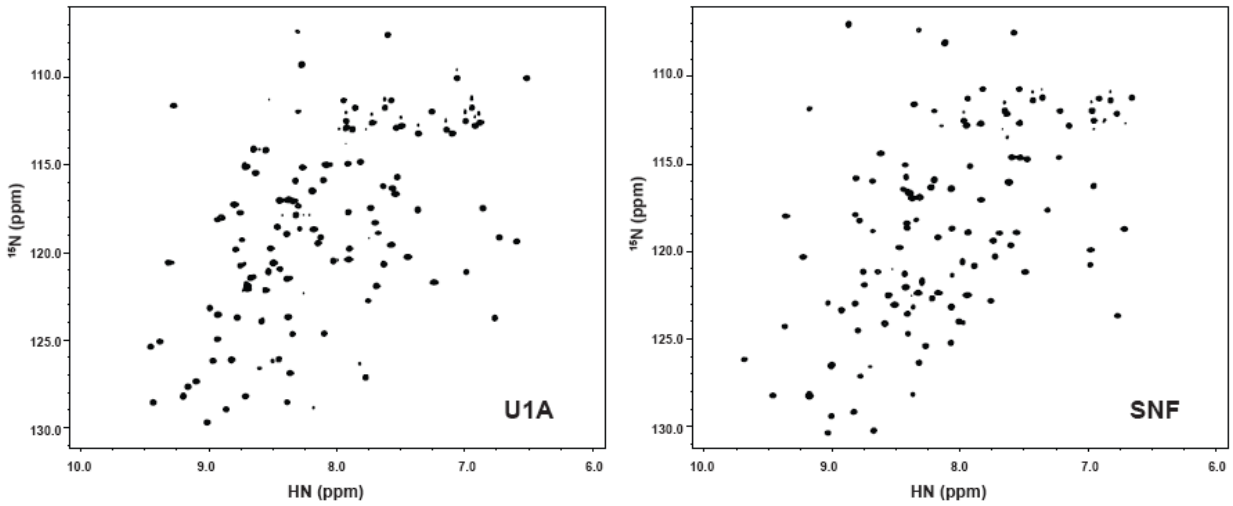

Figure S2. ^{1}H -^{15}N HSQC spectra of the RRM1 domains of wild-type U1A and SNF proteins. Data were collected at 22.5°C with 300 μM protein in 20 mM cacodylate (pH 6.5), 50 mM KCl, 2 mM EDTA, and 10% $^{2}H_2O$.

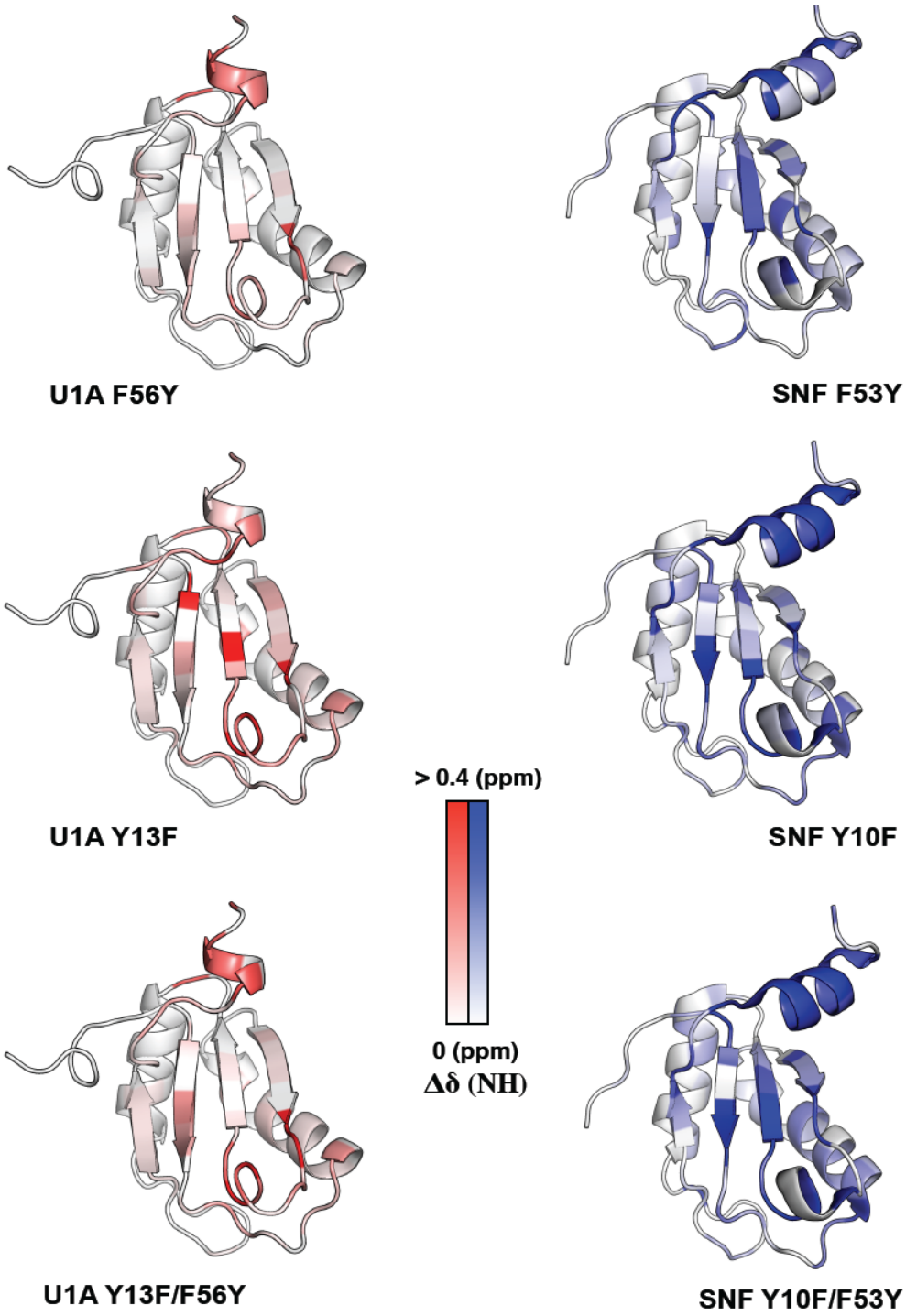

Figure S3. Chemical shift changes as a function of mutations in U1A and SNF RRM1. Data are plotted on the structures of U1A (1urn) and SNF (2k3k). In this U1A crystal structure (10), the third α-helix was truncated, while in the SNF NMR structure, it is shown in one of its possible orientations (13).

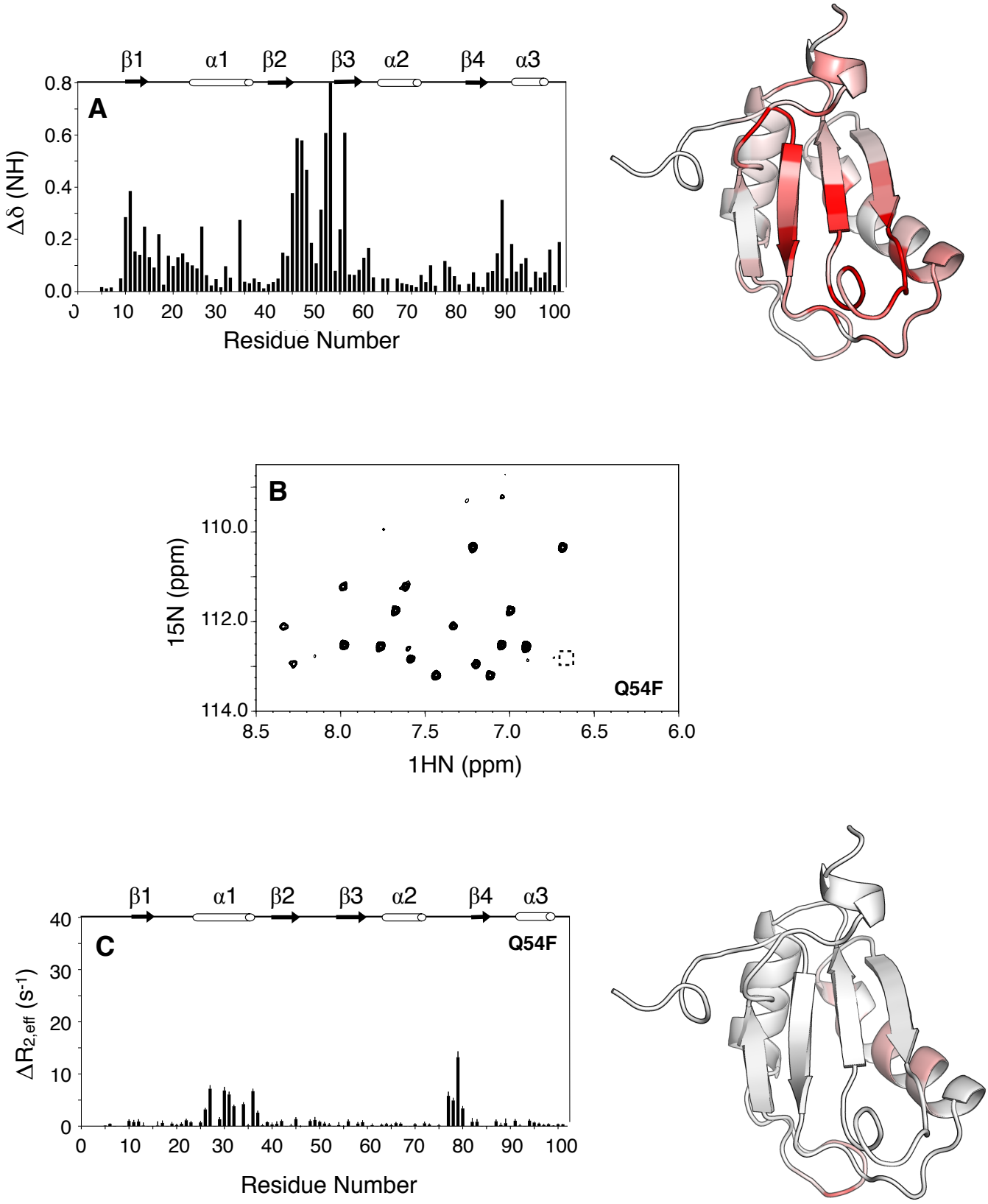

Figure S4. U1A mutant Q54F data. A) Backbone amide chemical shift differences from wild type (compare to Figure 3). B) {1H}-15N-HSQC spectrum of GlnAsn NH2 sidechain peaks. C) Intermediate exchange deltaR2 plot. Data are mapped onto the crystal structure (1urn) of U1A RRM1.

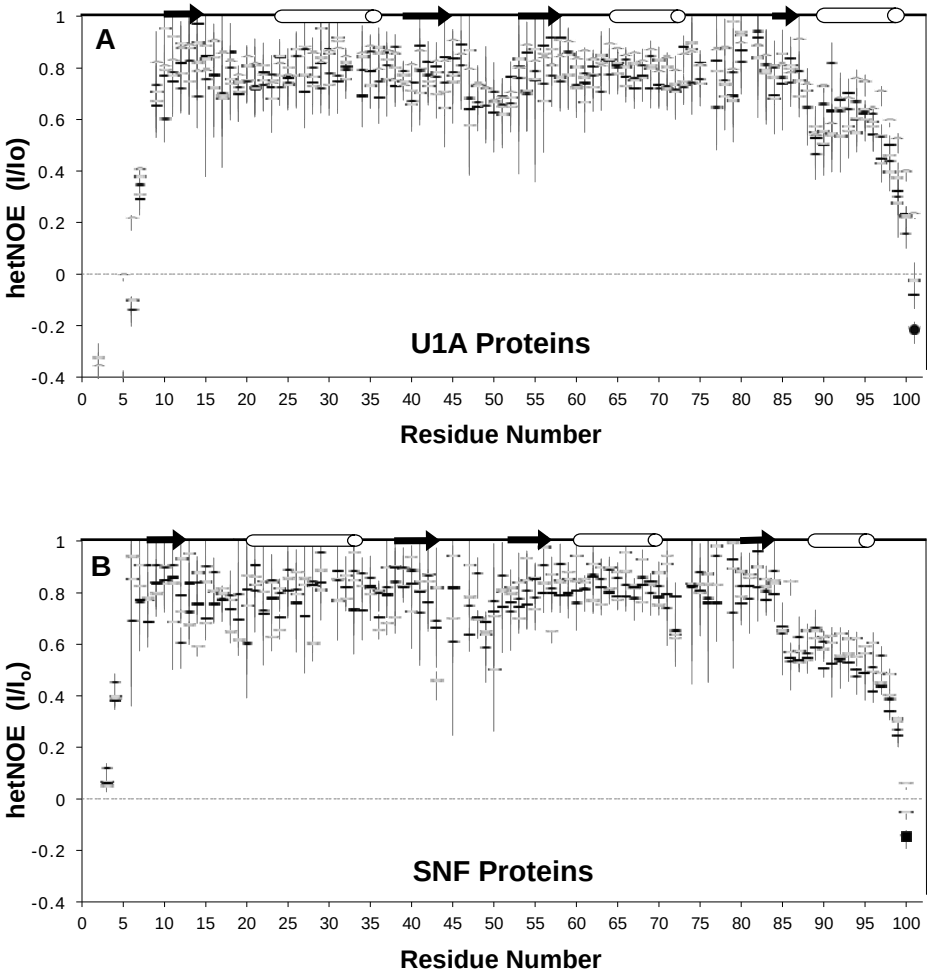

Figure S5. Backbone amide ^{15}N-{^{1}H} heteronuclear NOE data for U1A and SNF RRM1. (A) U1A RRM1 proteins: wild type U1A ●, U1A(F56Y) ■, U1A(Y13F) ○, U1A(Y13F/F56Y) □, U1A(Q54F) Δ, and in (B) SNF RRM1 proteins: wild type SNF●, SNF(F53Y) ■, SNF(Y10F) ○, SNF(Y10F/F53Y) □. Secondary structural elements are shown above the panel for reference. Errors were determined from the propagation of baseplane noise. Larger errors occur in SNF regions of broadened exchange, particularly in loop 3 where the signal/noise is poor.

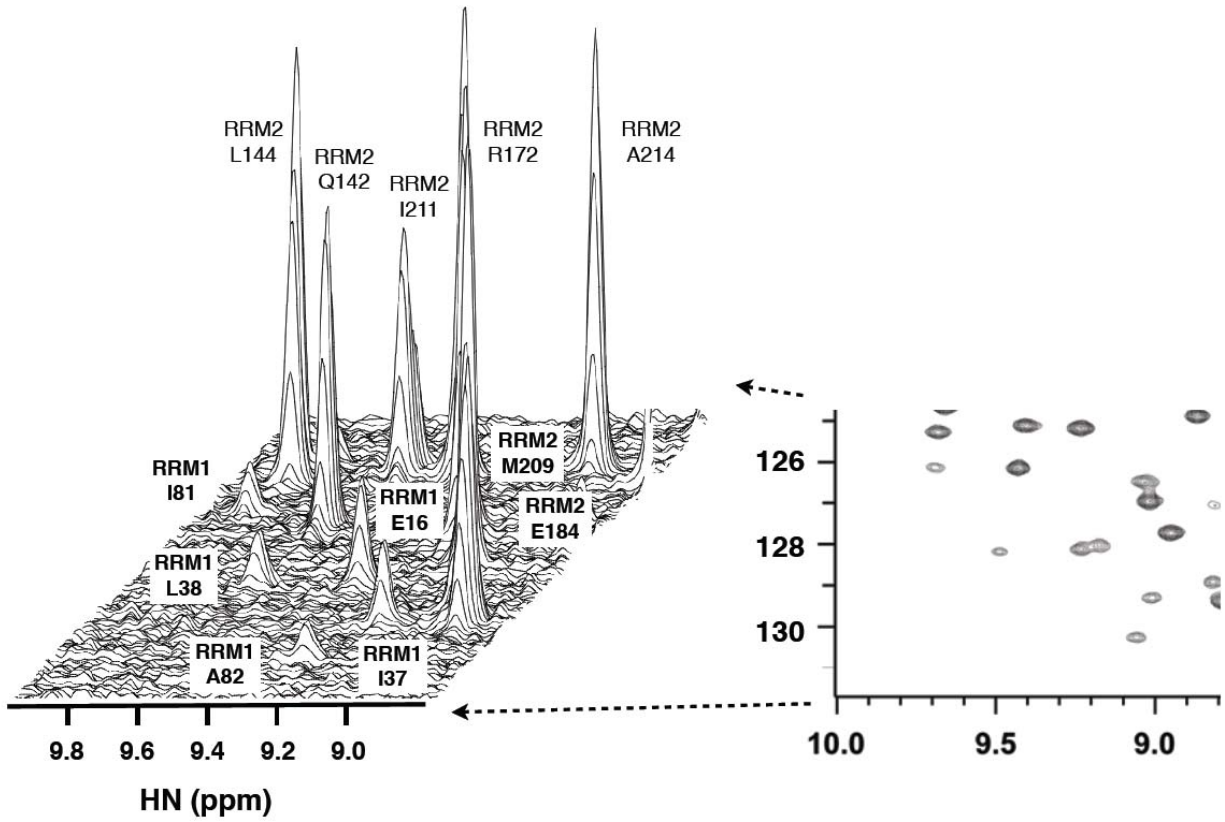

Figure S6. Intensity differences in full length SNF monitored by stacked plot. Left, is a stacked plot representation of the ^{1}H -^{15}N HSQC excerpt taken from the full length SNF spectrum (right). Peaks are labeled with the residue positions and RRM domain from the full length protein.

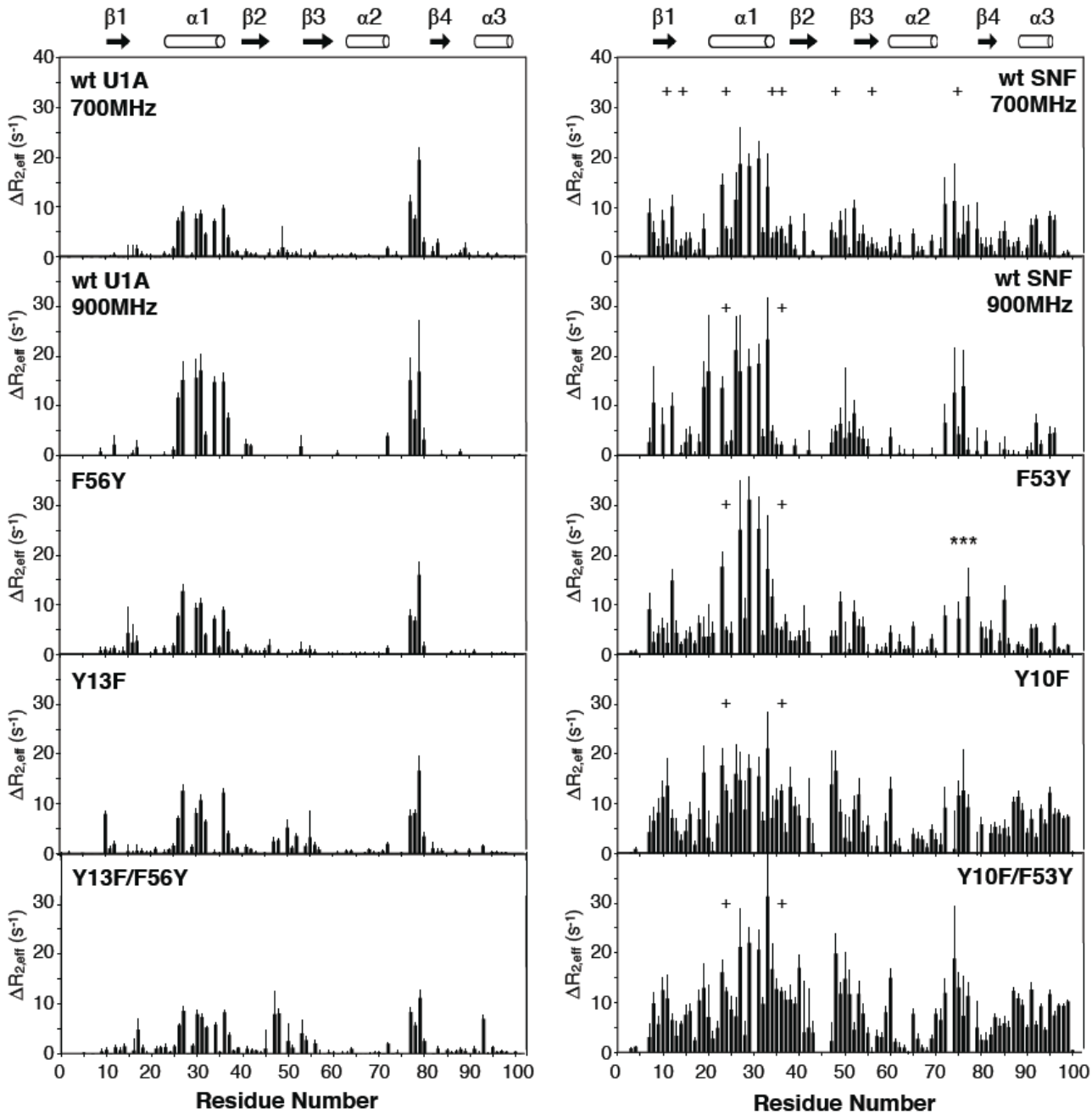

Figure S7. Intermediate exchange ΔR_2 for wild-type and mutant U1A (left column) and SNF (right column) RRM1 proteins. Secondary structural elements are above the plot for reference. Plus symbols (+) represent overlapped peaks that have $\Delta R_2 > 2$ sec^{-1}, which include residues I11/F56, E16/Y75, F34/M48 in wt SNF, and K24/Q36 in all SNF panels. Asterisks (*) represent missing ΔR_2 data broadened by exchange.