Supplementary information, Figure S3

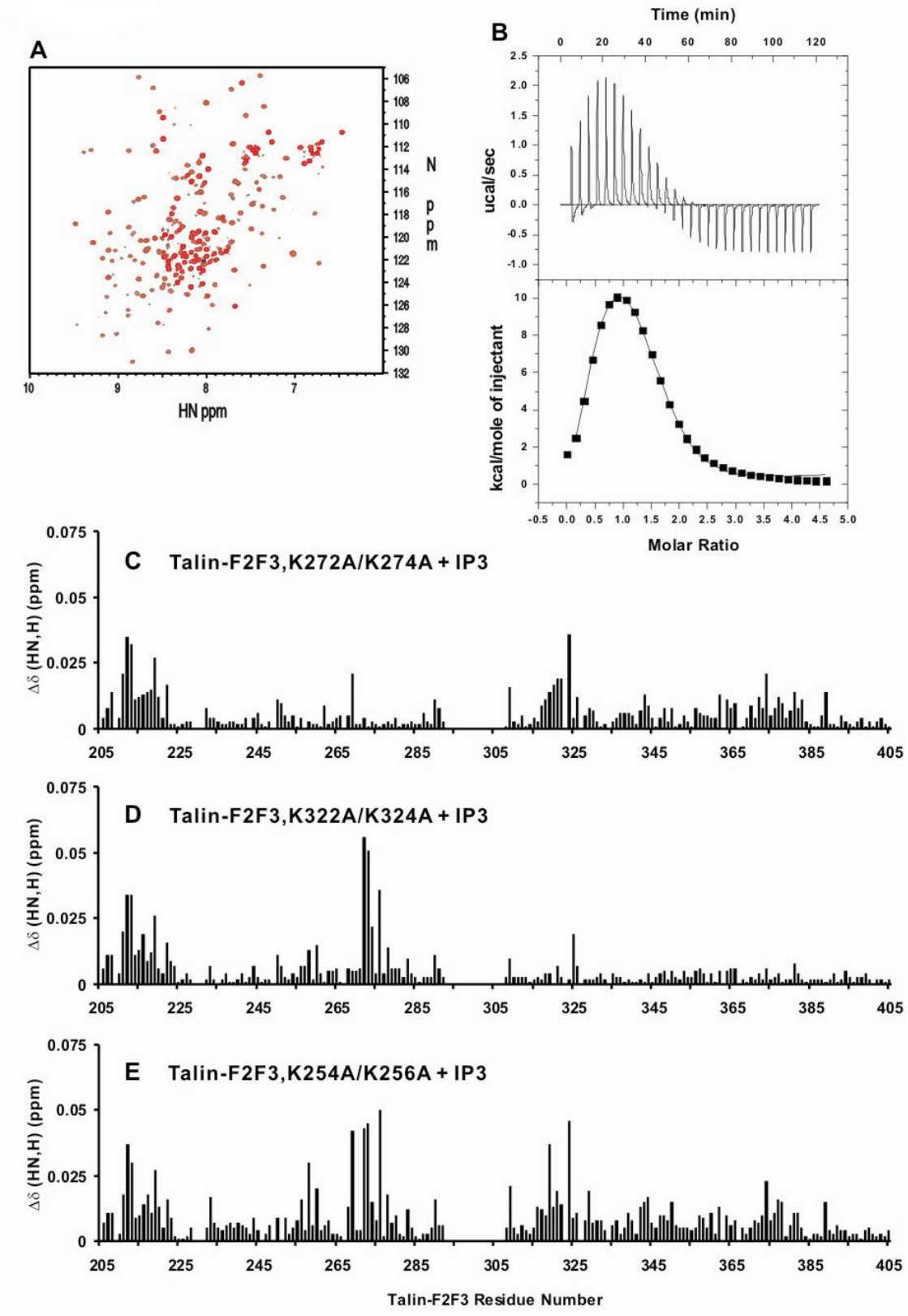

Figure S3 Properties of talin subdomain binding to PIP2. **(A)** Overlay of 0.04 mM ^{15}N-labeled talin-F0F1 in the absence (black) and presence of 0.2 mM IP4, showing no perturbation. Addition of 2.5 mM 19:1 POPC:PIP2 LUV also had little effect (not shown). **(B)** ITC measurement for Talin-H and PIP2-diC8 interaction. The upper panel shows the raw data and bottom panel shows that the data (square) fit very well with a 1:2 molar ratio binding event (curve), which is consistent with the NMR perturbation study (see the text) and recent functional analysis [15]. The K_D for first binding site is 0.4 ± 0.1 μM and 5.0 ± 0.5 μM for second binding site. The enthalpy and entropy changes are: ΔH_1 = –9.4 kcal/mol, ΔH_2 = 17.3 kcal/mol, ΔS_1 = –4.11 cal/(mol·K), and ΔS_2 = 82.2 cal/(mol·K). **(C-E)** Chemical shift plot of IP3 binding to various talin-F2F3 mutants. The patterns are similar to those for IP4 (see Figure 3C-3E).