Characterization of calmodulin and Fas death domain interaction: an integrated experimental and computational study

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Running Title: Interactions of CaM with Fas DD

Supporting Information

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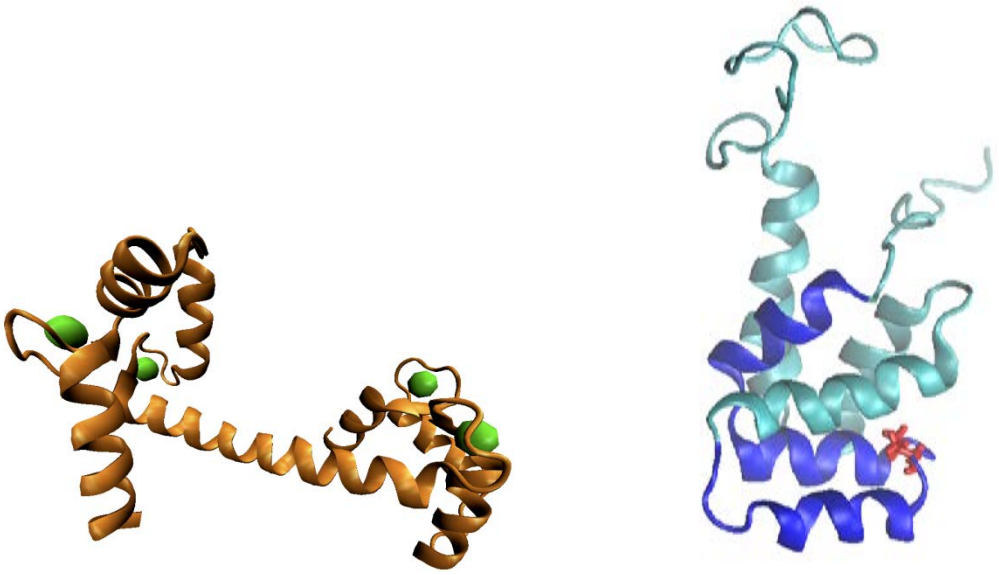


(a) (b)

Figure S1. (a) Structure of CaM (The four Ca^{2+} ions shown in green) (Ref. 7). (b) Structure of Fas DD (CaM binding region in Fas DD shown as blue, Val 254 residue shown as licorice in red) (Ref. 6 and 8). The images were made with VMD program.

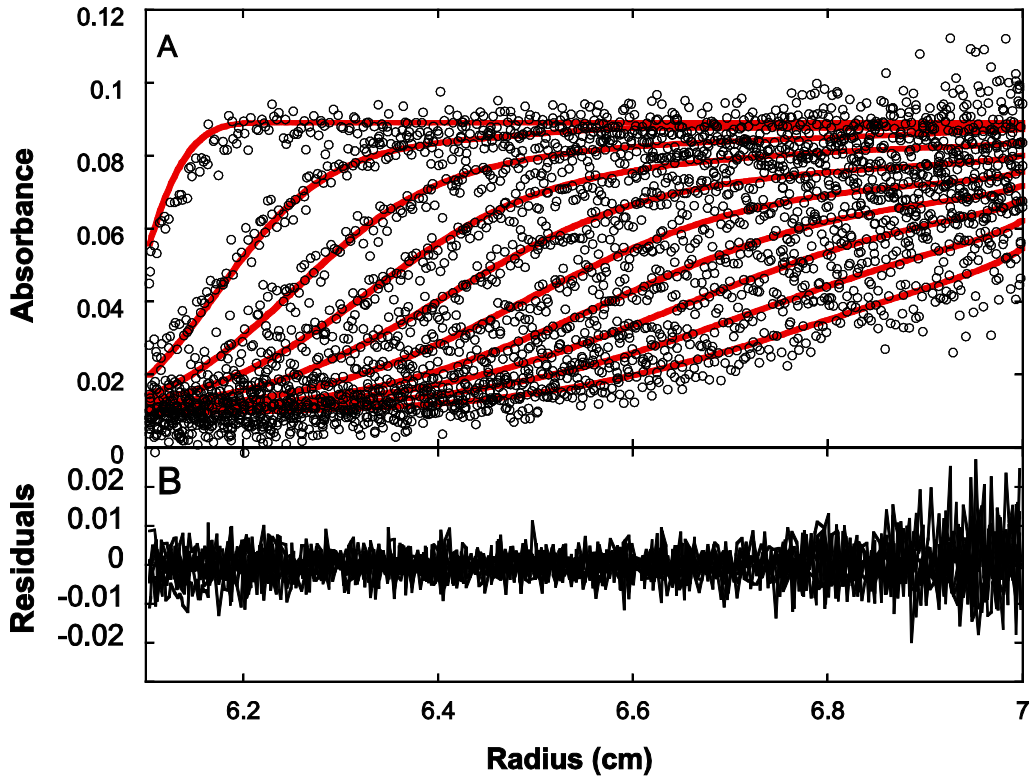


Figure S2. Sedimentation velocity experimental results. (A) Raw sedimentation velocity absorbance boundaries as a function of radial position for 10 □M Fas DD wild type in in a 50 mM sodium acetate pH 5, 150 mM NaCl, 20 mM CaCl2 and 0.5 mM Tris (2-carboxyethyl) phosphine (TCEP). The absorbance scans were collected every 1 minute 11 second at 280 nm, 37°C, every 15th scan is shown as example. Black open circles are data and red solid lines are the fit generated by c(s) analysis using Sedfit; (B) the residuals for the fit as a function of radial position.

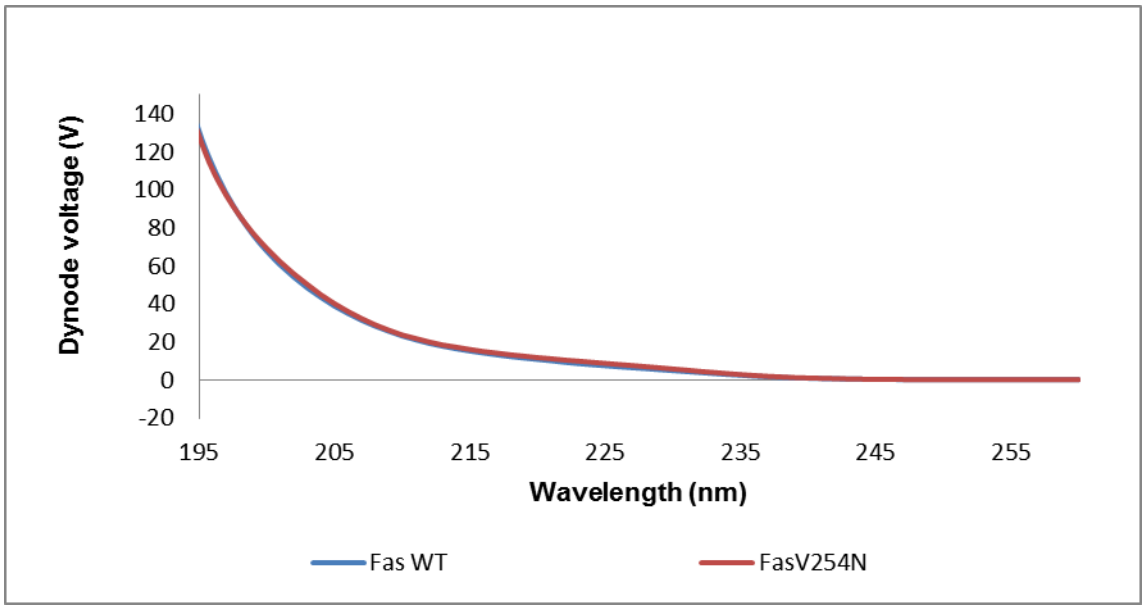


Figure S3. Plot of dynode voltage from Far UV CD spectra of Fas DD WT and Fas DD V254N. The dynode voltage traces for the Fas DD WT and Fas DD mutants overlay with no significant differences between traces, therefore differences in CD spectra of Fas DD WT and Fas DD V254N may not be attributed to differences in protein concentration between samples.

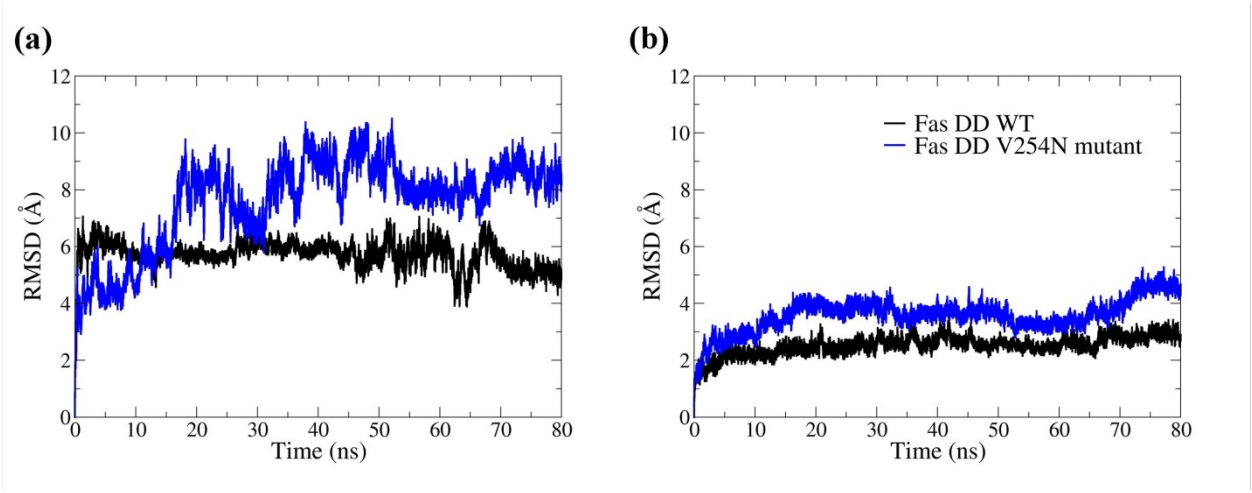


Figure S4. RMSD for Fas DD WT (black line) and Fas DD V254N mutant (blue line) (a), and for Fas DD protein core (residues 225~318) (b) over the 80 ns MD simulations.

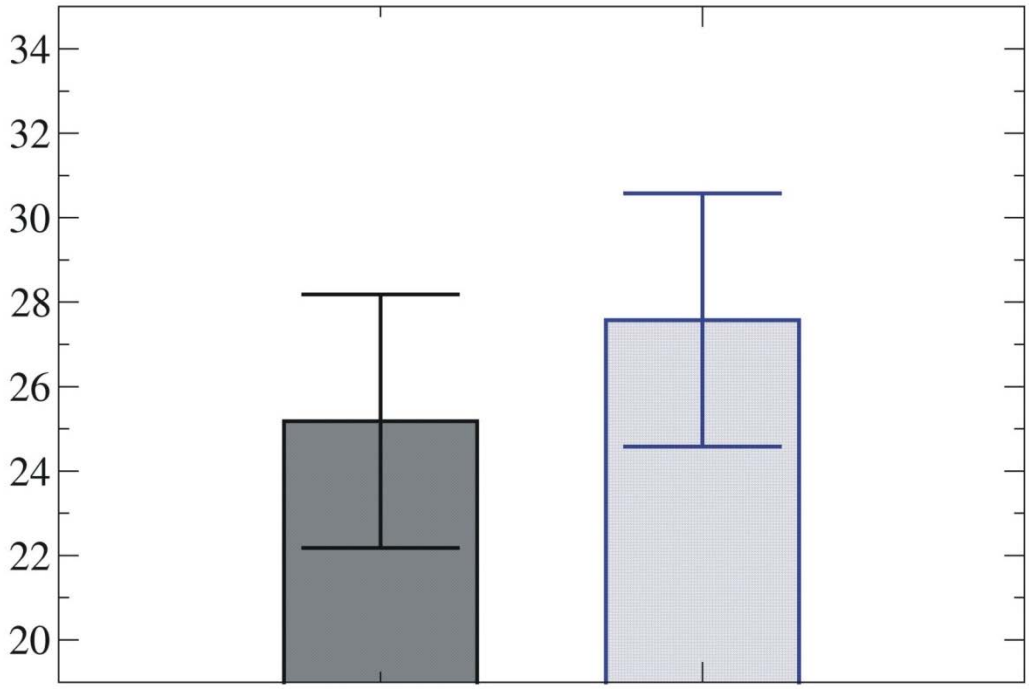


Figure S5. Comparison of the number of H bonds formed between CaM-binding region in Fas DD and the other part of Fas DD.

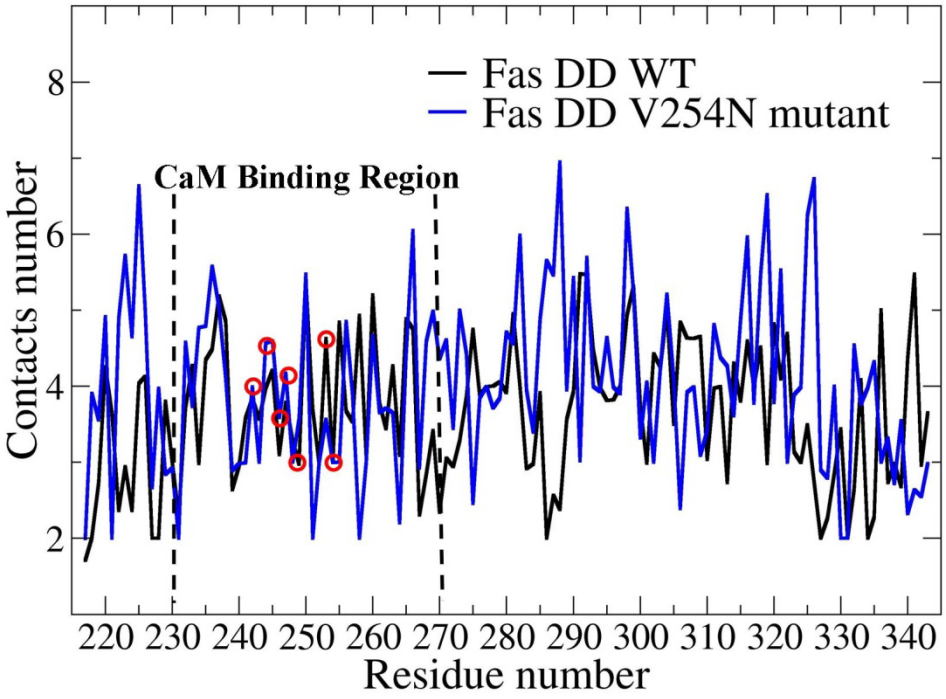
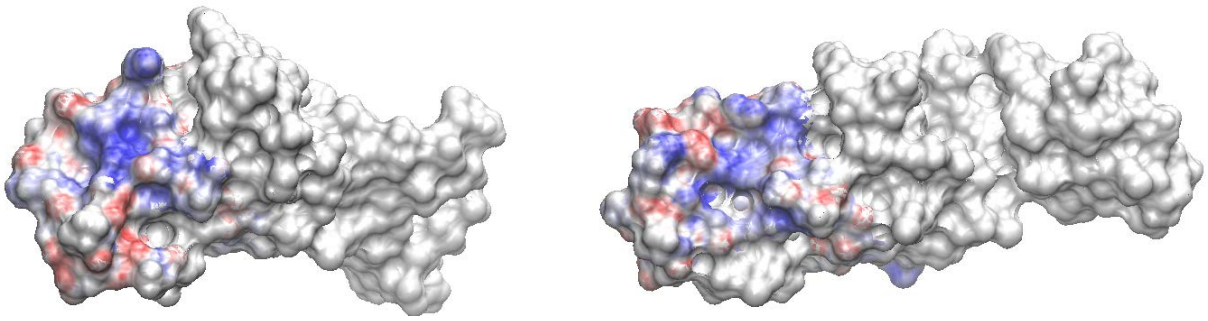


Figure S6. Contact numbers for residues of Fas DD. The CaM binding region of Fas is shown between two dotted lines, and the seven non-polar residues in the binding site are shown in red circles.

Fas DD WT

Fas DD V254N mutant



(a)

(b)

Figure S7. Electrostatic potential map for CaM-binding site in Fas DD WT and Fas DD V254N mutant (blue for positive electrostatic potential and red for negative electrostatic potential). Electrostatic potential map for CaM-binding site in Fas DD WT was different from that of Fas DD V254N mutant due to V254N mutation of the Fas DD.