Supplementary Data for

Bacterial aptamers that selectively bind glutamine

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Figure S1. The 67 *glnA* RNA binds D-glutamine. (**A**) In-line probing analysis of the 67 *glnA* RNA with various concentrations of D-glutamine, ranging from 100 µM to 10 mM. Other annotations are as described for Figure 2B. (**B**) Plot of normalized fraction of band modulation verses the logarithm of the concentration of D-glutamine. The solid line is a hypothetical binding curve for a standard one-to-one interaction with a K_D of 7 mM. Data points were derived from the numbered regions in (A).

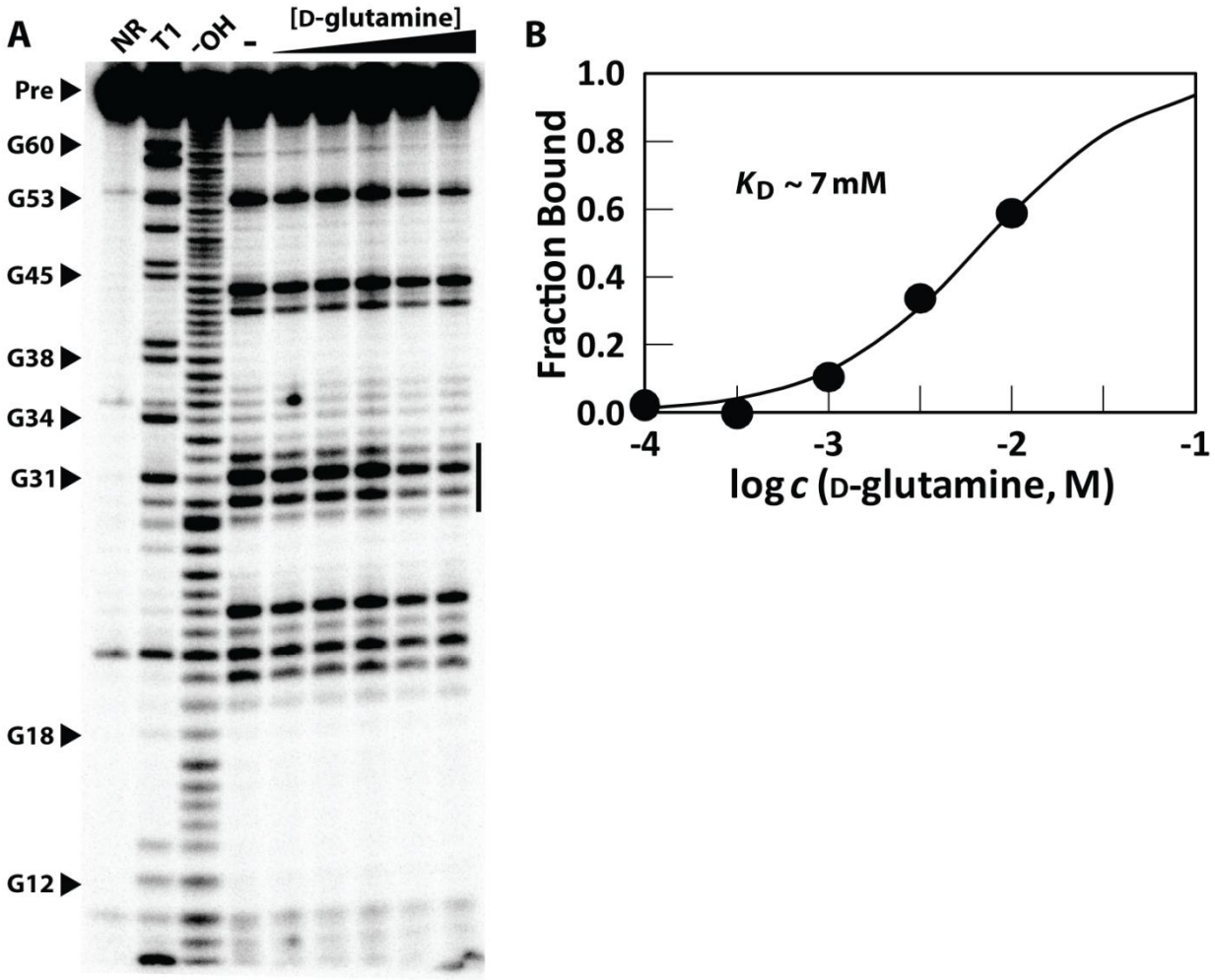

Figure S2. Disruptive mutations to the 67 *glnA* secondary structure interfere with binding L-glutamine, while compensatory mutations restore binding. (**A**-**D**) In-line probing analyses for RNA constructs M1, M2, M3, and M4 respectively. Lane annotations are similar to those used in Figure 2B. Concentrations of L-glutamine used in the in-line reactions ranged from 1 µM to 10 mM. (**E**-**F**) Plots of the logarithm of L-glutamine concentration versus fraction of RNA bound for the M3 and M4 RNAs. Points on the graphs were obtained from the gel regions in B and D labeled with numbered vertical lines. The solid lines on the plots show hypothetical binding curves for standard one-to-one interactions using the K_D values shown on the graphs.

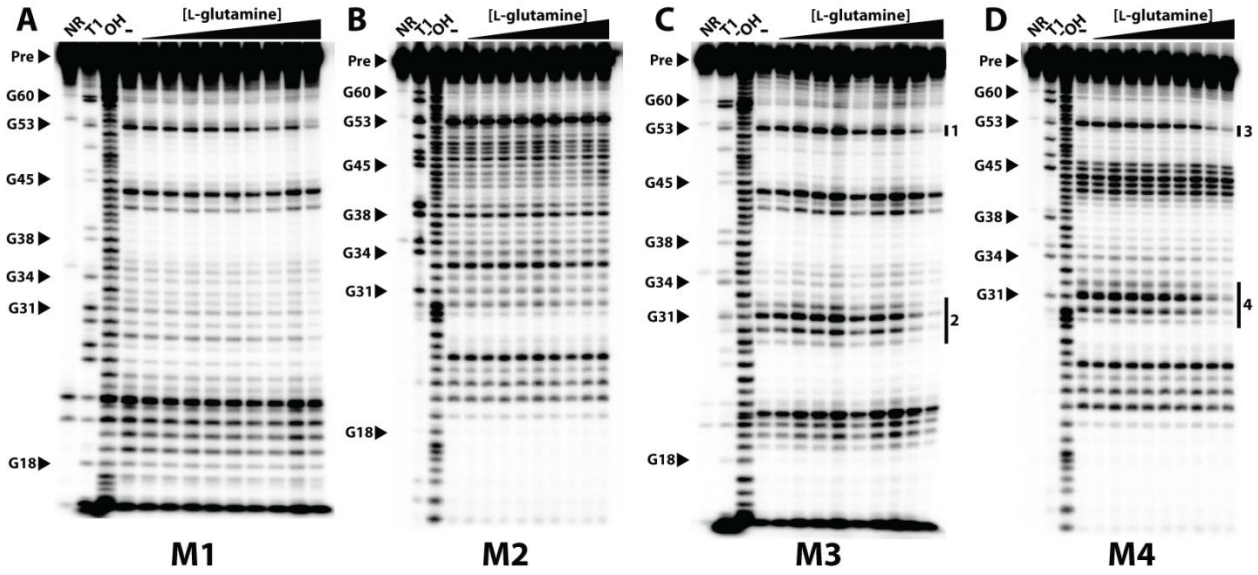

Figure S3. The 67 *glnA* RNA does not bind L-glutamate. In-line probing analysis of the 67 *glnA* RNA with L-glutamate. Annotations for the first four lanes are the same as those on Figure 2B. L-gln designates an in-line probing reaction where 10 mM L-glutamine was added, and L-glu designates a reaction containing 100 mM L-glutamate. Note that in reactions containing L-glutamate the buffer conditions were modified from the standard in-line probing conditions. See the materials and methods section in the main text for details.

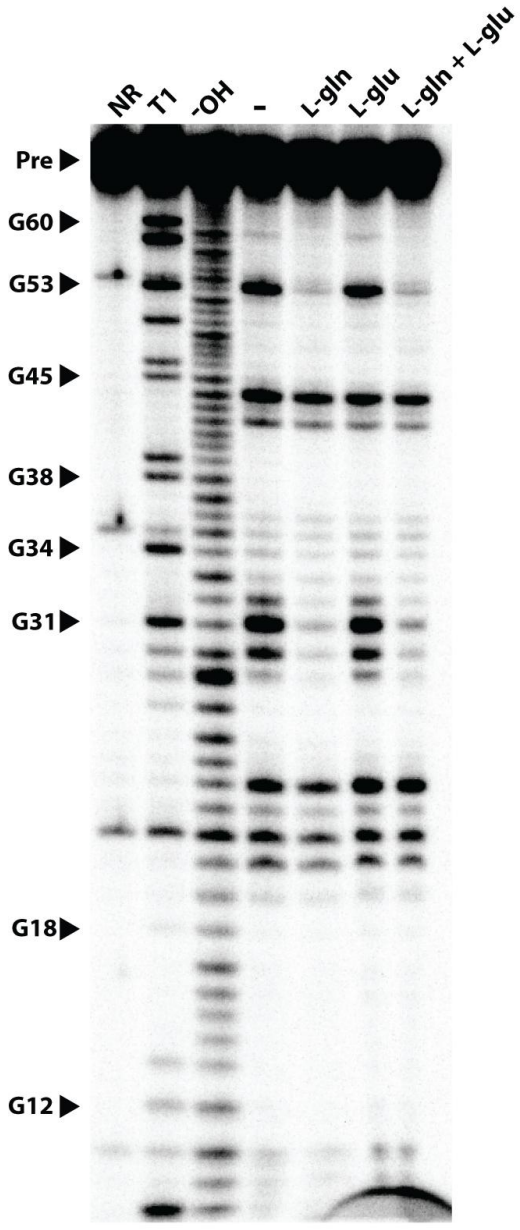

Figure S4. The 83 DP RNA rejects a variety of L-glutamine analogs. In-line probing analysis of the 83 DP RNA with a variety of compounds. Lane annotations are similar to those used in Figure 3A, with the addition of the dark wedge representing lanes where the RNA was tested with D-glutamine, using concentrations ranging from 100 µM to 10 mM.

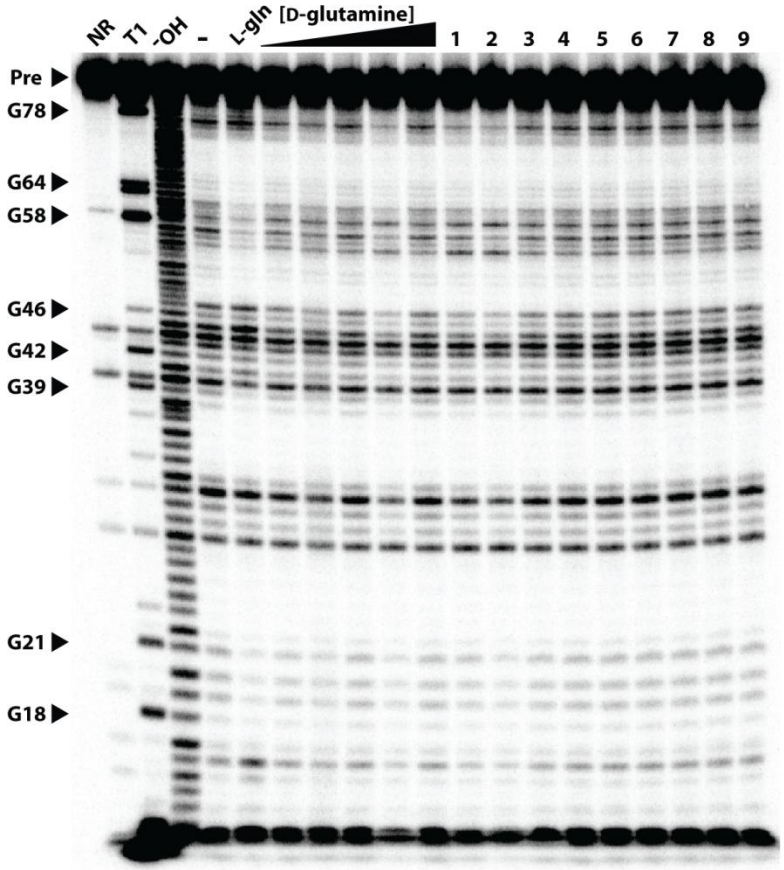

Figure S5. Mismatch mutations in the predicted pseudoknot of the 83 DP RNA interfere with ligand binding. (**A**, **B**) In-line gels testing RNAs M5 and M6 with L-glutamine. Concentrations of ligand used ranged from 10 µM to 10 mM. Numbered arrows indicate bands quantified and plotted in C. Other lane annotations are similar to those for Figure 2B. (**C**) Plot of the normalized band modulation versus the log of the concentration of L-glutamine. The solid line shows a hypothetical K_D curve for a standard one-to-one interaction with a disassociation constant of 3 mM.