Supplementary information

Conformational heterogeneity in the dGsw purine riboswitch: role of Mg^{2+} and 2'-dG in aptamer folding

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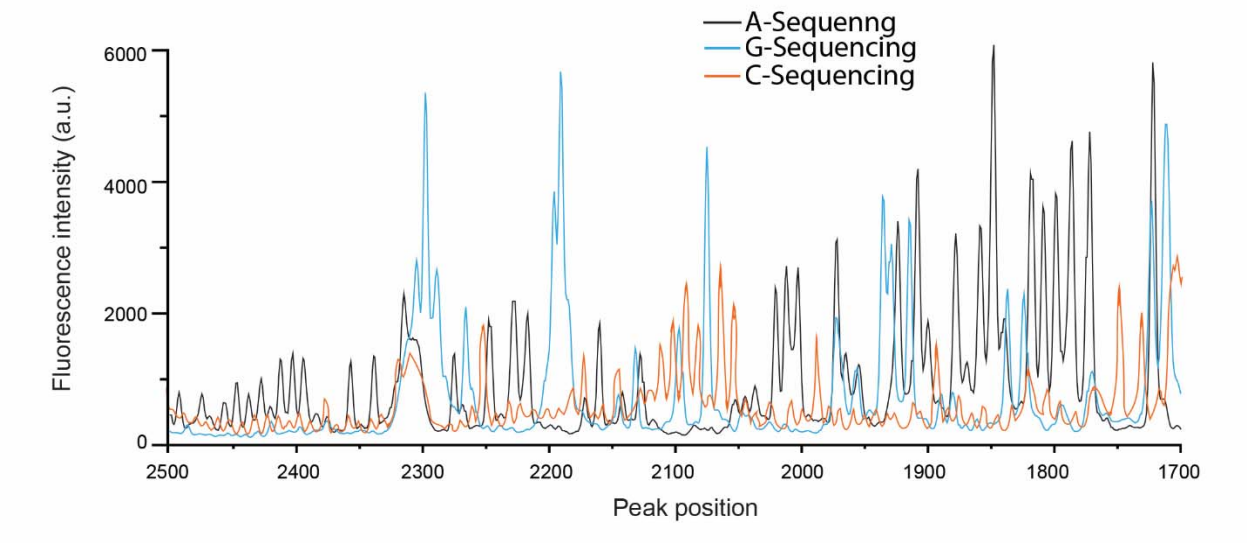
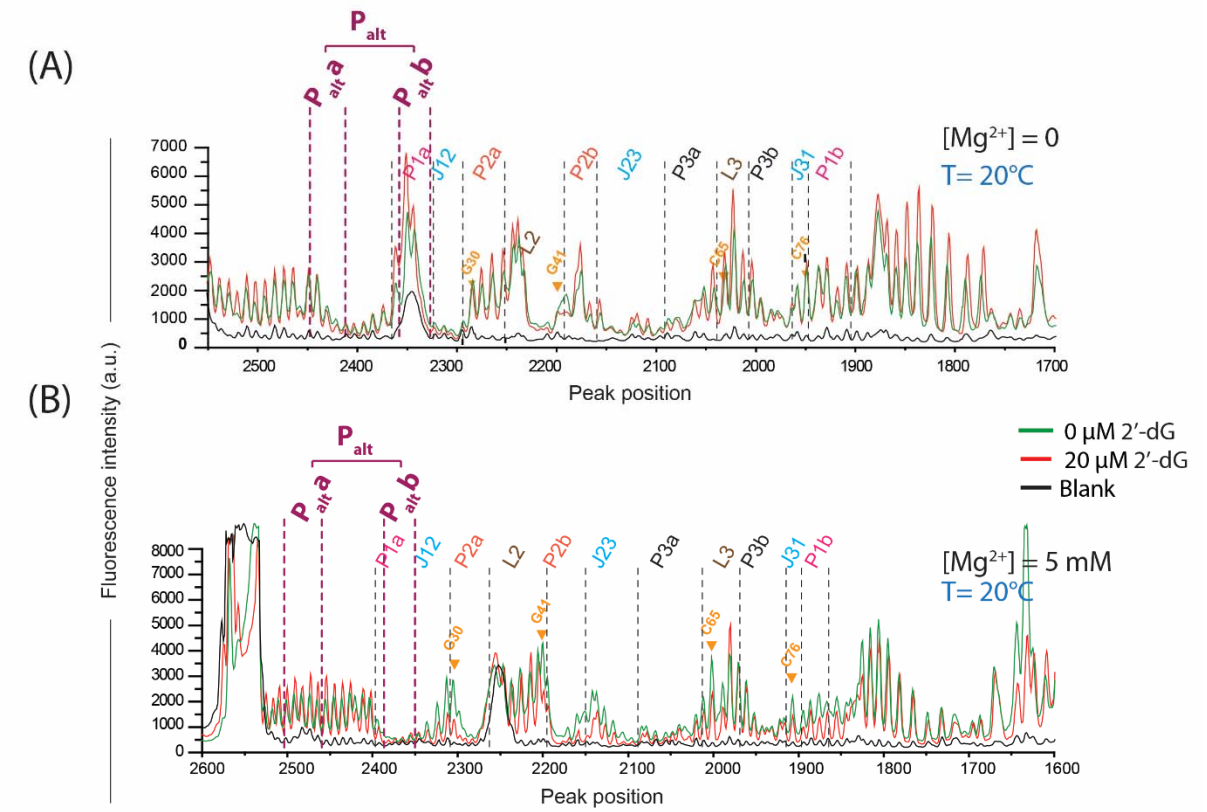
SI Figure 1



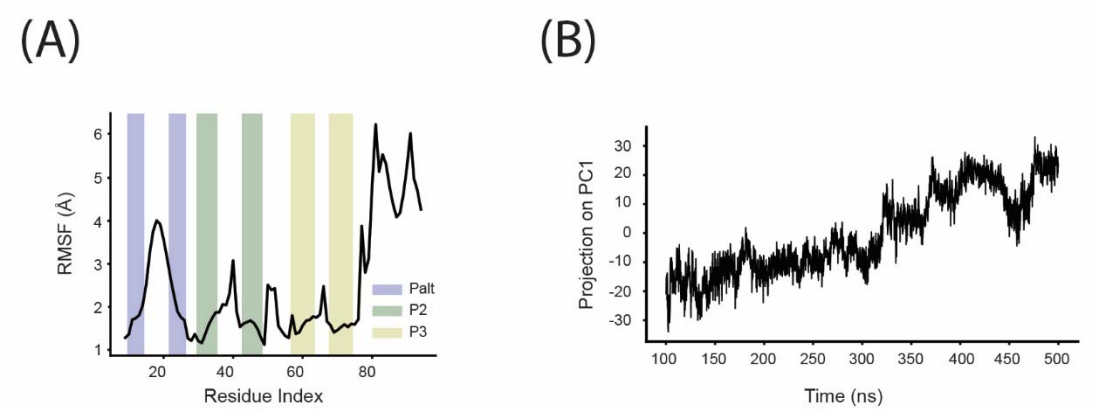
SI Figure 1: Sequencing Data of dGsw-apt RNA. A primer extension reaction for the sequencing ladder was also performed by dideoxy sequencing on unmodified RNA. Each sequencing reaction was supplemented with complementary ddNTPs

SI Figure 2



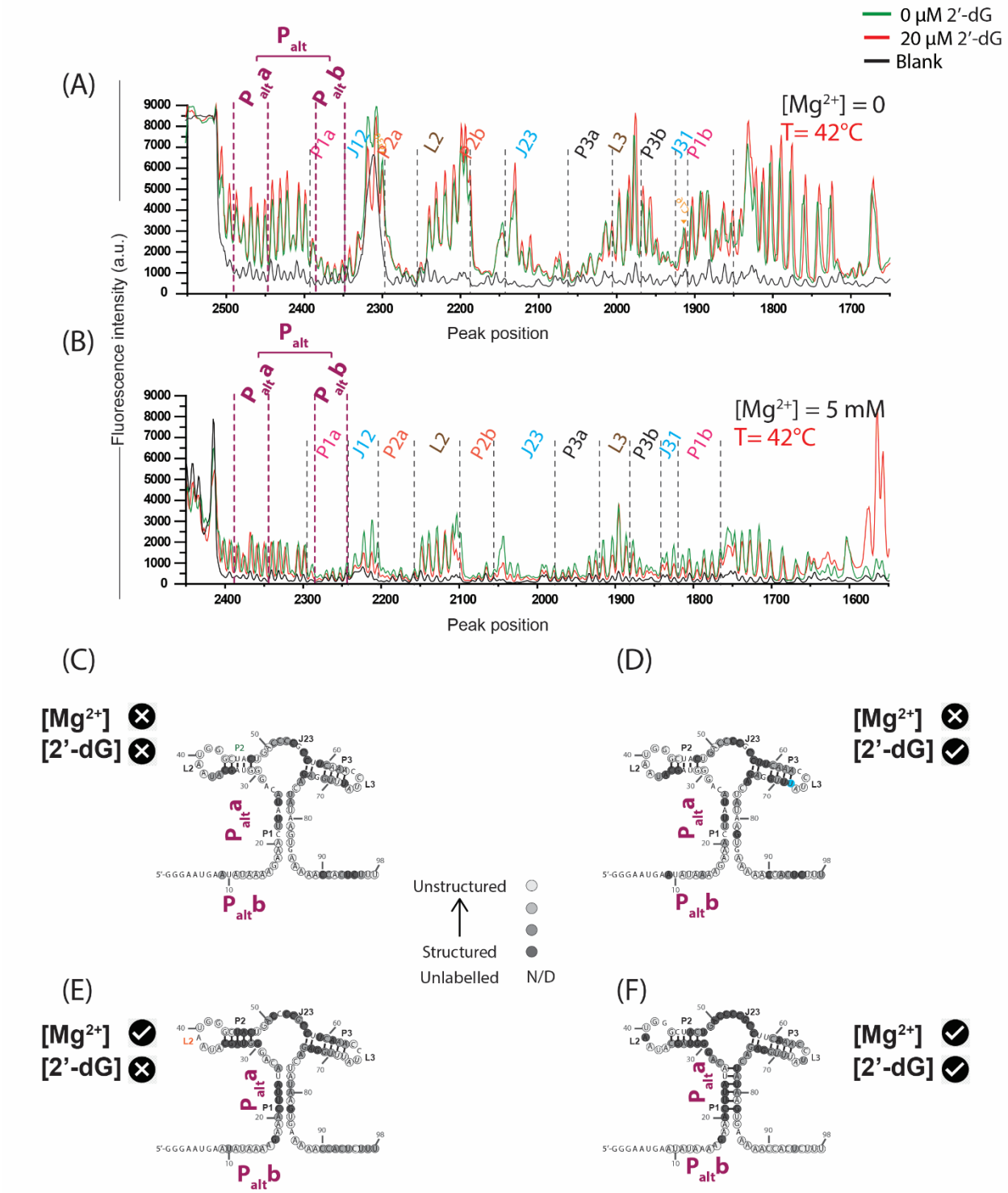
SI Figure 2: SHAPE probing data for the Mg^{2+} and 2′-dG titrations. Representative capillary electrophoresis traces are shown for 2′-dG titrations at 20°C (A) without Mg^{2+} and (B) 20µM 2′-dG.
The overlaid traces depict the data without 2′-dG (green) and with 20µM2′-dG (red). The nucleotides from the different structural regions analyzed here are indicated.

SI Figure 3



SI Figure 3 Computational method checks the stability of the P_{alt} helix and possible transition. (A) RMSF at nucleotide level in the case of 100 mM K^+ and dG[-] for the last 400 ns of the simulation. Due to the unstructured loop region, the P_{alt} helix shows a well-preserved structure while fluctuating more than the P2 and P3 helices. The significant fluctuation in the tail region suggests that it does not have a stable structure. (B) The simulation's first principal component (PC1) of the simulation with the first 100 ns of the trajectory and the free tail of RNA molecule removed. Trajectory projected on its first principal component, tending to increase.

SI Figure 4



SI Figure 4: SHAPE probing data for the Mg^{2+} and 2′-dG titrations. Representative capillary electrophoresis traces are shown for 2′-dG titrations at 42°C (A) without Mg^{2+} and (B) 20µM 2′-dG.

The overlaid traces depict the data without 2′-dG (green) and with 20µM2′-dG (red). The nucleotides from the different structural regions analyzed here are indicated. Overlay of SHAPE reactivities onto a

secondary structure model portraying a 76 nt dGsw-apt RNA. The color code serves to distinguish reactive (and thus unconstrained) nucleotides in C-F. An absence of markings denotes unanalyzable nucleotides. The lightest gray denotes nucleotides with the highest SHAPE reactivity, while the darkest gray indicates the lowest reactivity (C) 0mM Mg^{2+} without 2'-dG; (D) 0mM Mg^{2+} with 20 μM 2'-dG; (E) 5 mM Mg^{2+} without 2'-dG; and (F) 5 mM Mg^{2+} with 20 μM 2'-dG.