Supporting Information for:

Structure and dynamics of the HIV-1 frameshift element RNA

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Two figures and supporting text.

Supplemental Figure 1. Frameshift domain SHAPE reactivities for extracted *ex virio* RNA and refolded heat denatured RNA using three SHAPE reagents.

Supplemental Figure 2. Formamide-induced destabilization of the frameshift domain. (A) SHAPE reactivity profiles for formamide-denatured RNA (red and green) compared with *ex virio* reactivities (blue). (B) Average SHAPE reactivities for each helical strand as a function of formamide concentration. Helices defined by both the SHAPE-directed and conventional models are shown.

Supplemental Figure 2 – Discussion: Nucleotides on one side of the slippery sequence helix (1604-1611) had low reactivity in 10% (vol/vol) formamide but showed markedly increased reactivity at formamide concentrations of 20% and above. Only modest reactivity changes were apparent in 60% formamide on the partner strand (1629-1636) (Supplemental Figure 2A, yellow). These data indicate that the slippery sequence helix unfolds at 20% formamide, although the partner was protected from SHAPE reagent in the presence of high denaturant concentration. Nucleotides 1590-1595 form the alternate lower stem and were unreactive at up to 10% formamide, but became very reactive at 20% formamide and greater. Partner nucleotides 1678-1683, which can pair with either the alternate or conventional lower stem, were unreactive in up to 30% formamide but modestly reactive at higher formamide concentrations. Nucleotides 1638-1645, involved in the conventional lower stem, exhibited little change in reactivity in up to 30% formamide but became reactive at higher concentrations. These data suggest that the alternate lower stem unfolds and some of these nucleotides base pair to form the conventional lower stem conformation in 20% formamide (Figure 3B). With further increases in formamide concentration, the conventional lower stem also unfolds (Figure 3C). The low reactivity of the 1629-1636 region to SHAPE in 60% formamide presumably reflects formation of alternative semi-stable structures as the RNA is globally denatured.



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Low et al / Supp Figure 2