Supplementary Figure 1. Predicted secondary structures of RNAs that do not bind. Top: $GABA_A$ γ2 Exon4. Middle: c-src N1 exon hairpin. Lower: HCV X SL2. Secondary structure predictions and folding free energies are by *mfold* (28,29).

Supplementary Figure 2. Structure probing and footprinting of PTB1 with c-src 5′ and 3′ RNAs. Lane (-) is RNA alone. OH is base hydrolysis ladder. T1L is nuclease T1 cleavage under denaturing conditions to sequence the strand. Two lanes of nuclease T1 and two of RNase A are cleavage reactions at two concentrations of enzyme. Nuclease T1 cleaves single-stranded Gp↓N sites, and RNase A cleaves single-stranded (Py)p↓N sites. Note that in one lane of RNase A cleavage the reaction has gone too far and digested the RNA. Fig 2a) c-src 3′ RNA alone and with 10 nM and 1 μM PTB1 probed with nuclease T1 and RNase A. The cleavage pattern seen is consistent with the predicted RNA structure shown, in which nearly all GpN sites are in duplex regions and not accessible to nuclease T1. The pyrimidines marked * are cleaved by RNase A in the free RNA and at 10 nM PTB1, but become protected at 1 μM PTB1. Note that addition of PTB1 does not enhance cleavage at any site, indicating that the protein does not melt the duplex regions. Fig 2b) c-src 5′ RNA alone and with 10 nM and 1 μM PTB1. The cleavage pattern is consistent with the predicted structure shown (which is not the lowest free energy structure). Addition of 1 μM PTB1 protects nt 10 – 30 from cleavage, but does not significantly reduce nuclease T1 cleavage of G43/44/47/48 in the loop.