attC_{aadA7}- attC_{ereA2} synthetic cassettes

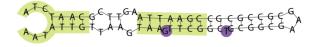
attC_{aadA7}

top strand (ts) sequence:

5'-TTATAACAATTCATCAAGCCGACGCCGCTTCGCGGCGCGCGTTAATTCAAGCGTTAGAT

 ΔG_{bs} value of the most stable structure without constraint= -15.5 kcal/mol ΔG_{bs} value of the most stable structure with constraints (recombinogenic)= -15.5 kcal/mol $attC_{bs}$ pfold value calculated with constrained vs. unconstrained folding= 0.30 The most stable structure is the recombinogenic one:

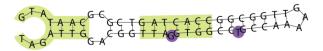
attC_{aadA7}bs



attC_{ereA2}

ts sequence: 5'-ATCTAACCTGCCAATCCACCGGACGGTTTTCAACCGCCGGTGATCAGCGCGTTATAC ΔG_{bs} value of the most stable structure without constraints= -14.0 kcal/mol ΔG_{bs} value of the most stable structure with constraints (recombinogenic)= -14.0 kcal/mol $attC_{bs}$ pfold value calculated with constrained vs. unconstrained folding= 0.44 The most stable structure is the recombinogenic one:

attC_{ereA2}bs



attC_{aadA7}- VCR₁₂₆ synthetic cassettes

attC_{aadA7}

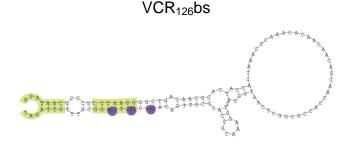
 ΔG_{bs} value of the most stable structure without constraints= -15.5 kcal/mol ΔG_{bs} value of the most stable structure with constraints (recombinogenic)= -15.5 kcal/mol $attC_{bs}$ pfold value calculated with constrained vs. unconstrained folding= 0.30 The most stable structure is the recombinogenic one:

attC_{aadA7}bs



VCR₁₂₆

 ΔG_{bs} value of the most stable structure without constraints= -24.1 kcal/mol ΔG_{bs} value of the most stable structure with constraints (recombinogenic)= -16.7 kcal/mol $attC_{bs}$ pfold value calculated with constrained vs. unconstrained folding= 1.04×10^{-5} The most stable structure is a non-recombinogenic one (improperly folded). The recombinogenic structure is:

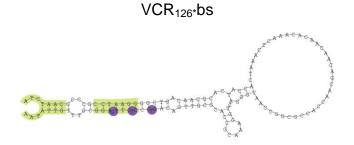


*VCR*_{126*}- *VCR*_{126**} synthetic cassettes

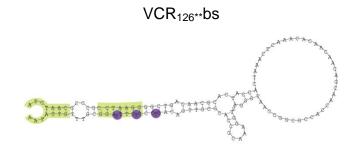
VCR_{126*}

 ΔG_{bs} value of the most stable structure without constraints= -20.4 kcal/mol ΔG_{bs} value of the most stable structure with constraints (recombinogenic)= -16.7 kcal/mol $attC_{bs}$ pfold value calculated with constrained vs. unconstrained folding= 3.09×10^{-3} The most stable structure is a non-recombinogenic one (improperly folded).

The recombinogenic structure is:



VCR_{126**}



VCR_{16*}- VCR₆₄ synthetic cassettes

VCR_{16*}

ts sequence: 5'- TTATAACAAACGGCTCAAGAGGGACTGTCAACGCGTGGCGTTTCCAGTCCCAAGAACTGC GGCGTTTACGGTTGCTGTGTTTGAGTTTAGTGTTAATGCGTTGCCAGCCCCTTAGCCGGGCGTTAGAT ΔG_{bs} value of the most stable structure without constraints= -19.6 kcal/mol ΔG_{bs} value of the most stable structure with constraints (recombinogenic)= -19.6 kcal/mol $attC_{bs}$ pfold value calculated with constrained vs. unconstrained folding= 0.35 The most stable structure is the recombinogenic one:

VCR_{16*}bs



VCR₆₄

VCR₆₄bs



Structures were determined by the RNAfold program from the ViennaRNA 2 package (Methods).