Effects of i-Motifs and G-quadruplexes on Bacterial Gene Transcription

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i-Motifs and G-quadruplexes are unusual DNA secondary structures that have been implicated as regulatory elements for eukaryotic gene expression. i-Motifs are intercalated ladder formations made up by intrastrand C-C base pairing, and G-quadruplexes are stacked intrastrand G-G base pairings. The aim of my research project was to study the effects of i-motifs and G-quadruplexes on bacterial gene transcription. I used Golden Gate Assembly to insert i-motif- and G-quadruplex-forming DNA sequences into a promoter located upstream of a Red Fluorescent Protein (RFP) reporter gene. I transformed the resulting constructs into E. coli , along with two control sequences. I conducted both in vivo and in vitro gene expression experiments and measured RFP expression using fluorometry. I attempted footprinting on the DNA sequence with T7 Endonuclease I and S1 Nuclease, but was unable to measure any difference between the control sequences and the i-motif and G-quadruplex sequences. However, the in-vivo and in-vitro results showed significant gene expression differences between promoter sequences with and without i-motif and G-quadruplex structural modifications. My results showed that the i-motif and/or G-quadruplex affect bacterial gene transcription, and have implications for improving the understanding of the regulatory role that these unusual DNA secondary structures might play in all cells.

Awards Won:
Fourth Award of $500