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Functional Analysis of a Putative Homing Endonuclease

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Poster Presentation P35

FUNCTIONAL ANALYSIS OF A PUTATIVE HOMING ENDONUCLEASE

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Endonucleases are enzymes that cleave phosphodiester bonds within polynucleotide chains such as DNA and RNA. A subgroup, homing endonucleases (HEase) are also able to propagate their encoding genes by transforming host genes to incorporate the homing endonuclease gene (HEG). This process is initiated by the expression of the HEase from the HEG, which then cleaves a homolog. The homolog subsequently undergoes a homologous recombination event as a repair mechanism, and in the process integrates a copy of the HEG. Based on predicted sequence, gene product (gp126) from the mycobacteriophage Gizmo is thought to encode for a HEase. Previous work constructed a vector for gp126 from which protein was expressed and purified. The goal of this study was to demonstrate that the DNA of the mycobacteriophage Shrimp was a viable target for the HEase and to characterize the function of the homing endonuclease. Shrimp has a very similar sequence to Gizmo, which contains the putative homing endonuclease, making it a viable substrate for experimentation. Demonstration of DNA binding will be a first step in the characterization of the interactions between gp126 and the Shrimp DNA.