Exploring Species Specificity of Lambda Red Recombination

Gabriel Filsinger¹* (filsinger@g.harvard.edu), Tim Wannier,¹ Xavier Rios,¹ Chris Gregg,¹ Marc Lajoie,² George M. Church¹

¹Harvard Medical School, Department of Genetics, Boston, MA; ²University of Washington, Seattle, WA

http://arep.med.harvard.edu

Project Goals: To extend recombineering methods to organisms other than E. coli by identifying and overcoming the host-specificity of Beta recombinases.

Recombination using the lambda red protein Beta has been extensively used in E. coli to incorporate insertions, deletions, and point mutations into the genome at chosen loci. Although Beta increases the rate of ssDNA recombination in E. coli 10,000 fold, this catalytic recombination activity has not been observed in other bacteria such as Lactobacillus or Corynebacterium. Interestingly, the family of single-stranded annealing proteins that includes Beta has been found to have species-specific activities and unpredictable efficiencies, making it difficult to design a generalized method for porting Beta recombination across organisms. In order to explore mechanisms of generalizing the recombinase activity, we use Lactobacillus and E. coli as two model organisms with orthogonal recombination systems to probe the source of species specificity.

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