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Biophysical Analysis of Homeodomain Transcription Factors

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Biophysical Analysis of Transcription Factors

Greg Amador '22– (Sponsor: Dr. Donald Spratt)

Abstract

Homeodomain transcription factors in *Drosophila Melanogaster* are essential to proper embryogenesis. This multidisciplinary project utilizes the expertise of several labs here at Clark including the Spratt, Drewell, and Dresch labs to identify the specific mechanisms of transcription factors that identify and bind to DNA segments. While there has been research surrounding these homeodomains, their ability to selectively recognize and bind to target DNA in the cell is largely unstudied. The impact of this research has the potential to bring greater understanding to how these transcription factors affect the development of an embryo. Due to their heavy expression in the early embryo, as well as their subsequent control of transcription of important development genes, it is likely that the research being conducted in this project will shed some light on the importance of these homeodomains. For this project, the transcription factor *even-skipped (eve)* is researched.

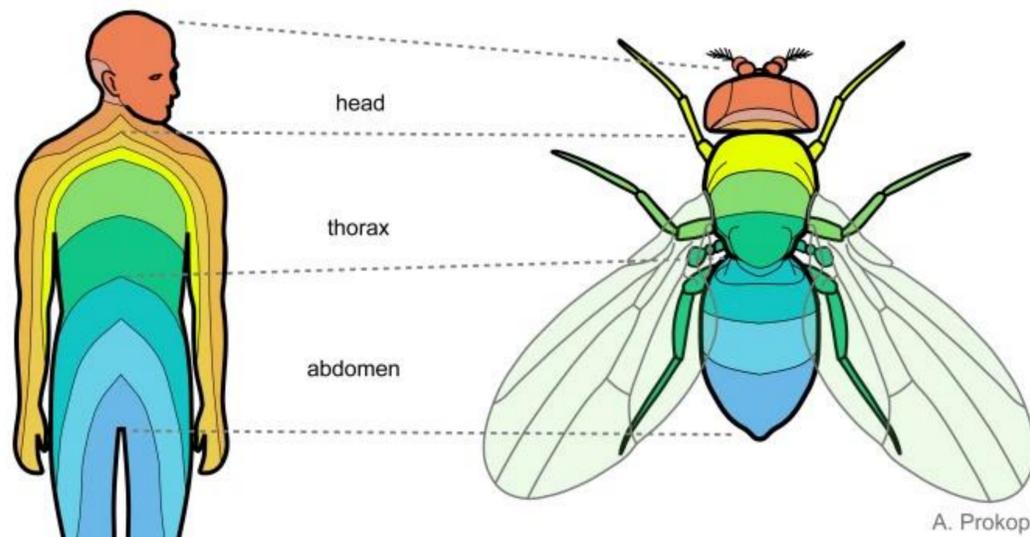


Figure 1. *Eve* homeodomain transcription factor is important in segmentation of embryogenesis.

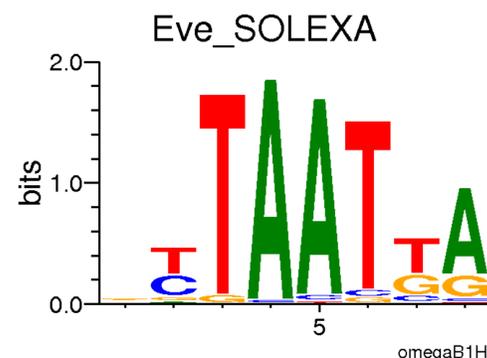


Figure 2. Most common base pair in each binding site position in *Eve*.

Acknowledgements. I'd like to thank Dr. Spratt for his guidance, as well as Sean Munroe, Tyler Vincent, and Mia Advocate for continued support on this project.

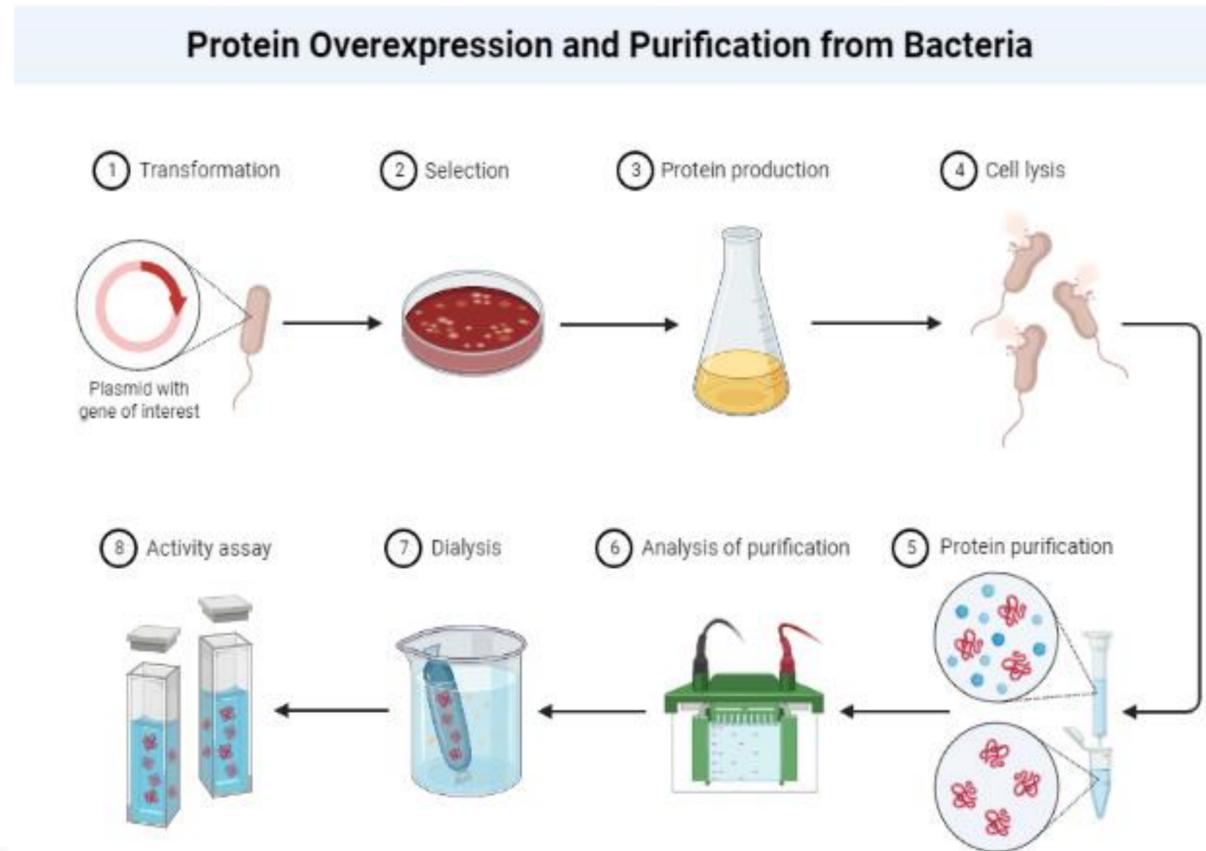


Figure 4. Overview of *Eve* overexpression and purification protocol in *E. coli* cells.



Figure 3. *Eve* HD binding interactions with DNA helix.

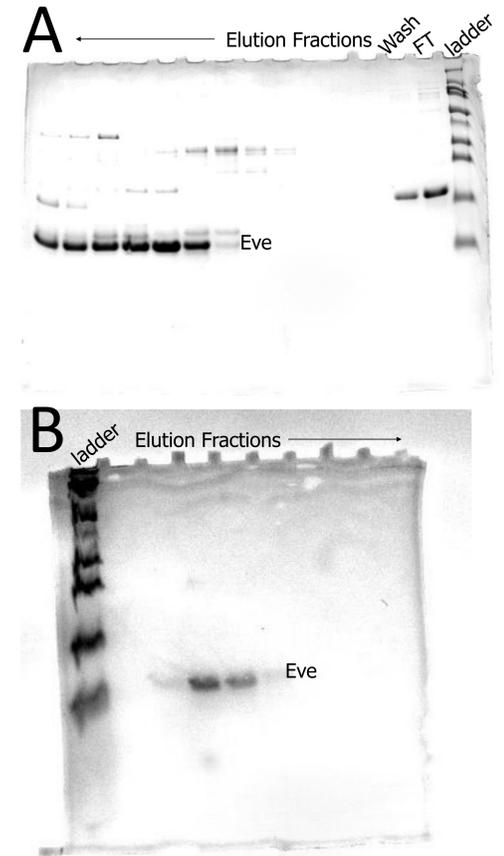


Figure 5. A) Impure protein sample following nickel and heparin affinity chromatography. B) Pure *eve* protein achieved following size exclusion chromatography.

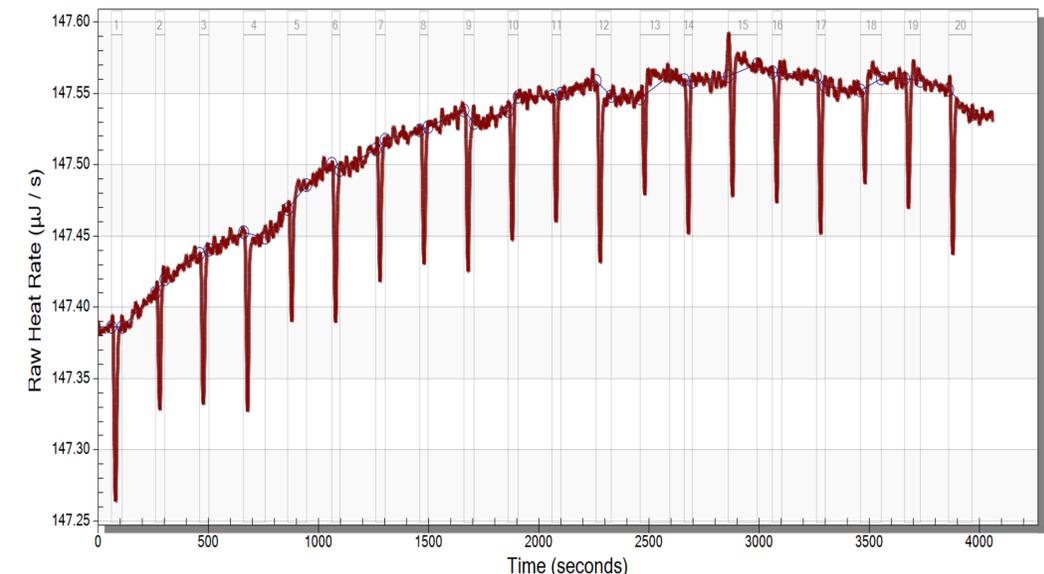


Figure 6. Future work will center around Isothermal Titration Calorimetry (ITC) and NMR resonance spectroscopy.