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Proposal Draft

# Binding affinity between HP1-HOAP interacting protein (HipHop) and telomere maintenance proteins Modigliani & Verrocchio in *drosophila melanogaster* through co-immunoprecipitation and Western blot analysis*.*

I. Introduction

Telomeres are repetitive nucleotide sequences located at the end of our chromosomes used as protection from chromosome fusion and general maintenance of said chromosomes. In humans the protein complex shelterin is used as protection against double-strand breaks in our DNA. Without these extraordinary sequences humans may undergo chromosome fusion, which can lead to many dangerous and cancerous outcomes (Cenci et al. 2002). The function of telomere capping in vertebrates and invertebrates is important for protection from chromosome fusion. Raffa et al (2013) have listed terminin and non-terminin genes that encode proteins for telomere capping/protection. HipHop is a protein that is needed in *drosophila* for the prevention of telomere fusion (Gao et al. 2010). Telomere maintenance proteins HipHop, HOAP, Verrocchio (ver), and Modigliani (moi) show signs of interaction through various studies (Raffa et al. 2013).

These proteins listed are known as terminin, which is a protein complex. These proteins are known to be rich in content specifically at *drosophila* telomeres (Raffa et al. 2013). Little research has been done to examine binding affinity between these terminin proteins, and it is also presently unknown if the HipHop protein interacts with moi and ver in *drosophila* telomeres. Raffa et al. (2013) speculate that *drosophila* telomere analysis is an extraordinary gateway to uncovering viable components and information regarding telomere protection and maintenance in human counterparts.

II. Experiment

The goal of this experiment is to test the expression of interaction between terminin proteins moi and ver and their binding affinity to the HP1-HOAP interacting protein (HipHop). To do so I will be using the Protein complex immunoprecipitation method (Co-IP). Co-IP is a method that isolates targeted protein complexes for analysis. I will be using the Co-IP method to isolate the terminin complex that includes HipHop, ver, and moi. Takada et al. (2000) used the immunoprecipitation method to discover any association between the polymerase III transcription factor BRF and TBP or TRF1 in *drosophila*. Takada, as well as other microbiology articles regarding immunoprecipitation in *drosophila* inspired the current Co-IP experiment. By using a specific antibody against the protein complex of interest (terminin) I will conduct an experiment using the Co-IP protocol.

The specific antibody will bind to the targeted protein complex (terminin) and will separate from the remaining proteins in the sample mixture. To test which proteins have a strong binding affinity with each other within the complex I will be using the western blot process to test binding affinity and protein-protein interaction between moi-ver and HipHop in the newly formed and extracted complex-antibody combination.

III. Discussion

If the experiment goes as planned I will expect to see high binding affinity between moi and HipHop. I’m unsure if ver will have much interaction with HipHop. I will also expect see little to no binding affinity between ver and HipHop due to the data analysis from Raffa et al. (2013) stating that HOAP and moi bind to Heterochromatin Protein 1 (HP1), but ver does not. HP1 is useful in *drosophila* as a transcription factor and heterochromatin regulation. On the other side of the same token, it can be speculated that ver may very well have a high binding affinity to HipHop due to all of these terminin proteins being localized exclusively at *drosophila* telomeres (Raffa et. al 2013). This leads me to believe that although ver may not bind to HP1, it must have some slight expression in the binding affinity with HipHop due to the concentration of location and exclusiveness of said proteins (Raffa et. al 2013).

Works Cited

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