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# Investigating Chromatin Interactions with Mediator Subunits through Chromatin Immunoprecipitation (ChIP)

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## **Investigating chromatin interactions with Mediator subunits through Chromatin Immunoprecipitation (ChIP)**

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Human Adipose-Derived Stem Cells (hASCs) are multipotent stem cells with the potential to self-renew, differentiate, and suppress inflammation. As hASCs continue to be tested in clinical trials for their therapeutic potential, scientists seek a more complete understanding of how stem cell state is maintained and how cell fate is determined. The behavior of hASCs is determined by signaling pathways, transcription factors, co-factors, and the gene expression profile that they regulate. The kinase domain of the Mediator complex is a critical regulatory element responsible for activating cell-type specific transcription factors that control gene expression. Through these mechanistic interactions, we aim to further investigate Mediator's potential role in transcriptional regulation of hASCs. Chromatin Immunoprecipitation (ChIP) is a multi-step process that isolates and enhances the genomic regions upon which transcription factors and other proteins bind. Each step of the ChIP protocol requires optimization and step-by-step validation to confirm interaction of target proteins with specific regions of the genome; specifically, genomic regulatory regions where Mediator is responsible for directing transcription. Further optimization must be conducted in order to determine the efficiency of binding and the location within the genome that the kinase domain interacts with. Our goal in this research is to better understand what role the Mediator kinase domain has in stem cell regulation. This information contributes towards our larger body of understanding on how stem cell's differentiate into distinct lineages, bringing us closer to a well-controlled stem cell therapy.