

## **Comparison of the Human and Mouse NF-kappaB Heterodimer Proteins**

Grace Kim, Isabelle Velasco, Monathysak Keo & Hannah Baughman

NFκB is a transcriptional factor that responds to strong inflammatory signals *in vivo*. Previous research utilized mouse proteins to investigate the biochemical mechanisms of the p50/RelA heterodimer of NFκB. The goal of the research conducted within this study was to compare p50/RelA heterodimers within human protein sequences, utilizing mouse proteins as a positive control. Doing so provides insight into the role of NFκB within the human body system and its potential impacts on human health. Over the course of three weeks, human and mouse plasmids were transformed utilizing competent *E. coli* cells, then protein expression was induced before being lysed, run through Nickel Affinity Chromatography and placed in a dialysis chamber. The samples collected throughout the process then underwent SDS-PAGE analysis to determine the presence of the p50/RelA heterodimers. Upon analysis, we hypothesized that increasing the concentration of Imidazole within the wash buffer utilized for Nickel Affinity Chromatography would increase the purity of the final SDS-PAGE results. The previous procedures were then implemented for the following three weeks, with the Imidazole concentration of the wash buffer increased from 20mM to 35mM. The increased Imidazole concentration did not successfully produce a higher level of purity within the SDS-PAGE results, making identification of the RelA and p50 heterodimers within the human sample harder than previous procedures. Further optimization of the purification protocol will enable future biochemical experiments investigating these target proteins.