The Identification and Characterization of PRDM1 Co-factors in HEK Cells

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Systemic Lupus Erythematosus is a multifactorial autoimmune disease. SLE is difficult to treat due to its various causes and progressions. The PRDM1 pathway has previously been implicated in SLE, it is a significant pathway because it is common to all patients and occurs early in the disease cascade. PRDM1 encodes the Blimp-1 protein which is implicit in B cell maturation and gene regulation. Blimp-1 is dependent on a largely undiscovered co-factor network. A recent study conducted by the lab identified Blimp-1 candidate co-factors. TP53BP1 was selected from the identified candidates on the basis of its previous clinical relevance. The lesser known HNRPM gene was also selected from this gene pool. To prepare for protein binding analysis the two target genes were cloned using the Blimp-1 expression vector, then transfected into a HEK 293 cell line using lipofectamine. Samples underwent a PLA assay before being imaged to view protein binding. The presence of red binding signals in the PRDM1/TP53BP1 and PRDM1/HNRPM samples confirmed the binding of both target molecules to Blimp-1. CoIP assay was also carried out to confirm protein binding. CoIP results were viewed on a western blot, showing the successful pull-down of Blimp-1 and HNRPM. Functional assays can be conducted in the future to assess their biological impact in terms of symptom progression and inflammation. By identifying which co-factors interactions are pathogenic and which are necessary to support metabolic processes, pinpointed gene therapies can be engineered for the treatment of SLE.