Instituto de Medicina Molecular

**Master Project Proposal**

**Title: The Role of NRARP in T-cell Acute Lymphoblastic Leukemia**

**Synopsis:** T-cell acute lymphoblastic leukemia (T-ALL) is an aggressive hematological malignancy that accounts for 15% and 25% of pediatric and adult acute lymphoblastic leukemia, respectively. Although the outcome of T-ALL patients has improved over recent years, the high frequency of relapse is an important clinical problem. Moreover, patients with relapsed disease often develop resistance to chemotherapy and have an extremely poor prognosis. Thus, the current relapse rates and the high toxicity of the existing therapies underline the need to develop more specific and effective therapeutic strategies.

Although NOTCH is a known driver in T-ALL development and maintenance, its inhibition cannot be efficiently achieved with the drugs currently available, due to their weak therapeutic effects and severe toxicity. Thus, a better understanding of the mechanisms downstream of NOTCH oncogenic signaling in T-ALL is necessary in order to develop NOTCH targeting therapies more effective and less toxic.

We have shown that loss of *mir-181ab1* blocks T-ALL development induced by the human NOTCH oncogenes partly by de-repressing the expression of NRARP, a transcriptional target of NOTCH and a negative regulator of NOTCH signaling (1). Interestingly, *Nrarp* over-expression in murine hematopoietic stem/progenitor cells impairs T-cell lineage commitment and early thymocyte differentiation. Since T-ALL originates from a block during T-cell development, these results suggest that de-regulation of NRARP expression can contribute to the pathogenesis of T-ALL. Importantly, and consistent with a role in T-ALL pathogenesis, we find NRARP expression de-regulated in T-ALL cells. Moreover, DNA sequencing data of T-ALL cells show alterations in the NRARP gene. All together, these data suggest that NRARP de-regulated or mutated expression may impair its function as a negative regulator of NOTCH signaling, contributing to T-ALL development. Therefore, NRARP may represent a new therapeutic approach to target NOTCH oncogenic signaling in T-ALL.

Therefore, using in vitro cellular assays we will investigate the role of NRARP in T-ALL pathogenesis and its therapeutic potential to target NOTCH oncogenic signals. In particular, the candidate will modulate NRARP expression in T-ALL cell lines (by lentiral transduction) and evaluate the functional effects of NRARP expression in T-ALL cells proliferation and viability (by flow cytometry analyses) and in NOTCH signaling activation (by real-time PCR and western blot analysis of NOTCH and downstream targets of NOTCH). The candidate will further evaluate the impact of NRARP expression modulation in other T-ALL / NOTCH-related pathways such as the WNT signaling pathway.

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**Bibliography:** R. Fragoso *et al.*, Modulating the strength and threshold of NOTCH oncogenic signals by mir-181a-1/b-1. *PLoS genetics* **8**, e1002855 (2012).

**Remunerated or volunteer training:** Volunteer training