MELANOMA AND MELANOCYTIC NAEVI

DISCOVERING ESSENTIAL AND FITNESS GENES IN MELANOMA USING A GENOME-SCALE CRISPR-CAS9 SCREEN

Anthony B Mak [1] - Amy HY Tong [2] - Katherine Chan [2] - Andrea Habsid [2] - Aurelio Lorico [3] - Jason Moffat [2]

University of Toronto, Dermatology, Toronto, Canada [1] - University of Toronto, Donnelly Centre for Cellular and Biomolecular Research, Toronto, Canada [2] - Roseman University of Health Sciences, Pathology, Las Vegas, United States [3]

Background: Current treatment options for patients with metastatic melanoma include targeting the mitogen-activated protein kinase (MAPK) pathway and checkpoint proteins. However, given the cases of poor response, the high rates of therapeutic resistance seen in MAPK signaling inhibitors, and the toxicities in check point immunotherapies, many patients with melanoma continue to have dismal outcomes in the form of relapses and metastases.

Objective: CRISPR-Cas9 technology has emerged as an efficient and targeted editing approach to knockout genes in order to study their function. We are using pooled genome-scale CRISPR-Cas9 screens to comprehensively survey genes involved in melanoma cell proliferation and survival (also known as fitness or essential genes, respectively).

Materials and Methods: We have established a high-throughput human genome editing platform based on state-of-the-art CRISPR-Cas9 library development, lentiviral pooled screening approaches and novel analytics. Using our highly optimized second generation lentiviral CRISPR-Cas9 library, which contains 71,090 guide RNAs (gRNAs) targeting 18,053 genes in the human genome, we transduced multiple melanoma lines of interest. Sample populations were harvested at various time points and gRNA abundance was determined by PCR amplification of genomic DNA followed by deep sequencing. The relative representation of gRNAs and their corresponding genes across different samples were ranked based on an algorithmic scoring system

Results: We have identified a significantly higher number of essential and fitness genes in melanoma than previous studies. Bioinformatic analyses revealed that a set of core fitness genes are required across multiple melanoma cell lines, which also overlaps significantly with other cancer types that we have studied.

Conclusions: This research project has identified fitness and essential genes in melanoma that have the potential to serve as molecular targets for melanoma treatment.