Exploring new roles for ADAR1 in cancer; generation of novel neoantigens, links with p53 and the effects of cancer treatment on ADAR1 activity

Reasons why did we choose this topic:

RNA editing by ADAR1 is among the 170 types of RNA modifications and it can generate different protein isoforms. It is one of the best studied and most widespread type of RNA modification and has been observed to increase in cancer. ADAR1 deaminates adenosines that are present in double-stranded (ds) RNA hairpins to inosines. This RNA editing mechanism plays an essential role in discrimination self and non-self RNA such as viruses; inosine in dsRNA is recognized as 'self' by innate immune dsRNA sensors. Thus, when cells lack ADAR1, non-edited dsRNAs present in cells are recognized as viral genetic material. This launches an innate immune response, which when chronic can kill the cell. Lack of ADAR1 can also result in an increase in the expression of endogenous retro-elements as well as changes in splicing patterns. On the other hand, when ADAR1 is highly expressed as seen in many cancers or induced by interferon (IFN), translation of transcripts with aberrantly high levels of editing could lead to re-wiring of the mutant cancer proteome as well as the production of neo-peptides expressed by the major histocompatibility complex (MHC) Class I system. These, if presented on the cell surface, can attract CD8+ T cells. Thus, either the absence of or increased expression of ADAR1 could result in novel ADAR1-dependent neoantigens which could potentially be used as biomarkers in cancer or as therapeutic vaccines. Currently, the total source of neoantigens are not defined; they can come from exons, introns, proteasome splicing, and post translational modifications. We plan to analyze the effects of RNA editing on a global map of neoantigen source; with a specific focus on the contribution by ADAR1 editing.

Aim of the project:

The experiments we plan will first of all define the global RNA editing targets that are ADAR1-dependent. In addition, the data will be used to define the immunopeptidome that is derived from RNA editing in a subset of samples. This data will be extremely informative as it is currently unknown if novel neopeptides generated due to either the excess or lack of RNA editing are presented as neoantigens. Secondly, we will determine which RNA editing events are p53-dependent since this tumor suppressor is a major sensor of altered stressed states of a cell. Finally, we will define ADAR dependent RNA editing changes that occur as a result of exposure to therapeutic DNA damaging treatments. This is an important question as variability in RNA editing after DNA damaging cancer treatments or chromosomal instability due to loss of p53 during cancer evolution would in turn alter neoantigen production that could have a heterogenous effect on anti-tumor immunity.

Research description:

During the first year of the project, the panel of 8 cell lines with ADAR1 high and low level will be generated. These will be processed for RNA-Seq. Once data from the RNA seq is available then we will focus on this. Cells will also be generated for MS and analyzed for neopeptides generated by RNA editing (AIM 1). During the second year, the p53 cell lines will be generated and sent for RNA-Seq and then analyze the data (AIM 2). During the third year, the panel of cell lines will be treated with cisplatin and ionizing radiation. These cells will then be processed for RNA-Seq and the analysis will be performed (AIM 3). Of the key cell models (ADAR high/low), finally, one time point and DNA damaging dose in this subset will be chosen for measuring changes in RNA editing levels.

Most important results we expect to obtain:

We will test the theory that alterations of ADAR1 RNA editing leads to changes in the presentation of tumor neoantigens. Moreover, we will possess the knowledge on the ADAR dependent RNA editing changes that occur as a result of exposure to therapeutic DNA damaging treatments and how p53 gene status impacts on RNA editing landscapes.