IDENTIFICATION OF NOVEL VULNERABILITIES OF VPS4B-DEFICIENT CANCER CELLS

One of the key challenges in oncology is to effectively kill cancer cells while leaving healthy cells intact. To meet this goal, precision oncology aims to tailor anti-tumor therapies to individual genetic changes in cancer cells of a given patient. Mutations that enable cancer cell growth often confer specific vulnerabilities that normal cells lack. For example, loss of a chromosome arm bearing a tumor suppressor (a gene inhibiting tumor cell growth) allows cancer cells to hyperproliferate, but at the same time it often results in unintended loss of another neighboring gene, e.g. encoding a protein X participating in an important cellular process. In such case, cancer cell survival may depend on a product of gene Y that compensates for the lost function of X. Such situation (loss of X) creates a weakness of a cancer cell and its specific dependency on Y. Then, if a selective inhibitor of Y protein product is available, this vulnerability might be exploited therapeutically. Therefore, to provide new targets for precision medicine we need to understand the relationship between the genetic alterations of cancer cells and the dependencies (vulnerabilities) they cause.

The reason for many failures of anti-tumor therapies are mutations in cancer cells leading to their resistance to apoptosis – a type of programmed cell death that normally protects the organism from cancer. However, apoptosis is not the only kind of cell death and activation of other programmed cell death types is an important alternative for killing apoptosis-resistant cancer cells.

VPS4A and VPS4B enzymes together with the Endosomal Sorting Complex Required for Transport (ESCRT) machinery are involved in membrane remodeling during e.g. endocytosis, cell division, and plasma membrane repair. In my previous project, we identified VPS4B deficiency as a selective weakness of colorectal cancers (CRC) with chromosome 18q deletion. We also demonstrated that survival of VPS4B-depleted cancer cells depends on the presence of VPS4A and characterized the molecular consequences of simultaneous depletion of VPS4 proteins that lead to cell death.

Very probably, VPS4B deficiency makes cancer cells more vulnerable not only to VPS4A loss but also to other perturbations affecting gene(s) cooperating with VPS4B in cellular processes essential for life. Based on our published results, new preliminary data and analysis of the Cancer Dependency Map Project (DepMap) datasets (Broad Institute, USA), we hypothesize that: i) VPS4B may cooperate with several gene products involved in motility of cells in an organism and/or intracellular vesicle trafficking, therefore VPS4B-depleted cancer cells might be selectively vulnerable to perturbation of expression of these genes; ii) VPS4B may have a protective role against non-apoptotic cell death, therefore cancer-associated VPS4B depletion might selectively sensitize cells to treatment with drugs inducing this cell death type.

The general aim of the project is to identify novel vulnerabilities of VPS4B-depleted cancer cells. To reach this goal we plan to use two approaches, in which we will verify the vulnerability of VPS4B-deficient cancer cells to: 1) inhibition of expression of candidate genes selected based on the DepMap analysis, 2) treatment by drugs inducing non-apoptotic cell death.

For the analyses within the first approach, we prioritized candidates whose protein structures have ability to bind drugs (thus are “druggable”). Using CRC and pancreatic cancer cell models grown *in vitro* and xenografted to mice, we will verify whether the selected candidates inhibit cancer cell growth when perturbed simultaneously with VPS4B. Additionally, to unravel the molecular mechanisms underlying vulnerability of VPS4B-depleted cancer cells to the candidate activity perturbation, we will assess whether there is a cooperation between VPS4B and a candidate in mediating selected cancer-relevant processes, like endocytosis, cell division (cytokinesis), cell migration and adhesion (attachment). In the second approach, we will verify whether VPS4B depletion sensitizes cancer cells to treatment with drugs inducing non-apoptotic cell death and we will investigate the molecular mechanisms underlying this vulnerability. Finally, we will perform microscopy-based immunohistochemical analysis of CRC samples from patients to evaluate whether VPS4B abundance correlates with the levels of proteins involved in non-apoptotic cell death.

The findings of this project should uncover molecular mechanisms underlying cancer cell vulnerabilities and provide novel targets for precision oncology, which is of great interest for both basic and translational research.