ID: 03339

Topic: DRUG TOLERANCE

Unveiling the role of BRCA2 in prostate cancer progression and resistance to therapy

Lidia Ramírez[1] , Marta Barber[1] , Ana Sarrias[1] , David Casajuana[1] , Juan Morote[1,2] , Olga Méndez[1]

1) Biomedical reasearch in Urology, Vall Hebron Research Institute (VHIR), Universitat Autònoma de Barcelona (UAB), Barcelona, Spain 2) Department of Urology, Vall d´Hebron Hospital, Barcelona, Spain.

Introduction: Prostate cancer (PCa) is the solid tumor most frequently diagnosed in adult men and the third leading cause of cancer death in this population. Alterations of genes associated with DNA damage response (DDR) are enriched in patients with metastatic castration-resistant PCa (mCRPC), being mutations in BRCA2 the most frequently detected. Patients with localized and locally advanced PCa with BRCA2 germline mutations experience more rapid progression to metastatic spread, suggesting a more aggressive phenotype.

Treatment options for patients with advanced PCa have largely focused on the pillars of hormonal blockade and chemotherapy. However, the recent discovery that a significant proportion of patients with advanced PCa, carry or develop alterations in DDR proteins has uncovered a new potential therapeutic area. Tumors that harbor DDR defects, particularly BRCA defects, are sensitive to PARP inhibitors (PARPi) through a synthetic lethality mechanism.

BRCA2 mutation carriers become resistant to androgen deprivation therapy faster than non-carriers. However, the mechanisms by which loss of BRCA2 might promote aggressive PCa and confer resistance, faster than non-carriers, to the therapeutic options available nowadays, including PARPi, are not clear. We proposed the proteome interrogation as a tool to get deeper knowledge of BRCA2 role in PCa. The analysis of the secreted fraction of the proteome from cells with BRCA2 mutations, treated with the PARPi Olaparib, will allow us to better understand the changes that are occurring at the molecular level, and unveil potential new biomarkers of response and resistance. The analysis of the secretome is especially relevant when working with cancer cells because tumoral cells need to interact with its surrounding microenvironment in order to adapt and be able to survive and grow in the organ of origin but also in distant places. Cancer cells interact with the surrounding tissue, through the secretion of different factors such as cytokines, growth factors and exosomes among others, facilitating the crosstalk between tumoral cells and the surrounding microenvironment. One characteristic especially relevant of the tumor microenvironment is the oxygen level. All solid cancers contain regions with very low cellular oxygen concentrations. Hypoxia in cancer cells is associated with DNA damage and metastasis, and tumors with constant high levels of hypoxia have a poor prognosis.

Altogether, the proteomic analysis of secretomes derived from PCa cells treated with the current therapeutic approaches for PCa patients will allow us to get deeper comprehension on how resistance mechanisms work in the context of DDR mutations.

Objectives: Advance in the understanding of BRCA2-prostate tumor's physiology and unveil the mechanisms of resistance to current therapies in the context of mutations in DDR genes.

Methods: Ø Generation of 22RV1 (PCa cell line with BRCA2 mutations) resistant to Olaparib (OLAP-R) by treating cells overtime with increasing doses of the drug until reaching 20mM. Ø Cellular and molecular characterization of OLAP-R cells in comparison with parental cells by analyzing: proliferation, migration, colony formation capacity and response to chemotherapeutic agents such as cisplatin. Ø Study the effect of hypoxia in the biological response of OLAP-R cells and the corresponding control cells. Ø Set up of the optimal conditions to obtained secretomes from OLAP-R cells and the corresponding control cells, in both normoxic and hypoxic conditions. Ø Proteomic interrogation of PCa cells with BRCA2 mutations resistant to PARPi (on going).

Results: Our results indicate that OLAP-R cells migrate more, have more capacity to form colonies and respond worse to the use of other therapeutic options such as cisplatin. Our preliminary results also indicate that under hypoxic conditions OLAP-R cells have higher proliferative and migratory capacity than the corresponding control cells and also respond worse to treatments such as cisplatin. The proteomic analysis is not yet finished but we expect to find some differentially secreted proteins that might help us to unveil potential new biomarkers of response and resistance.

Conclusions: OLAP-R cells are more malignant than their control counterparts. Acquired resistance to Olaparib results in higher migratory capacity, higher ability to form colonies and also worse response to other therapeutic options such as cisplatin. Moreover hypoxia, one key aspect of tumor microenvironment enhances the migratory capacity of OLAP-R cells and reduces the biological effect of other treatments such as cisplatin.