Establishment and Characterisation of a Preclinical Bladder Model of Resistance to Immune Checkpoint Inhibitors

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Immune checkpoint inhibitors (ICI), such as anti-PD1 or anti-PDL1 antibodies, are one of the most important recent breakthroughs in oncology. These molecules have a double originality: on one hand they stimulate the patient’s immune system antitumor activity, and on the other they are not specific of a tumor antigen and therefore can be efficient in multiple types of cancer. Unfortunately, resistance to these therapies has increasingly been observed and needs to be better understood.

In the present project, to study mechanisms of adaptive resistance, we developed an in vivo syngeneic model of bladder cancer resistant to anti-PD1 and anti-PDL1 therapies. This model was developed using the syngeneic MB49 cell line, grown in fully immunocompetent C57Bl/6 mice, exposed weekly to anti-PD1 or anti-PDL1 at at dose of 12 mg/kg, followed by serial reimplantation and reexposure to anti-PD1 or anti-PDL1, until a resistant phenotype was obtained. Following five serial reimplantation and reexposure cycles, the results showed no effect of antibody therapy on tumor growth in resistant tumors, in comparison with the unexposed group.

We then analysed the tumor immune microenvironment by flow cytometry, in both the sensitive and resistant phenotypes, at day 4 after the first anti-PD1 or anti-PDL1 administration in mice bearing established tumors. In resistant tumors (n=6), we first observed a decrease of the total leukocytic infiltrate, defined by expression of CD45. We also observed an increase of M2 immunosuppressive cells subpopulation, as well as an increase of M-MDSC. These findings suggest that altered expression of alternative checkpoints may be associated with adaptive resistance to PD1 or PDL-1 blockade.

To go further, we will analyse whether other immune checkpoints expressions such as TIM3 or LAG3 are upregulated. If so, these immune checkpoints will be blockade in combination with Anti-PD-1 or Anti-PD-L1 in vivo, to highlight potential cross-resistance or sensitivity on resistant tumors.

Another part of this work is to assess the efficacy of immune checkpoint antibodies in combination with standard chemotherapy MVAC in the WT model of bladder, in order to improve the antitumor effects of monoclonal a-PD1 and a-PDL-1 in vivo. As a result we observed a synergistic effect of MVAC with a-PDL-1. Immune infiltrate characterization by flow cytometry showed an increase of CD45+ total cells in the combination group MVAC + a
PD1, and a decrease of Treg cells with MVAC + a PDL1. We also describe a large decrease of TIGIT+ tumor cells in groups comprising chemotherapies.