ImmunoTools *special* Award 2020

Marc Bach Griera, PhD-student

Supervisor: Dr. Esther Julián Gómez

Department of Genetics and Microbiology
Autonomous University of Barcelona
Cerdanyola del Vallès, Spain

Manipulating tumor-associated macrophages through mycobacteria infection

Bladder cancer (BC) is the ninth most common type of cancer worldwide. Concretely, non-muscle invasive bladder cancer (NMIBC) represent the most frequent form of BC with around 80% of total diagnosis. After being diagnosed, transurethral resection of bladder tumor followed by weekly intravesical instillations of the bacterium *Mycobacterium bovis* Bacillus Calmette-Guérin (BCG), represent the gold standard treatment for NMIBC. Despite BCG benefits in the control of recurrence and progression of the disease, this treatment present side effects. Approximately 50% of patients suffer from non-severe side effects, while 1 to 5% of patients suffer from severe BCG-related events, including some cases of BCG infections. To avoid the possibility of BCG infection, different strategies involving mycobacteria or mycobacteria-derived agents are currently under investigation. *Mycobacterium brumae*, a nontuberculous mycobacterium, represent one of these alternatives. *M. brumae* has shown outstanding antitumoral and immunomodulatory capabilities in previous *in vitro*, *ex vivo* and *in vivo* experiments (Noguera-Ortega, 2016).

Although the precise mechanism of action of mycobacteria is not completely known, several studies have highlighted their capacity to modulate the tumor microenvironment (TME), a complex network of cellular and noncellular components which together promote the tumor progression and dissemination. Both mycobacteria, BCG and *M. brumae*, interact with BC cells and inhibit their proliferation. But, most relevant, mycobacteria are able to trigger an immune response both locally inside the bladder cavity and at systemic level (Noguera-Ortega, 2018). Therefore, we aimed to identify the precise role of mycobacteria in

TME, and specifically in this project their ability to modulate tumor associated macrophages (TAMs).

Our project consists to stablish different sets of macrophages: M1 macrophages with described antitumoral activity and M2 macrophages that are found in TME and are closely related with tumorigenesis. Following, the obtained macrophages will be infected with mycobacteria, aiming to analyse the possible capacity of both mycobacteria to polarize these cells.

In order to elucidate the macrophage polarization, we will combine two different techniques. On the one hand, we will use flow cytometry to assess the expression of M1 markers and M2 markers. Concretely, antibodies against CD14, CD80, HLA-DR and CD86 surface cell markers. On the other hand, we will evaluate cytokine secretion of th1 cytokines (M1) and th2 cytokines (M2) by ELISA assays. In this case, we will detect the production of IL-6, IL12-p40, MIP-4/CCL-18 and IL-10.

Thus, the **ImmunoTools** award will give us the opportunity to perform this project, elucidating the contribution of BCG and the promising *M. brumae* in the TME describing the role of mycobacteria in macrophages polarization. This work will provide new information about the immunomodulatory and antitumoral mechanisms of mycobacteria.

ImmunoTools *special* AWARD for **Marc Bach Griera** includes 23 reagents:

FITC - conjugated anti-human CD14, IgG1
PE - conjugated anti-human CD80, IgG1
PerCP - conjugated anti-human HLA-DR, IgG1
APC - conjugated anti-human CD86, IgG1

Human ELISA-sets: IL12-p40, MIP-4/CCL-18 and IL-10.

Recombinant human cytokines: IL-4, IL-10 and IL-13 .

DETAILS more AWARDS

References

1. Noguera-Ortega E., Secanella-Fandos S., Eraña H., Gasión J., Rabanal R.M., Luquin M., Torrents E., and Julián E. (2016) The non-pathogenic Mycobacterium brumae inhibits bladder cancer growth in vitro, ex vivo, and in vivo. European Urology FOCUS. 2(1): 67-76. DOI: 10.1016/j.euf.2015.03.003.
2. Noguera-Ortega E., Rabanal RM., Gómez-Mora E., Cabrera C., Luquin M., and Julián E. (2018) Intravesical Mycobacterium brumae triggers both local and systemic immunotherapeutic response against bladder cancer in mice. Scientific Reports. Oct 10;8(1):15102. DOI: 10.1038/s41598-018-33253-w