ImmunoTools *special* Award 2014

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Anti-inflammatory Role of Bone Marrow Mesenchymal Stem Cells Microvesicles in Murine Models of Brain Disease

My PhD project is focused on the study of the inflammatory processes occurring during the initiation and progression of some neurodegenerative diseases such as Alzheimer's disease and Hypoxia and I aim at assessing whether treatments with Microvesicles derived from Bone Marrow Mesenchymal Stem Cells are capable of reducing or modulating neuro- inflammation.

The rationale for this investigation is based on the evidence that several neurodegenerative diseases are associated with significant increase of the inflammatory load and on the recent studies showing Bone Marrow Mesenchymal Stem Cells (BM- MSCs) were successfully used as a new therapeutic approach for the treatment of Multiple Sclerosis, other neurodegenerative disease characterized by a strong inflammatory component. BM-MSCs, on the other hand, are known as releasing cells, and its shedding microvesicles (MVs), containing mRNA or microRNA mainly, mediate many anti-inflammatory effects.

In Alzheimer and Hypoxia diseases are acting different cytokines as TNFα, usually initiator cytokine of the cascade in the inflammatory process, IL-6, IL-1beta; in particular IL1beta is the cytokine that up regulate many processes of inflammation maintaining damage conditions in Alzheimer's disease; in Hypoxia another involved cytokine is INFgamma.

First, to characterize BM-MSCs population I choose some markers indicated in literature as stemness and hematopoietic markers: CD 90, CD73 and CD105, for which cell population have to be positive to indicate specific stemness of BM- MSCs; CD19, CD45 and CD 11b will be negative in my population to own a pure BM- MSCs culture.

I'll perform this characterization with FACS, so I could use ImmunoTools anti-mouse antibodies for flow cytometry (FITC - conjugated anti-mouse CD11b, CD19,CD45, or PE - conjugated anti-mouse CD11b, CD19, or APC - conjugated anti-mouse CD11b, CD19, CD45). I want to characterize BM-MSC MVs, and I'll search on them for some of previous cell markers (FITC - conjugated anti-mouse CD11b, CD19,CD45, or PE - conjugated anti-mouse CD11b, CD19, or APC - conjugated anti-mouse CD11b, CD19, CD45). In addiction I'll research other markers as CD9 and VLA4, and Sca1, which presence is knowing on cell shedding MVs.

In order to study anti- inflammatory effects of MVs BM- MSCs derived, I will perform in vitro experiments on neuron and microglial cultures administering inflammatory cytokines (rm TNFα, rm IL-6, rm IL-1beta, rm IFNgamma, ImmunoTools recombinant

mouse cytokines) to them and I'll study possible alterations of dendritic spine morphology and changes in pre and post-synaptic markers in the different in vitro models.

Progressively, in order to access what anti-inflammatory cytokines are produced after the treatments, I will study the release of inflammatory and anti-inflammatory cytokines from different cell types in culture medium by ELISA assay.

Moreover, it is interesting administer to BM-MSCs the cytokines involved in inflammatory process, in order to analyze MVs mRNA or microRNA contents changes.

In order to do that I will need a substantial amount of cytokines making this box really valuable. Results from these experiments will tell us whether microvesicles BM-MSCs derived directly impacts in specific inflammatory context and can give results to access experiments in vivo, in murine models of Alzheimer's disease and in murine Hypoxia model.