MOLECULAR EVALUATION OF THE NEUROPROTECTIVE AND REGENERATIVE PROPERTIES OF WJ-MSCS AFTER TRANSPLANTATION INTO INJURED RAT BRAIN

Ischemic strokes result in rapid impairment of tissue homeostasis, leading directly to focal necrosis surrounded by a region of delayed cell degeneration with successive disruption of the proper cellular interactions. There is currently a lack of effective treatments for stroke. Therefore, alternative therapies, including stem cell-based therapy, are required.

Mesenchymal stem cells (MSCs) are able to respond to the surrounding microenvironment by changing the factors secretion profile that affect on the proliferation, differentiation, and maturation of various progenitor cells in the brain. The neuroprotective effect of transplanted MSCs is associated with the secretion of neurotrophins and stimulation of endogenous cells to secretion of those factors.

Currently, when regenerative medicine based on stem cells enters the clinic, there is a need for establishing precise protocols for culturing, isolation, phenotypic verification, and biosafety of the acquired cells, including the enhancement of their therapeutic properties and survival after transplantation. Stem cells are exposed to a strong recipient immune response after transplantation, which can significantly hinder their survival and regeneration of the damaged tissue. Specific carriers or scaffolds, which protect transplanted cells against a reaction from the recipient immune system and enhance the regenerative properties of stem cells, are being developed to overcome this problem.

Accordingly, the main goal of this project is the molecular evaluation of the host nervous tissue response to the transplantation of human stem cells isolated from umbilical cord (WJ-MSCs) in an experimental model of brain injury.

We assume, that WJ-MSCs preconditioned in "physiological normoxia" (5% O_2) (an environment closest to those encountered in their natural niche) and transplanted in 3D hydrogel scaffolds show increased neuroprotective effect and stimulate the paracrine effects of host endogenous tissue, which modulate the inflammatory response and enhance the regenerative process, compared to the WJ-MSCs cultured in 21% O2 and transplanted in resuspension (2D). We want to verify this hypothesis by conducting an experiment where we will evaluate the neuroprotective, regenerative, and immunomodulatory properties of WJ-MSCs cultured in standard conditions (21% O_2) or preconditioned in physiological normoxia and then transplanted into the injured rat brain in resuspension and in 3D hydrogel scaffolds.

We will analyze the effect of WJ-MSCs on the damaged rat brain by determining the recipient's response to the transplant, based on the expression of a panel of pro- and anti-inflammatory cytokines, neurotrophins, and growth factors. We will also analyze the survival and migration ability of the transplanted cells at various time points post-transplantation. The experiments will be conducted on Wistar rats. After transplantation, we will collect animal brains and cerebrospinal fluid for further analysis. The location and migration ability of the transplanted cells will be analyzed by magnetic resonance imaging at various time points post-transplantation.

The novelty of the proposed project is the combination of WJ-MSCs preconditioning by culturing in "physiological normoxia" (5% O_2) with the use of 3D hydrogels for the transplantation of the WJ-MSCs to increase the therapeutic potential of those cells.

The results obtained from this project will help to explain how transplanted cells can potentially promote neuroprotection, modulate the inflammatory response, and stimulate recipient tissue for regeneration. Such studies may allow us to answer the following question: does the application of biomimetic conditions with a restrictive culture system ("physiological normoxia") have a positive effect by increasing the neuroprotective, regenerative, and immunomodulatory properties of WJ-MSCs?

Moreover, answering the above questions will allow the establishment of appropriate protocols for the culturing (biomimetic conditions) and transplantation of stem cells for the development of an innovative advanced therapy medicinal product (ATMP) for the treatment of central nervous system disorders.