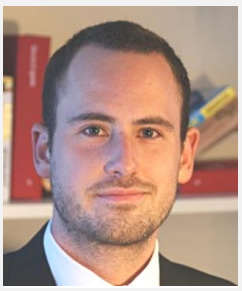
ImmunoTools special Award 2015



Morten Juhl, PhD-student

Supervisor: Dr. Jens Kastrup

Cardiology Stem Cell Centre, University Hospital Rigshospitalet,
Juliane Maries Vej 20, afd. 9302, 2100 Copenhagen E, Denmark

Phenotypic Identity of Immunomodulatory Adipose-tissue derived Stromal Cells

We have recently achieved positive results from clinical trials of treatment of ischemic heart disease using stromal cells from adipose tissue. (Haack-Sørensen et al., 2013; Mathiasen et al., 2012; Qayyum et al., 2012). An interesting attribute of these cells, besides apparent regenerative potential, is the ability to actively suppress immune responses, an attribute with few identified mechanisms of action. As part of ongoing research at the Cardiology Stem Cell Centre, we aim to gain knowledge of how our cell affects immune reactions and how to exploit such potential feats.

Among the first encounter a Adipose-tissue derived Stromal Cell (**ASC**) makes upon entry to donor is most likely the innate immune system. Part of the innate immune system is carried out by complement activation, and common host strategies to counter potential self-harm by members of the complement cascade include a number of surface proteins such as membrane cofactor protein (CD46) and decay-accelerating factor (CD55). Protectin (CD59) expression on mesenchymal stromal cells (**MSCs**) such as ASCs has recently been reported as crucial for impeding formation of membrane attack complexes to an even larger extent (Moll et al., 2011). Together, these surface markers offer some protection against complement deposition and subsequent opsonization or lysis, and can be identified by flow cytometry.

Following complement encounter, cellular immunity is the next central part of the immune system to take into consideration. Serving as an intermediary between innate and adaptive immunity are phagocytosing and antigen-presenting cells. CD9, which is expressed to a varying degree on MSCs, have been demonstrated to affect phagocytosis, inflammation, migration, and cell-cell signalling. Likewise, CD47 is expressed on MSCs and is involved in a plethora of cell processes; for purpose of our studies, phagocytosis, proliferation, and inflammation are the most intriguing associated terms.

Among possible mediators between MSCs and adaptive immunity are numerous factors. Of importance is CD54 expression on ASCs which can potentially affect T-cell differentiation and regulation through hitherto unidentified pathways. The expression of CD39 and CD73 may prove to be of significant importance for the immunosuppressive function of ASCs, as these markers are expressed in tandem on regulatory T cells.

MSCs express high levels of CD10, and CD10 expression on bone marrow stromal cells is believed to dampen B cell maturation, which might also be an advantage of ASC. Other potential markers of interest include CD44, CD50, CD62L, CD63, CD71, HLA-ABC, HLA-DP, and HLA-DR.

Some attributes of immunosuppression can be associated with secreted factors, such as cytokines, chemokines, and growth factors. While the list of identified secreted factors is ever increasing, we plan to focus on a few using flow cytometry-based technologies, but also ELISA analysis for e.g. IL-6. Besides elucidating the factors secreted by ASCs, we also flip the paradigm upside-down and investigate how various cytokines affects function and immunobiology of ASCs. For that purpose IFNα-1b, IL-1β, IL-15, IL-17F, CCL18, RANKL, and TRAIL are of special interest.

In vitro findings may potentiate allogeneic therapeutic use of ASCs for a wide range of diseases, and an ImmunoTools award could be a welcome addition to such experiments.

References:

Haack-Sørensen, M., Friis, T., Mathiasen, A. B., Jørgensen, E., Hansen, L., Dickmeiss, E., … Kastrup, J. (2013). Direct intramyocardial mesenchymal stromal cell injections in patients with severe refractory angina: one-year follow-up. *Cell Transplantation*, *22*(3), 521–8. doi:10.3727/096368912X636830

Mathiasen, A. B., Jørgensen, E., Qayyum, A. A., Haack-Sørensen, M., Ekblond, A., & Kastrup, J. (2012). Rationale and design of the first randomized, double-blind, placebo-controlled trial of intramyocardial injection of autologous bone-marrow derived Mesenchymal Stromal Cells in chronic ischemic Heart Failure (MSC-HF Trial). *American Heart Journal*, *164*(3), 285–91. doi:10.1016/j.ahj.2012.05.026

Moll, G., Jitschin, R., von Bahr, L., Rasmusson-Duprez, I., Sundberg, B., Lönnies, L., … Nilsson, B. (2011). Mesenchymal stromal cells engage complement and complement receptor bearing innate effector cells to modulate immune responses. *PloS One*, *6*(7), e21703. doi:10.1371/journal.pone.0021703

Qayyum, A. A., Haack-Sørensen, M., Mathiasen, A. B., Jørgensen, E., Ekblond, A., & Kastrup, J. (2012). Adipose-derived mesenchymal stromal cells for chronic myocardial ischemia (MyStromalCell Trial): study design. *Regenerative Medicine*, *7*(3), 421–8. doi:10.2217/rme.12.17

ImmunoTools *special* AWARD for **Morten Juhl** includes 25 reagents

FITC - conjugated anti-human CD9, CD46, CD47, CD54, CD55, CD71, HLA-ABC, HLA-DP, HLA-DR,

PE - conjugated anti-human CD44, CD50, CD62L, Annexin V,

APC - conjugated anti-human CD10, CD59, CD63, Annexin V,

recombinant human cytokines: rh IFNa1b, rh IL-1beta /IL-1F2, rh IL-15, rh IL-17F, rh MIP-1α / CCL3, rh MIP-4 / CCL18, rh sRANKL, rh TRAIL / CD253

DETAILS more AWARDS