## ImmunoTools IT-Box-139 Award 2012



## Rocio Ramos Medina

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Evaluation of the effect of intravenous immunoglobulin on gestational success in couples with recurrent reproductive failure associated to expansion of natural killer cells. Identification of new biomarkers.

Recurrent reproductive failure (RRF) is a relevant health problem affecting up to 10% of couples and with increasing incidence worlwide. RRF comprises women with three or more miscarriages of unknown cause and/or who have failed after three in vitro fertilization with transfer of 2 embryos of good quality (Stirrat, G. M. (1990); Margalioth and et al, 2006).). A significant percentage of these patients show immunological abnormalities that could account for failure implantation and miscarriage, being the most significant expansion of circulating natural killer (NK) cells. Indeed, our group has described that NK cells expansion is a independent risk factor of RRF and that could benefit of immunomodulation with intravenous gammaglobulin therapy (IVIg)

The main goal of this project consists of evaluating and comparing the safety and clinical effect of IVIg versus non-IVIg-treated patients withRRF associated with NK cells expansion. Secondly, to identify women's own genetic biomarkers associated with this immunological alteration and RRF. There is a real need to better define biomarkers in this group of patients that could better identify women susceptible of immunomodulatory strategies, with clear impact on their health care.

For this purpose, we will study the peripheral blood (PB) lymphocyte populations, such as NK and NKT cells and effector and regulatory mediators of immunity in a group of healthy women as controls. This point is essential for determining cutoff points in our population of patients.

- Immunophenotyping of subpopulations of NK cells: CD3<sup>-</sup>CD56<sup>bright</sup>CD16<sup>-</sup>, CD3<sup>-</sup>CD56<sup>dim</sup>CD16<sup>+</sup>, CD3<sup>-</sup>CD56<sup>-</sup>CD16<sup>+</sup>, and CD3<sup>+</sup>CD56<sup>+</sup> in day 1 and day 14 of the menstrual cycle

- Study of T CD4<sup>+</sup>, CD8<sup>+</sup> and NK lymphocyte subpopulations present in PB in study groups, in particulary (CD4<sup>+</sup>HLA-DR<sup>-</sup>CD38<sup>+</sup>), naive CD8<sup>+</sup>CD45RO<sup>-</sup>CD27<sup>+</sup>, lymphocytes T CD8<sup>+</sup> memory CD8<sup>+</sup>CD45RO<sup>+</sup>CD27<sup>-</sup>, CD8<sup>+</sup> memory effector (preeffectors) CD8<sup>+</sup> CD45RO<sup>+</sup>CD27<sup>-</sup> and CD8<sup>+</sup> effectors CD8<sup>+</sup>CD45RO<sup>-</sup>CD27<sup>-</sup>. Memory, main memory, and effector.
- Analysis of Th1/Th2/Th17 proinflammatory and anti-inflammatory cytokines and chemokines in serum.

ImmunoTools IT-Box-139 for Rocio Ramos Medina include 100 antibodies
FITC - conjugated anti-human CD1a, CD3, CD4, CD5, CD6, CD7, CD8, CD14, CD15, CD16, CD19, CD21,
CD25, CD29, CD35, CD36, CD41a, CD42b, CD45, CD45RA, CD45RB, CD45RO, CD49d, CD53, CD57, CD61,
CD63, CD80, CD86, HLA-DR, IL-6, Control-IgG1, Control-IgG2a, Control-IgG2b, Annexin V

PE - conjugated anti-human CD3, CD4, CD8, CD11b, CD15, CD14, CD18, CD19, CD20, CD21, CD22, CD31, CD33, CD38, CD40, CD45, CD45RB, CD50, CD52, CD56, CD58, CD62p, CD72, CD95, CD105, CD147, CD177, CD235a, HLA-ABC, IL-6, Control-IgG1, Control-IgG2a, Control-IgG2b, Annexin V

PE/Dy647 -tandem conjugated anti-human CD3, CD4, CD8, CD14, CD19, CD20, CD25, CD54

APC -conjugated anti-human CD2, CD3, CD4, CD8, CD10, CD11a, CD11c, CD14, CD16, CD27, CD37, CD42b, CD44, CD45, CD59, CD62L, CD69, CD71, IL-6, Control-lgG1, Control-lgG2a, Control-lgG2b, Annexin V

**DETAILS**