ImmunoTools *special* Award 2014

Carolina Jancic, PhD

Laboratorio de Inmunidad Innata, IMEX CONICET- Academia Nacional de Medicina, Pacheco de Melo 3081,
C1425 AUM Buenos Aires, Argentina

Regulation of γδ T cell responses by neutrophils: study of the mechanisms involved

γδ T cells constitute a functionally specialized subset of T lymphocytes that play an important role in linking the innate and adaptive immunity. γδ T cells expressing the Vγ9Vδ2 TCR are only found in higher primates and humans. They represent the vast majority of γδ T cells in human peripheral blood. In healthy adults, they comprise about 0.5–5% of circulating T cells, however, the number of Vγ9Vδ2 T cells can dramatically increase during the early response to many viral, bacterial, and parasitic infections, at times comprising up to >50% of all circulating T cells within a few days. Vγ9Vδ2 T cells acquire a preactivated phenotype early in their development allowing the rapid induction of a wide variety of functions upon stimulation in a non-MHC restricted manner by phosphoantigens such as (E)-1-hydroxy-2-methylbut-2-enyl 4-diphosphate (HMBPP). These functions include a cytotoxic response against infected and transformed cells, the production of a range of cytokines and chemokines, the recruitment and the activation of neutrophils, the differentiation of monocytes into a proinflammatory profile, the phenotypic maturation of dendritic cells, the polarization of $CD4^+$ T cells into a Th1 profile, the promotion of B-cell activation, and the presentation of antigenic peptides to both $CD4^+$ and $CD8^+$ T cells.

Other cells that play a critical role in the immune response are neutrophils. They ingest and destroy microbes through the action of a large array of antimicrobial weapons, which include reactive oxygen species, proteolytic enzymes, and antimicrobial proteins. These toxic weapons do not discriminate self from nonself; hence they are also able to induce host tissue damage in different pathologic conditions. Moreover, neutrophils are the first immune cells infiltrating the infected tissues and growing evidence supports a critical role of γδ T cells in the recruitment and activation of neutrophils at the sites of infection and inflammation. γδ T cells can promote the inflammatory activity of neutrophils not only by inducing their activation

but also by providing potent survival signals that rescued them from undergoing apoptosis.

Surprisingly, there were not previous reports showing the role of neutrophils on γδ T cell function. Taken this into account, we decided to analyze whether neutrophils were able to modulate the phenotype and function of human γδ T cells. For this purpose, we isolate human γδ T cells and neutrophils from peripheral blood samples obtain from healthy donors. Then, we evaluate the capacity of neutrophils to modulated γδ T cells in co-cultures. To this aim, we analyze the activation and maturation state of γδ T cells by using specific antibodies (CD25, CD69) and flow cytometry analysis. Additionally, we evaluate the proliferative capacity of γδ T cells in the presence of HMBPP and IL-2, and we measure the production of different cytokines (TNF-α, IFN-γ, IL-6, IL-17A). Our results, demonstrate that neutrophils effectively suppress the activation of γδ T cells stimulated by the phosphoantigen HMBPP. Our data show for the first time the existence of a bidirectional cross-talk between γδ T cells and neutrophils. While γδ T cells promote the recruitment and the activation of neutrophils to fight invading pathogens, the ability of neutrophils to inhibit the activation of γδ T cells might contribute to the resolution of inflammation and the restoration of tissue homeostasis. (Sabbione *et al*. *Eur. J. Immunol*. *2*013).

Currently, we are studying the effect of neutrophils on γδ T cell phenotype and function when γδ T cells are stimulated with agonists other than phosponatigen. In this project, we want to extend the characterization of γδ T cell phenotype by analyzing more surface markers such as CD2, CD27, CD45, CD62L, CD44, CD95, and LFA1; to go further on the description of the maturation state of these cells. As well, we want to analyze the migratory capacity of γδ T cells using CCL17 as stimulus.

Get the ImmunoTools Award will be a great help to our research and will allow us to move forward with the project.

ImmunoTools *special* AWARD for **Carolina Jancic** includes 23 reagents

PE - conjugated anti-human CD11a, CD25, CD27, CD44, CD62L, CD95, CD45RB, Annexin V, IL-6, Control-IgG2a,

PerCP - conjugated anti-human CD3, CD4, CD8, CD20, CD45,

human IL-6 ELISA-set for 96 wells, human TNFa ELISA-set for 96 wells (each 3 reagents),

recombinant human cytokines: rh IL-2, rh TARC DETAILS more AWARDS