ImmunoTools IT-Box-139 Award 2012

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HIV-Host interactions - Cell surface markers modulations by wild type and mutated HIV-1 Nef in primary human haematopoietic cells.

The interactions between host and viruses are dynamic processes in which host immune system tries to combat the infection while virus struggles to survive and spread. Retroviruses including Human Immunodeficiency Virus (HIV) can infect specialized antigen-presenting cells and influence their maturation and functional properties. Multiple mechanisms are used by HIV to interfere with host-cell immune effector functions. HIV-1 Nef protein is required for the maintenance of high viral loads, accelerates AIDS disease progression and $CD4^+$ T cells depletion. Nef expression in T cells leads to CD4 and major histocompatibility complex class I modulation and either enhancement or suppression of T cell activation. Nef accelerates the endocytosis rate of several human cell surface receptors in infected cells. Recent evidences have shown that Nef can be transferred to uninfected cells from the infected one via cellular protusion, cell to cell contact and exosomes release. In addition studies carried out on human derived non infected macrophages (MDMs) treated with Nef showed that the protein is rapidly internalized and hijacks specific cellular signaling pathways. The aim of my research is to better understand modulation of cell surface markers by Nef treatment of primary hematopoietic cells and a large antibody screening panel would be required for this purpose. In our experiments, Peripheral Blood Mononuclear Cells (PBMCs) will be isolated from buffy coats of healthy donors. Primary monocytes, B and T lymphocytes will be isolated using supermagnetic microbeads conjugated with specific anti-cell surface marker. The purity of the recovered cell population will be assayed by cytofluorimetric analysis. In addition primary monocytes will be induced to differentiate to macrophages or dendritic cells. All the different cell cultures will be treated with recombinant myristoylated HIV- 1 Nef and several mutants, and later analyzed for cell surface antigens modulation by Flow cytometry and Confocal Microscopy. Moreover, apoptotic effects and IL-6 production will be tested using Anexin-V and anti human IL-6 antibodies. In addition signal transduction pathways involved in pro-inflammatory response will be analyzed.
Therefore, specific fluorochrome coniugated antibodies are required to follow and characterize the differentiation profile of cells, the modulation of cell surface receptor expression and pro-inflammatory or proapoptotic cellular response.

ImmunoTools IT-Box-139 for Ali Muhammad 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-IgG1, Control-IgG2a, Control-IgG2b, Annexin V

DETAILS