A study of CD4-positive perforin-expressing cytotoxic T cells subpopulation's specificity in patients with chronic lymphocytic leukaemia

Anna Natriashvili
Nino Porakishvili
E-mail : ana.natriashvili504@ens.tsu.edu.ge
Department of Biology, Iv. Javakhishvili Tbilisi State University, University street.13, 0143, Tbilisi, Georgia

$CD4^+$ cytotoxic T cells (CTLs) play a significant role in immune responses in patients with chronic lymphocytic leukaemia (CLL). As a result of T cell dysfunction CLL patients suffer from recurrent infections and the expansion of CMV-specific $CD8^+$ $CD45RA^+CD27^-$ cytotoxic T cells has been observed in CLL . It has been demonstrated that there is an enhanced response to CMV containing cell lysed by $CD4^+PF^+$ T cells in seropositive CLL patients compared to normal seropositive controls measured by the expression of interferon γ (IFNγ) .

During the proposed study the response to the panel of MHC class II-restricted CMV peptides constituting pp65 (Affinity research products, UK) will be measured to evaluate clonal expansions of responding $CD4^+PF^+$ T cells. Irradiated autologous PBMC pulsed with the pools of overlapping pp65 peptides will be used as target cells and the killing will be measured by the changes in mitochondrial membrane potential using $DiOC_6$ dye.To identify possible targets for anti-CMV $CD4^+$CTLs the most likely cells to harbor CMV - monocytes and dendritic cells, generated from monocytes by stimulation with IL-4 and G-CSF for a week, will be loaded with CMV peptides and targeted by autologous $CD4^+$T cells. The perforin mediated MHC class II-dependent mechanism of killing by $CD4^+$ T cells will be confirmed by blocking with anti-HLA-DR mAb and Concanamycin A.

The proposed study should result in identifying the specific CMV epitopes - targets for anti-CMV $CD4^+$CTLs. We anticipate that the results of the project will ultimately lead to the establishment of the clinical approaches to the treatment of immunodeficiency and immunodeficiency-related infections in patients with CLL.