known as eccentric ventricular hypertrophy, which leads to impaired contraction of the ventricles due to volume overload and the result to heart failure. An inflammation of the myocardium caused by infection and/or autoimmune reactions plays a crucial role in dilated cardiomyopathy and heart failure. The annual incidence of HIV associated dilated cardiomyopathy was 15,9/1000 before the introduction of highly active antiretroviral therapy (HAART). However, in 2014, a study found that 17,6% of HIV patients have dilated cardiomyopathy (176/1000) meaning the incidence has greatly increased. HIV-1 virions appear to infect myocardial cells in a patchy distribution with no direct association between the presence of the virus and myocyte dysfunction. Immune cells infiltrating myocardium in dilated cardiomyopathy seem to play a significant pathogenetic role by activating multifunctional cytokines such as tumor necrosis factor-α and the inducible form of nitric oxide synthase that contribute to progressive and late myocardial tissue damage. The mode of entry of HIV into myocytes remains unclear since they are CD4 receptor negative.

Tumour necrosis factor-α (TNF-α) induces cardiomyocyte dysfunction and pathological changes. TNF-α is produced by infected macrophages and by lymphocytes infiltrating myocardium in dilated cardiomyopathy. The intensity of the stains for TNF-α and inducible nitric oxide synthase (iNOS) of the myocardium was greater in patients with HIV associated cardiomyopathy (as opposed to idiopathic cardiomyopathy), myocardial viral infection and was inversely correlated with CD4 count with antiretroviral therapy having no effect.

TNF-α has been recognized as important physiopathogenetic factor in the initiation and continuation of inflammatory cardiomyopathies. Experimental and human studies have demonstrated that TNF-α plays a crucial role in viral-induced myocarditis. Myocardial expression of TNF-α was correlated with different clinical and pathologic findings. Among TNF-α-positive cases, the greater TNF-α mRNAs, the more impaired was cardiac function. Some findings suggest that the expression of TNF-α may play an important role in the pathogenesis of viral myocarditis and cardiomyopathy of any etiology and may influence the severity of cardiac dysfunction. Whether myocyte damage in the early phase of the disease is primarily linked to the viral presence or to immunomediated damage is still under discussion; however, it is now accepted that the progression of the disease is mainly sustained by immunomechanisms. Previous clinical works have demonstrated that there is an association between depressed myocardial function and elevated TNF-α mRNA and protein levels either in plasma or in the myocardium of patients with myocarditis and dilated cardiomyopathy.TNF-α elicits its biological effects by binding to two distinct cell surface receptors with approximate molecular masses of 55 kDa (TNFRI) and 75 kDa (TNFRII), both expressed in human cardiac myocytes. Cytokine expression was then correlated with the main clinic hemodynamic and pathologic parameters. The direct effect of TNF in human cardiomyocytes has not been investigated yet. In our study we investigated pathological changes induced by inflammatory cytokine TNF-α in human cardiomyocytes.

Human fetal cardiomyocytes were stimulated with inflammatory agent TNF-α for up to 24 h and time-dependent changes in the expression of established markers of pathological hypertrophy were assessed by quantitative PCR. mRNA levels of ANF, marker of hypertrophy were increased following treatment with the inflammatory cytokine TNF-α. Induction of hypertrophic marker genes was gradual (peaking at 24 h). Elevated ANF expression levels are associated with pathological hypertrophic cardiomyocyte changes.

Our results demonstrate that TNF-α directly induces cardiomyocyte pathological changes

TNF-α produced by cardiomyocytes themselves as a response to TNF-α produced by lymphocytes might have an autocrine effect, thus contributing to the progression of cardiomyocyte damage. The role of intramyocardial TNF-α in the disease provides not only new insights to the pathogenesis of dilated cardiomyopathy but also suggests new targets for therapeutic intervention.

The search of informative biomarkers for early immunological diagnosis of tuberculosis in patients with HIV Infection

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Introduction. According to estimates of the World Health Organization, 13% of the 9 million people become ill with tuberculosis (TB) in 2013 were to have been infected with HIV. Since the human immunodeficiency virus affects the immune system, weakening it, to people with HIV causative agent of tuberculosis (*Mycobacterium tuberculosis*) is transmitted seven times more, often occurs more malignant, it has a tendency to generalize and much more goes into the active form. Therefore, early diagnosis of tuberculosis infection in these patients is critical in controlling the spread of the disease.

Own data. Previously we, like many other authors, have shown that QuantiFERON®-TB Gold In-Tube, is effective in the diagnosis of tuberculosis in patients without HIV. We also showed that the chemokine IP-10 (IFNγ — indu-

cible protein — 10, CXCL10) may be an alternative biomarker of tuberculosis infection, as it provides a greater range of measured concentrations compared with IFNγ and a higher threshold that is its unquestionable advantage. But these results can not be approximated for HIV patients because it is known that immune deficiency caused by HIV due primarily to a decrease in the number and the functions loosening of CD4 T-lymphocytes, which leads to a reduction of production and biological action of other cytokines, especially IFNγ.

In view of this, in the our last study we enrolled patients with HIV infection and pulmonary tuberculosis (n=40) and patients with HIV infection without evidence of active tuberculosis (n=47). All persons included in the study were carried out QuantiFERON®-TB Gold In-Tube and we determined the content of the antigen-induced (AG) and the spontaneous production (NIL) of IP-10. For IP-10 we reached the sensitivity of detection of tuberculosis infection 67% and specificity of 92%, whereas QuantiFERON-TB Gold was comparable sensitivity of 70%, but significantly lower specificity 73% (p=0,021, chi-square). And our results showed the possibility of using IP-10 as an alternative biomarker to IFN for the detection of tuberculosis in patients with HIV infection. Prospects for further development of the theme.

Actual is further study of the features of immunopathogenesis of TB in patients living with HIV, search the informative biomarkers of HIV, the development of approaches to assess the effectiveness of specific anti-tuberculosis therapy. At present we obtained blood from 29 patients of TB+HIV+ patients and 24 HIV−TB+. Also we are forming the following groups of patients: TB−HIV− and TB−HIV+.

In addition to the QuantiFERON®-TB Gold In-Tube, by flow cytometry, we assess the degree of maturity of T lymphocytes and NK-cells from the peripheral blood of patients with severe immunosuppression (HIV+) in combination with pulmonary tuberculosis, in the evaluation of effector cell populations.

Peripheral blood samples would be stained with antibodies to CD3, CD4, CD8, CD27, CD28, CD45, CD45RA, CD62L, as well as CD56 and CD57. Samples will be acquired using Navios flow cytometer, equipped with 405, 488 and 635 lasers (Beckman Coulter, USA). T-helpers will be identified as CD3+CD4+ and cytotoxic T-cells as CD3+CD8+. Based on initial expression of CD27 and CD28 and on the following analysis of CD45RA and CD62L expression on CD27+CD28+ subset CD3+CD4+ and CD3+CD8+ lymphocytes will be divided into the following subpopulations: «naïve» CD27+CD28+ CD45RA+CD62L+, central memory CD27+CD28+ CD45RA−CD62L+, transitional memory CD27+ CD28+CD45RA−CD62L−, as well as effector memory cells and effector cells (CD27+CD28− and CD27−CD28−, respectively). For T-cell subsets identification we also plan to use the «alternative» gating strategy based on initial analysis of CD45RA and CD62L expression. So CD3+CD4+ and CD3+ CD8+ cells will be divided into «naïve»» (CD45RA+ CD62L+), central memory (CD45RA−CD62L+), effector memory (CD45RA−CD62L−,) and «terminally differentiated» effector memory (CD45RA+ CD62L−) cells. Based on the expression of CD27 and CD28 in EM and TEMRA further populations will be distinguished, which within EM cells will be CD27+CD28+, CD27+CD28− CD27−CD28− and CD27−CD28+. While within TEMRA CD27+ CD28+ (pE1), CD27+CD28− (pE2) and CD27− CD28− (E) subpopulations will be isolated. The frequencies of all these T-cell subsets will be expressed in percentages from the total lymphocytes and in absolute counts. Furthermore, the presence of CD56 and CD57 — the key molecules, that characterize the effective capacity of cytotoxic T-cells and, in some measure, of T-helpers — was studied on all the above mentioned T-cells subsets. NK-cells will be purified from peripheral blood as CD3-CD56+. In order to identify the main subpopulations of NK-cells the expression of CD8, CD27, CD62L and CD57 will be measured.

The results allow to conclude that, if the level of immunosuppression effect on the functional activity of T-lymphocytes, which are the most useful cell populations in HIV detection and in diagnosis of tuberculosis in patients with HIV infection.

The evaluation of immune dysregulation during opioid withdrawal treatment and opioid abstinence among HIV infected injection drug users

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Introduction: Neurocognitive impairment (NCI) is an important complication of chronic HIV infections. Previous research found that Injection drug use (IDU) of opioids increase risk of NCI. We hypothesize that dysregulation of the neuroendocrine (hypothalamic pituitary axis, HPA and sympathoadrenalmedullary, SAM) and immune systems during opioid withdrawal and opioid addiction treatment may contribute to heightened neurological risk in HIV infected IDU. Our overall concept is that this dysregulation represents one of the mechanisms that sets the stage for HIV associated brain injury as evidenced by NCI.

Methods: This study will recruit 80 HIV infected IDU who are HAART naive during last month and entering opioid addiction treatment (detoxification). Participants