Summary:

Background: *Francisella tularensis* (F.t.) is a facultative intracellular bacteria, enrolled at the list of Centre for Disease Control (CDC) as a high risk bioterrorism agent, category A. There is a long-term effort to understand to the immunopathogenesis of F.t. infection. The aim of our study was focused on phenotype analysis (CD54, CD16/32 and CD86) and nitric oxide (NO) production analysis of murine macrophage-like cell line J774.2 durig F.t. live vaccine strain (LVS) *in vitro* infection. J774.2 cells were either untreated or stimulated either before or after F.t. infection by interferon gamma (IFNγ), lipopolysaccharide (LPS) separately or in its combination.

Method: We followed up the expression of cell surface markers and NO production 3, 6, 9, 12 and 24 hours after initiation of infection with or without stimulation using flow cytometry and Griess method respectively. The expression was followed as either absolute value of mean fluorescence index (MFI) or as relative change of MFI (ΔMFI). Murine macrophage-like cells (J774.2) were incubated in cultivation flasks ($2x10^6$ cells/10ml of medium Dulbecco´s MEM with Glutamax-1 with 10% BSA). The cells were activated with 10 or 50 ng of LPS / 1ml of medium or with 100 or 1 000 I.U. of IFNγ / 1ml of medium separately or in combination (10 ng/ml and 100 IU/ml or 50 ng/ml and 1000 IU/ml) and infected by *F. tularensis* LVS with multiplication of infection 1:100 in particular time schemes. F.t. proliferation was assessed by colony forming units (CFU) counts after 24 hours cultivation of cell supernatant on McLeod agar.

Results: Both absolute values and relative changes of mean fluorescences (MFI) of CD54, CD86 and CD16/32 and concentration of NO in cell supernatant are very sensitive predictors of effective macrophages activation and subsequent infection elimination. It was especially pronounced in the case of J774 cells stimulation with combination of IFNγ and LPS regardless time scheme of stimulation.

Conclusion: Measurement of expression of surface markers CD54, CD86 and CD16/32, as well as NO production seem to be very usefull approach to predict with high sensitivity and specificity the outcome of *in vitro* F.t. infection.