Interleukin 2 helps to detect the allergen responsive lymphocytes in whole blood cultures

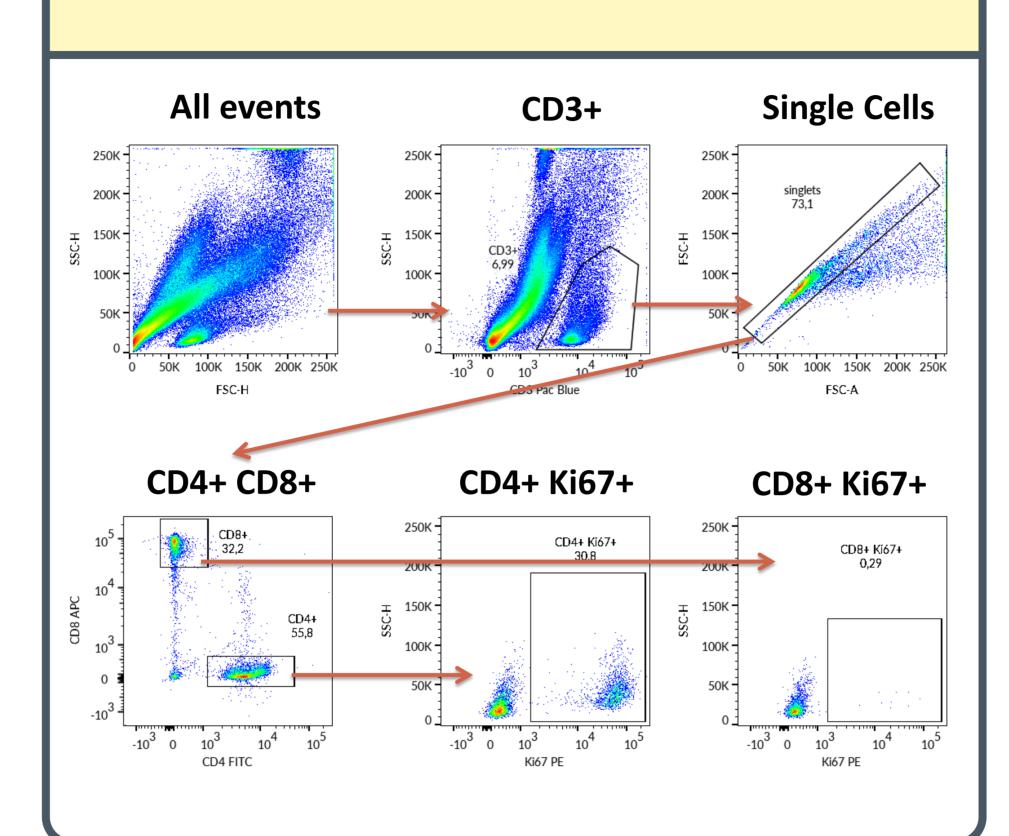


P. Jinoch, M. Prouza EXBIO Praha, Vestec, Czech Republic

Introduction

The *in vitro* tests for non-IgE mediated hypersensitivity are based on lymphocyte proliferative response to stimulation with the specific allergen. To simplify the methodology of the test we perform whole blood cultures in single use cytometry tubes with flow cytometry detection of the marker Ki-67. Initially IL-2 supplementation corrected our variable response rate to the tetanus vaccine stimulations.

Gating strategy and the staining profile of stimulated whole blood

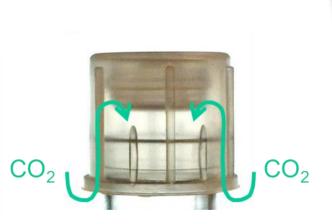


Objective

Evaluate the use of IL-2 in whole blood long term cultures (5-7 days) for the detection of lymphocyte proliferative responses to allergens in blood samples from allergic (BAT positive) patients and from patients diagnosed with non-lgE allergy.

Methods





Whole blood was diluted 1:10 in

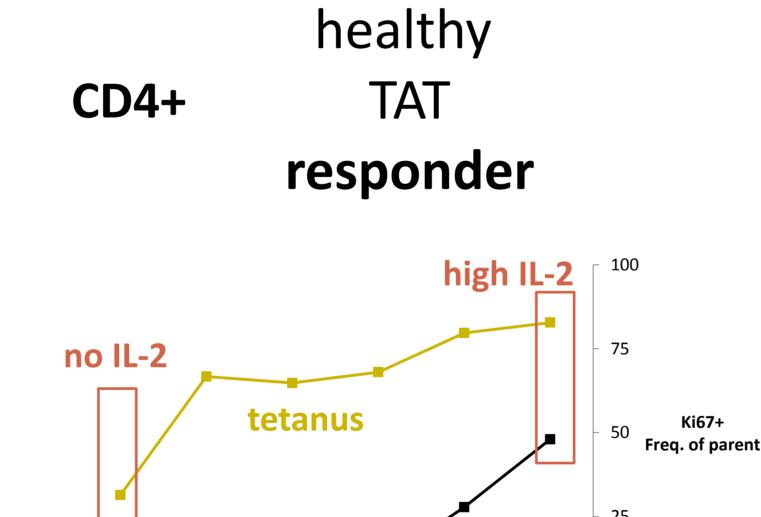
X-VIVO culture medium and incubated with food allergen extracts (wheat, cow milk, carrot) in cytometer compatible tubes. Cultures were supplemented with recombinant interleukin-2 to promote the proliferation of activated lymphocytes.

After 5-7 days the cultures were processed with T-cell BlastoFlowEx Kit (EXBIO) and stained with anti-CD3/CD4/CD8/Ki-67 antibodies.

Basophil activation test was perfomed with BasoFlowEx Kit (EXBIO) with the same allergen extracts.

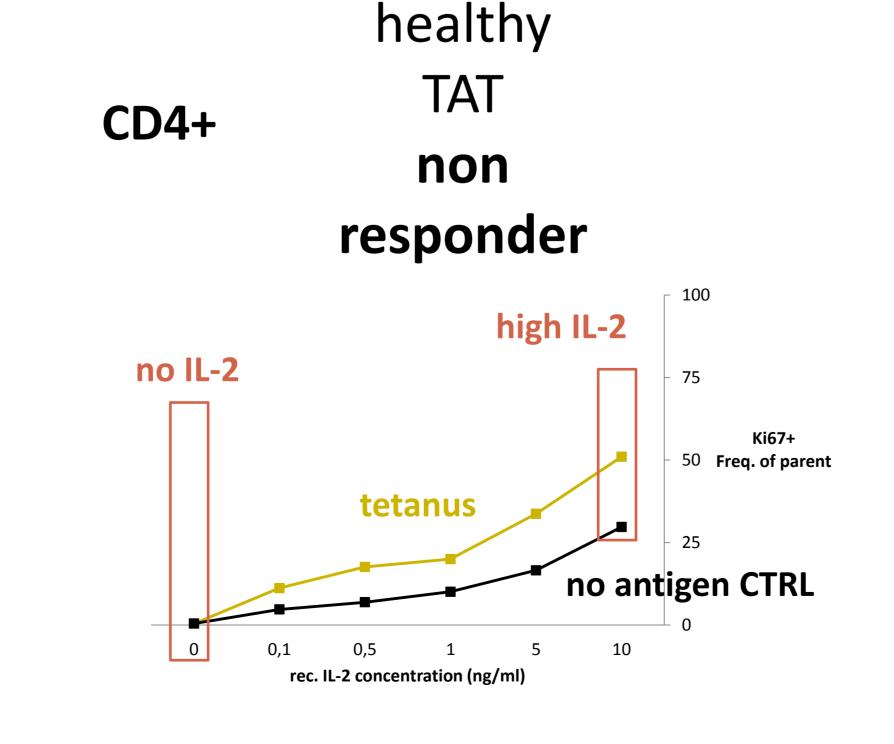
Results (I)

Low concentrations of IL-2
enhance the response *in vitro*to the Tetanus vaccine in non
responding blood donors



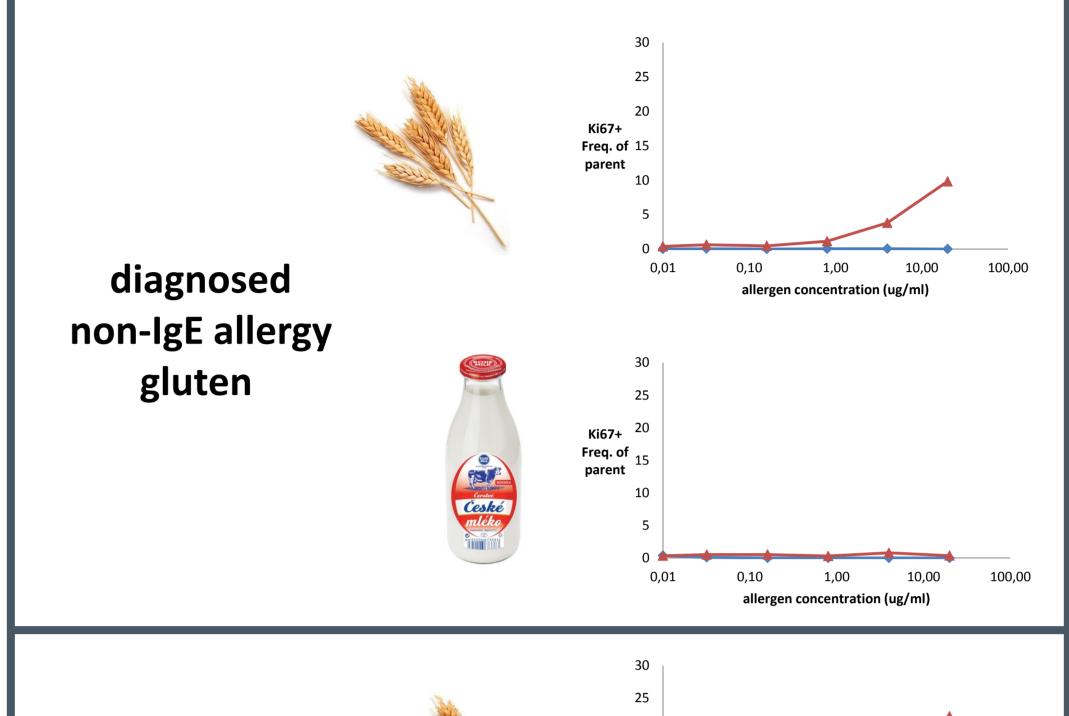
rec. IL-2 concentration (ng/ml)

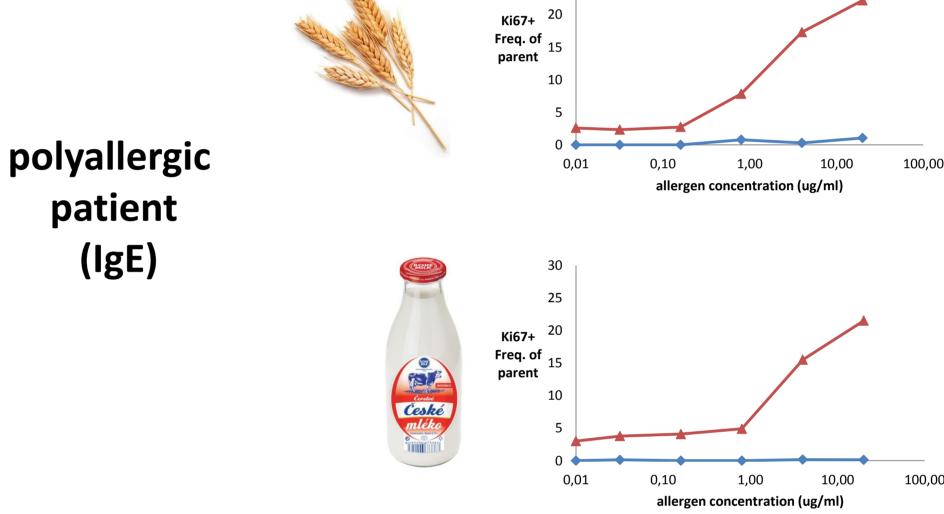
no antigen CTRL

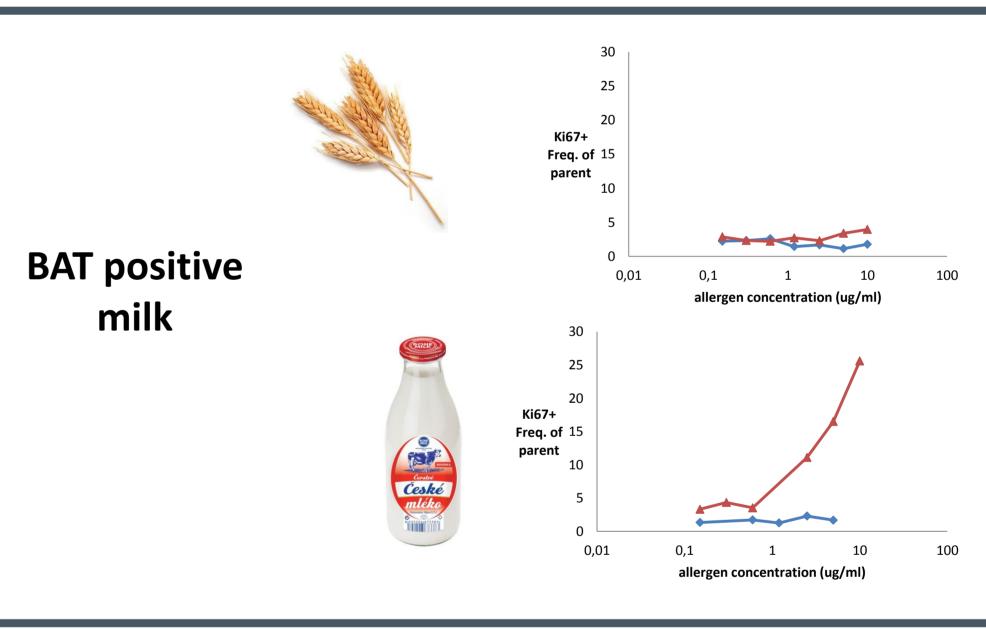


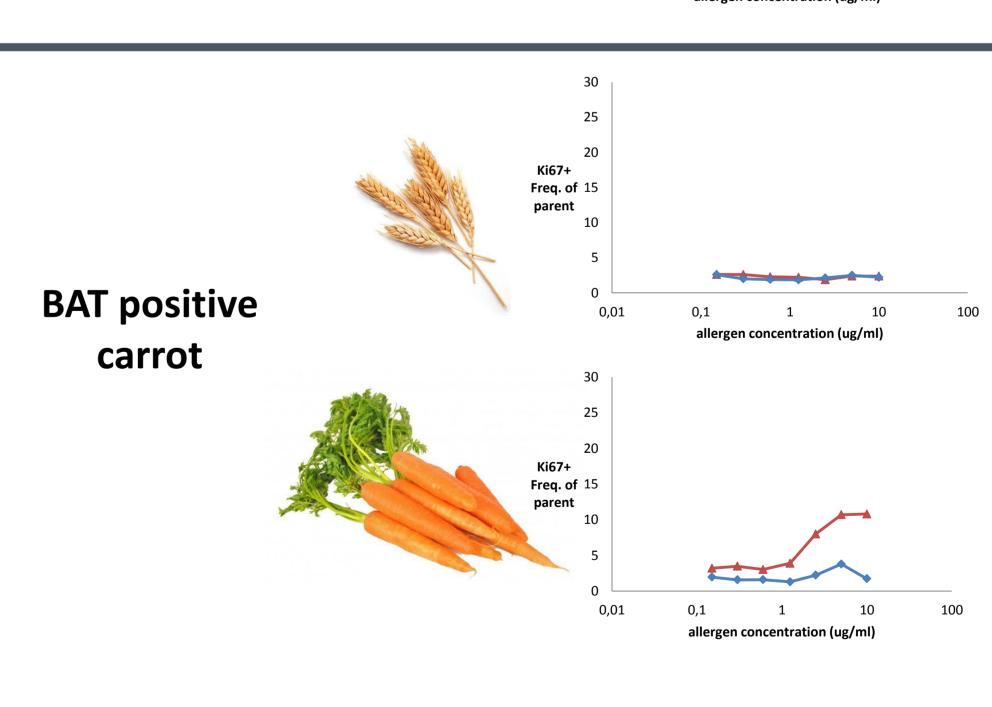
Results (II)

IL-2 allows the detection of response to food allergens









Conclusion

Interleukin-2 enhanced the detection rate of antigen/allergen specific lymphocytes in whole blood cultures.

Patients with IgE food allergy (BAT positive)
had positive response to the same allergens in
lymphocyte proliferation test.