

## **BAT as useful method for monitoring the effectiveness of specific allergen immunotherapy**

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### **Background**

The Basophil activation test (BAT) is a reliable and sensitive method in IgE-mediated diagnosis allergy. This method is based on stimulation of basophils in vitro with the appropriate allergen associated with their follow up degranulation. Activation of basophils is monitored by changing the expression of activation markers, most often expression of CD63, using a flow cytometer. BAT is currently the subject of intensive research, and it has been gradually introduced into diagnostic workup of allergic diseases. It appears to be a sensitive tool to measure changes in allergen response during specific immunotherapy which aims to suppress histamine release from basophilic granulocytes, resulting in less serious immediate allergic reactions.

### **Method**

For two years specific immunotherapy was observed in the patient with an allergy to Timothy grass pollen and yet another year will be monitored. We used commercially available flow cytometry detection kit based on combination CD63FITC/CD203cPE (BasoFlowEx® Kit, EXBIO) to monitor changes in basophil specificity and reactivity of the patient undergoing immunotherapy against Timothy grass (*Phleum pratense*) pollen. A twelve-point concentration range of Timothy grass pollen extract and recombinant molecules (rPhl p 1, rPhl p 5 and rPhl p 7) was used to stimulate basophils in freshly taken heparinized blood sample. Stimulated blood samples were assayed by BasoFlowEx® Kit followed by analysis using flow cytometry (BD FACSCanto SORP). Basophil specificity and reactivity changes were calculated from dose-response curves as an area under the curve (AUC) parameter for the comparison purpose. A correlation between the changes of basophil specificity and reactivity and changing levels of Timothy grass specific IgEs during immunotherapy was also checked.

### **Results**

During 2 years years of Timothy grass pollen immunotherapy there was a decrease in activation of basophils from 92,6 % to 2,1 %. Changes in basophil specificity and reactivity correlate with patient symptoms described during allergen specific immunotherapy as well as with changes of levels of allergen specific IgEs (measured with the ImmunoCAP Lab assay). The Basophil Activation Test thus can be very convenient method to evaluate efficacy of the disease treatment during allergen-specific immunotherapy.

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