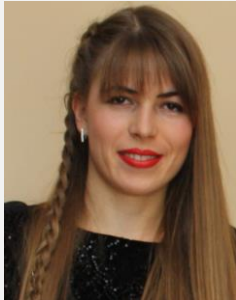


# ImmunoTools *special* Award 2013



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## **Dietary polyphenols modulate immune response to food allergens**

**Background:** Polyphenols, including phenolic acids, flavonoids, proanthocyanidins and resveratrol, are a large and heterogeneous group of phytochemicals abundantly present in foods and beverages. These compounds have a wide range of biological activities which include anti-inflammatory, antioxidant, cardiovascular protective and anti-cancer actions [1]. The interaction of polyphenols with allergens and immune cells can modulate the process of allergic sensitization and following re-exposure to the allergen; resveratrol and apple polyphenol extract inhibit symptoms of food allergy in mice [2, 3]. Although many data on the immunomodulatory effects of polyphenols have been collected, the underlying mechanisms are still not fully understood.

**Objective:** To investigate how polyphenolic compounds from natural sources (green tea, coffee, cocoa, soy and turmeric) influence allergen: (1) epithelial transport; (2) uptake and processing in DCs and subsequent presentation to T cells.

**Preliminary results:** Green tea polyphenol extract reduced the cell-surface expression of MHC-II and co-stimulatory molecules on monocyte derived dendritic cells (MDDCs). Polyphenols from obtained extracts formed non-covalent interactions with known milk, egg and peanut allergens as judged by fluorescence quenching analysis. Epigallocatechin-3-gallate from green tea impaired ovalbumin uptake by human monocytes and ovalbumin-induced basophil activation.

**Research plan- Implementation of ImmunoTools reagents:** Since polyphenols and food allergens enter our system through gastrointestinal tract it is of crucial importance to investigate their bioavailability and interactions with epithelium. Whether polyphenols modify allergen absorption will be examined in a time course transport study through Caco-2 cell monolayer, a model of intestinal barrier. Also, extensive Caco-2 cell phenotyping (CD10, CD13, CD14, CD35, CD63, CD18, CD61, CD21, CD47, CD59, CD25 and CD28) upon the incubation with polyphenols and allergens will be carried out by flow cytometry.

Next part of the project focuses on the effects of polyphenol extracts on the interactions between the allergens and the key players in the development of allergic immune response- dendritic cells. It is important to emphasize that experiments will be carried out with cells derived from the blood of both non-allergic and allergic donors. MDDCs will be generated from CD14<sup>+</sup> monocytes in the presence of

recombinant human IL-4 and GM-CSF. Uptake of fluorescently labeled allergens by MDDCs in the absence and presence of phenolic extracts will be monitored in time by flow cytometry. We will examine the effect of polyphenol extracts on the allergen induced maturation of MDDCs by flow cytometric analysis of surface expression of DC markers (CD1a, CD14, HLA-DR, HLA-DP, CD40, CD80, CD83, CD86, CD209, CD11a, CD11c, CD50, CD54, CD58 and CD102). Also, presence of TNF- $\alpha$ , IL-1 $\beta$ , IL-6, IL-10 and IL-12 in the culture supernatants will be assayed by ELISA. LPS will be used as a positive activator of DC maturation. Stimulated MDDCs will be co-cultured with naive CD4<sup>+</sup> T cells isolated from peripheral blood mononuclear cells of blood donors (characterized by flow cytometry as CD4<sup>+</sup>CD45RA<sup>+</sup>CD45RO<sup>-</sup>CD25<sup>-</sup> T cells). T cell proliferation is going to be monitored with CFSE and levels of secreted IL-4, IL-5, IL-13 and INF- $\gamma$  will be measured by ELISA.

The obtained results will help shed light on the anti-allergic and immunomodulatory effects of polyphenolic compounds.

#### References:

- [1] Singh A, Holvoet S, Mercenier A. Dietary polyphenols in the prevention and treatment of allergic diseases. *Clinical & Experimental Allergy* 2011; **41** (10): 1346-1359.
- [2] Okada Y, Oh-oka K, Nakamura Y, Ishimaru K, Matsuoka S, Okumura K, et al. Dietary Resveratrol Prevents the Development of Food Allergy in Mice. *PLoS ONE* 2012; **7** (9): e44338.
- [3] Zuercher AW, Holvoet S, Weiss M, Mercenier A. Polyphenol-enriched apple extract attenuates food allergy in mice. *Clinical & Experimental Allergy* 2010; **40** (6): 942-950.

**ImmunoTools special AWARD for Marija Stojadinovic** includes 25 reagents

**FITC** - conjugated anti-human CD1a, CD35, CD45RA, CD47, CD54, CD58, CD63, CD86, HLA-DR

**PE** - conjugated anti-human CD11a, CD18, CD25, CD50, CD80,

**APC** -conjugated anti-human CD4, CD11c, CD14, CD59,

recombinant human cytokines rh GM-CSF, rh IL-4

human IL-4 ELISA-set, human IL-6 ELISA-set, human IL-8 ELISA-set, human IL-12p40 ELISA-set, human TNFa ELISA-set,

[DETAILS](#)