

## In-vitro-induced cytokine secretion to characterise effector cell response to environmental allergens

There is a large body of scientific literature available that indicates involvement of cytokines in environmental exposure and diseases as well as allergies. Relevant medical conditions include fibromyalgia syndrome, chronic fatigue syndrome (CFS), multiple chemical sensitivity syndrome (MCS) as well as allergies, silicosis, asbestosis and various neurodegenerative processes. Cytokine patterns are extremely variable due to their kinetics, however, meaning that their measurement in blood only has diagnostic value in exceptional cases. For justified questions, on the other hand, 'in vitro allergen-induced cytokine secretion' can make an important contribution to understanding the pathogenesis of an environmental medical condition.

### What are cytokines?

Cytokines are proteins that are secreted by immune cells and other nucleated cells in the body. They trigger specific signals via receptor interactions either in the same cell (autocrine), neighbouring cells (paracrine) or systemically in remote target cells. They are the central regulators of the immune response, initiate every inflammatory reaction and trigger inflammatory-associated symptoms such as fever, swelling and pain. The more than 50 known cytokines include the interleukins (IL-1 to IL-32), the interferons (IFN- $\alpha$ , - $\beta$  and - $\gamma$ ) as well as various growth hormones (more information can be found on our homepage [www.imd-berlin.de](http://www.imd-berlin.de) under Diagnostic Information / Cytokine Diagnostics).

In particular, classification into pro- and anti-inflammatory cytokines of Th1- and Th2-associated type which are associated with T helper cells is clinically relevant. The Th1 cytokines include interferon-gamma (IFN- $\gamma$ ) and IL-2, TNF $\alpha$  and - $\beta$  and IL 12. These primarily pro-inflammatory mediators initiate cytotoxic immune responses. Th2 cells, on the other hand, induce humoral responses (antibody synthesis including IgE) via the production of IL-4, IL-5, IL-10 and IL-13 and can counteract the cytotoxic response with 'tolerance induction'.

### Clarification of pathomechanisms

Testing for the formation of cytokines by cells of the immune system as part of environment-associated processes is currently of interest. The test of whether particular environmental allergens and substances induce either monocytes/macrophages cytokines, Th1 or Th2 cytokines in an individual case may predict or indicate possible response patterns to these substances. These tests are available in the field of medication allergies but also contact allergies (Sachs 2002, Jakobson 2002, Lindemann 2003). Using nickel sensitisation as an example, several studies have shown

that a predominance of IFN- $\gamma$  synthesis (Th1 dominance) compared to IL-10 is associated with a pro-inflammatory, clinically manifest immune response and that with predominance of IL-10 (Th2 dominance) a tolerogenic response (without a cytotoxic immune response) can often be observed.

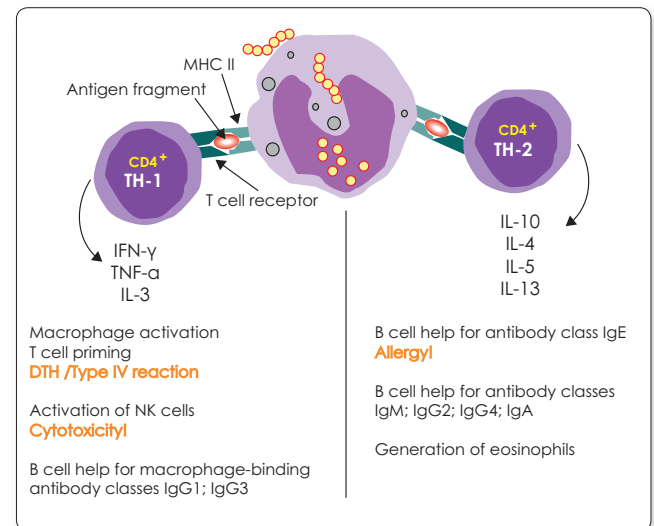


Fig. 1

### In-vitro-induced cytokine secretion

A suitable method to characterise the effector mechanisms is in-vitro stimulation of immune cells with the 'suspected' allergen/hapten. For this purpose immune cells from the blood of the patient are isolated in specialised laboratories and stimulated with the harmful substances being investigated for a few hours to several days under defined conditions. The cytokines formed are then quantified using highly sensitive methods (ELISA, ELISpot). A positive result indicates an immune response. The pattern of Th1/Th2 cytokines measured may also provide information about the existing effector cell response in light of the clinical situation.

### Indication for the test

The method answers the following questions for the given indication:

- Does a cytokine response to the specific agent (allergen, hapten) indicate an immunological sensitisation in the patient?
- Can this cytokine response be classified as a Th1 (cytotoxic, IFN- $\gamma$ ) or a Th2 (humoral, IL-10) immune response?

**Do you have questions? Our serviceteam will be happy to support you: +49 (0)30 770 01-220.**

The most important indication in the practice of environmental medicine is based on the fact that the results can indicate the priorities for necessary avoidance of exposure. Because this analysis is very elaborate due to the necessity of monitoring and dilution series as well as expensive cytokine analytics and interpretation is only possible with consideration of the clinical question and other laboratory data, the procedure is reserved for specific diagnostic questions.

IMD Labor Berlin		medical report	
Test	Result	Unit	Reference range
<b>Antigen-induced Th1-Th2 cytokine profile</b>			
IFN- $\gamma$ (preparation 1)	<b>2,8</b>	IU/ml	< 0,3
IL-10 (preparation 1) (1) Nickel	<b>126,8</b>	pg/ml	< 5,0
IFN- $\gamma$ (preparation 2)	<b>347,7</b>	IU/ml	< 0,3
IL-10 (preparation 2) (2) Gold	<b>12,6</b>	pg/ml	< 5,0
<p>The results indicate a significant Th1 cytokine induction by gold with almost complete absence of IL-10 release. A pro-inflammatory sensitisation to gold must thus be assumed in this case in contrast to nickel sensitisation.</p>			

**Fig. 2** Sample results: Allergen-induced cytokine pattern of a 38-year-old patient with type IV sensitisation to nickel and gold confirmed by LTT and epicutaneous testing.

## Material

5 ml heparin blood per to be tested allergen/hapten

Sample receipt within 24 hrs has to be ensured. The sample should be stored and transported at room temperature. Within the Berlin city area, we offer a courier service (+49 (0)30 7701- 250). For collections beyond Berlin, please contact our complimentary courier service (+49 (0)30 77001-450).

## Invoicing

The costs per allergen are about 46.92 €(+ a one-off fee of 26.81 € for the cell separation).

## Literature

- Romagnani S. TH1 and TH2 in human diseases 1996; Clin. Immunol Immunopathol. 80; 225-235
- Bedeutung von Zytokinbestimmungen in der umweltmedizinischen Praxis. Mitteilung der Kommission „Methoden und Qualitätssicherung in der Umweltmedizin“. Bundesgesundheitsblatt 2004; 47: 73-79.
- Lindemann M et al. ELISpot: a new tool for the detection of nickel sensitization. Clin. Exp. Allergy 2003; 33: 992-8.
- Sachs B, Erdmann S, Malte Baron J, Neis M, al Masaoudi T, Merk HF. Determination of interleukin-5 secretion from drug-specific activated ex vivo peripheral blood mononuclear cells as a test system for the in vitro detection of drug sensitization. Clin Exp Allergy. 2002; 32:736-44.
- Jakobson E, Masjedi K, Ahlborg N, Lundeberg L, Karlberg AT, Scheynius A. Cytokine production in nickel-sensitized individuals analysed with enzyme-linked immunospot assay: possible implication for diagnosis. Br J Dermatol. 2002 ; 147:442-9.