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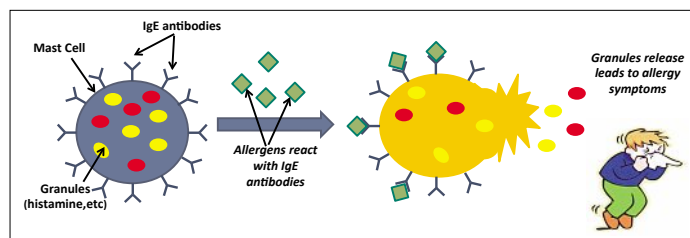
Allergy Diagnostics Using Magnetic Beads in a GRAVI™-Cell Microfluidic Device

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Allergy is a widespread immunological disorder often related with Western lifestyles in which sterile environments deprive the immune system of factors stimulating its proper development. Allergic diseases, for example allergic rhinitis and asthma, are mediated by histamine (and other substances) released from mast cells due to their interaction with special type of antibody, so-called IgE antibodies, in response to normally harmless substances, allergens. IgE antibodies recognize allergens and activate mast cells, thus a high level (>200 µg/L for adults) of IgE in blood serum is an allergy indicator.

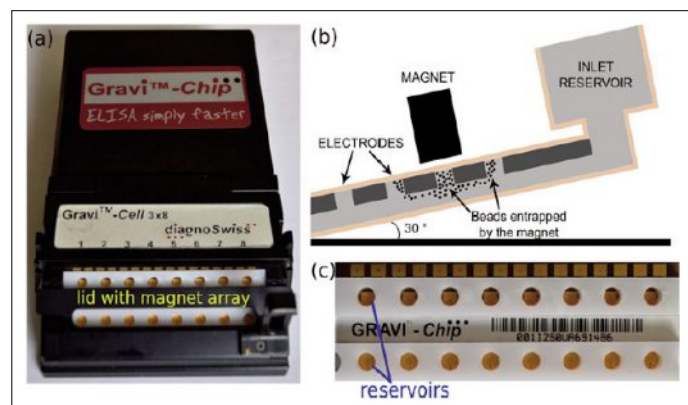


Schematic representation of allergic reaction: IgE antibodies fixed on the surface of mast cell recognize the allergen and provoke the release of histamine and other mediators leading to the development of allergy symptoms.

Measurement of total IgE concentration is one of the methods of allergy diagnostics in clinics. Analytical techniques employed for such routine analysis should therefore be fast and cheap. Herein, a fast immunoassay of IgE antibodies in human blood serum was developed using magnetic beads and the commercial device GRAVI™-Cell from DiagnoSwiss (Monthey, Switzerland).

A GRAVI™-Chip placed inside the device at an angle of 30° allows introduced liquids to flow under gravity and capillary forces only. It comprises of eight microchannels and each microchannel possesses two reservoirs and a microelectrode for electrochemical detection. IgE quantification is performed as one-step sandwich ELISA (enzyme-linked immunosorbent assay) with the immunocomplex formed on the surface of the magnetic beads which are trapped inside the microchannels by integrated permanent magnets.

The developed analysis shows good sensitivity (limit of detection 17.5 µg/L) suitable for effective allergy diagnosis.

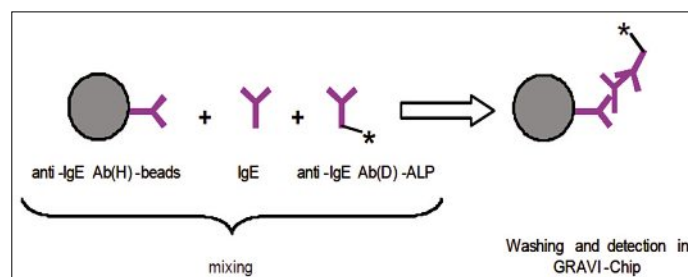


a) GRAVI™-Cell device with an 8-microchannel chip tilted at 30°. b) Cross-section along the microchip showing magnetic beads trapping. c) GRAVI™-Chip comprising 8 microchannels, inlet reservoirs at the top and outlet ones at the bottom.

Due to the miniaturized format and the application of magnetic beads as a support for the immunoassay the entire analysis can be performed in less than 1 hour and only 1.5 µl of sample is required. As each GRAVI™-Chip possesses eight parallel microchannels it is possible to perform calibration and sample analysis simultaneously with good inter- and intra-chip reproducibility (RSD <10%).

The presented technique for IgE analysis combines the sensitivity and selectivity of immunoassay performed on magnetic beads with advantages of microfluidics, of gravity force-driven fluidics and of electrochemical detection, offering an opportunity of process automation for cheap and high throughput clinical analysis.

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One-step sandwich ELISA: immunocomplex is formed by secondary anti-human-IgE antibodies Ab(H) immobilized on magnetic beads surface, IgE antibodies present in the sample, and by secondary anti-human-IgE antibodies Ab(D) labeled with enzyme alkaline phosphatase (ALP). The electrochemically active substrate of ALP is used for detection.

Reference

G. Proczek, A. L. Gassner, J. M. Busnel, H. H. Girault, *Anal. Bioanal. Chem.* **2012**, *402*, 2645.

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