

# Mass spectrometric and peptide chip epitope analysis on the RA33 autoantigen with sera from rheumatoid arthritis patients.

El-Kased RF, Koy C, Lorenz P, Drynda S, Guthke R, Qian Z, Koczan D, Li Y, Kekow J, Thiesen HJ, Glocker MO (2010) Mass spectrometric and peptide chip epitope analysis on the RA33 autoantigen with sera from rheumatoid arthritis patients. *Eur J Mass Spectrom (Chichester, Eng)* 16(3), 443-451.

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## Abstract

As the potential of epitope chips for routine application in diagnostics relies on the careful selection of peptides, reliable epitope mapping results are of utmost interest to the medical community. Mass spectrometric epitope mapping in combination with peptide chip analysis showed that autoantibodies from patients who suffered from rheumatoid arthritis (RA) were directed against distinct surface structures on the full-length human autoantigen RA33 as well as against partial sequences. Using the combined mass spectrometric epitope extraction and peptide chip analysis approach, four sequence motifs on RA33 emerged as immuno-positive, showing that epitopes were not randomly distributed on the entire RA33 amino acid sequence. A sequential epitope motif ((245)GYGGG(249)) was determined on the C-terminal part of RA33 which matched with the Western blot patient screening results using the full-length protein and, thus, was regarded as a disease-associated epitope. Other epitope motifs were found on N-terminal partial sequences

((59)RSRGFGF(65), (111)KKLFVG(116)) and again on the C-terminal part ((266)NQQPSNYG(273)) of RA33. As recognition of these latter three motifs was also recorded by peptide chip analysis using control samples which were negative in the Western blot screening, these latter motifs were regarded as

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## **Leibniz-HKI-Autor\*innen**



**Reinhard Guthke**

[Details](#)

## Identifier

**doi:** 10.1255/ejms.1046

**PMID:** 20530829