Results RAMOS cells presented exon 4 skipping (ratio inclusion/exclusion: 6:8) after minigene transfection. Following co-transfection of the minigene with coding plasmids for splicing proteins, only the overexpression of SC35 showed effect in the splicing of exon 4, promoting exon 4 inclusion (ratio: >30). Incubation of different cell lines with several cytokines showed that IFN-γ was able to induce A4BAFF-transcript. Thus, after IFN-γ stimulation in the minigene model, the ratio inclusion/exclusion markedly decreased (1.5). IFN-γ modifies the balance between SC35 and another member of hnRNP family (hnRNP C1/C2) favouring the alternative splicing of exon 4.

Conclusions These results demonstrated that IFN-γ induces A4BAFF, modifying the function of SC35 protein and increasing the hnRNPC1/C2. Our study provides an expanded conceptual view of BAFF gene regulation, and contributes to a better understanding of the mechanisms involved in BAFF up-regulation in autoimmune diseases.

Background and Objectives Anti-citrullinated protein/peptide antibodies (ACPAs) are the most sensitive and specific serological markers of RA. To identify the optimal epitopes that detect different subgroups of RA patients with high sensitivity and specificity, we have investigated citrulline and arginine containing peptides derived from filagrin, collagen or vimentin. We have identified a citrulline-containing peptide panel that was recognised by RA sera with high specificity. Our aim was to compare this peptide panel with the conventionally used serological assays and to detect peptide-specific ACPA producing B-cells in vitro cultures.

Materials and Methods Previously selected citrulline- and arginine-containing filagrin, vimentin and collagen peptide epitopes were investigated. We compared the recognition of these peptides by RA and control sera using indirect ELISA. B-cells were purified from peripheral blood by negative selection, IgG production was stimulated by B-cell activators (R848 and recombinant human IL-2) provided with the human ELISPOT kit. Antibody producing cells were enumerated after 4 days culture by using peptide-specific ELISPOT assay.

Results Sera samples from 247 RA and 148 age-matched (57 ± 14 years) healthy controls were collected. The citrulline peptide panel detected approximately 80% of RA patients, including 20% of seronegative/CCP negative patients as well. Individual peptides detected different subgroups of RA patients. The more peptides recognised by a particular RA serum sample, the more severe the disease of the patient was. In vitro cultured B-cells from selected RA patients synthesised multiple citrulline-containing peptide-specific antibodies after polyclonal stimulation, while B-cells from healthy blood donors did not.

Conclusions The citrulline peptide panel can detect 20% of ACPA negative RA patients thus may have a prognostic value. Furthermore, the panel is suitable to detect citrulline peptide-specific antibody producing cells, thus enables us to study ACPA producing B-cells of RA patients.