according with the international criteria for the diagnosis of AIH. From 45 subjects identified as having HCV/AIH overlap, 31 were enrolled after giving their informed consent. Control group was constituted by 29 consecutive patients with hepatitis C without autoimmune feature. Both groups were comparable regarding to gender and age. HLA typing was performed in DNA extracted from peripheral leukocytes, by DTAB/CTAB techniques, followed by SSCP with Micro SSPTM HLA DNA Typing kit (One Lambda Inc., CA, USA). Statistical analysis was done with Pearson's $\chi 2$ test, with Yates correction or Fisher exact test when appropriate.
Results: Class II HLA results are listed in the table. The allele DRB1*04 was detected in 45.1% patients with HCV/AIH overlap syndrome, and in 3.4% of control group ($p = 0.0006$).
Conclusions: These results support the hypothesis that the allele DRB1*04 confers susceptibility to AIH features in patients with hepatitis C.

860 HNRNP A2/B1 AS ANTINUCLEAR ANTIBODIES TARGET: EPITOPE MAPPING AND DIFFERENTIAL REACTIVITY IN AUTOIMMUNE HEPATITIS AND CONNECTIVE TISSUE DISEASES

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The hnRNP A2/B1 (heterogeneous nuclear ribonucleoprotein) protein was identified by serological proteome analysis as one of the targets recognized by 52% of anti-nuclear antibodies (ANA) detected in autoimmune hepatitis type 1 (AIH-1). However, this molecular target is not specifically recognized by AIH-1 sera and can be found in connective tissue diseases.
Aims: (i) To predict, by bioinformatic analysis, the linear epitopes of the hnRNP A2/B1 molecule. (ii) To estimate, by ELISA tests, the differences in epitope recognition of the hnRNPA2/B1 between patient sera with AIH-1 compared to other connective tissue diseases, rheumatoid arthritis (RA) and systemic lupus erythematosus (SLE).
Methods: Epitope mapping was carried out with the NPS@ and Geno3D servers and the AnTheProt software taking into consideration physico-chemical profiles, the research of functional sites, as well as homology modeling. Two peptides corresponding to putative epitopic sites were synthesized and used as antigens in ELISA tests. Populations tested were: 47 AIH-1 sera, 10 RA and 10 SLE. All sera were positive for ANA. As negative controls, 129 blood donors sera.
Results: (i) Eight linear epitopes were predicted. Two peptides were synthesized and used as antigens: the linear sequence hnRNP A2/B1 41–53 exposed on the surface of the native protein as predicted by modecular modeling, and the hnRNP A2/B1 288–303. (ii) By ELISA, the hnRNPA2/B1 41–53 peptide reacted with the AIH-1 patient sera with a frequency significantly higher than negative controls (21.2% vs. 3.8% $p = 0.0008$). Twenty percent of RA patients were also positive, while no SLE reacted with this peptide. Taken into consideration the hnRNP A2/B1 288–303 peptide, no significant difference was noted (2.4% of controls, 10.6% of AIH-1). But 20% of the RA patients were positive and no SLE reacted.
Conclusions: These results indicate that other epitopes exist on the molecule hnRNP A2/B1 than those used and recognized by AIH-1 patient sera. ANA anti-hnRNP A2/B1 found in AIH-1 sera react differently than sera from connective tissue diseases. The use of other epitopic peptides and the study of conformational epitopes will give a more complete reactivity profile.

861 A COMPLEX IMMUNOREGULATORY DEFICIENCY CHARACTERISES TYPE 1 AUTOIMMUNE HEPATITIS

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Background: Immuno-tolerance is maintained through the action of distinct populations of regulatory T cells such as CD4+CD25+, CD8+CD28−, γδ, and CD3+CD56+ cells. CD4+CD25+ cells, the best known regulatory subset, are numerically and functionally impaired in children with autoimmune hepatitis (AIH) but little is known about CD4+CD25+ and other regulatory populations in adults with AIH.
Aim: To determine the frequency of different regulatory T cell populations in adults with AIH.
Patients and Methods: Twenty-four patients with ANA/SMA positive AIH (median age 50 years, 20 females) including 3 with active disease [A] (onset or relapse) and 21 during drug-induced remission [R] were studied. Eighteen healthy subjects (median age 35 years, 13 females) served as controls. The following surface markers were evaluated by flow cytometry on peripheral blood mononuclear cells: CD4, CD25, CD8, CD28, γδ T-cell receptor, CD3, CD56. CD4+CD25+ cells were purified using immunomagnetic beads. The mean fluorescence intensity (MFI) of CD4+CD25+ functional markers FOXP3 and CTLA-4 was evaluated by flow cytometry.
Results: CD4+CD25+ and CD3+CD56+ cells were fewer in AIH patients (5.2% and 10.8%) than in controls (10.0% and 18.1%; $p = 0.028$ and 0.012 respectively), and fewer still in [A] (1.2% for CD4+CD25+ and 6.8% for CD3+CD56+) than in [R] (5.8% and 11.3%; $p = 0.0006$ and 0.042 respectively). In contrast, CD8+CD28− and γδ percentages were higher in patients (33.3% and 4.8%) than in controls (23.8% and 2.5%; $p = 0.032$ and 0.037 respectively), and higher still in [A] (58.7% for CD8+CD28− and 10.3% for γδ) than in [R] (29.3% and 3.9%; $p = 0.004$ and 0.018 respectively). Azathioprine-treated patients tended to have fewer CD4+CD25+ (3.9%) than those on steroid mono-therapy (7.4%, $p = 0.082$). CD4+CD25+ FOXP3 and CTLA4 MFI was lower in AIH (28.3 and 76.6) than in controls (39.4 and 95.8; $p = 0.010$ and 0.036 respectively).
Conclusion: Adults with AIH not only have impaired CD4+CD25+ cells, as reported in children, but also a numerically reduced CD3+CD56+ pool, indicating a complex immuno-regulatory deficiency, likely to predispose to autoimmunity. The paradoxical behaviour of the CD8+CD28− and γδ populations requires further investigation.

862 DOES AUTOIMMUNE PANCREATITIS EXIST IN CHILDHOOD? A TWENTY-YEAR SINGLE CENTRE RETROSPECTIVE REVIEW

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Aim: Autoimmune pancreatitis (AIP), characterized by elevated serum IgG4 levels and response to immunosuppression, has been recently described in adults.
Patients and Methods: We have reviewed the clinical course, biochemical and radiological features of children with chronic pancreatitis (CP) referred to our unit between 1987 and 2007. Serum levels of immunoglobulins,