Background and Aims: Genes regulating butyrophilins are located in a juxtaposed-telomeric position of HLA-class-I locus on the short arm of chromosome 6. These genes form a cluster with several families: BTN, BTN2A1, BTN2A2, BTN2A3, BTN3A1 and BTN3A3. Previous studies have demonstrated that BTN3A2 could select HCV infection depending on viral genotype (Del Campo et al., AEEH 2013). Aim: To perform a fine mapping study of gene region encoding the butyrophilin family in HCV patients to uphold as genetic signals associated with infection from the different genotypes.

Methods: We included 1413 patients with HCV. Mean age: 46±11 years old; 60% (852/1410) were males; HCV-genotype-1 79% (1115/1413), genotype-2 4% (56/1413), genotype-3 11% (155/1413) and genotype-4 6% (90/1413). IL28B-CC: 33% (457/1394) and IL28B-TT: 67% (937/1394). Advanced fibrosis (F3–F4): 22% (224/1001) and genotype-4 6% (90/1413). IL28B-CC: 33% (457/1394) and IL28B-TT: 67% (937/1394), genotype-2 4% (49/1413), genotype-3 11% (159/1413) and genotype-4 6% (90/1413). Previous studies have demonstrated that BTN3A2 could select HCV infection depending on viral genotype (Del Campo et al., AEEH 2013). Aims: To perform a fine mapping study of gene region encoding the butyrophilin family in HCV patients to uphold as genetic signals associated with infection from the different genotypes.

**SNPs**

<table>
<thead>
<tr>
<th>SNPs</th>
<th>n</th>
<th>Genotype 1</th>
<th>Genotype 3</th>
<th>p</th>
<th>O.R. (95%CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>rs10946818</td>
<td>436</td>
<td>GG</td>
<td>CT/TT</td>
<td>0.01</td>
<td>2.17 (1.11–4.25)</td>
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<tr>
<td>rs6929846</td>
<td>731</td>
<td>CC</td>
<td>CT/TT</td>
<td>0.032</td>
<td>0.64 (0.46–0.99)</td>
</tr>
<tr>
<td>rs6456729</td>
<td>721</td>
<td>AC/AA</td>
<td>AG/AA</td>
<td>0.013</td>
<td>1.79 (1.09–2.96)</td>
</tr>
<tr>
<td>rs471108</td>
<td>587</td>
<td>CC</td>
<td>CT/TT</td>
<td>0.034</td>
<td>0.53 (0.29–0.97)</td>
</tr>
</tbody>
</table>

**Results:** Univariate analysis of the distribution of SNPs was carried out according to viral genotype (1&3) and the presence of steatosis. GG genotype of rs10946818 (BTN2A1) might be involved in the selection of hepatocyte steatosis in patients with chronic hepatitis C.

**Conclusions:** Baseline polymorphisms and emerging mutations were determined using population sequencing. In vitro SMV susceptibility was assessed in replication assays. Results: In the Phase 2b/3 studies, Q80K polymorphism was present in 76/1254 (6.1%) of GT1 patients from Europe: 19.4% (73/377) GT1a and 0.3% (3/977) GT1b. Consistent with limited effect of Q80K on SMV in vitro activity (~10 fold reduction), in studies C208/C216, 150 mg SMV/PR resulted in a mean change in HCVRNA from baseline to week 1 of −4.1 and −4.6 log10 IU/mL in GT1a patients with and without Q80K, respectively, compared to −1.1 log10 IU/mL with placebo/PR. Following the pronounced initial decline, treatment failure occurred more frequently in SMV-treated patients with Q80K than those without.

**Background and Aims:** Simeprevir (SMV) is an oral HCV NS3/4A protease inhibitor. Here we describe the in vitro/in vivo effect of NS3 Q80K polymorphism and emerging mutations in patients with genotype 1 (GT1) who failed treatment with 150 mg SMV plus peginterferon/ribavirin (PR).

**Methods:** Baseline polymorphisms and emerging mutations were determined using population sequencing. In vitro SMV susceptibility was assessed in replication assays. Results: In the Phase 2b/3 studies, Q80K polymorphism was present in 76/1254 (6.1%) of GT1 patients from Europe: 19.4% (73/377) GT1a and 0.3% (3/977) GT1b. Consistent with limited effect of Q80K on SMV in vitro activity (~10 fold reduction), in studies C208/C216, 150 mg SMV/PR resulted in a mean change in HCVRNA from baseline to week 1 of −4.1 and −4.6 log10 IU/mL in GT1a patients with and without Q80K, respectively, compared to −1.1 log10 IU/mL with placebo/PR. Following the pronounced initial decline, treatment failure occurred more frequently in SMV-treated patients with Q80K than those without.

**Conclusion:** Minority baseline polymorphisms did not impact treatment outcome of SMV with peginterferon/ribavirin. Emerging mutations became undetectable over time by PS and DS after SMV was stopped. These results suggest no added value of DS for clinical usage of SMV.
and/or 168 (mainly R155K in GT1a; D168V in GT1b) at failure conferring high-level resistance in vitro (fold change: ~50–2000). Emerging mutations were no longer detectable at end of study in 90/180 (50%) of patients (median follow-up: 28 weeks).

**Conclusions:** In SMV/PR studies, treatment failure was associated with emerging high-level resistance mutations. Presence of Q80K at baseline had minor effect on initial response to SMV/PR treatment but might have facilitated the emergence of additional mutations.

**P222**

**DISTINCT ESCAPE PATHWAY BY HCV GENOTYPE 1A FROM A DOMINANT CD8+ T CELL RESPONSE BY SELECTION OF ALTERED EPITOPE PROCESSING**

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**Background and Aims:** Antiviral CD8+ T-cells are a key component of the adaptive immune response against HCV, however, mutational escape by the virus contributes to viral persistence. Highly conserved immunodominant epitopes are attractive for T-cell based prophylactic vaccines. The HLA-B*51-restricted CD8+ T-cell epitope IPYGKAILT31-38 is highly conserved across most HCV genotypes. Here, we characterize the immune response and the selected escape pathways in the most prevalent genotypes 1a, 1b and 3a.

**Methods:** The frequency of CD8+ T-cell responses against the epitope IPYGKAILT31-38 was determined in 32 HLA-B*51-positive patients including 9 with spontaneous immune control of HCV infection. The epitope region was sequenced in >600 HCV-RNA-positive patients and the influence of viral polymorphisms on T-cell targeting, antigen processing, proteasomal digestion and replication capacity was determined.

**Results:** Responses against the epitope were exclusively detectable in patients infected with genotype 3a and in individuals with spontaneous immune control. Sequence analysis suggests that escape mutations are selected inside the epitope in genotype 1b and 3a. Interestingly, in genotype 1a a distinct substitution in the N-terminal flanking region located 5 residues up-stream of the epitope was selected (S1368P; p <0.0001). Functional assays revealed that the S1368P substitution impaired recognition of target cells presenting the endogenously processed epitope. Impaired recognition was the consequence of differential proteasomal degradation producing precursor peptides poorly cleavable by ER-aminopeptidases.

**Conclusions:** The mutational escape pathways from an immunodominant epitope in HCV NS5 depend on the viral genetic background. In genotype 1a a distinct substitution is selected associated with altered proteasomal digestion and epitope processing.

**P223**

**HLA-B*27 IS PROTECTIVE AGAINST HCV GENOTYPE 1 AND 3 AND ASSOCIATED WITH TARGETING OF DISTINCT GENOTYPE-SPECIFIC CD8+ T CELL EPITOPES**

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**Background and Aims:** Particular HLA class I-alleles influence the outcome of HCV infection. Alleles such as HLA-B*27 and HLA-B*57 were reproducibly protective from chronic HCV infection. The beneficial effect of these alleles depends on the presence of preeminent epitope sequences in immunodominant targets of the CD8+ T cell response. Accordingly, sequence differences between viral genotypes can be detrimental to the protective effect. Here, we analyzed the impact of HLA class I-alleles on the outcome of HCV-infection in a high-risk group exposed to HCV genotype 1 and 3.

**Methods:** A total of 310 anti-HCV positive individuals was recruited from a local cohort of people who inject drugs (PWID). HLA class I-alleles associated with absence of HCV-RNA were identified by multiple logistic regression analysis. Novel CD8+ T cell epitopes in HCV genotype 3a were identified by screening of predicted HLA-B*27-binding peptides.

**Results:** HLA-B*27 was significantly enriched in HCV-RNA negative PWID (8.8%) compared to PWID with genotype 1 (3.8%) or 3 (2.1%). The well-described immunodominant epitope in HCV genotype 1 (ARMILMTHF) was targeted in 3 of 13 HLA-B*27-positive patients. CD8+ T cells directed against the genotype 3a-specific sequence of this epitope (VRMVMTTHF) were nearly undetectable. We therefore screened the CD8+ T cell response against 28 predicted HLA-B*27 epitopes in genotype 3a and identified two novel vigorously targeted epitopes in NS2 (GRLIWWNQY and GRWFNTYLY).

**Conclusions:** In highly exposed PWID HLA-B*27 is protective against HCV genotype 1 and 3. The epitope repertoire differs between both genotypes indicating that different CD8+ T cell responses contribute to the protective effect.

**P224**

**DIFFERENT PROFILE OF NATURAL RESISTANCE TO NS3 PROTEASE INHIBITORS IN HEPATITIS C SUBTYPES 1A AND 1C**

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**Background and Aims:** Different rate of response to protease inhibitors based treatment has been observed in individuals infected by HCV-subtype 1a (S1a) and 1b, while no data are available on individuals infected by HCV-S1c.

**Methods:** In the present study were included 35 HIV/HCV-S1a (study group, SG) coinfected individuals. A group of 60 protease sequences, retrieved from GenBank, belonging to naive HCV-S1a monoinfected pts enrolled in clinical studies of telaprevir was used as sequence control (CG). Infecting subtype and resistance associated mutations (RAM) were characterized by analysing protease sequences in both groups.

**Results:** In the SG, 46% of sequences clustered with S1c. In the CG, 62% of sequences clustered with S1c. In the SG RAM were detected in 52.6% of S1a infected pts. and 18.7% of pts. harbouring