Association between the IL28B genotype and hepatitis C viral kinetics in the early days of treatment with pegylated interferon plus ribavirin in HIV/HCV co-infected patients with genotype 1 or 4

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Objectives: To evaluate the effect of the interleukin 28B (IL-28B) genotype on hepatitis C virus (HCV) viral kinetics in the first 4 weeks from start of treatment with pegylated interferon plus ribavirin (PEG-IFN/RBV) in HIV/HCV co-infected patients.

Methods: HIV/HCV co-infected patients naive to PEG-IFN/RBV treatment were enrolled in a prospective study. HCV RNA plasma viral loads were measured at baseline and at weeks 1, 2 and 4 after commencement of treatment. Patients were grouped by HCV genotype (genotype 1/4 versus 3) and by IL-28B genotype (CC versus non-CC). Differences in viral load reduction were evaluated by IL-28B genotype between baseline, week 1, week 2 and week 4.

Results: One hundred and nineteen HIV/HCV patients were included in the study. HCV patients with genotype 1/4 and bearing the IL-28 CC genotype showed the greatest reductions in HCV RNA plasma levels between baseline and weeks 1 (B-1), 2 (B-2) and 4 (B-4) than did those with non-CC genotypes (B-1: 1.06 ± 0.89 versus 0.48 ± 0.48 log IU/mL, P = 0.009; B-2: 1.36 ± 0.72 versus 0.77 ± 0.66 log IU/mL, P = 0.01; and B-4: 1.91 ± 0.64 versus 1.38 ± 0.96 log IU/mL, P = 0.03). However, differences between weeks 1 and 2 (W1-2) and between weeks 2 and 4 (W2-4) were not associated with the IL-28B genotype (W1-2: CC 0.48 ± 0.42 versus non-CC 0.38 ± 0.38 log IU/mL, P = 0.62; W2-4: CC 0.32 ± 0.23 versus non-CC 0.39 ± 0.31 log IU/mL, P = 0.67). No differences in decline of HCV RNA viral load were found in HCV genotype 3 patients.

Conclusions: The IL-28B genotype impacts on viral kinetics during the first week of treatment with PEG-IFN/RBV in patients with HCV genotype 1/4.

Keywords: interleukin 28B, HIV, HCV, pharmacogenetics, viral kinetics

Introduction

The hepatitis C virus (HCV) is the cause of one of the most common blood-borne infections worldwide.1 It is considered to be the leading cause of cirrhosis and liver cancer in Europe and the USA.1 In HIV co-infected patients, the current standard of care is pegylated interferon (PEG-IFN) plus ribavirin (PEG-IFN/RBV), although rates of sustained virological response (SVR) vary significantly according to virus, disease and a host of other related factors.1

Several studies have provided information about the clinical applicability of early viral kinetics in predicting SVR. Rapid virological response (RVR), defined as an undetectable serum HCV RNA level at week 4 of treatment with PEG-IFN/RBV, is a reliable predictor of SVR.2 At the same time, it has been demonstrated that polymorphisms near the interleukin 28B (IL-28B) gene on chromosome 19 predict SVR and RVR in both HCV mono-infected and HIV/HCV co-infected patients carrying genotype 1/4 and treated with PEG-IFN/RBV.3,4

Several studies suggest that the effect of the IL-28B genotype on HCV viral kinetics can be seen in the first few days from start of treatment.4,5 However, there is limited information about the IL-28B effect after treatment has started, and no data about its effect on HIV/HCV co-infected patients.6,7 The objective of this study was to evaluate the effect of the IL-28B genotype on HCV viral kinetics in the first few weeks after starting treatment with PEG-IFN/RBV in HIV/HCV co-infected patients.
IL28B CC genotype is associated with a greater viral load reduction

Methods

Patients
Caucasian HIV-infected patients with chronic hepatitis C, naive to HCV treatment and receiving a PEG-IFN/RBV combination therapy, were included in this prospective study. The criteria used to determine hepatitis C therapy were in accordance with international guidelines. Host, clinical and virological characteristics were collected.

Treatment regimens
All individuals were treated with either PEG-IFN-α2a or PEG-IFN-α2b, at doses of 180 μg or 1.5 μg/kg per week, respectively, in combination with a weight-adjusted dose of oral ribavirin (1000 mg/day for <75 kg, 1200 mg/day for ≥75 kg).

Virological evaluation
Plasma HCV RNA load measurements were taken at baseline and at weeks 1, 2 and 4 using a quantitative PCR assay (Cobas TaqMan, Roche Diagnostic Systems Inc., Pleasanton, CA, USA) with a detection limit of 15 IU/mL.

Determination of the IL-28B genotype
The rs129879860 single-nucleotide polymorphism (SNP) was genotyped using a custom TaqMAN genotyping assay (Applied Biosystems) on DNA isolated from whole blood samples. The DNA was genotyped according to manufacturer’s instructions on an MX3005 thermocycler using MXpro software (Stratagene). Researchers responsible for genotyping procedures were unaware of other patient data. The IL-28B genotype was defined as CC or non-CC (TT/CT).

Statistical analysis
Continuous variables are expressed as mean± standard deviations or medians (Q1–Q3), and were analysed by Student’s t-test or the Mann–Whitney U-test. Categorical variables are expressed as the number of cases (percentage). Frequencies were compared using the χ² test or Fisher’s exact test. Significance was defined as a P value <0.05. The subpopulation of patients with HCV genotype 4 was evaluated together with the HCV genotype 1 subpopulation. Reductions in plasma HCV RNA were evaluated by the IL-28B genotype between baseline, week 1, week 2 and week 4. Patients who presented an undetectable HCV viral load at any timepoint during the study were excluded when calculating HCV RNA reduction at any later point. Patients were classified as slow or fast responders, according to whether the reduction of HCV RNA viral load was above or below the median. The decline in HCV RNA was calculated according to the stage of liver fibrosis, HCV baseline viral load was above or below the median. The decline in HCV RNA was calculated according to the stage of liver fibrosis, HCV baseline viral load and PEG-IFN type. The analysis was performed using the SPSS statistical software package, version 15.0 (SPSS).

Ethical aspects
The study was designed and performed according to the Helsinki Declaration and approved by the ethics committee of the Reina Sofia University Hospital, Cordoba, Spain. All patients provided written informed consent before participating in this study.

Results
One hundred and nineteen HIV/HCV co-infected patients were included in the study. The baseline characteristics of the patients are summarized in Table 1.

Table 1. General population characteristics

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<th>Characteristics</th>
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<tr>
<td>Log HCV RNA baseline, mean± SD</td>
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<td>1, 2 and 4 using a quantitative PCR assay (Cobas TaqMan, Roche Diagnostic Systems Inc., Pleasanton, CA, USA) with a detection limit of 15 IU/mL.</td>
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HCV genotype 1/4 patients carrying the IL-28 CC genotype showed a greater reduction in plasma HCV RNA levels between baseline and weeks 1 (B-1), 2 (B-2) and 4 (B-4) than did non-CC genotype carriers (B-1: 1.06±0.89 versus 0.48±0.48 log IU/mL, P=0.009; B-2: 1.36±0.72 versus 0.77±0.66 log IU/mL, P=0.01; and B-4: 1.91±0.64 versus 1.38±0.96 log IU/mL, P=0.03) (Figure 1a). However, no differences in HCV RNA reduction were found by the IL-28B genotype between weeks 1 and 2 (CC 0.48±0.42 versus non-CC 0.38±0.38 log IU/mL, P=0.62) or between weeks 2 and 4 (CC 0.32±0.23 versus non-CC 0.39±0.31 log IU/mL, P=0.67) in HCV genotype 1/4 patients.

In HCV genotype 3 patients, there were no observable differences of viral load reduction found between B-1, B-2 or B-4, irrespective of whether the IL-28B genotype was CC or non-CC (B-1: 1.81±1.06 versus 1.65±0.93 log IU/mL, P=0.64; B-2: 2.73±0.88 versus 3.15±1.4 log IU/mL, P=0.28; and B-4: 3.75±0.88 versus 4.04±1.06 log IU/mL, P=0.45) (Figure 1b).
Median HCV RNA reductions for HCV genotype 1/4 patients at week 1 were 1.28 log IU/mL (0.89–1.37), respectively. The IL-28B CC genotype was more common in patients who were fast responders than in slow responders at all timepoints analysed [B-1: 39/51 (76.4%) versus 10/58 (17.2%), P<0.001; B-2: 42/54 (77.7%) versus 7/55 (12.7%), P<0.001; and B-4: 42/54 (77.7%) versus 7/55 (12.7%), P<0.001].

Figure 1. Mean reduction in viral load from baseline according to IL-28B genotype in HCV genotype 1/4 (a) and genotype 3 (b) patients.

A total of 37 (31.1%) patients attained RVR, and at significantly lower rates for genotype 1/4 than for genotype 3 [16/81 (19.7%) versus 21/38 (55.2%), P=0.002]. Among patients bearing genotype 1/4, the RVR rate was significantly higher for IL-28B CC genotypes than for non-CC [9/31 (29.03%) versus 7/50 (14%), P=0.01]. However, in patients bearing genotype 3, there were no observable differences in RVR rate between IL-28B CC and non-CC genotypes [10/18 (55%) versus 11/20 (55%), P=0.985].

No differences in HCV RNA reduction were found by liver fibrosis stage, baseline viral load or PEG-IFN type.

Discussion

In this study, the host IL-28B genotype was a pre-treatment predictor of HCV RNA kinetics in HIV/HCV co-infected patients carrying genotype 1/4 and who started therapy with PEG-IFN/RBV. The IL-28B CC genotype was associated with a greater decline in on-treatment viral load compared with patients who harboured the IL-28B non-CC type. This difference was observed as early as the first week from start of therapy and was maintained during the first 4 weeks of treatment.

As previously reported, a small but not clinically significant difference in median plasma HCV RNA load at baseline has been noted for the IL-28B genotype, with higher levels in CC patients. It has been suggested that, as the product of the IL-28B gene is IFN-λ3, patients harbouroing the IL-28B rs12979860 allele C might exhibit lower immune activity, permitting higher HCV replication. Consequently, such patients might retain an enhanced susceptibility to exogenous PEG-IFN-based therapy.

Viral decline during IFN therapy is biphasic. The first phase occurs during the first 72 h from start of therapy, with a slower second phase. In one clinical trial, HCV viral load was measured at baseline and 24 h following a test dose of 9 MU IFN-α2a, and HCV RNA reduction after 24 h was used to randomly stratify patients carrying genotype 1. The decline in plasma HCV RNA load after 24 h was much greater in IL-28B CC genotype than in non-CC genotype carriers. Similar data were obtained for patients carrying genotype 4. In another study, differences in HCV RNA load reduction between IL-28B CC, CT and TT genotypes were detectable at week 2, the earliest timepoint evaluated. In our study we found differences in HCV RNA load reduction between IL-28B CC and non-CC genotypes from baseline at every timepoint analysed, although no difference in HCV RNA load reduction was found between week 1 and week 2, or week 2 and week 4. This is an important issue because it implies that the effect of the IL-28B genotype occurs during the first phase of viral decay, thought to be due to the process of free virion clearance and the inhibition of new viral production rather than the slower second phase, whose gradient is thought to mirror the process of infected cell clearance.

The fastest reductions in viral load correlated with increased rates of the appropriate on-treatment virological endpoint, such as RVR or complete early virological response. At the same time, RVR has been demonstrated to be a critical predictor of SVR, independent of the host IL-28B genotype. Our observations suggest that the major effect of this polymorphism is to increase the rate of early viral decline, leading to a higher RVR rate.

We found that the magnitude of viral decline for patients carrying genotype 1 or 4 was lower than for genotype 3 at all timepoints in the study. Differences in viral load reduction across the HCV genotypes were detectable as early as week 1, confirming that genotype is the most important factor for predicting an early response to antiviral therapy.

Data concerning the relevance of the IL-28B genotype in patients carrying HCV genotype 3 are emerging. Mangia et al. observed that genotype 2/3 patients with the IL-28B CC genotype who failed to achieve RVR were more likely to achieve SVR than their non-CC-type counterparts. In our study, the host IL-28B genotype in genotype 3 carriers was not found to have a significant impact on HCV viral kinetic response in the first few days of treatment. On-treatment viral kinetics provides a direct measurement of treatment response. The virological response to treatment for HCV genotype 3 is usually very strong and early, and this might limit the advantage conferred by a favourable IL-28B genotype.
A better-powered cohort is necessary to find statistically significant associations.

In summary, in HIV co-infected patients, SNP rs12979860 in the region of the IL-28B gene has an impact on early viral kinetics in response to PEG-IFN/RBV therapy for chronic hepatitis C caused by HCV genotypes 1 and 4. At present, as with viral genotype, information about IL-28B status is being used to inform patients about the likelihood of obtaining a response to PEG-IFN/RBV combination therapy. Whether determining viral kinetics during the first week from start of treatment will be a better predictor of response to HCV combination therapy, and whether this should be used for making decisions about treatment in such patients, remains to be studied.

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Transparency declarations
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References