IFNL4 ss469415590 Variant Is a Better Predictor than ILF3 (IL28B) rs12979860 of PegIFN-α/RBV Therapy Failure in HCV/HIV-1 Coinfected Patients

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BACKGROUND

Genome-wide association studies showed that single nucleotide polymorphisms (SNPs) nearby IL28B (principally rs12979860) are strongly associated with spontaneous[a] and pegIFN-α/RBV treatment-induced clearance of hepatitis C virus (HCV) infection[b]. We and numerous other investigators confirmed the importance of IL28B SNPs in the treatment of HCV infection in HCV/HIV-1 coinfected patients[c]. Recently, it has been discovered, upstream of the interferon-λ3 protein (IFNL3) (also termed IL28B) on chromosome 19, a new transiently induced region that harbors a dinucleotide variant ss469415590 (TT or ΔG)[d], which is in high linkage disequilibrium with rs12979860. ss469415590 (ΔG) is a frameshift variant that creates a novel gene, designated IFNL4, which is moderately similar to IFNL3. Importantly, ss469415590 may have a functional role in the innate immune response to HCV. Since there are no data about the impact of IFNL4 ss469415590 polymorphisms on treatment outcomes in HCV/HIV-1 coinfected patients, we decide to investigate the role of the ss469415590 SNP in a well characterized cohort of HCV/HIV-1 coinfected patients that were treated with pegIFN-α/RBV. This information may be essential to better predict the response to therapy and potentially to enable better selection of patients for treatment.

METHODS

ss469415590 and rs12979860 genotyping was performed using the ABI TaqMan allelic discrimination kit and the ABI7500HT Sequence Detection System (Applied Biosystems). The association of ss469415590 and rs12979860 polymorphisms with response to treatment was performed by univariate and multivariate logistic regression as implemented in the SNPStats software[e].

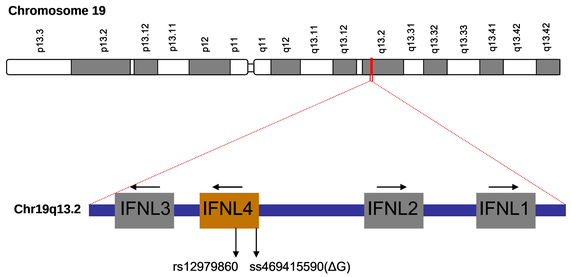


Figure 1. Schematic representation of both SNPs on the chromosome 19. (*Modified from Horner SM and Gale M Jr, Nat Med 2013; 19(7): 879-888.)*

RESULTS

ss469415590 and rs12979860 polymorphisms were in strong linkage disequilibrium (R2 = 0.95) and both had a minor allele frequency of 0.32. The ss469415590 minor allele frequency was identical to that found for Europeans in the HapMap samples (0.07 frequency in Asians, 0.32 frequency in Europeans and 0.73 frequency in Africans) (Table 1).

Table 1. Allele frequencies of both SNPs compared with HapMap samples.

	Alleles rs12979860		Alleles ss469415590	
	C	T	G	TT
HapMap-CEU	0.683	0.317	0.322	0.678
HapMap-YRI	0.278	0.722	0.783	0.217
HapMap-CHB JPT	0.933	0.067	0.067	0.933
Our HCV/HIV-1 cohort	0.684	0.316	0.319	0.681

HapMap samples from dbSNP (www.ncbi.nlm.nih.gov/projects/SNP): CEU (European), YRI (Africans), CHB JPT(Asians); Allele frequency was calculated with SNPStats software[e].

The prevalence of the ss469415590 TT/ΔG substitution, as well as the rs12979860 polymorphism, was analyzed in a cohort of 207 patients from our clinic with chronic HCV/HIV-1 coinfection and with a known response to treatment with pegIFN-α/RBV at 24 weeks post-treatment (Table 2). A significant higher HCV RNA viral load was observed in patients who failed treatment. Likewise, age and HCV genotype were significantly associated with treatment failure (Table 2).

Table 2. Clinical characteristics of 207 patients with chronic HCV/HIV-1 coinfection and treated with pegIFNα/RBV.

	Treatment success	Treatment failure	*p-value*
Patients, n (%)	73 (35%)	134 (65%)	-
Age (mean ± SEM)	43.73 ± 0.7553	41.04 ± 0.5023	0.0035
Gender, n (%)			0.0001
Female	23 (39%)	36 (61%)	
Male	50 (34%)	98 (66%)	
CD4+ (cell counts/ul) (mean ± SEM)	621 ± 33.68	566 ± 23.74	ns
HCV genotype, n (%)			0.0002
1	29 (27%)	79 (73%)	
3	34 (58%)	25 (42%)	
4	10 (26%)	28 (74%)	
Fibrosis stage, n (%)			ns
0-1	27 (35%)	49 (64%)	
2	17 (38%)	28 (62%)	
3	16 (33%)	33 (67%)	
4	6 (29%)	15 (71%)	
ALT (U/L) (mean±SEM)	102 ± 10.47	88 ± 5.035	ns
AST (U/L) (mean±SEM)	63 ± 5.457	66 ± 3.371	ns
HCV RNA (IU/mL) (mean±SEM)	5.684 ± 0.09192	5.921 ± 0.04415	0.0093
Undetectable HIV-1 RNA, n (%)	52 (70.3%)	89 (66.4%)	ns

Age, Mann-Whitney U test; Gender, Chi-square test; CD4+T cell count, Mann-Whitney U test; HCV genotype, Chi-square test; Fibrosis stage, Chi-square test; ALT and AST, Mann-Whitney U test; HCV RNA, unpaired t test; Undetectable HIV-1 RNA, Chi-square test.

We next correlated the ss469415590 and rs12979860 genotypes with the pegIFN-α/RBV treatment response. Both polymorphisms were associated with response to treatment. However, ss469415590 had a more significant association with SVR (Table 3). ss469415590 had also a stronger association in a multivariate model, after adjustment for age, sex, fibrosis stage and HCV RNA load than rs12979860 (Table 3).

Table 3. Association of ss469415590 and rs12979860 polymorphisms with HCV SVR in chronic HCV/HIV-1 coinfected patients.

				Association with SVR		Association with SVR (adjusted by sex, age, fibrosis stage, and HCV RNA load)	
Variant	**Genotype**	**n**	**SVR n (%)**	**OR[a] (95% CI)**	***p-value***	**OR[a] (95% CI)**	***p-value***
All HCV genotypes		207					
ss469415590	TT/TT	99	48 (66)				
	TT/ΔG	84	21 (29)				
	ΔG/ΔG	24	4 (5)	3.12 (1.72-5.67)	0.0001	3.56 (1.80-7.03)	0.0002
rs12979860	CC	99	46 (63)				
	CT	85	23 (32)				
	TT	23	4 (5)	2.60 (1.45-4.69)	0.0012	2.99 (1.53-5.84)	0.001
HCV genotype 1 and 4		146					
ss469415590	TT/TT	58	26 (67)				
	TT/ΔG	69	11 (28)				
	ΔG/ΔG	19	2 (5)	4.69 (2.14-10.27)	0.0001	5.18 (2.11-12.71)	0.0002
rs12979860	CC	60	26 (67)				
	CT	69	12 (31)				
	TT	17	1 (3)	4.29 (1.97-9.37)	0.0002	4.94 (2.02-12.07)	0.0003
HCV genotype 3		59					
ss469415590	TT/TT	40	22 (65)				
	TT/ΔG	15	10 (29)				
	ΔG/ΔG	4	2 (6)	0.71 (0.23-2.19)	0.55	0.62 (0.14-2.67)	0.51
rs12979860	CC	38	20 (59)				
	CT	16	11 (32)				
	TT	5	3 (9)	0.56 (0.18-1.68)	0.29	0.42 (0.10-1.79)	0.23

CI, confidence interval; HCV, hepatitis C virus; OR, odds ratio; SVR, sustained viral response.

[a]ORs, 95% CIs and p-values are for a dominant model in which s single copy of the minor allele is sufficient to modify the risk.

Similar results were observed when samples were stratified by HCV viral genotypes (Table 3). In patients infected with HCV genotypes 1/4, the strongest association with SVR was also found for ss469415590 either in the univariate or in the multivariate model.

DISCUSSION AND CONCLUSION

This is the first study that analyse the association of IFNL4 ss469415590 genotype variants with the response to pegIFN-α/RBV treatment in HCV/HIV-1 coinfected patients. These results demonstrate that ss469415590 is a better marker of sustained virological response than rs12979860 in HCV/HIV-1 coinfected patients. IFNL4 genotype may be a useful tool to select patients for IFN-α-based therapies.

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