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ITPA Gene Polymorphisms Significantly Affect Hemoglobin Decline and Treatment Outcomes in Patients Coinfected With HIV and HCV

Anu Osinusi[1,2,*], **Susanna Naggie**[3], **Seerat Poonia**[1], **Martin Trippler**[4], **Zonghui Hu**[5], **Emily Funk**[1], **Joerg Schlaak**[4], **Dawn Fishbein**[2], **Henry Masur**[6], **Michael Polis**[1], and **Shyam Kottilil**[1]

[1]Laboratory of Immunoregulation, National Institute of Allergy and Infectious Disease, NIH, Bethesda, Maryland [2]CMRP, SAIC-Frederick Inc, NCI-Frederick, Frederick, Maryland [3]Duke Clinical Research Institute, Durham, North Carolina [4]University Hospital of Essen, Essen, Germany [5]Biostatics Research Branch, NIAID, NIH, Bethesda, Maryland [6]Critical Care Medicine Dept, NIH, Bethesda, Maryland

Abstract

Published studies have described a strong association with a single-nucleotide polymorphism (SNP) in the inosine triphosphate pyrophosphatase (ITPA) gene and ribavirin (RBV)-induced hemolytic anemia in HCV-infected patients receiving pegylated interferon (pegIFN) and RBV. This study sought to evaluate the effect of these polymorphisms on anemia, hemoglobin reduction, HCV kinetics, and treatment outcomes. Sixty-three patients coinfected with HIV and HCV and 58 patients infected with HCV only were treated with pegIFN/RBV were genotyped using the ABI Taq-Man allelic discrimination kit for the 2 ITPA SNP variants rs1127354 and rs7270101. A composite variable of ITPA deficiency using both SNPs was created as previously reported. Statistical analysis was performed using Mann-Whitney test or Chi square/Fishers exact test for categorical data and mixed model analysis for multiple variables. Thirty-five patients (30%) were predicted to have reduced ITPA activity. ITPA deficiency was found to be protective against the development of hemoglobin reduction >3 g/dl over the course of treatment. The rates of hemoglobin reduction >3 g/dl decreased in correlation with the severity of ITPA deficiency. ITPA deficiency was associated with slower hemoglobin decline early in treatment (week 4, $P = 0.020$) and rapid virologic response (RVR) at week 4 ($P = 0.017$) in patients coinfected with HIV and HCV. ITPA polymorphisms are associated with hemoglobin decline and in patients coinfected with HIV and HCV it is also associated with early virologic outcomes. Determination of ITPA polymorphisms may allow prediction of RBV-induced anemia and earlier initiation of supportive care to ensure optimal therapeutic outcomes.

Keywords

ribavirin-induced hemolytic anemia; ITPA; HIV/HCV; pharmacogenomics

[*]Correspondence to: Anu Osinusi, Bldg10, Rm.6A-33A 10 Centre Drive, Bethesda, MD 20892. osinusia@niaid.nih.gov.
Anu Osinusi and Susanna Naggie contributed equally to this work.

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INTRODUCTION

Hepatitis C virus (HCV) affects an estimated 180 million individuals worldwide and over 4 million in the United States [Armstrong et al., 2006]. Hepatitis C is the principal cause of death from liver disease and remains the leading indication for liver transplantation in the United States [Alter, 1999; Kim, 2002]. Of the 1 million people infected with HIV in the United States, about a third are co-infected with HCV [Sherman et al., 2002]. While the successful implementation of antiretroviral therapy (ART) has decreased the number of AIDS-related opportunistic infections, morbidity, and mortality rates related to HCV-related liver disease has become a leading cause of death in this population [Selik et al., 2002; Jain et al., 2003]. Furthermore, patients coinfected with HIV and HCV have an accelerated progression of liver disease and therapeutic response rates with pegylated interferon (pegIFN) and ribavirin (RBV) that are lower than patients infected with HCV only [Sulkowski, 2001; Soriano et al., 2004].

Treatment with peg-IFN/RBV for hepatitis C is characterized by high rates of toxicities including psychiatric illness, constitutional side effects, and cytopenias. Furthermore, in patients coinfected with HIV and HCV ART and prophylactic medications contribute to cytopenias making patients more susceptible to hematologic toxicity from pegIFN/RBV. The major adverse drug events associated with the use of RBV are anemia due to extracellular hemolysis and suppression of the bone marrow release of erythroid elements [Gilbert and Knight, 1986; Dusheiko et al., 1996]. RBV-induced anemia can be severe enough to warrant the addition of erythropoietin (EPO) to treatment, RBV dosage reduction, or even discontinuation of treatment, which may affect overall efficacy of treatment. In HCV treatment studies, RBV dose adjustment was required in 22–40% of patients with rates as high as 50% observed in patients coinfected with HIV and HCV [Jeffers et al., 2004; Alvarez et al., 2006; Conjeevaram et al., 2006; Jacobson et al., 2007a,b; Opravil et al., 2008].

A genome wide association study (GWAS) showed a strong association between single-nucleotide polymorphisms (SNP) of the inosine triphosphate pyrophosphatase (ITPA) gene and treatment-induced anemia in a cohort of patients infected with HCV genotype 1 [Fellay et al., 2010]. The identified SNP was found on chromosome 20 and is in linkage disequilibrium with two SNPs rs1127354 (missense variant in exon 2) and rs7270101 (splicing-altering SNP in second intron) described previously. Similar findings have also been reported in various ethnicities of Caucasian, African-American, and Asian patients and in the setting of telaprevir use, with limited reports in patients coinfected with HIV and HCV [Sakamoto et al., 2010; Thompson et al., 2010; Hitomi et al., 2011; Suzuki et al., 2011]. The proposed mechanism of this genetic variation is one of conferred protection against RBV-induced reduction of ATP, by substituting ITP for GTP, which is depleted by RBV and results in hemolytic anemia [Hitomi et al., 2011].

This study sought to confirm and extend the understanding of this genetic association in a cohort of patients coinfected with HIV and HCV in comparison to patients infected with HCV only. This study explored the association between ITPA variants and the development of anemia, hemoglobin decline and the use of growth factor supplementation and explored the relationship between ITPA variants and therapeutic response.

MATERIALS AND METHODS

Study Patients

Group A: Patients coinfected with HIV and HCV—Three prospective, pilot, single center, open label trials were performed at the National Institute of Allergy and Infectious Diseases (NIAID), National Institutes of Health from 2001 to 2010. Sixty-five treatment naive patients coinfected with HIV and HCV were treated with either weekly injections of pegIFN-α-2a (Pegasys, Genentech, South San Francisco, CA) or pegIFN-α-2b (Peg-Intron, Merck & Co., Inc, Whitehouse Station, NJ) or biweekly albinterferon-α-2b with weight-based RBV daily (at 1,000 mg dose <75 kg, 1,200 mg dose for >75 kg). Patients who had less than a 2-log reduction in HCV RNA by week 12 or a detectable HCV RNA at 24 weeks were considered to be non-responders and therapy was stopped. EPO therapy was permitted to maintain hemoglobin over 10.0 g/dl. Therapy with RBV was modified if anemia (hemoglobin <10 g/dl) did not respond to EPO by decreasing RBV by 200–400 mg. No patients required a reduction in the dose of RBV in these studies. All patients gave written informed consent including provision of genetic material as approved by the NIAID Institutional Review Board prior to enrollment in the studies.

Group B: Patients infected with HCV only—Fifty-eight patients infected with HCV only were treated at the University of Essen, Essen, Germany as part of standard of care with either weekly pegIFN-α-2a or pegIFN-α-2b and weight based RBV daily (at 1,000 mg dose <75 kg, 1,200 mg dose for >75 kg). Specific dose-adjustments for RBV were per clinic protocol, which excluded the use of erythropoietin stimulating agents (ESAs). In summary the dose of RBV was reduced by 200 mg decrements once hemoglobin dropped below 10 g/dl and was repeated until hemoglobin stabilized. All patients gave written informed consent approved by the local ethics committee prior to enrollment.

Laboratory Evaluations

Laboratory tests and immune profiles including $CD4^+$ T cell counts were obtained at baseline and frequently during the entire study. HCV RNA was measured during all study visits, however, these were more frequent in the patients coinfected with HIV and HCV at days 0, 1, 3, 7, 10, week 2, week 3, week 4, week 6, week 8, and then every 4 weeks until week 48). For both cohorts HCV RNA concentration in plasma was measured by Versant HCV RNA 3.0 assay [quantitation range of 615–7.7×10^6 HCV RNA IU/ml] (Bayer Diagnostics, Puteaux, France).

ITPA Genotyping

Genotyping was performed by the Duke Centre for Human Genome Variation. The two functional variants in the ITPA gene, rs1127354 and rs7270101 were genotyped with the ABI TaqMan allelic discrimination kit and the ABI7900HT sequence detection system (Applied Biosystems, Carlsbad, CA), as described by Thompson et al. [2010]. The possible genotypes for each biallelic polymorphism are as follows: C/C, A/C, and A/A (A is the minor allele) for rs1127354 and A/A, A/C, and C/C (C is the minor allele) for rs7270101.

ITPase activity was defined by the presence of minor alleles at the respective polymorphic sites as described previously [Thompson et al., 2010, 2011]. ITPA deficiency has been grouped into none; mild (>60% activity); moderate (30–60% activity); and severe (<30% activity) (Table I).

IL28B Genotyping

This cohort was also tested for the *IL28B* SNP rs12979860 on chromosome 19 in a blinded fashion on DNA specimens collected from each individual, using the 5′ nuclease assay with allele-specific TaqMan probes (ABI TaqMan allelic discrimination kit and the ABI7900HT Sequence Detection System (Applied Biosystems, Carlsbad, CA). Genotyping calls were inspected and verified prior to release.

Study Objectives

The primary objective was to assess the association of ITPA variants with a composite variable of anemia (hemoglobin <10 g/dl) at week 4. Other objectives included the association with hemoglobin decline >3 g/dl over the course of treatment; HCV viral load decline over time and treatment response including rapid virological response [undetectable HCV RNA at week 4], early virological response [>2log$_{10}$ decline in HCV RNA at week 12] and sustained virological response [undetectable HCV RNA 6 months after the end of treatment].

Statistical Analysis

For descriptive statistics, continuous variables were reported as median (25th to 75th percentiles). Categorical variables were represented as frequencies and percentages. Comparisons between groups for demographic and clinical data were performed using the *t*-test for continuous variables and Fisher's exact test or Chi square test for categorical variables. Statistical significance was defined as $P < 0.05$ with a two-tailed test. Statistical tests were done using Graphpad Prism 5.0 software. Logistic regression models were used to evaluate the association between a binary response and other variables, where the estimated odds ratios and *P*-values were reported. Linear mixed-effects models were used for estimating the time course of hemoglobin and HCV, where the slopes of the changes from baseline were reported and compared between ITPA normal and ITPA deficient groups.

RESULTS

Patient Characteristics

The study consisted of 123 patients whose clinical characteristics are represented in Table II. Of the study population 58 (47.2%) were patients infected with HCV only and 65 (52.8%) were patients coinfected with HIV and HCV. The monoinfected cohort was largely Caucasian, while patients coinfected with HIV and HCV was majority African-American and male. Patients coinfected with HIV and HCV had higher baseline HCV RNA and more advanced fibrosis. Both cohorts were similar with regards to age, *IL28B* genotype, baseline hemoglobin, platelet counts, HCV genotype, and creatinine levels. Patients coinfected with HIV and HCV had well-controlled HIV infection with >80% treated with HAART of which 87% were virologically suppressed. ITPA genotyping was not successful in five patients.

Population Distribution of ITPA Variants

The ITPA polymorphisms rs1127354 and rs7270101 have been well characterized and validated as functional variants in studies of patients with ITPase deficiency [Fellay et al., 2010]. The majority of patients (70%) were predicted to have wild-type ITPAse activity but a significant minority (30%) was predicted to have reduced ITPAse activity (12% mild, 16% moderate, 2% severe; Table I). There was no significant difference in the prevalence of ITPA deficiency between HCV monoinfected and patients coinfected with HIV and HCV although moderate to severe deficiency (≤30% activity) occurred more often in HCV monoinfected patients (22% vs. 14%, $P > 0.05$) possibly due to the different racial components of both cohorts.

Anemia Occurs at Higher Rates in Patients Coinfected With HIV and HCV

Anemia (hemoglobin <10 g/dl) was more common in patients coinfected with HIV and HCV occurring in 29 (46%) of patients compared to 7 (13%) patients infected with HCV only ($P = 0.0001$). This was seen at all relevant clinical endpoints (week 4: 19% vs. 4%, $P = 0.011$; week 12: 32% vs. 7%, $P = 0.001$). Patients coinfected with HIV and HCV with normal ITPA activity experienced significantly more anemia than similar patients infected with HCV only (50% vs. 14% $P = 0.0010$) compared to patients with ITPA deficient variants (HIV/HCV: 30% vs. HCV 11%, $P = 0.1122$).

ITPA Deficiency Protects Against Hemoglobin Decline

Week 4

Hemoglobin reduction >3 g/dl: Both ITPA variants and the composite ITPA deficiency variables were evaluated for an association with hemoglobin reduction >3g/dl. In patients infected with HCV only, there was a lower incidence of hemoglobin reduction >3 g/dl in patients with ITPA deficiency compared to normal ITPA activity at week 4 (5% vs. 37%, $P = 0.0098$) which was not significant in patients coinfected with HIV and HCV (27% vs. 46%, $P = 0.2382$; Fig. 1A). This association of ITPA deficiency with hemoglobin reduction >3 g/dl at week 4 was independent of HCV genotype.

Anemia: While there was a lower incidence of anemia in both patients coinfected with HIV and HCV and patients infected with HCV only with ITPA deficiency variants [HIV/HCV: 13% vs. 21% ($P = 0.714$); HCV: 0% vs. 6% ($P = 0.529$)] at week 4, this was not significant (Fig. 1B).

Course of treatment

Hemoglobin reduction >3g/dl: ITPA deficiency variants were protective against the development of hemoglobin reductions >3 g/dl through the course of therapy in both cohorts (Fig. 1A). Increased severity of ITPA deficiency with resulting enzyme activity ≤30%, was associated with the greatest protection against hemoglobin reduction in both cohorts (Fig. 1C). Protection against anemia by ITPA deficiency variants was also observed throughout the course of therapy but this did not meet statistical significance (Fig. 1B).

AZT use: Among patients coinfected with HIV and HCV, there were 10 patients (15.4%) receiving AZT while on HCV treatment (6 (12%) with normal ITPA activity and 4 (26%) with ITPA deficiency). There was a trend towards increased hemoglobin reduction >3 g/dl and the development of anemia on treatment in patients who were exposed to AZT (hemoglobin reduction >3 g/dl: 80% vs. 66%, $P = 0.384$; anemia 70% vs. 46% $P = 0.097$).

Rate of Hemoglobin decline: There was a slower hemoglobin decline over the course of therapy in patients with ITPA deficient variants; this was most notable in the first 4 weeks in patients coinfected with HIV and HCV and first 12 weeks in patients infected with HCV only (Fig. 2A). Overall ITPA deficient variants were significantly associated with hemoglobin decline and reduction >3 g/dl, over the course of treatment on univariate analysis. However, this significance was lost in multivariate analysis due to small sample size. In multivariable analysis of patients coinfected with HIV and HCV, baseline hemoglobin was the only variable associated with the development of anemia. Variables such as age, gender, BMI, baseline creatinine, platelets, baseline CD4 count, baseline HIV viral load, baseline HCV viral load, fibrosis, and AZT use were not associated with the development of anemia (Table III).

ITPA Deficiency Is Associated With RVR in Patients Coinfected With HIV and HCV

Treatment outcomes—ITPA deficiency variants were tested for an association with treatment outcomes using a logistic regression model including *IL28B* genotype and other known baseline predictors of treatment response. In patients coinfected with HIV and HCV genotype 1, ITPA deficiency was associated with rapid virologic response (RVR; $P = 0.017$) but not early virologic response or sustained virologic response ($P > 0.05$; Table IV). This was not seen in non-genotype 1 patients or patients infected with HCV only. The only other variable associated with RVR was baseline HCV viral load. *IL28B* was associated with EVR but no other outcomes.

HCV viral kinetics—Due to frequent early viral kinetic sampling, the impact of ITPA deficiency on early viral decay in patients coinfected with HIV and HCV was assessed which showed no significant difference in viral kinetics based on ITPA polymorphisms (Fig. 2B).

DISCUSSION

This study demonstrates that ITPA deficiency renders protection against the development of RBV-induced hemoglobin decline in patients infected with HCV only and in patients coinfected with HIV and HCV. Furthermore, ITPA variants were also associated with early virologic responses in patients coinfected with HIV and HCV suggesting possible pharmacogenomic value for this variant. These results provide valuable data that can be of clinical use for predicting the occurrence of anemia and possible erythropoetin use among patients coinfected with HIV and HCV treated with RBV containing regimens.

Anemia remains a substantial risk with pegIFN and RBV therapy. A postulated mechanism of development of anemia is the accumulation of triphosphorylated RBV in erythrocytes causing eventual oxidative damage to erythrocyte membranes and extravascular hemolysis by the reticuloendothelial system [De Franceschi et al., 2000; Russmann et al., 2006]. ITPA also accumulates in erythrocytes of individuals with ITPA genetic mutations resulting in a benign red-cell enzymopathy. The expression of ITPA is controlled and reduced in individuals with mutations in the ITPA gene[Cao and Hegele, 2002; Sumi et al., 2002]. Although the exact mechanism, whereby ITPA deficiency protects against RBV-induced hemolysis is yet to be resolved, it has been suggested that ITPA deficiency confers protection against RBV-induced ATP reduction by substituting for erythrocyte guanosine triphosphate (GTP) which is depleted by RBV in the biosynthesis of ATP [Hitomi et al., 2011].

This study is of particular importance in patients coinfected with HIV and HCV with typically higher rates of anemia and other treatment-related adverse events. This study shows that significant hemoglobin decline of >3 g/dl occurs quicker and earlier in the treatment course in patients with normal ITPA activity. These results also demonstrated an association between ITPA deficient variants and clinical outcome at week 4 in patients coinfected with HIV and HCV although this benefit did not translate to improved sustained virologic response. There are several reasons why this discrepancy occurred including small sample size resulting in type II error and unfavorable baseline demographics in this difficult-to-treat population.

ITPA deficient variants were found to be protective against the development of significant hemoglobin decline >3 g/dl at week 4 and over the course of treatment, consistent with prior studies [Fellay et al., 2010; Ochi et al., 2010; Sakamoto et al., 2010; Thompson et al., 2010, 2011]. There was also a trend towards less anemia in ITPA deficient patients which did not achieve statistical significance unlike prior studies in patients infected with HCV only likely

due to the small sample sizes. There was a higher incidence of hemoglobin decline in patients coinfected with HIV and HCV with a majority (>75%) developing a hemoglobin decline of >3 g/dl over the course of treatment. Patients with moderate to severe ITPase deficiency appeared to have the greatest protection against development of anemia with maximal benefits early in therapy which were sustained over the course of therapy. The impact of ITPA variants on hemoglobin decline could be seen by week 4 in patients coinfected with HIV and HCV and week 12 in patients infected with HCV only even though baseline hemoglobin were similar in both cohorts. A well-recognized cause of anemia in patients coinfected with HIV and HCV is administration of Zidovudine (AZT), however, in this study no significant differences in both ITPA deficient and normal ITPA activity patients based on AZT exposure were observed. This may have been due to the small sample size, since this is a well-recognized cause of anemia in patients coinfected with HIV and HCV.

Although ITPA polymorphisms are significantly associated with RBV-induced anemia the effect on therapeutic outcome is unclear. Two previous studies from the United States [Fellay et al., 2010; Thompson et al., 2010] showed no association with treatment outcomes in patients infected with HCV only, while in contrast two studies from Japan [Ochi et al., 2010; Azakami et al., 2011] reported a possible association between ITPA polymorphisms and treatment outcome. This study shows a possible association with RVR at week 4 only in co-infected patients but no association with final outcomes. This finding is similar to the Japanese study [Sakamoto et al., 2010] and while the etiology is unclear, it is plausible that since patients with ITPA deficient variants are somewhat refractory to RBV-induced anemia, they are therefore able to maintain full RBV dosing as well as higher adherence to RBV resulting in improved treatment outcomes.

There are several limitations to this study. The small sample size and the significant difference in ethnicity from the populations studied could have contributed to the loss of significance observed. Furthermore, the impact of different degrees of ITPA deficiency could not be fully assessed as the patients coinfected with HIV and HCV were largely African-American with a very low incidence of moderate to severe deficiency. Furthermore, due to the retrospective nature of the study design, there was loss of some data points in patients infected with HCV only, which could have introduced type II errors.

In conclusion, polymorphisms in the ITPA gene were associated with protection from RBV associated hemoglobin reductions in patients infected with HCV only and in patients coinfected with HIV and HCV being treated with a course of pegIFN and weight based RBV. In this study, ITPA deficient variants were also associated with early hemoglobin declines as well as early treatment outcomes in patients coinfected with HIV and HCV. It may play a role in the pretreatment assessment of patients at high risk of RBV associated anemia such as patients coinfected with HIV and HCV or in future studies of direct acting antiviral drugs such as HCV protease inhibitors that are associated with exacerbation of anemia.

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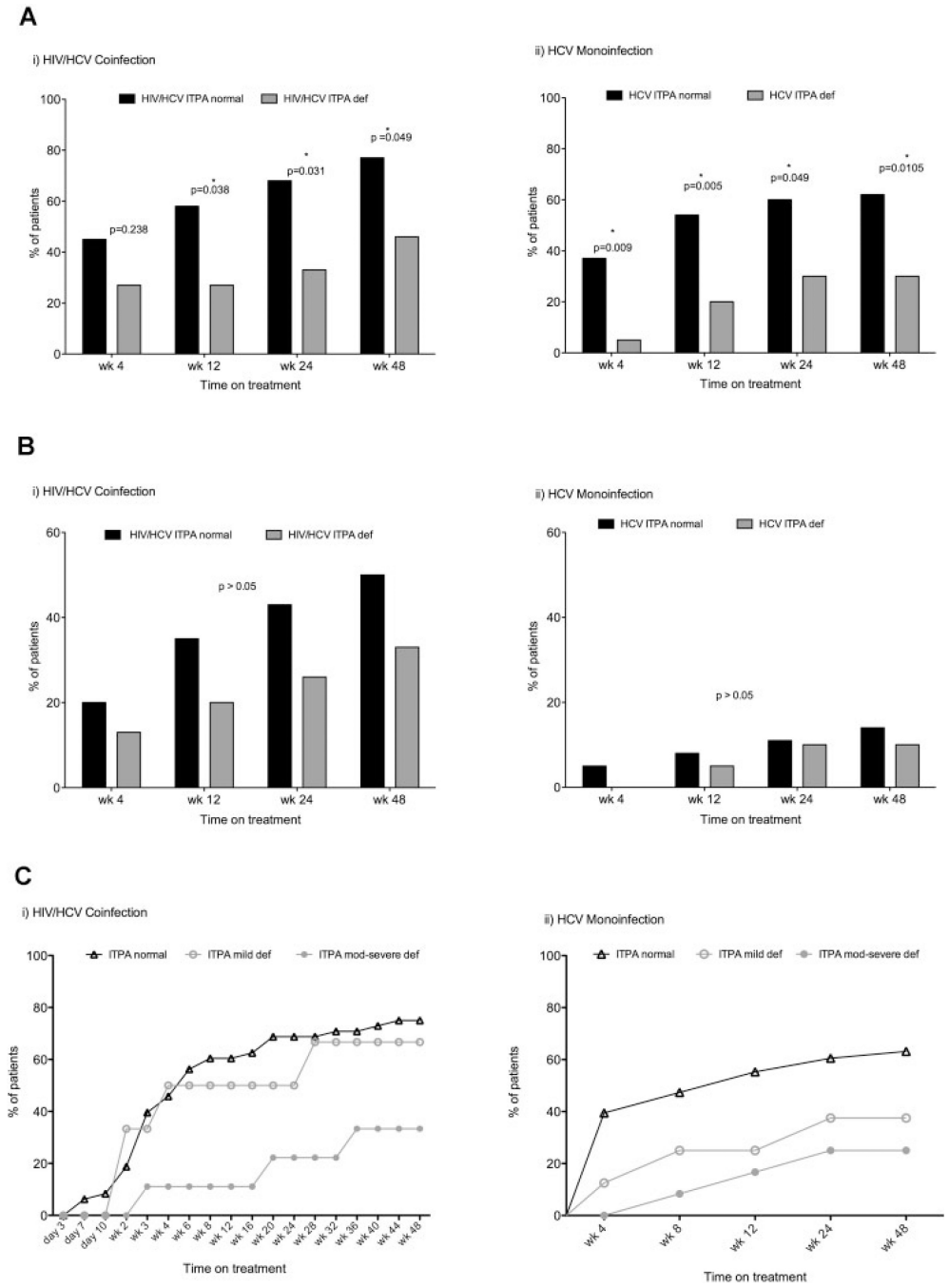
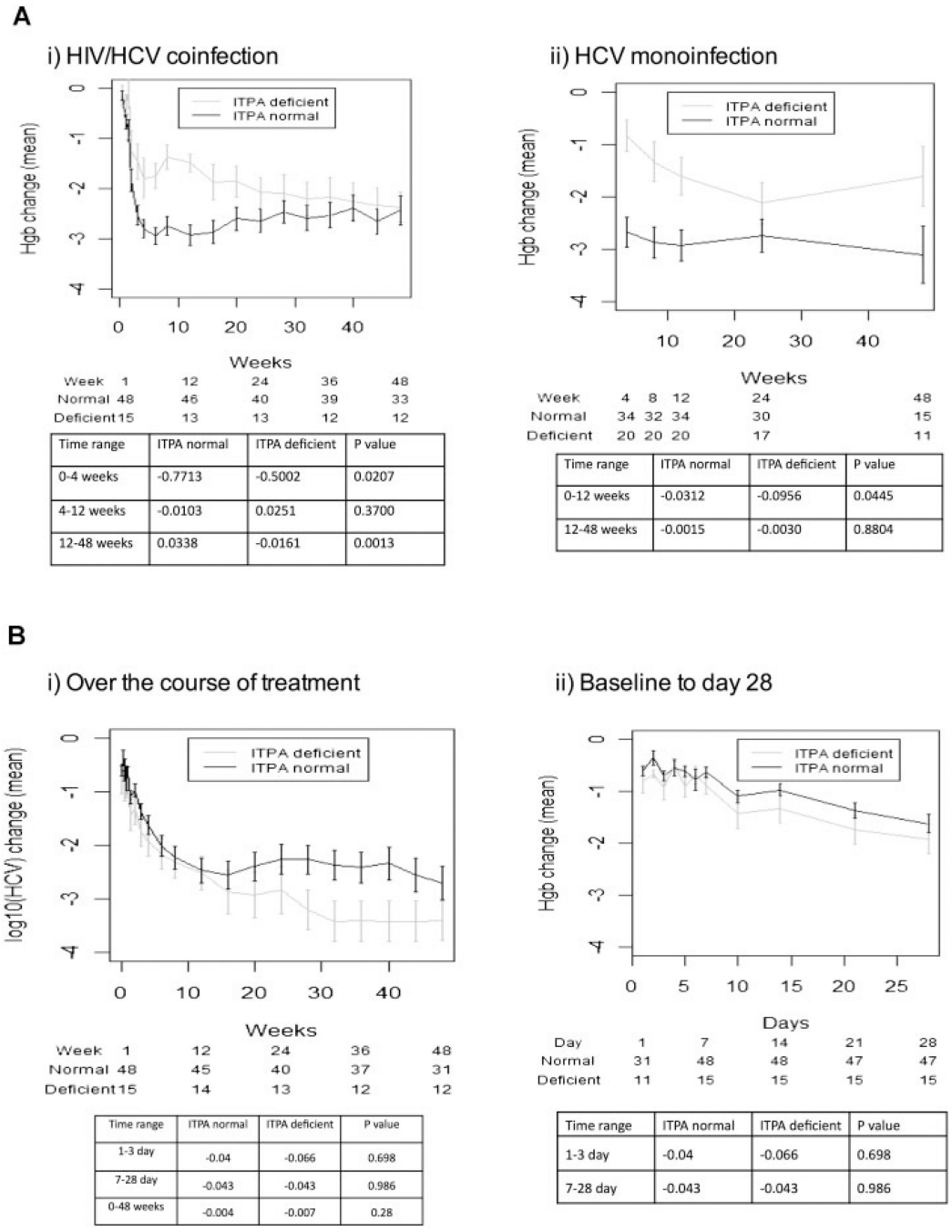


Fig. 1.

A: Prevalence of Hgb reduction >3 g/dl. **B**: Prevalence of anemia Hgb <10 g/dl. **C**: Cumulative frequency of Hgb reduction >3 g/dl by ITPA deficiency severity.



Time range	ITPA normal	ITPA deficient	P value
0-4 weeks	-0.7713	-0.5002	0.0207
4-12 weeks	-0.0103	0.0251	0.3700
12-48 weeks	0.0338	-0.0161	0.0013

Time range	ITPA normal	ITPA deficient	P value
0-12 weeks	-0.0312	-0.0956	0.0445
12-48 weeks	-0.0015	-0.0030	0.8804

Time range	ITPA normal	ITPA deficient	P value
1-3 day	-0.04	-0.066	0.698
7-28 day	-0.043	-0.043	0.986
0-48 weeks	-0.004	-0.007	0.28

Time range	ITPA normal	ITPA deficient	P value
1-3 day	-0.04	-0.066	0.698
7-28 day	-0.043	-0.043	0.986

Fig. 2.

A: Median hemoglobin change from baseline. **B**: Median change in HCV Viral load (log_{10}) in HIV/HCV coinfected patients.

TABLE I

Population Distribution of ITPA Deficiency

rs1127354	rs7270101	Predicted ITPase activity	Predicted ITPase deficiency	Overall, n (%)	HCV, n (%)	HIV/HCV, n (%)
Wild type (C/C)	Wild type (A/A)	100	−	83 (70.3)	35 (63.6)	48 (76.2)
Wild type (C/C)	Heterozygous (A/C)	60	+	14 (11.9)	8 (14.5)	6 (9.5)
Heterozygous (C/A)	Wild type (A/A)	30	++	14 (11.9)	6 (10.9)	8 (12.7)
Wild type (C/C)	Homozygous (C/C)	30	++	5 (4.2)	4 (7.3)	1 (1.6)
Heterozygous (C/A)	Heterozygous (A/C)	10	+++	2 (1.7)	2 (3.6)	0
Homozygous (A/A)	Wild type (A/A)	<5	+++	0	0	0

Severity of ITPase deficiency was predicted as absent, representing wild-type activity (−) and mild (+), moderate (++), or severe (+++) deficiency according to previous studies. ITPase activity was defined as the accumulated presence of minor alleles at the respective polymorphic sites [the minor alleles for both SNPs never occurred together on the same chromosome].
The prevalence of different alleles and the predicted levels of ITPAse deficiency according to the haplotypes.

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TABLE II

Patient Characteristics

Characteristics	HCV monoinfected (n = 58)	HIV/HCV co-infection (n = 65)	*P*-value*
Age (year)	43.9 (10.2)	47.0 (7.6)	NS
Male gender	32 (55.2)	55 (84.6)	0.0006
Race			
Caucasian	52 (89.7)	20 (30.8)	<0.0001
Black	1 (1.7)	37 (56.9)	
Other	5 (8.6)	8 (12.3)	
BMI	25.2 (22.5–29.2)	26.1 (23.3–28.8)	NS
Fibrosis			
0–2	45 (77.6)	40 (61.5)	0.0253
3–4	10 (17.2)	24 (36.9)	
HCV genotype			
Genotype 1	47 (81)	56 (86.2)	NS
Genotype 2 & 3	11 (19)	9 (14.8)	
ITPA deficiency			
No deficiency	35 (64)	48 (76)	NS
Mild deficiency (>60% activity)	8 (14)	6 (10)	
Minimal activity (<30%)	12 (22)	9(14)	
IL28B CC genotype	14 (24.1)	18 (27.7)	NS
Baseline HCV RNA (log_{10} IU/ml)	5.8 (5.4–6.2)	6.3 (5.8–6.7)	<0.0001
High viral load (>800,000)	30 (52%)	49 (75%)	0.0083
Baseline Hgb (g/dl)	14.9 (13.7–16.0)	14.2 (13.6–15.2)	NS
Baseline platelet count ($\times 10^3/mm^3$)	232 (180–279)	205 (165–249)	NS
Baseline creatinine (mg/dl)	0.96 (0.18)	0.90 (0.14)	0.038
Baseline CD4 count	NA	531 (356–772)	
Baseline HIV RNA (IU/ml)	NA	<50 (<50–107)	
Suppressed HIV RNA	NA	47 (72.3)	
On HAART	NA	54 (83.1)	
AZT containing HAART	NA	10 (15.4)	
Erythropoietin use	NA	22 (35%)	
Composite anemia at week 4	2/55 (3.6)	12/63 (19)	0.0106
Composite anemia at week 12	4/55 (7.2)	20/63 (31.8)	0.0011
Composite anemia at week 48	7/55 (12.7)	29/63 (46)	0.0001
Overall SVR rates (%)	20/51 (39)	22/63 (35)	NS

AZT, zidovudine; ABC, abacavir; HAART, highly active antiretroviral therapy; Hgb, hemoglobin.

Values are median (interquartile ranges) or (percentages). *P*-values: chi-square or Mann–Whitney tests.

TABLE III

Factors Associated With Anemia Over the Course of Therapy in Patients Coinfected With HIV and HCV

Baseline factor	Odds ratio	*P*-value
ITPA deficiency	0.5	0.263
Age	1.032	0.356
Gender	0.614	0.429
BMI	0.879	0.071
Baseline HIV VL (Log10)	1.08	0.823
Baseline HCV VL (Log10)	1.005	0.987
Baseline Hgb (g/dl)	**0.518**	**0.008**
Fibrosis	1.137	0.543
Creatine	0.398	0.616
Platelet count	0.998	0.475
CD4 count	0.99	0.502
CD4%	1.015	0.57
AZT use	1.957	0.339

P values < 0.05 are shown in bold.

TABLE IV

Association of ITPA With Treatment Response (RVR, EVR, and SVR) in Patients Coinfected With HIV and HCV

	RVR		EVR		SVR	
Baseline characteristics	**Odds ratio**	***P*-value**	**Odds ratio**	***P*-value**	**Odds ratio**	***P*-value**
[A] Association of the response with baseline characteristics based on univariate logistic regression						
ITPA deficiency	**4.952**	**0.012**	0.823	0.748	2.776	0.093
Age	0.959	0.276	0.973	0.436	0.936	0.076
Gender	0.684	0.569	1.032	0.96	1.384	0.617
BMI	0.991	0.661	0.988	0.4	0.976	0.607
Baseline HIV VL	0.791	0.578	0.989	0.974	1.764	0.113
Baseline HCV VL	**0.28**	**0.003**	0.8	0.508	0.783	0.463
Baseline Hgb	1.037	0.877	1.178	0.447	1.571	0.055
Fibrosis (≥2)	0.611	0.069	1.245	0.328	1.012	0.958
Creatine	4.121	0.485	8.469	0.281	5.966	0.35
Platelet count	1	0.979	0.998	0.455	0.997	0.467
CD4 count	1.001	0.377	1	0.558	1	0.571
CD4%	1.02	0.492	0.989	0.682	1.028	0.318
AZT use	3.417	0.085	88664074	0.993	3.469	0.08
IL28	1.231	0.745	**17**	**0.008**	2	0.236
[B] Multivariate logistic regression on the association of ITPA with the responses adjusting for IL28						
ITPA deficiency	**6.358**	**0.006**	0.881	0.851	3.383	0.054
IL28	1.402	0.627	**16.936**	**0.008**	2.215	0.192

There was no ribavirin reduction in HIV/HCV coinfected patients.
For RVR, only ITPA and baseline HCV are of significant association.
For EVR, only IL28 is of significant association.
For SVR, no baseline characteristics are of significant association.
After adjusting for IL28, ITPA is still significantly associated with RVR, not significantly associated with EVR, and associated with SVR with marginal significance, that is about the same pattern as the univariate logistic regression results.