Recipient *IL28B* Polymorphism Is an Important Independent Predictor of Posttransplant Diabetes Mellitus in Liver Transplant Patients With Chronic Hepatitis C

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IL28B **polymorphisms are strongly associated with response to treatment for HCV infection.** *IL28B* **acts on interferon-stimulated genes via the JAK-STAT pathway, which has been implicated in development of insulin resistance. We investigated whether** *IL28B* **polymorphisms are associated with posttransplant diabetes mellitus (DM). Consecutive HCV patients who underwent liver transplantation between 1–1995 and 1–2011 were studied. Genotyping of the polymorphism rs12979860 was performed on DNA collected from donors and recipients. Posttransplant DM was screened for by fasting blood glucoses every 1–3 months. Of 221 included patients, 69 developed posttransplant DM (31%). Twenty-two patients with recipient** *IL28B* **genotype TT (48%), 25 with** *IL28B* **genotype CT (25%) and 22 with** *IL28B* **genotype CC (29%) developed posttransplant DM. TT genotype was statistically significantly associated with posttransplant DM over time (log rank p = 0.012 for TT vs. CT and p = 0.045 for TT vs. CC). Multivariate Cox regression analysis correcting for donor age, body mass index, baseline serum glucose, baseline serum cholesterol, recipient age and treated rejection, showed that recipient** *IL28B* **genotype TT was independently associated with posttransplant DM (hazard ratio 2.51; 95% confidence interval 1.17–5.40; p = 0.011). We conclude that the risk of developing posttransplant DM is significantly increased in recipients carrying the TT polymorphism of the** *IL28B* **gene.**

Key words: Diabetes mellitus, HCV, liver transplantation

Introduction

More than 170 million people are chronically infected with hepatitis C. It has been hypothesized that the hepatitis C virus (HCV) might escape from eradication by the immune system by inhibiting interferon (IFN)-α-induced tyrosine phosphorylation and activation of STAT1 in hepatic cells (1), a process mediated by suppressors of cytokine signaling (SOCS)-1 and SOCS-3 which are important physiological regulators of the immune system (2). Thus, HCV infection may inhibit the positive feedback loop for the amplification of endogenous IFN-α. In addition, a clinical study of chronic HCV patients showed that SOCS-3 overexpression was associated with nonresponse to treatment of HCV with peginterferon-alpha (pegIFN-α; Ref. 3).

SOCS may not only be implicated in the persistence of HCV infection, but might also cause insulin resistance through inhibition of tyrosine phosphorylation of insulin receptor substrate (IRS) proteins (3,4).

Indeed, chronic HCV infection has been associated with a high prevalence of insulin resistance, both in the pre- and posttransplant setting (5,6). In addition, sustained virological response (SVR) to antiviral treatment has been shown to improve insulin sensitivity (7).

It has recently been shown that genetic *IL28B* polymorphisms coding for IFN-λ are strongly associated with SVR to pegIFN-α treatment for chronic HCV infection (8).

Because IFN-λ, in analogy with IFN-α, may act on IFN-stimulated genes (ISGs) via SOCS and the JAK-STAT pathway (9), we hypothesized that *IL28B* polymorphisms might also be associated with development of diabetes mellitus (DM) in HCV patients.

To investigate this hypothesis, we investigated the occurrence of posttransplant DM according to IFN-λ genotype in a large cohort of HCV patients during long-term follow-up after liver transplantation (LT).

Patients and Methods

Participants
Consecutive chronic hepatitis C patients who underwent LT between January 1, 1995 and January 1, 2011 in the Mayo Clinic in Rochester, Minnesota, were included.

Patients with a pretransplant diagnosis of DM were excluded
DNA was collected prospectively from all donors and recipients. Data were obtained according to a standard protocol and development of DM was screened by fasting blood glucoses every 1–3 months during long-term follow-up after LT. If there were longer lapses between follow-up visits or if follow-up data were missing, patients were considered non-DM. DM was defined as a fasting blood glucose level >126 mg/dL on at least two occasions (10).

A subset of the patients was treated with IFN-based regimens. Treatment start and duration of therapy was documented.

The study protocol was approved by the Institutional Review Board of the Mayo Clinic and was carried out in accordance with institutional guidelines. All participating patients gave informed consent.

Laboratory testing
Virological (genotype, viral load), biochemical and hematological data were measured in the certified Mayo Clinic laboratories. A sensitive qualitative assay was used to detect serum HCV RNA (COBAS Amplicor HCV Test, version 2.0 assay; Roche Molecular Systems, Branchburg, NJ, USA) with a sensitivity of 100 IU/mL. Genotypes were assigned using nucleotide primers specific for a 401 base pair target sequence within the NS5 (nonstructural protein; Ref. 5) region with sequence information compared with published HCV-type reference sequences using the FASTA algorithm (Wisconsin Genetics Computer Group, Madison, WI, USA).

DNA extraction and IL28B genotyping
DNA was extracted from stored paraffin-fixed liver tissue blocks using the QIAamp DNA Mini Kit (Qiagen, Valencia, CA, USA) assay. Tissue was used because whole blood was not available. Liver tissue from donors was obtained during the donor recovery procedure or from protocolled liver biopsies during follow-up after transplantation. Liver tissue from recipients was obtained from the explanted liver. Donor and recipient DNA was tested for the polymorphism rs12979860 using the ABI TaqMan allelic discrimination kit and the ABI7900HT Sequence Detection System (Applied Biosystems, Carlsbad, CA, USA). The possible genotypes for this biallelic polymorphism are: CC, CT and TT, where the CC variant has previously been associated with good response to pegIFN plus RBV therapy in patients infected with genotype 1 HCV (8,11).

Statistics
The statistical analyses were performed using SPSS, version 17.0.2 (IBM Corporation, Somers, NY, USA).

Baseline characteristics were compared using Mann–Whitney, Kruskal–Wallis and chi-square tests as appropriate. The p values in Table 1 were corrected for multiple testing, according to the Bonferroni method.

The Kaplan–Meier method was used to estimate the effect of recipient *IL28B* genotypes on occurrence of posttransplant DM over time. Univariate and multivariate Cox regression analyses were applied to assess risk factors for the development of posttransplant DM over time.

All of the following factors were evaluated in univariate Cox regression analysis: Age, gender, donor age, body mass index (BMI) corrected for the amount of ascites removed during the transplant procedure, glucose, cholesterol, triglycerides, albumin, bilirubin, international normalized ratio, aspartate aminotransferase, alanine aminotransferase, creatinine, warm ischemia time, cold ischemia time, tacrolimus level, cyclosporine level, prednisone dose, HCV RNA, genotype, rejection.

Subsequently, a multivariate model was built, using variables that were significantly associated with development of posttransplant DM in univariate analysis, with a p < 0.15. The following factors were included in the multivariate model, because they were associated with posttransplant DM with a p < 0.15 (Table 2): recipient *IL28B* genotype, glucose, cholesterol and donor age.

Gender, recipient age, BMI and early rejection episodes treated with steroid boluses were forced into the model because it has been previously shown in the literature that these factors are associated with DM (12,13). Importantly, in all models, recipient *IL28B* genotype was statistically significantly associated with posttransplant DM. We performed a separate analysis, only including patients with a diagnosis of DM beyond 4 months of follow-up after orthotopic LT, to avoid bias of patients with transient DM because of the initial steroid treatment. To avoid confounding by the effect of exogenous IFN on occurrence of DM, in the final model patients were censored at the start of IFN-based treatment.

Logistic regression analysis was used to assess the association of posttransplant DM, donor and recipient *IL28B* polymorphism with SVR in patients who were treated with IFN during follow-up after LT.

Results

Patient characteristics
Between January 1, 1995 and January 1, 2011, 329 LT were performed for chronic hepatitis C in our center. Donor and recipient liver tissue was available for *IL28B* genotyping in 289 patients. Genotype at the polymorphic site rs12979860 on chromosome 19 was suitable for analysis in 256 patients (89%). Thirty-five patients with a pretransplant diagnosis of DM were excluded.

Development of posttransplant DM
Of the remaining 221 patients, 69 developed posttransplant DM (31%). Baseline characteristics are shown in Table 1.

During a median follow-up of 5.5 years (interquartile range [IQR] 1.9–9.0), 22 out of 46 patients with recipient *IL28B* genotype TT (48%), 25 out of 99 patients with *IL28B* genotype CT (25%) and 22 out of 76 patients with *IL28B* genotype CC (29%) developed posttransplant DM.

Table 1: Patient characteristics (a) at the time of transplantation and (b) first month after transplantation

	Overall (n = 221)	Posttransplant DM (n = 69)	No DM (n = 152)	p-Value*
(a) Patient characteristics at the time of transplantation[1]				
Male gender (%)	153 (69)	52 (75)	101 (66)	0.183
Age (years)	52 (46–57)	51 (47–58)	52 (45–56)	0.80
HCV genotype (%)				0.24
1	123 (74)	47 (81)	76 (70)	
2	21 (13)	4 (7)	17 (16)	
3	17 (10)	5 (9)	12 (11)	
4	5 (3)	1 (2)	4 (4)	
Recipient *IL28B* polymorphism (%)				0.021
TT	46 (21)	22 (32)	24 (16)	
CC	76 (34)	22 (32)	54 (36)	
CT	99 (45)	25 (36)	74 (49)	
Donor *IL28B* polymorphism (%)				0.74
TT	18 (9)	6 (10)	12 (9)	
CC	101 (52)	30 (48)	71 (54)	
CT	74 (38)	26 (42)	48 (37)	
BMI (kg/m^2)	27 (24–32)	28 (23–32)	27 (25–32)	0.88
Glucose (mg/dL)	100 (88–115)	103 (90–130)	99 (87–112)	0.141
Cholesterol (mg/dL)	119 (86–148)	108 (74–139)	123 (96–153)	0.030
Triglycerides (mg/dL)	82 (57–119)	78 (55–123)	84 (62–116)	0.39
Albumin (g/dL)	3.1 (2.7–3.4)	3.1 (2.6–3.4)	3.1 (2.8–3.4)	0.47
Bilirubin (mg/dL)	2.9 (1.5–4.7)	2.9 (1.9–5.0)	2.9 (1.5–4.7)	0.45
Aspartate aminotransferase (U/L)	100 (70–149)	104 (71–170)	99 (70–142)	0.55
Alanine aminotransferase (U/L)	69 (45–107)	71 (37–105)	68 (47–108)	0.57
Creatinine (mg/dL)	1.1 (0.9–1.3)	1.1 (1.0–1.6)	1.1 (0.9–1.3)	0.049
Hepatitis C virus ribonucleic acid[2] (10^5 IU/mL)[3]	3.7 (0.4–15.3)	6.2 (0.6–23)	2.3 (0.1–12)	0.27
Donor age (years)	45 (28–55)	46 (33–56)	44 (24–55)	0.188
Warm ischemia (min)	44 (37–61)	46 (40–63)	44 (36–60)	0.61
Cold ischemia (min)	428 (364–504)	434 (379–516)	428 (364–494)	0.40
(b) Patient characteristics in the first month after transplantation[1]				
Mean tacrolimus level first month (ng/mL)	9.0 (7.9–10.7)	7.8 (9.0–10.2)	9.0 (7.9–10.8)	0.45
Tacrolimus versus cyclosporine (%)	204 (94)	64 (93)	140 (95)	0.46
Rejection treated with steroid boluses (%)	8 (4)	1 (1)	7 (5)	0.24

[1]Continuous variables are expressed as median (interquartile range).
[2]Pretreatment HCV RNA was available in 52 patients.
[3]Using the Bonferroni correction for multiple testing, p value of <0.02 was considered statistically significant.

Kaplan–Meier analysis showed that recipient TT genotype was statistically significantly associated with development of posttransplant DM over time, the 5-year DM-free survival being 42.9% (95% confidence interval [CI] 25.7–60.1) for recipient *IL28B* genotype TT, 71.7% (95% CI 61.5–81.9) for genotype CT and 65.9% (95% CI 52.9–78.9) for genotype CC (log rank p = 0.045 for TT vs. CC; p = 0.012 for TT vs. CT and p = 0.59 for CC vs. CT; Figure 1).

In contrast, donor *IL28B* genotype was not associated with development of posttransplant DM (log rank p = 0.78 for TT vs. CC and p = 0.71 for TT vs. CT).

There were no statistically significant differences in fibrosis stage at the time of diagnosis of posttransplant DM, according to recipient *IL28B* genotype. At the time of diagnosis of posttransplant DM, 14 patients with recipient *IL28B* genotype TT had Metavir fibrosis stage 0, one had fibrosis stage 1, six had fibrosis stage 2, one had fibrosis stage 3 and none had fibrosis stage 4. In comparison, 17 patients with recipient *IL28B* genotype CT had Metavir fibrosis stage 0, six had fibrosis stage 1, one had fibrosis

stage 2, one had fibrosis stage 3 and none had fibrosis stage 4. In addition, 14 patients with recipient *IL28B* genotype CC had Metavir fibrosis stage 0, four patients had fibrosis stage 1, three had fibrosis stage 2, none had fibrosis stage 3 and one had fibrosis stage 4 (Pearson chi-square p = 0.24).

The proportions of patients using tacrolimus as primary immunosuppression were similar for patients who developed posttransplant DM and those who did not. Sixty-four (93%) of patients who developed posttransplant DM were treated with tacrolimus, compared to 140 (95%) of patients who did not develop posttransplant DM (chi-square p = 0.46).

Levels of HCV viremia did not vary significantly with recipient *IL28B* genotype. The median pretransplant viral load was 14 (IQR 3.1–51) × 10^5 IU/mL for recipient *IL28B* genotype TT, 3.0 (IQR 0.5–18) × 10^5 IU/mL for genotype CC and 1.2 (IQR 0.3–10) × 10^5 IU/mL for genotype CT (overall Kruskal–Wallis p = 0.50 and Mann–Whitney p = 0.30 for TT vs. CC/CT).
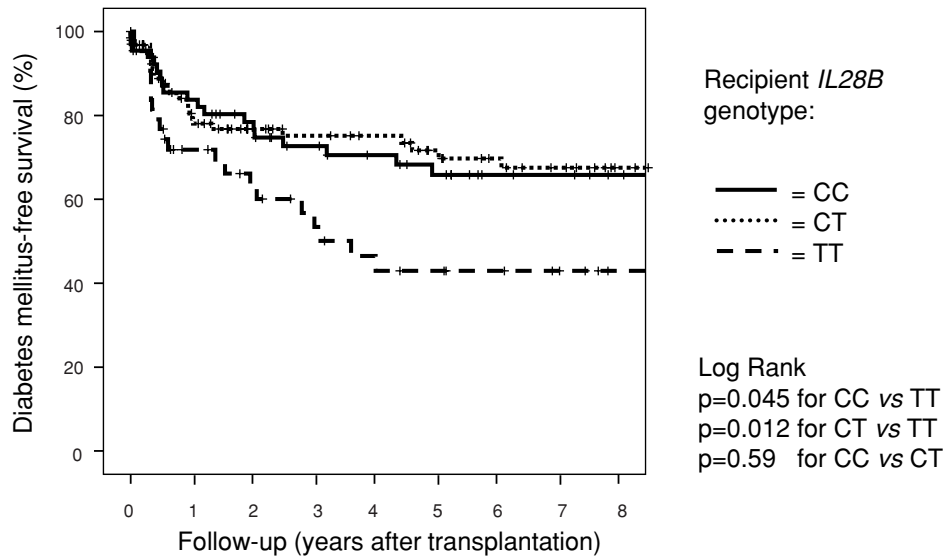
Figure 1: Kaplan–Meier curve showing the diabetes mellitus-free survival according to recipient *IL28B* genotype.

Antiviral treatment

A total of 93 patients received antiviral therapy for recurrent hepatitis C during follow-up. This antiviral treatment was started at a median of 1.6 years (IQR 0.8–3.3) after transplantation. The median duration of treatment was 44 weeks (IQR 23–54) and 39 patients (42%) achieved SVR.

Eighteen out of 31 patients (58%) with recipient *IL28B* genotype CC achieved SVR, 18 out of 38 patients (47%) with recipient *IL28B* genotype CT achieved SVR and 3 out of 24 patients (13%) with recipient *IL28B* genotype TT achieved SVR (OR 0.10; 95% CI 0.03–0.42) for TT versus CC and (OR 0.65; 95% CI 0.25–1.69) for CT vs. CC; p = 0.006. As described previously (14), donor *IL28B* polymorphism was also a predictor of SVR: 24 out of 43 patients (56%) with donor *IL28B* genotype CC achieved SVR, 13 out of 37 patients (35%) with donor *IL28B* genotype CT achieved SVR and 1 out of 7 patients (14%) with donor *IL28B* genotype TT achieved SVR (OR 0.13; 95% CI 0.02–1.19) for TT versus CC (OR 0.43; 95% CI 0.17–1.06) for CT versus CC; overall p = 0.061 and TT versus CT/CC (p = 0.002). Pretreatment DM was not associated with SVR (OR 1.18; 95% CI 0.51–2.75; p = 0.72) in our cohort.

Predictors of DM

Multivariate Cox regression analysis adjusting for gender, age, BMI (corrected for the amount of ascites removed during the transplant procedure), baseline serum glucose, baseline serum cholesterol, donor age and early rejection treated with steroid boluses showed that recipient *IL28B* genotype TT was independently associated with development of posttransplant DM (hazard ratio [HR] 2.51; 95% CI 1.17–5.40; p = 0.011). In this analysis, which starts at 1 month after transplantation, patients were censored at the start of IFN-based treatment (Table 2).

We performed a separate analysis, only including patients with a diagnosis of DM beyond 4 months of follow-up after LT, to avoid bias of patients with transient DM because of the initial steroid treatment (15).

Six patients with recipient *IL28B* genotype TT, 13 patients with genotype CT and nine patients with genotype CC developed posttransplant DM within the first 4 months after transplantation. In this analysis, the results for recipient *IL28B* genotype remained essentially unchanged (HR 1.86; 95% CI 0.93–3.72; p = 0.017 for recipient TT vs. CC; Table 2).

Graft survival

To verify that differences in time at risk are not responsible for the association we found between recipient *IL28B* genotype and development of posttransplant DM, we compared graft survival for the different recipient *IL28B* genotypes.

Indeed, there was no difference in overall graft survival according to recipient *IL28B* polymorphism.

Ten patients (4.5%) with recipient *IL28B* CC polymorphism experienced graft loss, 16 patients (7.2%) with recipient *IL28B* CT polymorphism experienced graft loss and 12 patients (5.4%) with recipient *IL28B* TT polymorphism

Table 2: Univariate and multivariate Cox regression analyses, evaluating risk factors for posttransplant diabetes mellitus

	Univariate Cox regression analysis: HR (95% CI)	p-Value	Overall multivariate Cox regression[2]: HR (95% CI)	p-Value	Overall multivariate Cox regression starting at 4 months after transplantation: HR (95% CI)	p-Value	Multivariate Cox regression[2] censoring patients at start of IFN-based treatment: HR (95% CI)	p-Value
Male gender	1.40 (0.80–2.45)	0.25	1.39 (0.75–2.86)	0.29	1.53 (0.79–2.96)	0.21	1.82 (0.85–3.87)	0.122
HCV genotype		0.35						
1	1 (ref)							
2	0.42 (0.15–1.17)							
3	0.76 (0.30–1.92)							
4	0.50 (0.07–3.62)							
Recipient *IL28B* polymorphism		0.037		0.018		0.017		0.011
CC	1 (ref)		1 (ref)		1 (ref)		1 (ref)	
TT	1.78 (0.97–3.26)		2.03 (1.04–3.96)		1.86 (0.93–3.72)		2.51 (1.17–5.40)	
CT	0.85 (0.48–1.52)		0.82 (0.44–1.56)		0.68 (0.35–1.33)		0.87 (0.41–1.84)	
Donor *IL28B* polymorphism		0.95						
CC	1 (ref)							
TT	1.12 (0.47–2.70)							
CT	0.98 (0.57–1.67)							
Age (years)	1.01 (0.98–1.04)	0.56	1.01 (0.98–1.04)	0.51	1.01 (0.98–1.04)	0.62	1.02 (0.98–1.06)	0.30
BMI (kg/m^2)	1.00 (0.96–1.04)	0.90	1.01 (0.96–1.06)	0.76	1.00 (0.96–1.06)	0.79	1.03 (0.97–1.08)	0.35
Glucose (mg/dL)	1.00 (1.00–1.01)	0.134	1.00 (1.00–1.01)	0.42	1.00 (1.00–1.01)	0.20	1.00 (0.99–1.01)	0.73
Cholesterol (mg/dL)	0.99 (0.99–1.00)	0.028	0.99 (0.99–1.00)	0.060	0.99 (0.99–1.00)	0.067	1.00 (0.99–1.00)	0.172
Triglicerides (mg/dL)	1.00 (1.00–1.00)	0.88						
Albumin, g/dL	0.92 (0.60–1.41)	0.70						
Bilirubin (mg/dL)	0.99 (0.97–1.02)	0.54						
Gamma GT (U/L)	1.00 (1.00–1.00)	0.72						
Aspartate aminotransferase (U/L)	1.00 (1.00–1.00)	0.75						
Alanine aminotransferase, U/L	1.00 (1.00–1.00)	0.65						
Creatinine (mg/dL)	1.06 (0.82–1.37)	0.66						
Hepatitis C virus ribonucleic acid (10^5 IU/mL)	1.00 (1.00–1.00)	0.186						
Donor age (years)	1.02 (1.00–1.03)	0.035	1.01 (1.00–1.03)	0.176	1.01 (1.00–1.03)	0.148	1.01 (0.99–1.03)	0.24
Warm ischemia (min)	1.00 (0.99–1.02)	0.68						
Cold ischemia (min)	1.00 (1.00–1.00)	0.66						
Mean tacrolimus level first month (ng/mL)[1]	0.90 (0.78–1.04)	0.152						
Mean prednisone dose first month (mg/day)[1]	1.00 (0.99–1.01)	0.57						
Tacrolimus versus cyclosporine (%)[1]	0.88 (0.35–2.19)	0.78						
Rejection treated with steroid boluses (%)[1]	0.37 (0.05–2.65)	0.32	0.57 (0.08–4.27)	0.59	0.70 (0.09–5.22)	0.73	0.80 (0.11–5.98)	0.83

[1]For these variables, the univariate Cox regression analysis starts at 1 month after transplantation.
[2]Because rejection treated with steroid boluses is included as a variable in the multivariate analysis, this multivariate Cox regression analysis starts at 1 month after transplantation.

experienced graft loss during follow-up (Pearson chi-square p = 0.174). Mean graft survival was 5.9 years (IQR 2.2–9.0) for patients with recipient *IL28B* polymorphism CC, 4.7 years (IQR 1.7–9.0) for patients with recipient *IL28B* polymorphism CT and 5.5 years (IQR 23–9.0) for patients with recipient *IL28B* polymorphism TT (Kruskal–Wallis p = 0.58).

Discussion

The most important result of this study is the novel observation that *IL28B* polymorphisms are strongly associated with the development of DM during following LT. Specifically, recipient *IL28B* TT genotype is associated with a more than twofold increase in relative risk for posttransplant DM. This observation has potentially important implications.

The fact that only recipient and not donor *IL28B* polymorphisms are associated with development of DM, suggests either of two possibilities: a role for antigen presenting cells and immune effector cells, rather than primary hepatocytes in this process, or that the reduced insulin sensitivity in HCV is not solely related to the hepatocytes. This is interesting because one of the most important features of insulin resistance in non-HCV liver cirrhosis lies within the liver itself, where cirrhosis causes impaired suppression of hepatic glucose production (16).

We hypothesize that similarities in downstream signaling of the IFN-α and the IFN-λ receptor might be responsible for the association between *IL28B* polymorphisms and development of DM in chronic HCV-infected patients. Both IFN-λ and IFN-α act on ISGs via SOCS (9). It has already been suggested that SOCS may not only be implicated in the persistence of HCV infection, but might also cause insulin resistance through inhibition of tyrosine phosphorylation of IRS proteins (3,4).

It is important to consider whether the observed association of recipient *IL28B* TT genotype and the increased risk of development of posttransplant DM is simply a reflection of the lower frequency of SVR seen among patients with the TT genotype. Although the mechanisms for HCV-associated insulin resistance are not fully understood, the HCV core protein downregulates IRS 1 and 2, promoting insulin resistance (17). In addition, eradication of HCV has been convincingly shown to improve insulin sensitivity. When compared to baseline (pretreatment), HOMA2-IR values fall by a mean of over 2 among patients with complete virological response, compared to increases of 0.2 in HOMA2-IR in nonresponders (18). Several observations in this study strongly suggest, however, that response to antiviral therapy, and thus effects of HCV on SOCS and IRS signaling, was not the basis of recipient *IL28B* genotype and development of posttransplant DM. Most importantly, the HR for recipient *IL28B* genotype TT for development of

posttransplant DM was the same if patients were censored at the start of IFN-based treatment. In addition, levels of HCV viremia did not vary significantly with *IL28B* genotype and fibrosis stage at the time of diagnosis of DM was similar between the different groups.

The fact that recipient TT *IL28B* genotype is associated with DM in our study raises some interesting questions about the possible mechanism by which IFN responsiveness varies with *IL28B* genotype.

IFN-λ-3, the product of *IL28B*, is considered to be a proinflammatory cytokine with antiviral properties. The exact role of *IL28B* single nucleotide polymorphisms (SNPs) in the biology of IFN-λ-3 remains unknown: There are conflicting data regarding a possible association between the favorable SNPs and expression of *IL28B* mRNA either in peripheral blood mononuclear cells (PBMC) or liver tissue (19–21), and one report failed to demonstrate a more potent antiviral activity (22). However, studies have consistently showed that the unfavorable genotypes (CT and TT for rs12979860 SNP, and GG and TG for rs8099917) are associated with an increased expression of ISGs (21–24). The SOCS family stands out as one of the many ISGs associated with failed viral eradication (25,26). Remarkably, SOCS-3 and SOCS-7 have been implicated as causing insulin resistance in the setting of hepatitis C (17,27,28). It has been described that HCV can induce both SOCS-1 and SOCS-3—either directly through the core protein or indirectly through tumor necrosis factor alpha/interleukin 6 induction—which impairs signaling of the insulin receptor pathway and causes insulin resistance (17,27,29). Irrespective of the mechanistic consequences of these SNPs in IFN-λ-3 biology, a possible upregulation of SOCS in patients with CT/TT genotypes could explain the link between *IL28B* polymorphisms and the development of DM.

As insulin induces expression of SOCS-3 and SOCS-3 is known to inhibit IFN signaling (3,4), induction of SOCS-3 by insulin could contribute to the inhibition of IFN-α signaling and lower SVR rate associated with *IL28B* TT genotype. Functional studies of variability of SOCS-3 signaling and insulin receptor activity with *IL28B* genotypes would be needed to test this hypothesis.

Given that the increased risk for developing insulin resistance and T2DM in hepatitis C is reversed after viral eradication, current findings pose a double benefit for HCV patients from the metabolic point of view. The inherent reduced risk for DM in all viremic patients with the CC/CT genotype, and a further decrease in risk after achieving viral eradication with antiviral therapy.

Our findings raise some practical considerations. In the nontransplant setting, DM has been shown to be a risk factor for development of hepatocellular carcinoma in patients with HCV (30) and liver transplant patients with HCV have been shown to have an increased risk of liver graft

fibrosis (31). As treatment with insulin sensitizing medications might improve the outcome of HCV patients with insulin resistance, consideration might be given to screening for insulin resistance in patients with the *IL28B* TT genotype.

Although the mechanisms through which *IL28B* polymorphisms influence insulin sensitivity remain hypothetical as long as the exact molecular pathways have not been identified, we believe that our observations may open important new directions for future research on insulin resistance and chronic HCV infection. The fact that we studied a large cohort of well-characterized patients with a long-term follow-up, supports the validity of our observations.

Conclusion

The risk of developing posttransplant DM is significantly increased in recipients carrying the TT polymorphism of the *IL28B* gene.

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Disclosure

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