Protective Effects of Statin Therapy in Cirrhosis Are Limited by a Common *SLCO1B1* Transporter Variant

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Complications of cirrhosis and portal hypertension (PH) can be reduced by statin therapy. The common loss-of-function variant p.V174A in the solute carrier organic anion transporter gene 1B1 (*SLCO1B1*) gene encoding the organic anion transporting polypeptide 1B1 results in decreased hepatic uptake of statins. Our specific aim was to assess the impact of this variant in patients with cirrhosis and statin treatment while controlling for the stage of cirrhosis and other potential confounders with propensity score matching (PSM), availing of a large cohort of genotyped study patients. In total, from 1,088 patients with cirrhosis in two German academic medical centers, PSM yielded 154 patients taking statins and 154 matched controls. The effect on PH was assessed by the liver stiffness–spleen size–to–platelet score (LSPS), and complications of cirrhosis were retrospectively recorded applying consensus criteria. As hypothesized, patients on statin treatment presented less frequently with signs of PH: Esophageal varices (41% vs. 62%; $P < 0.001$) were less common, and LSPS (4.8 ± 11.5 vs. 5.6 ± 6.4; $P = 0.01$) was reduced. Correspondingly, decompensation events were also reduced in patients on statins (odds ratio [OR] = 0.54, 95% confidence interval [CI] 0.32-0.90; $P = 0.02$). When the variant in *SLCO1B1* was present in patients on statins, esophageal varices (OR = 2.68, 95% CI 1.24-5.81; $P = 0.01$) and bacterial infections (OR = 2.50, 95% CI 1.14-5.47; $P = 0.02$) were more common as compared with wild type carriers on statins. *Conclusion:* In this cohort, signs and complications of PH were reduced in patients with cirrhosis treated with statins. Notably, this effect was diminished by the common loss-of-function variant in *SLCO1B1*. Further prospective studies in independent cohorts are warranted to confirm these genotype-specific observations. (*Hepatology Communications* 2021;5:1755-1766).

Cirrhosis, the common end stage of chronic liver diseases, is caused by destruction and remodeling of hepatic tissue. Starting with a compensated status, the disease can further develop into the decompensated stage, which is characterized by markedly elevated mortality with median survival of 2 years, as compared with more than 12 years in the compensated stage.[1] Decompensation is defined by the development of ascites, variceal bleeding, hepatic encephalopathy (HE), or hepatorenal syndrome (HRS). Portal hypertension (PH) is an important factor for the development of decompensation and the clinical course in patients with cirrhosis.

Abbreviations: ALT, alanine aminotransferase; AST, aspartate aminotransferase; CI, confidence interval; CK, creatine kinase; EHR, electronic health records; FIB-4, Fibrosis-4 index; HDL, high-density lipoprotein; HE, hepatic encephalopathy; HRS, hepatorenal syndrome; INR, international normalized ratio; LDL, low-density lipoprotein; LSM, liver stiffness measurement; LSPS, liver stiffness–spleen size–to–platelet score; MELD, model of end-stage liver disease; OR, odds ratio; PH, portal hypertension; PSM, propensity score matching; SLCO, solute carrier organic anion transporter.

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Data from both experimental and human studies consistently confirm that 3-hydroxy-3-methylglutaryl-CoA reductase inhibitors, also known as statins, lower PH and are associated with reduced mortality.[2] A prospective placebo-controlled hemodynamic study in humans reported that 1 month of simvastatin treatment (40 mg/day) lowers the hepatic venous pressure gradient by 8%.[3] This reduction of portal pressure could also be observed in preclinical models and prospective controlled studies.[4-6] A randomized controlled trial evaluated a 2-year therapy with simvastatin in the setting of secondary variceal bleeding prophylaxis. In this setting, a reduced mortality was observed in patients on simvastatin.[7] Several retrospective studies reported an impact of statin therapy on decompensation events, infections, and mortality[8] in patients with cirrhosis.[8,9]

These beneficial effects of statins are not uniform in all patients. The variability of the effects may be due to different drug concentrations resulting from variants of enzymes and/or transporters involved in statin metabolism. Statin-induced myopathy as a frequent side effect of statin treatment[10] has been suggested to be mediated by increased statin plasma concentrations. In a landmark genome-wide association study, the common single nucleotide variant c.521T>C in the solute carrier organic anion transporter gene *1B1* (*SLCO1B1*) has been associated with statin-induced myopathy,[11] a finding that has been validated in several follow-up studies.[12] This gene encodes the organic anion transporting polypeptide 1B1, which is localized in the sinusoidal membrane of hepatocytes and mediates the hepatic uptake of various metabolites, including unconjugated bilirubin and xenobiotics such as statins or methotrexate.[13] Among the variants associated with an impaired transporter function,[14] c.521T>C, which results in the amino acid substitution p.V174A and loss of function, decreases the reduction of low-density lipoprotein (LDL) cholesterol concentrations by simvastatin significantly.[11] Patients on statins carrying this common *SLCO1B1* variant exhibit a 3-fold elevated area under the curve of simvastatin metabolite levels in plasma.[15] These effects have also been reported for other statins[16] and are mediated by reduced intrahepatic statin concentrations.[17]

The aim of this study was to evaluate the effect of statin treatment on PH and other complications of cirrhosis, and to consecutively assess whether the common *SLCO1B1* loss-of-function variant p.V174A impacts the effects of statin treatment in patients with cirrhosis. To conduct this analysis, propensity score matching (PSM) within a large retrospectively characterized study cohort was chosen, which provides a sufficient number of patients carrying the risk variant for successful matching.

Patients and Methods

Overall, 1,088 patients with cirrhosis and genetic data on the *SLCO1B1* variant p.V174A were retrospectively included from two German academic medical centers in Halle and Homburg between April 2013 and April 2019 in the context of screening for the randomized controlled I*N*CA (Impact of *NOD2* genotype–guided antibiotic prevention on survival in patients with liver Cirrhosis and Ascites) trial.[18] Patients without confirmed diagnosis of cirrhosis and missing information about statin medication were excluded. Patients with severe comorbidities such as chronic heart failure (ejection fraction <20%), or nonresectable cancers except limited hepatocellular carcinoma (Barcelona Clinic Liver Cancer stages A-C) were excluded. The study was conducted according to the Declaration of

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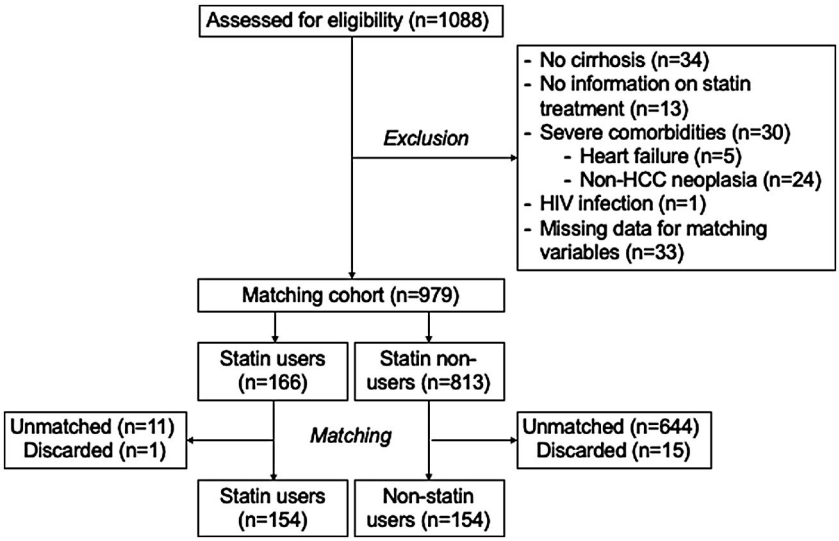

FIG. 1. Flow chart outlining the inclusion and exclusion of the patients for the PSM analysis. Abbreviations: HCC, hepatocellular carcinoma; HIV, human immunodeficiency virus.

Helsinki and the Good Clinical Practice guidelines of the European Medicines Agency. Institutional review board approval was obtained (Homburg #271/11, Halle #2017-85), and all participants provided written, informed consent. Electronic health records (EHRs) were retrospectively reviewed at the index date with uniform criteria, including all necessary information on diagnosis and stage of cirrhosis as well as decompensation events, as outlined subsequently (Fig. 1). Information from previous events of decompensations at other centers were questioned and considered. The index date was the inclusion in the analysis. To allow us to include as much data as possible, this provided data for different stages of cirrhosis. Laboratory parameters were compared as absolute values.

DIAGNOSIS OF CIRRHOSIS

Cirrhosis was defined by (1) biopsy, (2) a combination of clinical (jaundice, liver skin signs), laboratory (parameters of parenchymal damage and synthesis performance), ultrasound (ascites, narrow intrahepatic vessels, rounded edges, heterogeneous parenchyma, enlarged or small liver, enlarged caudate lobe, collaterals, and/or liver surface nodularity), and endoscopic examinations (presence of varices and signs of PH), or (3) liver stiffness measurement (LSM) > 13.0 kPa. In patients with LSM < 19.7 kPa,[19] diagnosis of cirrhosis was additionally confirmed by (1) or (2). EHRs were retrospectively reviewed for clinical data, including clinical decompensation before and at index date. Further information regarding medication use and laboratory parameters at the time of inclusion were also recorded.

STATIN EXPOSITION

Statin exposition was assessed retrospectively for all patients with a statin prescription at the index date based on EHR entries. Changes in medication were documented at all contacts in the department, both for inpatients and outpatients. Exact data on beginning of the statin exposure were not available for the full cohort. Patients receiving the first prescription at index date were considered as not exposed (n = 3, 1.8%). To compare different kinds of statins with variable potency, we calculated the simvastatin equivalent dose and additionally derived the effects on LDL cholesterol concentrations, defining three intensity groups.[20] Statin-induced myopathy was assessed by analyzing the EHR. Statin-induced myopathy was defined as change or discontinuation of statin therapy in the case of elevated creatine kinase (CK) activities (male > 174 U/L, female > 140 U/L) and/or clinical signs of myopathy.

DECOMPENSATION OF CIRRHOSIS

Decompensation was defined as the presence of ascites, HE, HRS, jaundice, and/or variceal bleeding. Diagnostic criteria correspond to the European Association for the Study of the Liver guidelines.[21] The presence of ascites was confirmed by physical examination and/or by abdominal sonography. Patients without clinical signs of ascites who took diuretics (e.g., spironolactone) were considered to have ascites. HE was assessed by the West Haven criteria.[22] Criteria for HRS were applied as currently recommended.[21] Jaundice was arbitrarily defined by a total bilirubin ≥3 mg/dL, or a bilirubin increase of ≥3 mg/dL from previous concentrations. Variceal bleeding was defined as hematemesis or melena leading to hospitalization and diagnosed by a physician and endoscopy.

BACTERIAL INFECTIONS

All episodes of previous or current bacterial infections, which took place after the diagnosis of cirrhosis, were recorded. Bacterial infections were defined according to the criteria outlined by Bajaj et al.[23] Inclusion criterion was treatment with antibiotics.

TRANSIENT ELASTOGRAPHY AND OTHER NONINVASIVE MARKERS

Transient elastography is an ultrasound-based noninvasive method with high accuracy to diagnose liver fibrosis, cirrhosis,[19] and PH.[24] Recent reports demonstrate that it is able to predict esophageal varices.[25] LSM with interquartile range ≤30% and success rate >60% were considered as reliable.[25]

Liver stiffness–spleen size–to–platelet score (LSPS) is an established noninvasive marker to assess clinically significant PH and esophageal varices.[24,26] The parameters composing LSPS are common, easy to access, and thus available for patients with cirrhosis who undergo LSM using elastography. To determine spleen size, the diameter crossing the hilus was measured in ultrasound or cross-sectional imaging (computed tomography or magnetic resonance imaging). Data closest to inclusion date were selected (maximum interval of 14 days).

The Fibrosis-4 (FIB-4) index was calculated as follows: age [years] × aspartate aminotransferase [AST; IU/L]/(platelet count × 10^9/L) × ALT [alanine aminotransferase; IU/L]. The ratio is a noninvasive marker for detecting advanced hepatic fibrosis.[27] In compensated cirrhosis, the score identifies patients with clinically significant PH and esophageal varices.[28]

GENOTYPING

Isolation of genomic DNA from ethylenediaminetetraacetic acid-anticoagulated blood was performed using a membrane-based extraction kit (Qiagen, Hilden, Germany). The *SLCO1B1* polymorphism rs4149056 (c.521T>C, p.V174A), which is in nearly complete linkage disequilibrium with rs4363657,[11] was genotyped using TaqMan polymerase chain reaction (assay C_30633906_10). The genotype distributions were compared with data stored in the gnomAD browser (https://gnomad.broadinstitute.org).[29]

STATISTICAL ANALYSIS

To minimize structural differences between patients on statins and patients without statins, such as different metabolic status, compliance, and cirrhosis development, PSM was applied. Propensity scores were set up to adjust for 19 confounders (Table 1). Most confounders are associated with the stage of liver disease. With respect to the application of PSM, we excluded all patients with missing data in matching variables. These patients represent only 3% of the collective and were excluded, as they could not be characterized because of lack of information. Hence, 979 patients underwent initial PSM. Selection of the parameters was based on a potential impact on the success of statin therapy and not on exposition to statins.[30] Propensity scores were matched 1:1 with a greedy matching algorithm, applying nearest-neighbor matching with a caliper of 0.15. In total, 154 statin-treated patients could be matched to 154 nonstatin users. Twelve statin users could not be matched and were excluded. PSM and subsequent analyses were performed with the "psmatching" module in SPSS v24.0 (IBM, Munich, Germany) under R-Plugin v3.2.5 and R-Essentials for SPSS. To compare the matched groups, conditional logistic regression models, *t* tests for two related samples, or Wilcoxon rank tests, depending on the distributions of the test variables, were used. Hardy-Weinberg equilibrium was tested with an exact test (https://ihg.gsf.de/cgi-bin/hw/hwa2.pl). Genetic subgroup analysis

TABLE 1. MATCHING VARIABLES AT BASELINE BEFORE AND AFTER PSM

	Before PSM			After PSM		
	No Statin (n = 813)	Statin (n = 166)	*P*	No Statin (n = 154)	Statin (n = 154)	*P*
Age (years)	59.7 ± 11.2	66.1 ± 9.6	**<0.001**	65.2 ± 10.3	65.3 ± 9.4	0.75
Sex						
Female	296 (36.4)	40 (24.1)	**0.002**	32 (20.5)	39 (25.3)	0.34
Male	517 (63.6)	126 (75.9)		122 (79.5)	115 (74.7)	
Etiology of cirrhosis						
Alcoholism	460 (56.6)	70 (42.2)		75 (48.7)	69 (44.8)	
Hepatitis B	27 (3.3)	3 (1.8)		4 (2.6)	3 (1.9)	
Hepatitis C	118 (14.5)	12 (7.2)		13 (8.4)	12 (7.8)	
Metabolic	49 (6.0)	23 (13.9)		12 (7.8)	22 (14.3)	
Cryptogenic	69 (8.5)	37 (22.3)		26 (16.9)	29 (18.8)	
Other	90 (11.1)	21 (12.6)		24 (15.6)	19 (12.3)	
Diabetes	221 (27.2)	88 (53.0)	**<0.001**	77 (50.0)	77 (50.0)	0.90
Medication						
β-blocker	383 (47.1)	100 (60.2)	**0.002**	95 (61.7)	88 (57.1)	0.39
Long-term antibiotic therapy	162 (19.9)	21 (12.7)	**<0.001**	20 (13.0)	21 (13.6)	0.87
Lactulose	331 (40.7)	43 (25.9)	**0.03**	46 (29.9)	42 (27.3)	0.56
PPI	517 (70.2)	129 (77.7)	0.05	114 (74.0)	118 (76.6)	0.56
Therapy of viral hepatitis	110 (13.5)	10 (6.0)	**0.007**	10 (6.5)	10 (6.5)	1.00
Bacterial infection 2 weeks before inclusion	126 (15.5)	16 (9.6)	0.05	14 (9.1)	15 (9.7)	0.85
Hospitalization 6 months before inclusion	441 (54.2)	70 (42.2)	**0.005**	70 (45.5)	69 (44.8)	0.91
Hospitalization ever	716 (88.1)	143 (86.1)	0.49	141 (91.6)	133 (86.4))	0.16
Treatment in ICU ever	151 (18.6)	39 (23.5)	0.15	35 (22.7)	35 (22.7)	1.00
Laboratory parameters						
Serum sodium (mmol/L)	138 ± 5	139 ± 4	**0.001**	138 ± 4	139 ± 4	0.08
Serum creatinine (mg/dL)	1.1 ± 0.8	1.2 ± 0.7	**0.003**	1.2 ± 0.8	1.2 ± 0.7	0.86
Total bilirubin (mg/dL)	2.8 ± 4.8	1.5 ± 1.8	**<0.001**	1.4 ± 1.3	1.5 ± 1.9	0.46
Serum albumin (g/dL)	34.8 ± 7.2	36.3 ± 8.0	**0.01**	35.5 ± 7.0	36.1 ± 8.3	0.28
Hemoglobin (g/dL)	11.9 ± 2.6	12.6 ± 2.3	**0.002**	12.3 ± 2.4	12.5 ± 2.4	0.68
INR	1.3 ± 0.4	1.3 ± 0.3	**0.01**	1.3 ± 0.3	1.3 ± 0.3	0.94

Note: Data are presented as frequency and percentage or means and SD. Significant *P* values are marked in bold. Decompensated patients can have multiple decompensation events.
Abbreviation: ICU, intensive care unit.

was conducted applying χ^2 tests for contingency tables, *t* tests, or Mann-Whitney U tests. Two-sided *P* values < 0.05 were regarded as significant. All variables are described as proportions or means with SD.

Results

PATIENT CHARACTERISTICS AND MATCHING

After exclusion and matching, we studied 308 patients in total (154 patients on statins and 154 nonstatin users) (Fig. 1). The median age of these patients was 65.3 ± 9.8 years, and 76.9% of the patients (n = 237) were men (Table 2). The most common etiology of cirrhosis in our cohort was alcohol-associated liver disease (n = 144; 46.8%). Child-Turcotte-Pugh scores were distributed as follows: 36.4% of patients were class A, 30.8% were class B, and 11.3% were class C. Mean Model for End-stage Liver Disease (MELD) score was 11.9 ± 4.8, and mean LSPS was 5.2 ± 9.4.

Table 1 summarizes the matching variables before and after matching. Before PSM, patients on statins were (1) younger and primarily male with (2) lower

TABLE 2. BASELINE CHARACTERISTICS OF ALL PATIENTS

	Number	Frequency or Mean ± SD
Age (years)	308	65.3 ± 9.8
Sex		
female	71	23.1%
male	237	76.9%
Body mass index (kg/m^2)	216	28.6 ± 5.8
Charlson index	226	5.9 ± 2.2
Etiology of cirrhosis		
Alcoholism	144	46.8%
Hepatitis B	7	2.3%
Hepatitis C	25	8.1%
NAFLD	34	11.0%
Cryptogenic	55	17.9%
Other	43	13.9%
Diabetes	155	50.3%
Decompensated stage	193	62.7%
MELD	308	11.9 ± 4.8
Child-Turcotte-Pugh score	242	7.1 ± 2.1
A	112	36.4%
B	95	30.8%
C	35	11.3%
LSM (kPa)	167	35.8 ± 21.3
LSPS	167	5.2 ± 9.4
FIB-4	228	5.1 ± 7.9
Spleen size (cm)	283	13.5 ± 2.8
Total bilirubin (mg/dL)	308	1.5 ± 1.6
Type of statin		
Simvastatin	87	63.0% (in statin group)
Atorvastatin	39	28.3% (in statin group)
Pravastatin	8	5.8% (in statin group)
Fluvastatin	4	2.9% (in statin group)
Simvastatin equivalent dose (mg/day)	154	35.2 ± 22.1
Intensity of statin therapy		
Low	13	10.5% (in statin group)
Moderate	93	75.0% (in statin group)
High	18	14.5% (in statin group)

Abbreviation: NAFLD, nonalcoholic fatty liver disease.

severity of liver disease, as reflected by lower total bilirubin concentrations as well as higher serum albumin and hemoglobin levels; (3) the prevalence rates of alcohol-associated liver disease as well as chronic hepatitis B and C infections were lower, whereas metabolic and cryptogenic causes of liver disease were more than twice as frequent; and (4) as expected, the patients on statins were more likely to have diabetes. After PSM, all standardized differences of matching parameters were reduced, and there was no difference with respect to comorbidities, as reflected by the Charlson indexes in both groups (Table 3).

Among the patients on statins, the most prescribed statin was simvastatin (63%, information available for 138 patients), followed by atorvastatin (28%). The mean simvastatin equivalent dose in our cohort was 35 mg ± 22 mg per day (Table 2).

EFFECTS OF STATIN TREATMENT ON CIRRHOSIS

Patients with cirrhosis treated with statins were less likely to present with events of decompensation (odds ratio [OR] = 0.54, 95% confidence interval [CI] 0.32-0.90; P = 0.02), primarily due to the significantly lower frequency of variceal bleeding (6% vs. 14%; P = 0.02) and ascites (48% vs. 60%; P = 0.04) (Table 3). Indeed, corresponding signs of PH such as the endoscopic presence of esophageal varices (41% vs. 62%; P < 0.001), LSPS (4.8 ± 11.5 vs. 5.6 ± 6.4; P = 0.01), FIB-4 index (4.8 ± 10.1 vs. 5.4 ± 4.7; P = 0.02), and spleen size (13.1 ± 2.5 cm vs. 13.8 ± 2.9 cm; P = 0.02) were significantly reduced in patients on statins. Furthermore, platelet counts were significantly higher (182 ± 104 × 10^9/L vs. 146 ± 87 × 10^9/L; P < 0.001) in statin-treated patients. On statin treatment, serum lipids (in particular total, LDL, and high-density lipoprotein [HDL] cholesterol concentrations) did not differ from patients without statins (Table 3).

Serum CK activities were significantly elevated in patients on statins (112 ± 91 IU/L vs. 96 ± 77 IU/L; P = 0.03), which could reflect the myotoxicity of statins. In the statin cohort, three cases of statin-induced myopathy were observed (1.9%), as compared with no cases in the nontreated patients. Serum ALT and AST activities did not differ between the groups (Table 3). Associations among the high, moderate, and low-intensity groups of statin therapy (see "Patients and Methods" section) and the effects described previously could not be detected either (Supporting Tables S1-S3).

ROLE OF THE SLCO1B1 TRANSPORTER VARIANT

All patients on statins were genotyped successfully for the *SLCO1B1* loss-of-function variant p.V174A. For 2 patients without statins, genotyping was not

TABLE 3. RESULTS IN PSM-MATCHED COHORT COMPARING 154 PATIENTS WITHOUT AND 154 PATIENTS ON STATINS

	No Statin	Statin	*P*	OR	95% CI
Body mass index (kg/m^2)	27.9 ± 6.0	29.2 ± 5.5	0.23		
Child-Turcotte-Pugh score	7.3 ± 2.2	7.0 ± 2.0	0.41		
Charlson index	6.0 ± 2.3	5.8 ± 2.2	0.18		
Decompensation	106 (68.8)	87 (56.5)	**0.02**	0.54	0.32-0.90
Variceal bleeding	22 (14.3)	9 (5.8)	**0.02**	0.35	0.15-0.83
HE	24 (15.6)	21 (13.6)	0.62	0.85	0.45-1.62
Jaundice	31 (20.1)	29 (18.8)	0.78	0.92	0.53-1.61
Ascites	92 (59.7)	74 (48.1)	**0.04**	0.61	0.38-0.97
HRS	9 (5.8)	4 (2.4)	0.18	0.44	0.14-1.44
PH					
Esophageal varices	95 (62.1)	63 (41.4)	**<0.001**	0.39	0.23-0.66
LSM (kPa)	36.3 ± 21.1	35.4 ± 21.6	0.29		
LSPS	5.6 ± 6.4	4.8 ± 11.5	**0.01**		
FIB-4	5.4 ± 4.7	4.8 ± 10.1	**0.02**		
Spleen size (cm)	13.8 ± 2.9	13.1 ± 2.5	**0.02**		
Hepatocellular carcinoma	54 (35.1)	43 (27.9)	0.16	0.69	0.42-1.16
Bacterial infection (yes)	43 (27.9)	46 (29.9)	0.71	1.10	0.67-1.80
Decompensated (yes)	37 (86.0)	32 (69.6)			
Compensated (yes)	6 (14.0)	14 (30.4)			
Laboratory parameters					
WBC (×10^9)	6.5 ± 0.8	7.3 ± 3.7	**0.03**		
Platelets (×10^9)	146 ± 87	182 ± 104	**<0.001**		
ALT (IU/L)	48 ± 67	72 ± 144	0.13		
AST (IU/L)	76 ± 117	67 ± 95	0.39		
AST (IU/L)	67 ± 95	76 ± 117	0.39		
CRP (mg/dL)	19 ± 27	16 ± 27	0.33		
LDL (mg/dL)	95 ± 37	91 ± 62	0.40		
HDL (mg/dL	41 ± 23	43 ± 23	0.18		
Cholesterol (mg/dL)	156 ± 48	151 ± 71	0.37		
Triglycerides (mg/dL)	128 ± 95	128 ± 68	0.77		
HbA1c (mmol/mol)	43.1 ± 16.9	46.1 ± 16.3	0.17		
CK (IU/L)	96 ± 77	112 ± 91	**0.03**		
Events of myopathy	0	3 (1.9)	0.39	65.80	

Note: Data is presented as frequency and percentage or means and SD. Significant *P* values are marked in bold. Decompensated patients can have multiple decompensation events.
Abbreviations: CRP, C-reactive protein; HbA1c, hemoglobin A1c; WBC, white blood count.

possible due to insufficient DNA samples. In this cohort, 225 wild type carriers, 76 heterozygous, and 5 homozygous *SLCO1B1* risk allele carriers were detected, which is consistent with the Hardy-Weinberg equilibrium (P = 0.813; exact test). Correspondingly, the minor allele frequency was 14.1%, similar to the frequency of 15.9% in a large European population (gnomAD database).[29]

The impact of the variant in *SLCO1B1* was analyzed separately in the statin and control groups to delineate the effects of statin therapy. Table 4 summarizes the results of this analysis. Applying a dominant model (wild type vs. heterozygous + homozygous carriers) in the statin group, decompensation events, PH, bacterial infections, and total bilirubin concentrations all indicate that the beneficial effects of statins are reduced by the presence of the *SLCO1B1* variant. Although some differences nominally did not reach significance, the frequencies consistently point in the same direction: Variceal bleeding, HE, jaundice, and ascites as clinical signs of decompensation (and rate of hepatocellular carcinoma) are markedly reduced in wild type carriers in comparison to patients with loss-of-function alleles,

TABLE 4. ANALYSIS OF THE DOMINANT MODEL OF THE *SLCO1B1* VARIANT p.V174A IN PATIENTS ON STATINS

	Wild Type	Heterozygous + Homozygous	*P*	OR	95% CI
Age (years)	64.9 ± 9.3	66.9 ± 9.8	0.29		
Sex (male)	91 (76.5)	24 (68.6)	0.35	1.49	0.65-3.42
Body mass index (kg/m^2)	29.3 ± 5.6	28.7 ± 5.2	0.65		
Charlson index	5.7 ± 2.2	6.3 ± 2.2	0.28		
MELD	11.3 ± 4.6	13.5 ± 5.8	**0.01**		
Child-Turcotte-Pugh score	6.8 ± 1.9	7.6 ± 2.2	0.19		
Simvastatin equivalent dose (mg)	35.8 ± 22.2	32.4 ± 22.1	0.41		
Decompensation	63 (52.9)	24 (68.6)	0.10	1.94	0.87-4.31
Variceal bleeding	5 (4.2)	4 (11.4)	0.11	2.94	0.75-11.60
HE	13 (10.9)	8 (22.9)	0.07	2.42	0.90-2.42
Jaundice	20 (16.8)	9 (25.7)	0.24	1.71	0.70-4.20
Ascites	53 (44.5)	21 (60.0)	0.11	1.87	0.87-4.02
HRS	1 (0.8)	3 (8.6)	**0.01**	11.07	1.11-109.97
PH					
Esophageal varices	42 (35.9)	21 (60.0)	**0.01**	2.68	1.24-5.81
LSM (kPa)	33.7 ± 21.0	43.0 ± 23.5	0.12		
LSPS	4.9 ± 12.6	4.5 ± 4.9	0.35		
FIB-4	4.8 ± 11.3	5.0 ± 3.5	0.08		
Spleen size (cm)	13.0 ± 2.5	13.5 ± 2.5	0.14		
Hepatocellular carcinoma	29 (24.4)	14 (40.0)	0.07	2.07	0.90-2.42
Bacterial infection (yes)	30 (25.2)	16 (45.7)	**0.02**	2.50	1.14-5.47
Decompensated (yes)	17 (56.6))	15 (93.8)			
Compensated (yes)	13 (43.3)	1 (6.2)			
Laboratory parameters					
Serum sodium (mmol/L)	139 ± 4	137 ± 5	0.07		
Serum creatinine (mg/dL)	1.2 ± 0.7	1.3 ± 0.9	0.47		
Urea (g/dL)	46 ± 35	49 ± 43	0.52		
Total bilirubin (mg/dL)	1.3 ± 1.7	2.0 ± 2.3	**0.007**		
ALT (IU/L)	75 ± 145	60 ± 139	0.06		
AST (IU/L)	72 ± 93	90 ± 175	0. 96		
Serum albumin (g/dL)	36 ± 9	35 ± 7	0.12		
CRP (mg/dL)	16 ± 27	17 ± 26	0.30		
WBC (×10^9)	7.2 ± 3.9	7.5 ± 3.0	0.31		
Hemoglobin (g/dL)	12.8 ± 2.2	11.4 ± 2.6	**0.007**		
Platelets (×10^9)	189 ± 112	156 ± 74	0.13		
INR	1.2 ± 0.4	1.3 ± 0.3	**0.02**		
PTT (seconds)	28.9 ± 5.5	32.2 ± 7.3	**0.02**		
Cholesterol (mg/dl)	152 ± 71	144 ± 72	0.70		
LDL cholesterol (mg/dL)	94 ± 66	78 ± 36	0.40		
HDL cholesterol (mg/dL)	44 ± 24	37 ± 20	0.36		
Triglycerides (mg/dL)	126 ± 67	133 ± 78	0.59		
HbA1c (mmol/mol)	46.6 ± 16.6	43.7 ± 14.4	0.36		
CK (IU/L)	111 ± 70	114 ± 164	**0.04**		

Note: Data is presented as frequency and percentage or means and SD. Significant *P* values are marked in bold. Decompensated patients can have multiple decompensation events.
Abbreviations: CRP, C-reactive protein; HbA1c, hemoglobin A1c; PTT, partial thromboplastin time; WBC, white blood count.

whereas HRS occurs significantly more often in the latter patients (OR = 11.07, 95% CI 1.11-110; P = 0.01). As a major sign of PH, the presence of esophageal varices is significantly higher in carriers of the variant alleles (OR = 2.68, 95% CI 1.24-5.81; P = 0.01). Accordingly, the FIB-4 index and other markers (LSM, spleen size) signal less advanced PH in patients without the variant (Table 4).

Of note, patients with *SLCO1B1* loss-of-function alleles are also more likely to present with bacterial infections (OR = 2.50, 95% CI 1.14-5.47; P = 0.02). Remarkably, there was an overall increased excretion capacity of the liver, as reflected by significantly lower total bilirubin concentrations (1.3 ± 1.7 mg/dL vs. 2.0 ± 2.3 mg/dL; P = 0.007) and MELD scores in patients without risk alleles (11.3 ± 4.6 vs. 13.5 ± 5.8; P = 0.01). Accordingly, lower international normalized ratio (INR; 1.2 ± 0.4 vs. 1.3 ± 0.3; P = 0.02) and shorter partial thromboplastin time (28.9 ± 5.5 vs. 32.2 ± 7.3 seconds; P = 0.02) indicate higher liver synthesis capacity. Serum sodium, ALT, and platelets showed consistent trends to levels indicating ameliorated hepatic injury (Table 4). In contrast, carriage of the risk allele was found to confer increased CK activities (114 ± 164 IU/L vs. 111 ± 70 IU/L; P = 0.04). Due to a low number of homozygous patients, the recessive model is not expressive, however, it is comparable to the described results (Supporting Table S5).

In the control group of patients not taking statins, the presence of the variant p.V174A did not have any consistent impact on the outcome of patients with cirrhosis (Supporting Table S4**)**. The results in the dominant model were inconsistent: HRS was more likely in wild type carriers (OR = 0.68, 95% CI 0.61-0.76; P = 0.04), and AST levels were lower in these patients (74 ± 103 IU/L vs. 51 ± 47 IU/L; P = 0.04). Other variables did not show corresponding trends. In contrast, in the recessive model, no consistent effects were observed (Supporting Table S6).

Discussion

Portal hypertension is a common consequence of cirrhosis, caused by the increase of the intrahepatic resistance due to destruction of the parenchyma of the liver, accumulation of extracellular matrix, and systemic arterial vasodilation. The ideal treatment of PH would be drugs that reduce hepatic resistance without affecting hepatic perfusion and display anti-inflammatory as well as antifibrotic effects.[31] Statins—in addition to their lipid-lowering effects—decrease hepatic resistance and increase hepatic blood flow and, and in contrast to β-blockers, do not negatively affect systemic hemodynamics in cirrhosis.[32] Therefore, as warranted by experimental data, the impact of statins was investigated in randomized trials and observational studies,[3,5,7,9,33,34] which indicated reduction of portal pressure and lower rates of infections in cirrhosis.

In this study, we investigated the effect of statin treatment in patients with cirrhosis by minimizing confounding factors such as stage of disease and degree of PH. Previous studies examined the statin effects on portal pressure using invasive measurements.[3,5] In this context, we analyzed whether these observations are reflected by noninvasive assessment of PH. Spleen size, the presence of esophageal varices, platelet counts, and FIB-4 indexes were reduced in patients on statins in the matched cohort, indicating decreased PH. These results could also be confirmed by elastography and calculating LSPS, as expressed by the reduction of the single components (liver stiffness, platelets, spleen size) as well as the full score. Consecutively, PH-associated events of decompensation such as ascites and variceal bleeding occurred less frequently in patients taking statins. Serum lipid concentrations did not differ between treated and nontreated patients, indicating that the overall prescribed statin doses are effective.

After confirming the previously described data, we specifically studied the effects of the *SLCO1B1* loss-of-function variant p.V174A (c.521T>C), which leads to a reduced hepatic uptake of statins.[17] We availed of a large cohort of patients with genetic information on p.V174A to establish the PSM cohort of patients with cirrhosis. This analysis revealed consistent differences between wild type and risk allele carriers in the group of patients with cirrhosis who took statins: Patients carrying the risk allele developed more complications of cirrhosis associated to PH. Notably, we found more bacterial infections in patients carrying the *SLCO1B1* risk variant. There are scarce data in patients with cirrhosis, but other studies described less frequent pulmonary infections,[35] sepsis,[36] as well as reduced overall mortality[37] because of lower frequency of infections in patients on statins. Taken together, our findings indicate that the *SLCO1B1* variant could lead to reduced therapeutic efficacy of statins in patients with cirrhosis. As

the same analysis in the control group did not reveal consistent associations, we suggest that in this cohort the genotype itself does not impact the clinical course of cirrhosis. Therefore, we suggest that the beneficial effects of statins in patients with cirrhosis might be limited in patients carrying the p.V174A risk allele. Even though we did not conduct functional analyses, we speculate that the higher frequency of bacterial infections and increased PH in patients with the *SLCO1B1* risk allele taking statins could be a consequence of the reduced intrahepatic effect of statins.

Statins are prescribed with caution in patients with chronic liver diseases due to the fear of side effects, and there is reluctance of many physicians to prescribe statins in patients with cirrhosis. In fact, pharmacokinetics of statins, as assessed by maximum concentration and area under the curve, are altered and *SLCO1B1* expression is decreased in patients with cirrhosis.[38] Of note, in our patients the levels of liver enzymes and bilirubin did not differ between statin users and nonusers. Additionally, no case of statin-induced liver injury was noted in any of our patients. Hence, our findings, together with the known cardiovascular and potential hepatic benefits, provide further support for the careful use of statins in this vulnerable population. This is in line with small prospective and larger retrospective studies that found statins to be safe in compensated cirrhosis.[39] Further studies are currently being performed assessing the safety and efficacy of different doses of statins in patients with decompensated cirrhosis.[40] In fact, we could not exactly determine the defined daily doses, and information on type or dose was not available in a subgroup of our patients. Most of the patients were treated with the lipophilic compound simvastatin; therefore, other types of more hydrophilic statins are less represented. Even though statins express a class effect, we could not assess pleiotropic mechanisms on liver and intestine or metabolic pathways either.

Our study design has certain limitations that are mostly inherent to the retrospective design. In our PSM model, we could have potentially missed additional confounders of statin therapy, and we might have created distortion because of the selection of variables. Due to the lack of data, we could not balance the duration, indication, or decision to initiate therapy (e.g., HDL, LDL), and accordingly we could not address the indication bias of statin therapy, which might also resemble the fact that patients with a more advanced stage of the disease might be prescribed statins less commonly. Therefore, we could not provide information on the dose or duration of statin treatment required for the effects. Because the overall size of our PSM cohort was moderate, some confidence intervals were wide, and we therefore might have missed small effects. We could robustly assess the dominant association model only. The study design was retrospective and observational, so we could not perform an analysis of survival or decompensation-free time. Our findings should be therefore confirmed in prospective controlled studies. To quantify the pharmacological associations, other study designs are required. Due to the cross-sectional study design, no assessment of causality can be made. Because the exposure periods of statin therapy might be missed in a few patients, events or bacterial infections could occur outside the exposure period in a small number of patients. Notwithstanding the potential bias of observational analyses on statins,[38] in our study the results after genetic stratification fully support the beneficial effects of statins on cirrhosis outcomes.

In summary, we assessed the effect of a loss-of-function variant in *SLCO1B1,* causing reduced hepatic uptake of statins on markers and signs of PH in a retrospectively propensity score–matched cohort of patients with cirrhosis. We found that signs of PH were reduced in patients with cirrhosis on statin therapy. Notably, carriers of the common loss-of-function *SLCO1B1* variant appeared to benefit less from statin therapy and developed more bacterial infections, increased PH, and associated complications. Future molecular studies and prospective assessment of genetic testing for *SLCO1B1* variants in patients with cirrhosis and PH are now warranted.

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Supporting Information

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