

# *SLCO1B1*\*5 Allele Is Associated With Atorvastatin Discontinuation and Adverse Muscle Symptoms in the Context of Routine Care

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The *SLCO1B1* genotype is known to influence patient adherence to statin therapy, in part by increasing the risk for statin-associated musculoskeletal symptoms (SAMSs). The *SLCO1B1*\*5 allele has previously been associated with simvastatin discontinuation and SAMSs. Prior analyses of the relationship between *SLCO1B1*\*5 and atorvastatin muscle side effects have been inconclusive due to insufficient power. We now quantify the impact of *SLCO1B1*\*5 on atorvastatin discontinuation and SAMSs in a large observational cohort using electronic medical record data from a single health care system. In our study cohort ( $n = 1,627$  patients exposed to atorvastatin during the course of routine clinical care), 56% ( $n = 912$  of 1,627 patients) discontinued atorvastatin and 18% ( $n = 303$  of 1,627 patients) developed SAMSs. A univariate model revealed that *SLCO1B1*\*5 increased the likelihood that patients would stop atorvastatin during routine care (odds ratio 1.2; 95% confidence interval (CI), 1.1–1.5;  $P = 0.04$ ). A multivariate Cox proportional hazards model further demonstrated that this same variant was associated with time to atorvastatin discontinuation (hazard ratio 1.2; 95% CI, 1.1–1.4;  $P = 0.004$ ). Additional time-to-event analyses also revealed that *SLCO1B1*\*5 was associated with SAMSs (hazard ratio 1.4; 95% CI, 1.1–1.7;  $P = 0.02$ ). Atorvastatin discontinuation was associated with SAMSs (odds ratio 1.67;  $P = 0.0001$ ) in our cohort.

## Study Highlights

### WHAT IS THE CURRENT KNOWLEDGE ON THE TOPIC?

☑ The *SLCO1B1* genotype is associated with simvastatin discontinuation.

### WHAT QUESTION DID THIS STUDY ADDRESS?

☑ Is the *SLCO1B1* genotype also associated with atorvastatin discontinuation?

### WHAT DOES THIS STUDY ADD TO OUR KNOWLEDGE?

☑ The *SLCO1B1*\*5 allele was associated with atorvastatin discontinuation (primary end point) and atorvastatin muscle side effects (secondary end point) in a practice-based data set.

### HOW MIGHT THIS CHANGE CLINICAL PHARMACOLOGY OR TRANSLATIONAL SCIENCE?

☑ These findings lay the groundwork for future comparative effectiveness studies assessing the impact of gene-based statin selection on side effect frequency and clinical efficacy.

The 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase inhibitors (statins) are among the most commonly prescribed medications in the industrialized world. Statins decrease cardiovascular event rate in the context of both primary and secondary prevention.<sup>1</sup> The combined 2018 American College of Cardiology / American Heart Association guidelines on the

treatment of blood cholesterol to reduce the atherosclerotic cardiovascular risk for adults recommend moderate to high intensity statin therapy for patients at high risk of cardiovascular events.<sup>1</sup> In patients with known cardiovascular disease, each 1 mmol/L reduction in low-density lipoprotein (LDL) cholesterol lowers the annual incidence of major vascular events by 20%.<sup>2</sup> Despite

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compelling data to support clinical efficacy, statins are still not optimally utilized.<sup>3–5</sup> Reasons for this discrepancy include provider underprescribing and patient nonadherence.

In general, statins are considered safe and well tolerated. Although they can cause asymptomatic elevation in hepatic transaminases, statin-induced liver injury is extremely rare.<sup>6,7</sup> Conversely, muscle pain (myalgias) and muscle damage (myopathy) occur quite often in the context of statin therapy. Statin-associated musculoskeletal symptoms (SAMSs) frequently lead to patient nonadherence or discontinuation.<sup>8,9</sup> In observational studies, myalgias have been reported in as many as 10% of patients taking statins. Most of these cases do not have laboratory evidence of muscle damage. Myopathy occurs much less frequently, ranging from 1% to fewer than 0.1%, depending upon the degree to which creatine kinase (CK) levels are elevated in the systemic circulation.<sup>10</sup> Rhabdomyolysis, the most extreme example of this adverse drug reaction (with CK levels exceeding 50 times the upper limit of normal), is rare with an incidence of 1:1,000,000 on statin monotherapy.<sup>9,11</sup>

Drug–drug interactions (DDIs) and drug–gene interactions (DGIs) increase the risk for statin-related muscle complications.<sup>12–14</sup> Concomitant medications influencing the absorption, distribution, metabolism, and elimination of statins have long been known to alter risk and severity for SAMSs. Cytochrome P450 (CYP3A4) inhibitors (e.g., macrolide antibiotics and grapefruit juice) increase myotoxicity risk by altering phase I metabolism (oxidation), and uridine diphosphate (UDP)-glucuronosyltransferase 1A1 (UGT1A1) inhibitors (e.g., gemfibrozil) increase the risk by altering phase II metabolism (conjugation). Solute carrier organic anion transporter family member 1B1 (SLCO1B1) inhibitors (e.g., cyclosporine) increase the risk by attenuating the hepatic uptake of statins, dampening first-pass elimination and increasing the delivery of parent drug to the systemic circulation.<sup>15</sup> These important DDIs implicate higher systemic statin exposure as a risk factor for SAMSs. For *SLCO1B1*, DGIs also lead to higher exposure for nearly all statins,<sup>13 (supplement)</sup> and the *SLCO1B1*\*5 allele has specifically been associated with increased risk of SAMSs due to simvastatin.<sup>10,16</sup> Conversely, the impact of this allele on the risk of SAMSs in patients using atorvastatin has been less well defined.

It is clear that SAMSs are a leading cause of premature statin discontinuation,<sup>10,17</sup> and for simvastatin the STRENGTH (Statin Response Examined by Genetic Haplotype Markers) trial has shown that *SLCO1B1* genotype influences a composite primary outcome of discontinuation for any reason and patient-reported musculoskeletal symptoms.<sup>16</sup> Although limited by statistical power, the STRENGTH trial also showed a trend toward significance for *SLCO1B1* genotype and patient discontinuation of atorvastatin.<sup>16</sup> To quantify the degree to which *SLCO1B1*\*5 impacts atorvastatin discontinuation in a practice-based data set, we now leverage electronic medical record data from a large observational cohort previously genotyped for *SLCO1B1* within the context of primary care.<sup>18–20</sup>

## METHODS

### Sanford healthcare system overview

The present study included retrospective analysis of clinical and genetic data from an observational cohort. The study protocol was approved

by the Institutional Review Board at Sanford Research. Study design included a retrospective assessment of prescribing pattern in a large observational cohort. Sanford Health and the Sanford Medical Center represent a fully integrated healthcare system in the upper Midwestern United States with a longstanding comprehensive electronic medical record, and the majority of patient records in this system have been electronic for over a decade.<sup>20</sup> This record includes comprehensive problem lists, detailed diagnoses and procedures, detailed prescription history, and access to over a decade of laboratory results. Since the majority of patients in the current study (> 90%) receive primary care through Sanford Health, chart fragmentation and patients leaving the health system are both minimal, and this record is therefore considered both longitudinal and comprehensive.

### Study subjects

Our study cohort consisted of patients aged 18 years or older who had been genotyped for *SLCO1B1* during the course of routine clinical care. At the time of data extraction, our electronic medical record (EMR) contained nearly 15,000 unique individual patient records (out of ~ 2 million individuals), wherein the *SLCO1B1* genotype had been obtained as part of a large array of genes tested pre-emptively in the context of primary care.<sup>19,20</sup> Since published guidelines for gene-based statin dosing have been limited to *SLCO1B1*\*5, \*15, and \*17,<sup>13,14</sup> our analyses focused on rs4145096 (Val174Ala) which is common to all three haplotypes. A total of 1,627 of these patients also had atorvastatin prescribed at some point prior to genotyping. To avoid confounding due to intervention, we censored all longitudinal data based on the date of genotyping.

### Data extraction

Clinical data extracted from the EMR were contained within multiple data frames (patient demographics, disease status, laboratory test results, prescription information, and drug allergies) with unique data identifiers linking the data sets. Prescription data entries contained dose, schedule, and route of administration. Drug allergies were extracted from the EMR as discrete fields, and statin “allergies” were restricted to one of the seven statins. Clinical indications for statin prescription (e.g., cardiovascular disease) and comorbid illness known to alter myopathy risk (e.g., thyroid disease) were collected using International Classification of Diseases, Tenth Revision codes. Medication data (including levothyroxine prescription) were also used to confirm the presence of hypothyroidism as previously described.<sup>21</sup> Clinical laboratory data reflected six unique components: thyrotropin, CK, aspartate aminotransferase (AST), alanine aminotransferase (ALT), creatinine, and estimated glomerular filtration rate (eGFR). To calculate eGFR, our clinical lab routinely uses the MDRD (Modification of Diet in Renal Disease Study) “modified 4-variable equation,” based on age, sex, race, and creatinine level. For all clinical variables used in our analytical model (e.g., age and eGFR), our analyses included the value closest to genotyping date.

Pharmacogenetic data included genotype results from the Fluidigm SNP Dynamic Array platform (San Francisco, CA, USA) and TaqMan Assays (Waltham, MA, USA) routinely run at our institution as part of the Sanford Chip.<sup>19,20</sup> Because this platform multiplexes most actionable pharmacogene loci with implementation guidelines published by the Clinical Pharmacogenetics Implementation Consortium (CPIC),<sup>19,20</sup> the *SLCO1B1* genotype was available for all study subjects in our study cohort. Published guidelines for gene-based statin dosing have specifically been limited to *SLCO1B1*\*5, \*15, and \*17.<sup>13,14</sup> The single nucleotide polymorphism rs4149056 is common to all three *SLCO1B1* haplotypes of interest, and it is therefore the only allele that we analyzed for association with atorvastatin discontinuation and SAMSs when assessing the role of the *SLCO1B1* genotype. This approach was the same approach that we used successfully in our prior study demonstrating association between *SLCO1B1* genotype, SAMSs, and simvastatin discontinuation in the STRENGTH Trial.<sup>16</sup> We have also previously shown that a loss-of-function variant in a second pharmacogene, the *CYP3A5*\*3 allele,

can influence the severity of statin myopathy (degree of CK elevation), although this variant did not predict who developed the muscle side effects.<sup>1,22</sup> To determine whether *CYP3A5\*3* is associated with SAMSs and atorvastatin discontinuation, we tested its defining polymorphism, *rs776746*, for association with our primary end points as well. The rate of genotyping no-calls for the pharmacogenetic variants in our analysis was < 1%.

### Outcome definitions and statistical analyses

Starting from a list of all patients with *SLCO1B1* genotyping, the data were filtered down to include those individuals with a history of having received a prescription for atorvastatin prior to pharmacogenetic testing. Each data string was then censored at the time of pharmacogenetic testing, and each individual patient was labeled as either continuing or discontinuing (primary outcome) based upon whether atorvastatin was present in their medication list on the date of pharmacogenetic testing. The *discontinuation* end point and the number of *SLCO1B1\*5* variant alleles were then analyzed using an additive genetic model to determine dependence of adherence on genotype, specifically at the date of pharmacogenetic testing.

Initially, logistic regression was used to assess the strength of association between atorvastatin discontinuation and genotype, using both a univariate and multivariate approach. Demographic covariates were age, sex, and race. Clinical covariates included the indication for statin therapy, as well as relevant comorbidities such as thyroid disease. Concomitant medications, relevant lab data, and atorvastatin dose were also included in the final model. Before fitting the full model, we verified that all variables roughly held to the assumptions for logistic regression (no serious multicollinearity, linear relationship of logit and predictors, and lack of influential values). Lastly, a Cox proportional hazards model was then used to illustrate the association of *SLCO1B1\*5* carrier status with (i) time to atorvastatin discontinuation, as well as (ii) time to onset of SAMSs (secondary outcome) represented as a composite variable: elevated CK (CK level) > 3 times the upper limit of normal, diagnosis codes for myalgia, myopathy, or rhabdomyolysis, and/or a new atorvastatin allergy.

### RESULTS

Our study cohort consisted of 1,627 unique individuals who had received an atorvastatin prescription prior to their date of genotyping within the context of routine care. As shown in **Table 1**, the mean age of our study subjects was  $64 \pm 11$  years, and there was a slight difference in the distribution of age between continuers and discontinuers ( $P = 0.02$ , Wilcoxon rank sum test (Mann-Whitney *U* test)); i.e., discontinuers were slightly younger. There was also a difference in the distribution of sex ( $P = 0.0004$ , Fisher exact test), and in the distribution of pre-existing thyroid disease ( $P = 0.02$ , Fisher exact test). The frequency of pre-existing vascular disease (coronary artery disease, peripheral artery disease, and cerebrovascular disease) was the same for continuers and discontinuers, indicating that confounding by indication for statin therapy was unlikely. Genotype distribution in this cohort was similar to the distribution of *SLCO1B1\*5* in other cohorts: homozygous major allele (TT) 71.7%, heterozygous (TC) 26.2%, and homozygous minor allele (CC) 2.2%.

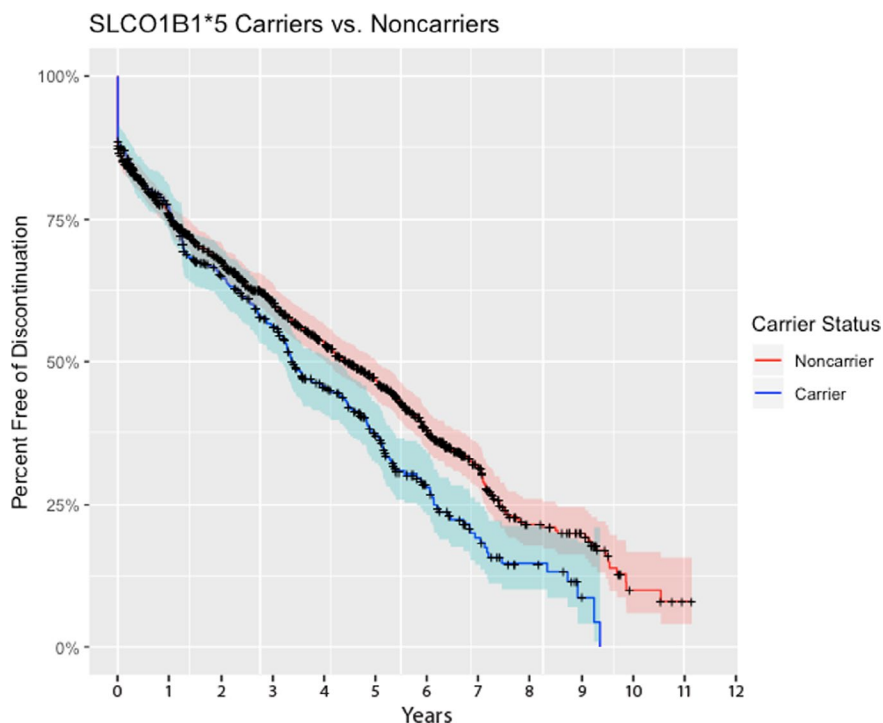
### Association with drug discontinuation

Our initial univariate test for genotype-phenotype association confirmed that *SLCO1B1\*5* is associated with atorvastatin discontinuation ( $P = 0.0444$ ). As shown in **Table 1**, logistic regression using an additive genetic model revealed an odds ratio of 1.2 (95% confidence interval (CI), 1.0049–1.4723). Within this cohort, ~ 1 in 2 patients that started atorvastatin eventually discontinued the drug over the 12-year time window illustrated in **Figure 1**. It is important to note that this represents the discontinuation rate for one statin—atorvastatin; i.e., 50% atorvastatin discontinuation does not mean 50% discontinuation of all statins. Many of these patients were then treated with a different statin, as well as diet

**Table 1** Baseline characteristics and univariate association with atorvastatin discontinuation

Characteristic	Overall cohort (n = 1,627)	Continuers (n = 715)	Discontinuers (n = 912)	P value continuers vs. discontinuers
Age, mean (SD)	63.8 (11.3)	64.6 (10.7)	63.2 (11.7)	0.0176
Female sex, %	44.3%	39.3%	48.1%	0.0004
Non-white race, %	1.9%	2.2%	1.6%	0.4656
Smoking, %				
Current	6.6%	6.2%	6.9%	0.6146
Former	43.9%	43.6%	44.1%	0.8800
Never	49.5%	50.2%	49.0%	0.6531
Comorbidities (n, %)				
Thyroid Disease	313, 19.2%	119, 16.6%	194, 21.3%	0.0191
Neuromuscular Disease	18, 1.1%	7, 1.0%	11, 1.2%	0.8125
CAD	257, 15.8%	124, 17.3%	133, 14.6%	0.1325
PAD	45, 2.8%	24, 3.4%	21, 2.3%	0.2239
CVA	9, 0.6%	5, 0.7%	4, 0.4%	0.5177
<i>SLCO1B1</i> genotype (%)				
TT	71.7%	74.4%	69.5%	0.0307
TC	26.2%	23.9%	28.0%	0.0692
CC	2.2%	1.7%	2.5%	0.3023

CAD, coronary artery disease; CVA, cerebrovascular accident; PAD, peripheral artery disease.



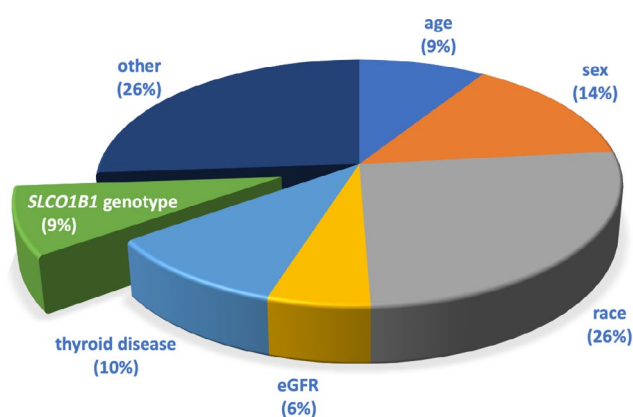
**Figure 1** Primary end point: atorvastatin discontinuation, stratified by genotype. Kaplan-Meier curves showing the influence of *SLCO1B1*\*5 over time for study subjects who discontinued atorvastatin ( $n = 912$ ). Carrier status for the *SLCO1B1*\*5 allele is associated with time-to-atorvastatin discontinuation (hazard ratio 1.2; 95% confidence interval, 1.1–1.4;  $P = 0.004$ ).

and exercise. These findings are consistent with our prior results in the STRENGTH Trial.<sup>16</sup>

Since other clinically actionable absorption, distribution, metabolism, and elimination gene variants were also available in our practice-based data set,<sup>20</sup> we also tested the *CYP3A5* genotype for association with atorvastatin discontinuation. While *CYP3A5*\*3 has previously been reported to influence the severity of SAMSs in patients using atorvastatin,<sup>22</sup> the *CYP3A5*\*3 allele was not associated with atorvastatin discontinuation in our current study cohort ( $P = 0.823$ ). This is consistent with prior reports of relative effect size (*CYP3A4* and *CYP3A5* variants have a modest effect in comparison with the large effect of *SLCO1B1*\*5, where the odds ratio can approach 20.0).<sup>10</sup>

To further understand the association between the *SLCO1B1* genotype and atorvastatin discontinuation, we used a Cox proportional hazards model to illustrate this relationship over time. The Kaplan-Meier curves are shown in **Figure 1** for discontinuers only ( $n = 912$ ). Due to the underrepresentation/rarity of patients who are homozygous for the loss-of-function variant (*SLCO1B1*\*5/\*5) in this cohort, we were unable to adequately evaluate the comparison between the \*5/\*5 and \*1/\*5 genotypes with sufficient power. Therefore, in our time-to-event analyses (the Kaplan-Meier plots), we have binned both heterozygotes and homozygotes together, reporting the collective impact of “carrier status.” The adjusted hazard ratio of 1.2 (95% CI, 1.1–1.4), confirms that the *SLCO1B1* genotype is also associated with atorvastatin discontinuation using a multivariate approach ( $P = 0.004$ ). The relative impact of each variable in our model is further illustrated in **Figure 2**. Based on the squared semipartial

correlations for each covariate, the relative contributions are as follows: *SLCO1B1* genotype (8.8%), age (9.0%), sex (14.3%), eGFR (5.5%), and the presence or absence of thyroid disease (10.2%). Race also appeared to contribute to the model (26.1%); however, small cell size for some subpopulations may have overestimated the role of race.



**Figure 2** Relative contribution of genotype. *SLCO1B1* genotype explains 9% of the variance in atorvastatin discontinuation (multivariate model). Age and estimated glomerular filtration rate (eGFR) were included in the model as continuous variables. Sex, race, and the presence or absence of thyroid disease were included as categorical variables. The contributions of each variable were as follows: *SLCO1B1* genotype (8.8%), age (9.0%), self-reported race (26.1%), self-reported sex (14.3%), eGFR (5.5%), and pre-existing thyroid disease (10.2%).



**Table 2 Multivariate Cox-proportional hazards model**

Characteristic	Hazard ratio	95% CI	P value
Age (per 10 years)	0.87337	(0.81576, 0.9350)	0.00010
Female sex (vs. male)	1.3136	(1.1363, 1.5185)	0.00023
Non-white race (vs. white)	0.76048	(0.2838, 2.037)	0.58607
Smoking status (vs. never)			
Current	1.22693	(0.93398, 1.612)	0.14176
Former	1.12616	(0.97714, 1.298)	0.10088
Comorbidities			
Thyroid disease	1.09447	(0.92513, 1.295)	0.29252
Neuromuscular disease	0.89730	(0.47508, 1.695)	0.73838
Coronary artery disease	0.88234	(0.71645, 1.087)	0.23880
Peripheral arterial disease	0.92835	(0.59357, 1.452)	0.74460
Cerebrovascular disease	0.72383	(0.26925, 1.946)	0.52183
Concomitant medications			
SLCO1B1 inhibitor	0.69356	(0.39668, 1.212)	0.19887
CYP3A4/5 inhibitor	0.96884	(0.79653, 1.178)	0.75140
Combined inhibitor SLCO1B1-CYP3A4/5	2.05678	(0.17335, 5.929)	0.18184
eGFR (per Standard Deviation)	1.01787	(0.94771, 1.093)	0.62696
Initial dose (per 10 mg)	0.94308	(0.90384, 0.984)	0.00688
SLCO1B1*5 carrier status	1.2350	(1.06823, 1.428)	0.00435

Multivariate Cox-proportional hazards model including SLCO1B1 genotype and relevant clinical covariates as a function of time to atorvastatin discontinuation. CI, confidence interval; CYP3A4/5, cytochrome P450 3A4/3A5 isozyme; eGFR, estimated glomerular filtration rate; SLCO1B1, solute carrier organic anion transporter family member 1B1.

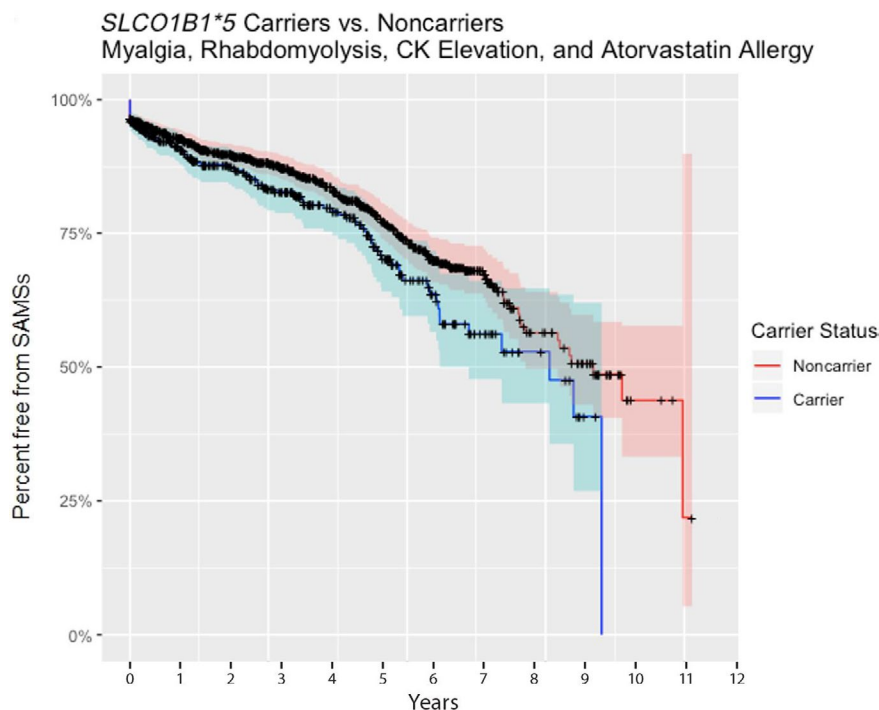
The full multivariate model is presented in **Table 2**. Disease variables were transformed for pre-existing vascular disease, neuromuscular disease, and thyroid disease, as well as for diagnostic codes representing myalgia, myopathy, and rhabdomyolysis. Direct measures of muscle toxicity (CK level) and liver toxicity (AST and ALT levels) were not obtained frequently enough in the context of routine clinical practice to include them in the final multivariate model. However, creatinine and eGFR were obtained much more frequently during routine practice (and eGFR was available for 99% of all study subjects within our cohort). To avoid redundancy, creatinine level was omitted and eGFR was used as a single measure of kidney function in the final model. Using this multivariate approach, *SLCO1B1* genotype was again strongly associated with our primary end point, atorvastatin discontinuation ( $P = 0.004$ ).

### Association with muscle side effects

Since our primary end point, “atorvastatin discontinuation for any reason,” was associated with the *SLCO1B1* genotype over time, we then sought to determine whether the *SLCO1B1* genotype was also associated with the presence of adverse muscle symptoms over time—the leading cause of premature statin discontinuation. To quantify the effect of *SLCO1B1*\*5 alleles on SAMSs as a secondary outcome, we built a composite variable containing a combination of diagnostic codes available longitudinally within the EMR (i.e., International Classification of Diseases, Tenth Revision codes for myalgia, myopathy or rhabdomyolysis), clinical laboratory data (threefold elevation in serum CK levels), and data contained in the allergy section of our EMR (presence of an “atorvastatin allergy”

entered into the EMR after a prescription of atorvastatin had been initiated but prior to genotyping). We chose to use a threshold of CK > 3 times the upper limit of normal in this composite because this was the CK threshold set for myopathy case definition in the landmark SEARCH (Study of the Effectiveness of Additional Reductions in Cholesterol and Homocysteine) Trial, wherein a GWAS revealed *SLCO1B1* as the only locus associated with simvastatin myopathy at a level that reached genome-wide significance.<sup>10</sup> While serum CK levels were not available for all individuals in our clinical practice-based study cohort (CK levels are not drawn routinely for medication surveillance), 30% of the cohort (495 of our 1,627 study subjects) did have CK data available. Allergy entries for any clinically available statin drug were available in 24% of the cohort (392 of our 1,627 study subjects).

Overall, the SAMSs composite occurred in a total of 303 study subjects within our entire study cohort of 1,627 unique individuals exposed to atorvastatin. The SAMSs variable was more frequently observed in patients with the *SLCO1B1*\*5 allele (95/461, 20%) than in patients without the *SLCO1B1*\*5 allele (208/1,166, 18%). The frequency of this composite SAMSs variable was similar to the frequency of SAMSs observed for patients using statins in the STRENGTH Trial.<sup>16</sup> The Kaplan-Meier curves representing this Cox proportional hazards model are shown in **Figure 3**. The adjusted hazard ratio of 1.4 (95% CI, 1.1–1.7), clearly indicates that *SLCO1B1* genotype is also associated with SAMSs ( $P = 0.02$ ). Further, within the entire cohort ( $n = 1,627$ ), 200 of the 912 discontinuers (22%) met criteria for our composite end point representing SAMSs, whereas only 103 of the 715 (14%) continuers met these criteria. Thus, there was a strong association between SAMSs and atorvastatin discontinuation (odds



**Figure 3** Secondary end point: statin-associated musculoskeletal symptoms (SAMSs). Kaplan-Meier curves showing the influence of *SLCO1B1*\*5 on rate of development for SAMSs (SAMSs composite occurred in 303 study subjects). Carrier status for the *SLCO1B1*\*5 allele is associated with SAMSs in a time-to-event analysis (hazard ratio 1.4; 95% confidence interval, 1.1–1.7;  $P = 0.02$ ). CK, creatine kinase.

ratio 1.67,  $P = 0.0001$ ; Fisher exact test). It is therefore likely that SAMSs had contributed, at least in part, to patient discontinuation of this important drug in the context of routine care.

## DISCUSSION

The *SLCO1B1* gene encodes a membrane transporter that facilitates the hepatic uptake and first-pass elimination of statins, including atorvastatin. The *SLCO1B1*\*5 variant represents a loss-of-function allele previously shown to attenuate the hepatic uptake of atorvastatin, resulting in an increase in the amount of parent drug circulating in the systemic circulation.<sup>13</sup> We now demonstrate that this loss-of-function allele is associated with SAMSs and atorvastatin discontinuation in longitudinal data from a clinical practice-based cohort.

### Atorvastatin discontinuation (primary end point)

In our univariate analysis ( $P = 0.0444$ ), the odds ratio for atorvastatin discontinuation per allele was 1.2 (95% CI, 1.0049–1.4723). Although the association between the *SLCO1B1* genotype and hepatic uptake of statins is a class effect, the magnitude of this pharmacokinetic change varies by statin.<sup>13</sup> The net effect appears to be largest for simvastatin, where a 220% increase in the area under the curve has been documented for patients homozygous for the *SLCO1B1*\*5 allele.<sup>13,14</sup> Over a decade ago, we demonstrated that, for simvastatin, the *SLCO1B1*\*5 carrier status is associated with discontinuation for any reason ( $P = 0.01$ ;  $n = 150$ ).<sup>16</sup> While the same study observed a trend (odds ratio = 1.2;  $P = 0.2$ ) toward association of *SLCO1B1*\*5 with discontinuation or side effects for atorvastatin as well, the statistical power of that trial was limited

due to the small number of participants ( $n = 150$ ) and short duration of follow-up (16 weeks). To date, this initial observation had not yet been replicated for atorvastatin. In the current study we tested the hypothesis that *SLCO1B1*\*5 is associated with atorvastatin discontinuation in a larger observational cohort of patients receiving atorvastatin in the context of routine clinical care. The larger sample size ( $n = 1,627$ ) and practice-based nature of our longitudinal data have allowed us to replicate the findings for atorvastatin from prior work<sup>16</sup> (discontinuation for any reason) in an adequately powered cohort with long-term follow-up, and they have positioned us to extend these findings by quantifying the impact of genotype alongside relevant covariates.

To define the relative impact of genotype (vs. covariates) within our current practice-based data set, we built a multivariate model which refined our understanding of each clinical risk factor influencing atorvastatin intolerance. As shown in Figure 2, the full multivariate model identified age, sex, and the presence or absence of thyroid disease as strong predictors of atorvastatin discontinuation risk in this cohort. Our model also identified race as a potential contributor to risk, when European ancestry, Asian ancestry, African ancestry, and Native American ancestry were considered separately; however, the small sample size for each of the cells containing patients of non-European ancestry raises the possibility that the association of race with atorvastatin discontinuation may be overestimated in this cohort.

Thyroid disease has long been known to increase patient risk for myopathy on statins. In the PRIMO (Prediction of Muscular Risk in Observational Conditions) Trial,<sup>23</sup> the odds ratio for statin-induced muscle pain was 1.74 in patients with a prior

diagnosis of hypothyroidism (95% CI, 1.19–2.55;  $P = 0.004$ ). Thyroid disease was a strong predictor of atorvastatin discontinuation in our current practice-based cohort (univariate analysis,  $P = 0.0191$ ). As shown in **Figure 2**, the presence of thyroid disease explains ~ 10% of the variance in atorvastatin discontinuation. The specific cellular mechanism underlying this interaction (the impact of thyroid disease on statin myopathy risk) remains poorly understood.<sup>24</sup> In animal models, extreme variation in tri-iodothyronine (T3) levels—either too high or too low—are known to alter binding of thyroid nuclear receptor- $\alpha$  (THRA) to its thyroid response elements, disrupting the normal control of gene expression networks regulating muscle mass as a function of age.<sup>25</sup> Derangements in thyroid homeostasis may therefore predispose some patients to develop SAMSs by altering the expression of THRA-dependent pathways involved in contractility (e.g., myosin isoforms) as well as the remodeling of extracellular matrix (e.g., integrin isoforms).

### Muscle side effects (secondary end point)

Since our primary end point was atorvastatin discontinuation, there are many factors which could have contributed to the association observed in our longitudinal data. These factors include potential lack of efficacy, toxicity (hepatotoxicity as well as myotoxicity), changes in provider prescribing patterns over time, changes in formulary, prescription cost (availability of generic alternatives), and patient adherence. We therefore also sought to define the relationship between the *SLCO1B1*\*5 allele and the frequency of adverse muscle symptoms while these patients were on atorvastatin. Our observation that the *SLCO1B1* genotype is also associated with SAMSs strongly suggests that muscle pain may have been a contributor to the association we observed between the *SLCO1B1*\*5 allele and drug discontinuation.

To quantify the magnitude of the relationship between the *SLCO1B1* genotype and adverse muscle symptoms related to the use of atorvastatin in this cohort, we constructed a composite end point representing SAMSs within longitudinal clinical data. This secondary end point included diagnostic codes (myalgia, myopathy, rhabdomyolysis), objective lab data already available in the chart (CK level > 3 times the upper limit of normal), and the presence of atorvastatin “allergy” (recorded as a flag in the allergy section of our EMR). As a composite end point, SAMSs was strongly associated with the *SLCO1B1* genotype in this cohort (odds ratio 1.67,  $P = 0.0001$ ). As shown in **Figure 3**, our data indicate that the increased risk of developing SAMSs may occur up to a year after starting the drug. Decision support is therefore needed to identify patients at risk.

Decision support is already commonly used to alert providers to patients who may be at increased risk of developing SAMSs, based on DDI. Cyclosporine, a competitive inhibitor at the level of the organic anion transporter encoded by *SLCO1B1*, is known to increase risk for SAMSs. Atorvastatin is a known substrate for this transporter, and coadministration of cyclosporine increases risk for atorvastatin-induced myopathy.<sup>26–28</sup> We have recently shown that longitudinal data from EMRs can be used to validate this DDI as well as identify other concomitant medications previously unrecognized to increase risk for atorvastatin intolerance.<sup>28</sup>

Our current data demonstrate that a similar approach can be leveraged to confirm the clinical actionability of DGI for statins as well. Decision support flagging *SLCO1B1*\*5 is already deployed in many EMRs for simvastatin<sup>18,29,30</sup> based on published guidelines from CPIC.<sup>13,14</sup> Our findings indicate that this decision support and the CPIC guidelines surrounding *SLCO1B1* should be expanded to include atorvastatin.

### Genotyping may improve adherence

Patients frequently discontinue statin use due to muscle side effects. We have previously demonstrated that genetic variation in *SLCO1B1* is associated with SAMSs due to simvastatin and patient discontinuation of simvastatin.<sup>16</sup> We now demonstrate that *SLCO1B1* genotype is associated with SAMSs due to atorvastatin and patient discontinuation of atorvastatin. Thus, our findings help to establish the clinical validity of the *SLCO1B1* genotype as a risk marker for atorvastatin-related muscle side effects, and they expand the scope of this DGI beyond simvastatin to include the most commonly prescribed drug in this class.

This marker, the *SLCO1B1* genotype, may also have utility in guiding choice of therapeutic alternatives to mitigate the risk of SAMSs. Rosuvastatin, for example, represents another high-potency statin capable of achieving the robust reduction in LDL cholesterol that is expected with atorvastatin. In the JUPITER (Justification for the Use of Statins in Prevention: An Intervention Trial Evaluating Rosuvastatin) trial, where patients were randomized and double-blinded to placebo or rosuvastatin 20 mg/day, a substudy of more than 8,700 participants demonstrated that carriers of *SLCO1B1*\*5 were at no higher risk of SAMSs with rosuvastatin than with placebo.<sup>31</sup> Although studies directly comparing the safety of rosuvastatin to the safety of simvastatin or atorvastatin have not been performed specifically in carriers of *SLCO1B1*\*5, many clinical centers are now implementing clinical decision support tools for *SLCO1B1*\*5 genotype based on its interactions with simvastatin.<sup>18,29</sup> We anticipate that our data will form the foundation for expansion of these tools to atorvastatin.

In general, statin discontinuation is common.<sup>32</sup> In patients with known cardiovascular disease, around 50% discontinue their initial statin within a year.<sup>5</sup> Although reasons for nonadherence to statins are multifactorial, side effects are cited as the main reason patients prematurely discontinue statin therapy.<sup>33</sup> Therefore, it is plausible that *SLCO1B1*-informed statin therapy with the aim of reducing muscle side effects may lead to improved adherence for this important class of drugs. CPIC already provides guidance related to the choice of specific alternative therapies in carriers of *SLCO1B1*\*5.<sup>13,14</sup> If a low-potency statin is acceptable, the area under the curve (AUC) is affected the least by *SLCO1B1*\*5 for fluvastatin.<sup>13</sup> If a moderate- or high-potency statin is required, the AUC for pravastatin and rosuvastatin are impacted less by the *SLCO1B1*\*5 variant than the AUC for simvastatin or atorvastatin.<sup>13,34</sup> If a high-potency statin is absolutely necessary, the JUPITER Trial has shown that rosuvastatin is safe in carriers of *SLCO1B1*\*5.<sup>31</sup>

We have previously demonstrated, in a longitudinal cohort of patients known to have been nonadherent with statins, that *SLCO1B1* genotyping—with communication of test results and the

alternative therapies described above to reduce side effect risks—can lead to lower LDL-cholesterol levels through improved patient acceptance of guideline-appropriate, new statin prescriptions.<sup>35</sup> This nonrandomized prospective study delivered simple, written, patient-facing and provider-facing educational materials regarding *SLCO1B1* genetic test results and recommendations delivered without the assistance of a pharmacist. More recently, we have confirmed the effectiveness of this approach using the same intervention in a randomized trial of 159 patients who had previously discontinued their statins due to myalgias, again demonstrating that *SLCO1B1* gene-based statin prescribing leads to an increased use of new statin prescriptions (55.4% vs. 38%,  $P = 0.040$ ) and lower LDL-cholesterol levels ( $132 \pm 42$  vs.  $144 \pm 43$  mg/dL at 3 months of follow-up;  $P = 0.048$ ).<sup>36</sup> Because participants were followed for < 1 year in these studies, the long-term effects of delivering *SLCO1B1*-guided statin therapy on adherence and LDL-cholesterol level remain unknown. Future comparative effectiveness studies are therefore needed to assess the impact of *SLCO1B1*-informed decision support for statins on their side effect frequency, adherence, and clinical efficacy in very large cohorts followed across multiple institutions.

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#### CONFLICT OF INTEREST

The authors declared no competing interests for this work.

#### AUTHOR CONTRIBUTIONS

D.V., J.B., A.M., S.N.G., R.A.W., A.N.M., C.H., and E.A.L. wrote the manuscript. D.V., J.B., A.M., and R.A.W. designed the research. D.V., J.B., A.M., S.N.G., R.A.W., A.N.M., C.H., and E.A.L. performed the research. D.V., A.M., R.A.W., C.H., and E.A.L. analyzed the data.

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