Association of genetic variations with pharmacokinetics and lipid-lowering response to atorvastatin in healthy Korean subjects

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Background: Statins are effective agents in the primary and secondary prevention of cardiovascular disease, but treatment response to statins varies among individuals. We analyzed multiple genetic polymorphisms and assessed pharmacokinetic and lipid-lowering responses after atorvastatin 80 mg treatment in healthy Korean individuals.

Methods: Atorvastatin 80 mg was given to 50 healthy Korean male volunteers. Blood samples were collected to measure plasma atorvastatin and lipid concentrations up to 48 hours after atorvastatin administration. Subjects were genotyped for 1,936 drug metabolism and transporter genetic polymorphisms using the Affymetrix DMET plus array.

Results: The pharmacokinetics and lipid-lowering effect of atorvastatin showed remarkable interindividual variation. Three polymorphisms in the SLCO1B1, SLCO1B3, and ABCC2 genes were associated with either the maximum concentration (Cmax) of atorvastatin or changes in total cholesterol or low-density lipoprotein cholesterol (LDL-C). Minor homozygotes (76.5 ng/mL) of SLCO1B1 c.-910G>A showed higher Cmax than heterozygotes (34.0 ng/mL) and major homozygotes (33.5 ng/mL, false discovery rate P=0.040). Cmax and the area under the plasma concentration curve from hour 0 to infinity (AUC0-∞) were higher in carriers of the SLCO1B1*17 haplotype that included c.-910G>A than in noncarriers (46.1 vs 32.8 ng/mL for Cmax; 221.5 vs 154.2 ng/mL for AUC0-∞). SLCO1B1 c.-910G>T homozygotes (63.0 ng/mL) also showed higher Cmax than heterozygotes (34.7 ng/mL) and major homozygotes (31.4 ng/mL, FDR P=0.037). A nonsynonymous ABCC2 c.1249G>A was associated with small total cholesterol and LDL-C responses (0.23% and −0.70% for G/A vs −11.9% and −17.4% for G/G). The Cmax tended to increase according to the increase in the number of minor allele of SLCO1B1 c.-910G>A and SLCO1B3 c.334G>T.

Conclusion: Genetic polymorphisms in transporter genes, including SLCO1B1, SLCO1B3, and ABCC2, may influence the pharmacokinetics and lipid-lowering response to atorvastatin administration.

Keywords: atorvastatin, pharmacokinetics, pharmacogenomics, SLCO1B1, SLCO1B3, ABCC2

Introduction

As therapeutic agents administered to reduce the risk of cardiovascular disease and manage hypercholesterolemia, statins upregulate low-density lipoprotein (LDL) receptors, increase plasma clearance of LDL, and reduce hepatic secretion of apolipoprotein B (ApoB)-containing lipoproteins, very low-density lipoprotein (VLDL), and LDL. Statins can reduce the plasma concentration of low-density lipoprotein cholesterol (LDL-C) by as much as 50% as well as triglycerides.

Atorvastatin is a potent competitive inhibitor of 3-hydroxy-3-ethylglutaryl-coenzyme A (HMG-CoA) reductase, an enzyme that catalyzes conversion of...
HMG-CoA to mevalonate, an early rate-limiting step in cholesterol synthesis. In spite of the beneficial effects of statin treatment in cardiovascular disease prevention, responses to statin therapy show considerable interindividual variation, and some patients may not achieve sufficient LDL-C reduction even with the most efficacious statins.

Genetic factors are expected to be part of the interindividual variation in the pharmacokinetic and pharmacodynamic response to statins. Various genes encoding for enzymes and transporters that influence pharmacokinetics and the targets of pathways on which a drug acts, as well as those involved in related disease conditions, have been evaluated in candidate gene studies and hypothesis-free genome-wide investigations. Genetic variations on drug transporter genes, ABCB1 and SLCO1B1; P450 system genes, CYP3A4, CYP3A5, and CYP2D6; and other genes encoding lipoproteins and enzymes of lipid metabolic pathways such as APOE and HMGACR, have been suggested to have associations with statin responsiveness. Genetic variations that affect pharmacokinetics of statins may modify atorvastatin disposition and hence its efficacy and toxicity. However, the effects of genetic variations have been inconsistently replicated, and there are relatively few data available in Asian populations. In addition, there are lots of variations in metabolic processes according to statin type, and in the frequency of genetic variations and responsiveness to statins according to ethnic background.

Accordingly, we investigated pharmacokinetic and pharmacodynamic changes in healthy Korean individuals after high-dose atorvastatin administration through serial plasma measurements of drug and lipid concentrations. We assessed associations between genetic variations and pharmacokinetics or lipid-lowering effects of atorvastatin using a predesigned gene panel including genes related to absorption, distribution, metabolism, and elimination.

Materials and methods

Subjects and study design

This study enrolled 50 healthy Korean male subjects. All subjects were from unrelated families and were ascertained to be healthy by medical history, physical examination, vital signs, electrocardiography, and routine clinical laboratory tests. Subjects were given a single oral dose of 80 mg atorvastatin calcium at 08:00 am with 240 mL of water in the overnight fasting state. Subjects fasted for 4 hours after atorvastatin administration, then lunch and dinner were served. Venous blood samples for pharmacokinetic analysis were collected via an intravenous catheter at 0, 0.25, 0.5, 0.75, 1, 1.5, 2, 3, 4, 6, 8, 10, 12, 24, 36, and 48 hours after dosing. Plasma concentrations of total cholesterol, LDL-C, and triglycerides were measured before and at 24 and 48 hours after atorvastatin administration. Blood sampling for genotyping was performed before drug administration. The study protocol was approved by the Institutional Review Board of Dankook University and Samsung Medical Center, Korea. Written informed consent was obtained from each participant.

Pharmacokinetic and pharmacodynamic measurements

Plasma concentrations of atorvastatin were determined by liquid chromatography–tandem mass spectrometry (LC–MS/MS) using a TSQ Quantum Discovery mass spectrometer (Thermo Electron, San Jose, CA, USA). The ion transitions monitored were m/z 559.2 → 440. Pharmacokinetic parameters were determined by BA-Calc software (Korea Food and Drug Administration, Korea) using actual sampling times. Plasma concentrations of the terminal phase were fitted to a log-linear line by the least squares method to obtain the elimination rate constant. The area under the plasma concentration curve from hour 0 to infinity (\(\text{AUC}_{\infty}\)) was calculated using a combination of the trapezoidal rule and extrapolation to infinity by the elimination rate constant. The maximum drug concentration in plasma (\(C_{\text{max}}\)) and time to \(C_{\text{max}}\) (\(t_{\text{max}}\)) were determined from observed values. Clearance (CL) of atorvastatin was adjusted according to the body weight of each subject. Plasma lipid concentrations were measured with an Hitachi 7600-110 chemistry analyzer (Hitachi, Tokyo, Japan).

Genotyping

Genomic DNA was isolated from peripheral blood samples using the Wizard Genomic DNA Purification Kit (Promega, Madison, WI, USA). Genotyping was performed using the Affymetrix drug-metabolizing enzyme and transporter (DMET) Plus array (Affymetrix, Santa Clara, CA, USA), which gauges 1,936 polymorphisms from 225 genes encoding phase I and phase II drug metabolism enzymes as well as drug transporters. Briefly, we determined the yields of pure double-stranded genomic DNA samples before genotyping. Samples were adjusted to concentrations of 60 ng/μL. Normalized genomic DNA (17 μL) was used as a template for DMET arrays. For loci that had pseudogenes and close homologs, initial genomic amplification using locus-specific primers in a multiplex polymerase chain reaction (mPCR) was performed. By hybridization of highly
selective molecular inversion probes (MIPs) to their complementary genomic templates, sequences containing polymorphisms of interest were amplified and then fragmented to improve hybridization onto DMET arrays. Hybridized DMET arrays were scanned with an Affymetrix GeneChip Scanner 3000 7G. Genotyping was performed according to the predefined software algorithms of the manufacturer using DMET Console version 1.0.16,18

Statistical analyses
Of 1,936 polymorphisms in 225 genes screened, 519 non-monomorphic polymorphisms in 181 genes were identified with a ≥90% call rate, ≥5% minor allele frequency, and nonsignificant deviation from Hardy–Weinberg equilibrium ($P$>0.001). Analysis of variance (ANOVA) or Kruskal–Wallis tests were used to test for associations between genotypes and pharmacokinetic parameters or lipid concentration changes from baseline to 48 hours after atorvastatin administration. Pharmacokinetic parameters included $\text{AUC}_{\infty}$, $\text{C}_{\text{max}}$, and clearance adjusted with body weight (CL$_{\text{adj}}$). An ANOVA test was applied for polymorphisms that satisfied the assumptions of normality and homogeneity of variances in phenotype distribution. A $P$-value less than 0.050 was considered statistically significant. For statistically significant associations, the Jonckheere–Terpstra test was performed to test for ordered differences among genotypes. Corrected $P$-values were obtained using the Benjamini–Hochberg false discovery rate (FDR) approach. Statistical analyses were conducted using R, version 2.9.1 (R Foundation for Statistical Computing, Vienna, Austria), and IBM SPSS Statistics version 18.0 (SPSS Inc., Chicago, IL, USA).

Results
Participant demographics
A total of 50 healthy individuals were included in this study. Baseline characteristics are presented in Table 1. All subjects were male with a mean age of 24 years (range, 20–27 years) and a mean body weight of 69 kg (50–98 kg). Mean plasma concentrations of total cholesterol, LDL-C, and triglycerides were 150, 72.6, and 110 mg/dL, respectively. The CVs of dose-per-body weight normalized AUC$_{\text{c}}$ and $\text{C}_{\text{max}}$ were 31.0 ng/mL per mg/kg and 31.0 ng/mL per mg/kg, respectively. The CVs of dose-per-body weight normalized AUC$_{\text{c}}$ and $\text{C}_{\text{max}}$ were 59.0% and 54.4%.

Atorvastatin pharmacokinetics and pharmacodynamics
Pharmacokinetic properties of atorvastatin are summarized in Table 2. The mean $\text{AUC}_{\infty}$, $\text{C}_{\text{max}}$, $t_{\text{max}}$, CL, CL$_{\text{adj}}$, and half-life ($t_{1/2}$) were 172 ng·h/mL, 36.2 ng/mL, 1.07 h, 603 L/h, 8.85 L/(h·kg), and 7.75 h, respectively. Pharmacokinetic parameters showed marked interindividual variability, with a coefficient of variation (CV) ranging from 33.3% to 81.8%. The mean and standard deviation (SD) of the plasma concentration–time profile for atorvastatin after a single oral administration in all participants are shown in Figure 1A. The mean dose-per-body weight normalized AUC$_{\text{c}}$ and $\text{C}_{\text{max}}$ were 148 ng·h/mL per mg/kg and 31.0 ng/mL per mg/kg, respectively. The CVs of dose-per-body weight normalized AUC$_{\text{c}}$ and $\text{C}_{\text{max}}$ were 59.0% and 54.4%.

Table 1 Demographics and baseline characteristics (n=50)

| Variable                  | Mean (range) |
|---------------------------|--------------|
| Age (years)               | 24 (20–27)   |
| Gender, n (%)             |              |
| Male                      | 50 (100.0)   |
| Female                    | 0 (0.0)      |
| Body weight (kg)          | 69 (50–98)   |
| Baseline lipid concentration (mg/dL) |          |
| Total cholesterol, n (%)  |              |
| ≥200                      | 150 (101–212)|
| <200                      | 2 (4.0)      |
| LDL-C, n (%)              |              |
| ≥130                      | 27.2 (42–108)|
| <130                      | 0 (0.0)      |
| Triglycerides, n (%)      |              |
| ≥150                      | 110 (52–233) |
| <150                      | 9 (18.0)     |

| Genotype, n (%)           |              |
|----------------------------|--------------|
| SLCO1B1 c.-910G>A         |              |
| G/G                       | 33 (66.0)    |
| G/A                       | 14 (28.0)    |
| A/A                       | 3 (6.0)      |
| SLCO1B3 c.334G>T          |              |
| G/G                       | 28 (56.0)    |
| G/T                       | 16 (32.0)    |
| T/T                       | 6 (12.0)     |
| ABC22 c.1249G>A           |              |
| G/G                       | 43 (86.0)    |
| G/A                       | 7 (14.0)     |

Abbreviation: LDL-C, low-density lipoprotein cholesterol.

Table 2 Atorvastatin pharmacokinetics in 50 healthy individuals

| Parameter                  | Mean     | SD       | CV      | Range |
|----------------------------|----------|----------|---------|-------|
| $\text{AUC}_{\text{c}}$ (ng·h/mL) | 172      | 94.9     | 55.3%   | 48.9–508.8 |
| $\text{C}_{\text{max}}$ (ng/mL)    | 36.2     | 19.3     | 53.3%   | 9.5–92.2  |
| $t_{\text{max}}$ (h)               | 1.07     | 0.89     | 81.8%   | 0.5–4.0   |
| CL (L/h)                        | 603      | 310      | 51.3%   | 157–1,636 |
| CL$_{\text{adj}}$ (L/(h·kg))     | 8.85     | 4.54     | 51.1%   | 2.5–25.2  |
| $t_{1/2}$ (h)                    | 7.75     | 2.56     | 33.3%   | 2.1–17.0  |

Abbreviations: $\text{AUC}_{\text{c}}$, area under the plasma concentration curve from hour 0 to infinity; CL, clearance; $\text{CL}_{\text{adj}}$, clearance adjusted with body weight; $\text{C}_{\text{max}}$, maximum drug concentration in plasma; CV, coefficient of variation; SD, standard deviation; $t_{1/2}$, half-life; $t_{\text{max}}$, time to $\text{C}_{\text{max}}$. 


Lipid concentration changes at 24 and 48 hours from baseline are summarized in Table 3. Mean total cholesterol and LDL-C concentrations at 48 hours after single atorvastatin administration were decreased by 10.2% and 15.1%, respectively. Triglyceride concentrations did not show any statistically significant change. There was no correlation between baseline plasma concentrations or pharmacokinetic parameters of atorvastatin and changes in lipid concentrations.

Genetic polymorphisms associated with pharmacokinetic/pharmacodynamic variables

Sixty-four polymorphisms from 47 genes were associated with pharmacokinetic variables or lipid concentration changes according to genotype ($P<0.050$ in ANOVA or Kruskal–Wallis tests; Table S1). Seventeen polymorphisms were associated with AUC$_{\infty}$, 26 polymorphisms with $C_{\text{max}}$, and 16 polymorphisms with CL$_{\text{adj}}$. In terms of a lipid-lowering effect, 16 polymorphisms were associated with LDL-C, and 12 polymorphisms with total cholesterol. The 13 genes related to variance in LDL-C lowering included ABCB11, ABCC1, ABCC2, AHR, CBR1, CYP19A1, CYP1B1, CYP4F11, NAT2, SLC10A2, SLC5A6, SLC7A8, and SULT1B1.

Among these 64 polymorphisms, 3 in the SLCO1B1, SLCO1B3, and ABCC2 genes showed ordinal associations with $C_{\text{max}}$ or changes in total cholesterol or LDL-C (Tables 4 and S2). The mean of the plasma concentration–time profile for atorvastatin according to each genotype is
shown in Figure 1B–D. For c.-910G>A (rs4149015) in the SLCO1B1 gene, 33 subjects were G/G homozygotes, 14 were G/A heterozygotes, and 3 were A/A homozygotes. The mean $C_{\text{max}}$ was 76.5 ng/mL for A/A, 34.0 ng/mL for G/A, and 33.5 ng/mL for G/G (Figure 2). In the analysis of haplotypes that included rs4149015, carriers possessing the SLCO1B1*17 variant allele (rs2306283, rs4149056, and rs4149015) showed higher $C_{\text{max}}$ and $AUC_{\infty}$ compared to noncarriers (46.1 vs 32.8 ng/mL, $P=0.032$ for $C_{\text{max}}$; 222 vs 154 ng/mL, $P=0.026$ for $AUC_{\infty}$). The SLCO1B3 c.334G>T (p.Ala112Ser, rs4149117) also influenced the $C_{\text{max}}$ of atorvastatin. Mean $C_{\text{max}}$ for 6 subjects with T/T (63.0 ng/mL) was higher than that in 16 with G/T (34.7 ng/mL) and 28 with G/G (31.4 ng/mL, FDR $P=0.037$). In genotype combination analysis of SLCO1B1 c.-910G>A and SLCO1B3 c.334G>T, major homozygous individuals of both polymorphisms showed the lowest mean $C_{\text{max}}$ (30.7 ng/mL), and the $C_{\text{max}}$ tended to increase according to the increase in the number of minor alleles ($P=0.011$, Figure 3); 39.3 ng/mL for 1 minor allele, 29.0 ng/mL for 2 minor alleles, 56.0 ng/mL for 3 minor alleles, and 76.5 for 4 minor alleles. ABCC2 c.1249G>A (p.Val417Ile, rs2273697) was associated with changes in total cholesterol and LDL-C at 48 hours after atorvastatin administration. In particular, the decrease in total cholesterol and LDL-C was smaller in those with G/A (n=7) than in the 43 subjects with G/G. There was no A/A homozygote identified. The mean percentage changes in total cholesterol and LDL-C in subjects with G/A were 0.23% and −0.70%, compared to −11.9% and −17.4% for those with G/G.

**Discussion**

This study investigated pharmacokinetic characteristics and lipid-lowering response following high-dose atorvastatin treatment in young, healthy Korean males in association with genotypes in genes related to absorption, distribution, metabolism, and elimination of drugs.

Statins are known to produce immediate biochemical changes.19,20 We showed that atorvastatin achieved $C_{\text{max}}$ at around 1.07 hours. Dose-per-body weight normalized $AUC_{\infty}$ and $C_{\text{max}}$ were comparable to the results from previous studies in Asians and Caucasians.14 Interindividual variability of pharmacokinetic parameters was observed in spite of the uniformity of the enrolled subjects, who were all young and healthy males, and controlled conditions. This finding suggests that much of the pharmacokinetic variability is caused by innate or underlying conditions such as genetic factors and the gut microbiome, instead of controllable environmental factors such as concomitant medicines and compliance.
Table 4 Genetic polymorphisms associated with pharmacokinetic parameters and lipid-lowering response

| Gene     | rs no      | Nucleotide change | MAF  | Variable | Mean ± Unit |  P-value | FDR P |
|----------|------------|-------------------|------|----------|-------------|----------|-------|
| SLCO1B1  | rs4149015  | c.-910g>A         | 0.177| C<sub>max</sub> | 33.5 ±34.0 | 76.5 ng/mL | 0.021 | 0.040 |
| SLCO1B3  | rs4149117  | c.334G>T          | 0.310| C<sub>max</sub> | 31.4 ±34.7 | 63.0 ng/mL | 0.010 | 0.037 |
| ABCC2    | rs2273697  | c.1249G>A         | 0.113| %ΔTC     | -11.9 ±0.23 | –% | 0.003 | 0.019 |
|          |            |                   |      | %ΔLDL-C  | -17.4 ±0.70 | –% | 0.023 | 0.048 |

Notes: A/A, major homozygote; A/B, heterozygotes; B/B, minor heterozygotes.

Abbreviations: C<sub>max</sub>, maximum drug concentration in plasma; FDR, false discovery rate; %ΔTC, percentage change in total cholesterol from baseline to 48 hours after atorvastatin administration; %ΔLDL-C, percentage change in low-density lipoprotein cholesterol from baseline to 48 hours after atorvastatin administration.

Figure 2 The pharmacodynamic and pharmacokinetic differences according to each genotype of SLCO1B1 c.-910g>A, SLCO1B3 c.334G>T, and ABCC2 c.1249G>A.

Notes: The box-and-whisker plots of the area under the plasma concentration curve from hour 0 to infinity (AUC<sub>∞</sub>), clearance adjusted with body weight (CL<sub>adj</sub>), maximum concentration (C<sub>max</sub>) of atorvastatin, and percentage changes in total cholesterol (%ΔTC) and LDL-C (%ΔLDL-C) from baseline to 48 hours after atorvastatin administration according to genetic polymorphisms, are presented. There were differences in C<sub>max</sub> according to SLCO1B1 c.-910G>A and SLCO1B3 c.334G>T genotypes, and %ΔTC and %ΔLDL-C between G/G and G/A genotypes of ABCC2 c.1249G>A.
The lipid-lowering effect of atorvastatin also showed interindividual variation. In agreement with previous studies, there were no pharmacokinetic parameters associated with the lipid-lowering effect of atorvastatin.19,20

Because the pharmacokinetic and pharmacodynamic changes in this study were as expected, we next inspected their association with multiple genetic polymorphisms. Polymorphisms in the SLCO1B1, SLCO1B3, and ABCC2 genes were ordinally associated with pharmacokinetic properties or lipid-lowering responses. SLCO1B1 c.-910G>A, identified in 17 subjects, was associated with C\textsubscript{max}, and the SLCO1B1*17 haplotype including this polymorphism was also related to C\textsubscript{max} and AUC\textsubscript{\textalpha}. The SLCO1B1 gene encodes the organic anion transporting polypeptide (OATP) 1B1, which facilitates hepatic uptake of statins on the sinusoidal membrane of hepatocytes.21,22 Variations in SLCO1B1, c.-910G>A and c.521T>C (rs4149056), and haplotypes, *5, *15, and *17, have been reported to be associated with pharmacokinetic and lipid-lowering responses in previous studies.8-12 In addition, a loss-of-function variation, c.521T>C, which reduced liver influx of the statins, has a potent effect on myalgia, one adverse effect of statins.8 Similar findings have been reported in individuals who received atorvastatin, including Asians.8,24,25

SLCO1B3 c.334G>T was associated with a higher C\textsubscript{max} in this study. The OATP 1B3 encoded by the SLCO1B3 gene is one of the major hepatic OATPs and has a potent function as an active transporter of atorvastatin, following the OATP 1B1.26,27 Several genetic polymorphisms in the SLCO1B3 gene have been investigated in previous in vitro studies.28,29 A preclinical study showed no effect of c.334G>T on cellular uptake of atorvastatin,28 which suggests the minor effect of the SLCO1B3 gene on the distribution of atorvastatin. As previous studies suggested the aggregate effect of top-associated polymorphisms,30,31 we evaluated the genotype combination effect. We observed the genotype combination effect of SLCO1B1 c.-910G>A and SLCO1B3 c.334G>T; thus, further in vivo analysis of the role of transporter enzymes on the metabolism of statins is needed to clarify the interaction.

c.1249G>A in the ABCC2 gene was associated with a small lipid-lowering response in this study. Multidrug resistance-associated protein 2 (MRP2/ABCC2) is an efflux transporter expressed in various types of cells, including hepatocytes, enterocytes, and proximal renal tubular cells,32 and plays an important role in reducing gastrointestinal absorption and facilitating the biliary and urinary excretion of its substrates, including pravastatin and fluvastatin.32,34 A polymorphism in the ABCC2 gene has been related to low plasma concentrations of pravastatin,33 as well as dose decreases or switches to other cholesterol-lowering agents during simvastatin and atorvastatin therapy.35 In addition, after atorvastatin administration, mRNA levels of transporters, including MRP2/ABCC2, are downregulated and positively correlated with the percentage of reduction in LDL-C.36 Collectively, these data indicate that the ABCC2 gene might affect the lipid-lowering response to atorvastatin treatment.

In this prospective study, we performed a pharmacokinetic and pharmacodynamic analysis in healthy Korean individuals following high-dose atorvastatin administration. However, we should acknowledge the limitation of our study. Because of the relatively small sample size, some associations may have been missed or noticed only by chance, and also the aggregate effect of polymorphisms was partially evaluated. Our study findings should be confirmed through future large prospective studies in various ethnic populations. The strength of our study is that we provided prospective data about pharmacokinetics and the lipid-lowering response after atorvastatin 80 mg administration for a hypothesis-free genetic association study in healthy, young male Asian subjects; a population that has not been studied in this context before.25 Our findings support the view of further studies investigating factors that affect interindividual atorvastatin treatment variability, such as genes related to pharmacodynamics, and the contribution to the risk of adverse effects of polymorphisms identified as associated with the C\textsubscript{max} of atorvastatin.

**Conclusion**

In conclusion, we genotyped multiple polymorphisms in genes related to phase I and II drug metabolism enzymes and drug transporters, and evaluated the association of these
with pharmacokinetic properties and lipid-lowering response following atorvastatin administration. Our findings describe pharmacokinetic and pharmacodynamic changes with variations among individuals after high-dose atorvastatin treatment in healthy Korean subjects. We also identified various genetic polymorphisms related to the response to atorvastatin treatment, including the association between polymorphisms in the transporter genes, SLCO1B1, SLCO1B3, and ABCG2, and either $C_{\text{max}}$ of atorvastatin or lipid-lowering response. These findings contribute to the understanding of interindividual variation in atorvastatin treatment.

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**Disclosure**

The authors report no conflicts of interest in this work.

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### Supplementary materials

**Table S1** Genotypes associated with atorvastatin pharmacokinetics and lipid-lowering response

| Category | Gene    | rs number       | Nucleotide (AA) change | Call rate (%) | MAF  | Major/minor allele | Associated variable | P-value |
|----------|---------|-----------------|------------------------|---------------|------|---------------------|----------------------|---------|
| Phase I  | ADH6    | rs1002894       | c.-931C>T             | 98.3          | 0.155| T/C                 |                      | 0.028   |
|          | ALDH2   | rs671           | c.1369G>A (p.Glu504Lys)| 100           | 0.076| G/A                 |                      | 0.010   |
|          | ALDH3A1 | rs2072330       | c.741T>A (p.Pro247=)  | 96.6          | 0.342| T/A                 |                      | 0.015   |
|          | CBR1    | rs1005695       | c.397+210G>C          | 98.3          | 0.379| G/C                 |                      | 0.011   |
|          |         | rs998383        | c.+559C>G             | 100           | 0.356| C/G                 |                      | 0.005   |
|          | CYP1B1  | rs1056837       | c.1347T>C (p.Asp449=)| 100           | 0.127| C/T                 |                      | 0.048   |
|          | CYP2B6  | rs2279344       | c.822+183G>A          | 100           | 0.393| A/G                 |                      | 0.024   |
|          | CYP2E1  | rs3813867       | c.-1295G>C            | 100           | 0.218| C/T                 |                      | 0.040   |
|          | CYP4F2  | rs2074900       | c.1029C>T (p.His343=)| 100           | 0.169| C/T                 |                      | 0.024   |
|          | CYP4F8  | rs4239614       | c.+52C>T              | 98.3          | 0.224| T/C                 |                      | 0.049   |
|          | CYP1B2  | rs4536          | c.873G>A (p.Ala291=)  | 100           | 0.441| A/G                 |                      | 0.032   |
|          | CYP1B2  | rs4536          | c.873G>A (p.Ala291=)  | 100           | 0.441| A/G                 |                      | 0.010   |
|          | CYP1B2  | rs700519        | c.790C>T (p.Arg264Cys)| 100           | 0.136| C/T                 |                      | 0.031   |
|          | CYP3A5  | rs1017940       | c.161T>G              | 100           | 0.093| G/A                 |                      | 0.011   |
|          | EPHX1   | rs1051740       | c.337T>C              | 100           | 0.234| G/C                 |                      | 0.026   |
|          | FMO3    | rs1736557       | c.769G>A (p.Val257Met)| 98.3          | 0.129| G/A                 |                      | 0.014   |
|          | GSTA3   | rs2266780       | c.923A>G              | 100           | 0.203| A/G                 |                      | 0.030   |
|          | GSTA4   | rs367836        | c.*137C>A             | 98.3          | 0.129| G/A                 |                      | 0.037   |
|          | NAT2    | rs1799931       | c.857G>A (p.Arg286Glu)| 100           | 0.144| G/A                 |                      | 0.007   |
|          | SULT1B1 | rs11731028      | c.376-2858G>A         | 100           | 0.220| G/A                 |                      | 0.037   |
|          | SULT1C2 | rs17036104      | c.796T>G              | 96.6          | 0.105| T/G                 |                      | 0.006   |
|          | UGT2B4  | rs1966151       | c.+225T>C             | 94.9          | 0.491| T/C                 |                      | 0.031   |
|          | UGT2B15 | rs3100          | c.*168C>T             | 100           | 0.212| C/T                 |                      | 0.004   |
|          | UGT8    | rs4148254       | c.677C>T (p.Pro226Leu)| 100           | 0.085| C/T                 |                      | 0.018   |

(Continued)
Table S1 (Continued)

| Category    | Gene   | rs number   | Nucleotide (AA) change | Call rate (%) | MAF  | Major/minor allele | Associated variable | P-value |
|-------------|--------|-------------|------------------------|---------------|------|--------------------|---------------------|---------|
| Transporter | ABCB1  | rs10276036  | c.1000-44G>A           | 98.4          | 0.451| G/A                | AUC∞               | 0.040   |
|             |        | rs2235033   | c.1554+24T>C           | 100           | 0.444| T/C                | AUC∞               | 0.028   |
|             | ABCB1   | rs2287622   | c.1331T>C (p.Val444Ala)| 100           | 0.271| C/T                | %ΔLDL-C            | 0.037   |
|             | ABCB4   | rs2302387   | c.175C>T (p.Leu59=)   | 100           | 0.220| C/T                | Cmax               | 0.029   |
|             | ABC2    | rs246221    | c.825T>C (p.Val275=)  | 100           | 0.441| T/C                | %ΔLDL-C            | 0.005   |
|             |         | rs4148380   | c.*1293G>A            | 100           | 0.093| G/A                | %ΔLDL-C            | 0.043   |
|             | ABCG2   | rs2231142   | c.1249G>A (p.Val417ile)| 100           | 0.113| G/A                | %ΔTC               | 0.003   |
|             | SLC5A6  | rs246221    | c.1442C>T (p.Ser481Phe)| 100           | 0.364| T/C                | %ΔLDL-C            | 0.023   |
|             | ABCC2   | rs2281677   | c.1442C>T (p.Ser481Phe)| 100           | 0.250| C/A                | CLhit              | 0.025   |
|             | SLC7A7  | rs7141505   | c.-1065G>T            | 98.3          | 0.155| T/G                | %ΔTc               | 0.034   |
|             | SLC7A1  | rs2286877   | c.152-1008T>C         | 98.3          | 0.371| T/C                | %ΔTc               | 0.038   |
|             | SLC10A2 | rs7987433   | c.-373C>G             | 100           | 0.085| A/G                | %ΔLDL-C            | 0.045   |
|             | SLC25A2 | rs9381468   | c.-457A>G             | 100           | 0.212| C/G                | %ΔTC               | 0.041   |
|             | SLC7A1  | rs2235033   | c.1554+24T>C           | 100           | 0.444| T/C                | AUC∞               | 0.029   |
|             | SLC7A7  | rs2235033   | c.1554+24T>C           | 100           | 0.444| T/C                | AUC∞               | 0.029   |
|             | SLC7A1  | rs2286877   | c.152-1008T>C         | 98.3          | 0.371| T/C                | %ΔTc               | 0.034   |
|             | SLC7A7  | rs7141505   | c.-1065G>T            | 98.3          | 0.155| T/G                | %ΔTc               | 0.038   |
|             | Others  | AHR         | rs2066853              | 100           | 0.415| G/A                | %ΔLDL-C            | 0.028   |
|             | ATP7B   | rs2277448   | c.-75C>A               | 100           | 0.390| A/C                | Cmax               | 0.031   |
|             | COMT    | rs4633      | c.36C>T (p.His12=)    | 100           | 0.254| C/T                | %ΔTC               | 0.006   |
|             | NR1I2   | rs3814055   | c.-1135C>T            | 98.3          | 0.302| C/T                | %ΔTC               | 0.007   |
|             | PGAP3   | rs2952151   | c.560T>C              | 100           | 0.458| T/C                | AUC∞               | 0.023   |

Abbreviations: AA, amino acid; AUC∞, area under the plasma concentration curve from hour 0 to infinity; CLhit, clearance adjusted with body weight; Cmax, maximum drug concentration in plasma; %ΔLDL-C, percentage change in low-density lipoprotein cholesterol from baseline to 48 hours after atorvastatin administration; MAF, minor allele frequency; %ΔTC, percentage change in total cholesterol from baseline to 48 hours after atorvastatin administration.
Table S2 Pharmacokinetic parameters and lipid-lowering response of SLCO1B1 c.-910G>A, SLCO1B3 c.334G>T, and ABCC2 c.1249G>A

| Variable                  | Unit            | Genotype | FC (95% CI) | P-value |
|---------------------------|-----------------|----------|-------------|---------|
| **SLCO1B1 c.-910G>A**     |                 |          |             |         |
| n                         |                 | 33       | 14          | 3       |
| AUC∞                       | ng/mL           | G/G      | 156 (51.5%) | 173 (46.0%) | 333 (54.4%) | 1.00 (0.77–1.31) | 0.077 |
|                           |                 | G/A      | (48.9–403)  | (86.2–334) | (147–509)   | 1.89 (0.39–9.10) | 0.099 |
|                           |                 | A/A      | 9.5 (1.4%)  | 8.0 (42.3%) | 5.1 (70.6%) | 0.77 (0.59–1.00) | 0.032 |
|                           |                 |          | (2.48–25.2) | (2.95–15.0) | (2.46–9.22) | 0.46 (0.08–2.46) |         |
|                           |                 |          | 33.5 (49.7%)| 34.0 (43.8%)| 76.5 (34.4%)| 0.92 (0.71–1.21) | 0.021 |
|                           |                 |          | (9.50–81.6)| (10.9–69.7) | (46.1–92.2) | 2.18 (0.81–5.84) |         |
|                           | %ΔTC            |          | –11.9 (8.2%)| –10.7 (–76.9%)| –10.1 (–73.1%)| 0.992 |
|                           | %ΔTG            |          | –14.3 (51.8%)| –16.6 (–208%)| 1.70 (98.6%) | 0.659 |
|                           | %ΔLDL-C         |          | –13.7 (–82.4%)| –17.6 (–74.0%)| –19.5 (–75.7%)| 0.948 |
| **SLCO1B3 c.334G>T**     |                 |          |             |         |
| n                         |                 | 28       | 16          | 6       |
| AUC∞                       | ng/mL           | G/G      | 151 (57.4%) | 172 (40.1%) | 267 (53.2%) | 1.06 (0.86–1.30) | 0.069 |
|                           |                 | G/T      | (48.9–403)  | (97.6–334) | (126509) | 1.57 (0.91–2.72) |         |
|                           |                 | T/T      | 10.1 (52.0%)| 7.7 (33.0%) | 5.85 (51.3%)| 0.72 (0.59–0.88) | 0.109 |
|                           |                 |          | (2.48–25.2) | (2.85–11.9) | (2.46–9.74) | 0.51 (0.29–0.90) |         |
|                           |                 |          | 31.4 (49.9%)| 34.7 (44.6%)| 63.0 (39.6%)| 1.01 (0.80–1.28) | 0.010 |
|                           |                 |          | (9.50–65.4)| (10.9–81.6)| (36.6–92.2) | 1.88 (1.21–2.87) |         |
|                           | %ΔTC            |          | –8.7 (147%) | –12.1 (–57.1%)| –13.9 (–64.1%)| 0.070 |
|                           | %ΔTG            |          | 12.5 (625%) | –0.79 (–5.470%)| –15.3 (–187%)| 0.770 |
|                           | %ΔLDL-C         |          | –11.4 (–227%)| –16.9 (–35.7%)| –27.3 (–51.2%)| 0.201 |
| **ABCC2 c.1249G>A**      |                 |          |             |         |
| n                         |                 | 43       | 7           |         |
| AUC∞                       | ng/mL           | G/G      | 172 (53.6%) | 170 (69.7%) | 0.80 (0.42–1.53) | 0.547 |
|                           |                 | G/A      | (48.9–509)  | (58.2–342) | (58.2–342) | 0.92 (0.47–1.81) | 0.412 |
|                           |                 |          | 8.6 (49.5%) | 9.8 (62.7%) | 0.9 (309) | 0.88 (0.43–1.81) | 0.642 |
|                           | %ΔTC            |          | –11.9 (–82.7%)| 0.23 (3.549%)| –8.7 (11.8) | 0.003 |
|                           | %ΔTG            |          | 1.3 (4789%) | 27 (283%) | 62.5 (156) | 0.452 |
|                           | %ΔLDL-C         |          | –17.4 (–117%)| –0.70 (–2.565%)| –26.6 (25.2) | 0.023 |

Notes: *Mean ± SD (CV) (range) of each variable according to genotype. Fold change in heterozygotes compared to major homozygotes. Fold change in minor homozygotes compared to major homozygotes. Abbreviations: AUC∞ area under the plasma concentration curve from hour 0 to infinity; CI confidence interval; CL-git clearance adjusted with body weight; Cmax maximum drug concentration in plasma; CV coefficient of variation; FC fold change; %ΔLDL-C percentage change in low-density lipoprotein cholesterol from baseline to 48 hours after atorvastatin administration; SD standard deviation; %ΔTC percentage change in total cholesterol from baseline to 48 hours after atorvastatin administration; %ΔTG percentage change in triglycerides from baseline to 48 hours after atorvastatin administration.

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