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ORIGINAL RESEARCH

Familial Resemblance in Low-Density Lipoprotein Cholesterol Response to Statins in the Danish Population

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BACKGROUND: Change in low-density lipoprotein cholesterol (LDL-C) level after statin initiation varies widely among individuals, and in part may be because of factors shared by family members.

METHODS AND RESULTS: We used the Danish national registers to identify 89 006 individuals who initiated statins between 2008 and 2018 and had LDL-C measured immediately before and after the start of treatment. Among these, we identified 5148 first-degree relatives and 3198 spouses. We decomposed the variation in attained LDL-C level after statin initiation by applying a mixed-effect model with 5 variance components (inter-family and inter-individual variance in pre-statin LDL-C level, inter-family and inter-individual variance in statin response, and residual variance). Results were presented as a percentage of the total variance explained by the different variance components. We found that half of the variation in attained LDL-C level after statin initiation consisted of variance in statin response, approximately one third of variance in pre-statin LDL-C level, and the remaining 10% to 15% of residual variance. While the inter-individual variance in statin response accounted for almost half of the LDL-C variation in both cohorts, the inter-family variance in statin response accounted for 3.3% among first-degree relatives and for 6.0% among spouses.

CONCLUSIONS: Individual factors account for most of the variation in LDL-C level after statin initiation; factors affecting statin response common within spouses and first-degree relatives account for a similar share of variation. These results suggest a modest influence of shared genetics and shared familial environment on statin response.

Key Words: familial environment ■ genetic ■ inheritance ■ LDL-C ■ statin response

Statins are the most frequently prescribed lipid-lowering drugs worldwide, and first-line treatment for dyslipidemia, familial hypercholesterolemia, and for prevention of cardiovascular disease.[1] Large, randomized controlled trials have consistently shown the effectiveness of statins in preventing major vascular events, with a treatment effect that is proportional to the reduction in low-density lipoprotein cholesterol (LDL-C).[2] There is, however, substantial inter-individual variability in the reduction of LDL-C achieved for any given dose of a statin.[3–5]

LDL-C levels in untreated individuals vary because of genetic factors, and are also affected by environmental factors, including cohabitation.[6,7] Few studies have investigated the impact of genetic and environmental factors on the response to statin treatment. Oni-Orisan et al reported that the response of LDL-C levels to statin treatment was modestly heritable among 1036 first-degree relatives.[5] Their study consisted of a small, selected sample, enrolled only first-degree relatives, and did not investigate the potential contribution of shared environment on response to statins.

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CLINICAL PERSPECTIVE

What Is New?

- Our study does not support a strong influence of genetic constitution and family-shared environment on low-density lipoprotein cholesterol response to statin initiation.

What Are the Clinical Implications?

- This study does not support use of family information about statin response in clinical decision making regarding statin treatment.

Nonstandard Abbreviations and Acronyms

ATC	anatomical therapeutic chemical classification system
LDL-C	low-density lipoprotein cholesterol

We used the Danish health and socioeconomic registries to evaluate the influence of familial and individual factors on the variation of LDL-C response to statin therapy in the general population by analyzing data from first-degree relatives and spouses who both initiated statin therapy.

METHODS

Data Availability Statement

The data are available for research upon request to Sundhedsdatastyrelsen and Danmarks Statistik and within the framework of the Danish data protection legislation and any required permissions from authorities. According to Danish law, ethics approval is exempt for such research.

Ethics

Because of the nature of this research, there was no involvement of patients or members of the public in the design or reporting of this study. Direct dissemination to study participants is not possible. The study was covered by records of data-processing activities under the responsibility of Statens Serum Institut (j.nr. 21/01486). The publication only contains aggregated results and no personal data. The publication is therefore not covered by the European General Data Protection Regulation. The study is fully compliant with all legal and ethical requirements and there are no further processes available regarding such studies.

Data Sources

This study was based on information from the nationwide Danish registries available for research. The Danish Central Person Registry contains continuously updated data on vital status, residency, marital status, and kinship for all individuals resident in Denmark since 1968, identified through the unique personal identification number that allows individual level linkage with a large number of other nationwide demographic and administrative registries.[8] The Danish Family Relations Database is based on kinship information in the Danish Central Person Registry, and includes information about first-degree relatives for individuals born since 1950, and second- and third-degree relatives for individuals born since 1985. The Danish Register of Medicinal Product Statistics holds information on all prescribed medicine redeemed outside of the hospital since 1995.[9] The Register of Laboratory Results for Research includes results from clinical biochemical analyses performed at Danish laboratories. Different laboratories started contributing data to this registry at different time points between 2008 and 2015; laboratories in 1 out of 5 regions in Denmark (Central Denmark Region) had not begun to contribute data to the registry at time of data extraction for this project. Only laboratory analyses identified by means of Nomenclature for Property and Unit (NPU) codes, the internationally used clinical laboratory terminology, are available for research (≈95% of all laboratory analyses).[10] The Danish National Patient Register contains information such as date of admission and discharge in relation to diagnoses and treatments for all inpatient and outpatient hospital contacts since 1977 and 1995, respectively.[11] Statistics Denmark provides time-varying socioeconomic data such as educational attainment (since 1980) and disposable household income (since 1990).[12,13]

Identification of Lipid-Lowering Treatment

Use of lipid-lowering medication was ascertained by filled prescriptions in the Danish Register of Medicinal Product Statistics. Statin initiation was defined as the first filled prescription of simvastatin or atorvastatin, identified using Anatomical Therapeutic Chemical Classification System (ATC) codes (C10AA01 and C10AA05). Dose was defined using the strength labeled on the package and the assumption of administration of 1 pill per day. The use of simvastatin or atorvastatin in combination with ezetimibe was identified as a filled prescription for the combined pill (ATC C10BA02 or C10BA05) or a filled prescription for ezetimibe (ATC C10AX09) within 1 week of the date of statin initiation. Use of lipid-lowering drugs other than simvastatin and atorvastatin was identified using ATC codes (C10* other than C10AA01 and C10AA05).

Identification of LDL-C Measurements

LDL-C measurements were identified in the Register of Laboratory Results for Research using codes NPU01568 and NPU10171. The pre-statin LDL-C level was defined as an LDL-C measurement in the 12 weeks preceding statin initiation. If more than 1 measurement was available, the latest (closest to statin initiation date) was chosen. The on-statin LDL-C level was defined as an LDL-C measurement from 2 to 24 weeks after statin initiation. If more than 1 measurement was available, the first (closest to statin initiation date) was chosen.

Study Population

After linking the health and socioeconomic Danish registries by means of the unique personal identification number, we identified adults (≥18 years old) who initiated treatment with either simvastatin or atorvastatin between January 2008 and March 2018. Cohort members were further required to have a pre-statin and an on-statin LDL-C measurement, as well as a registered address in the Danish Central Person Registry at statin initiation (to be able to define region of residence). Subsequently, we excluded individuals residing in the Central Denmark Region (region not represented in the Register of Laboratory Results for Research for the calendar period in question), previous users of other lipid-lowering medications (including statins not considered in this study), and individuals prescribed simvastatin or atorvastatin in combination with ezetimibe. We also excluded individuals with <1 year of data before statin initiation or <24 weeks of follow-up after statin initiation (to exclude potential prevalent users of statin and to minimize missing data on covariates) and, *a posteriori*, individuals initiated on simvastatin 80 mg, because of small numbers.

Using information from the Danish Family Relations Database, we identified families in which 2 or more first-degree relatives initiated statin therapy and had LDL-C measured before and after statin initiation. If both parents were identified together with 1 or more children, then only 1 parent, chosen randomly, was considered for analysis (38 father-mother pairs). An individual was only allowed to be a member of 1 family of first-degree relatives. The necessary exclusions were made randomly. Among families of first-degree relatives, we further distinguished between parent-offspring pairs and siblings. If 1 family of first-degree relatives included 1 parent and 2 or more offspring (43 families), then all offspring contributed as a family in the analysis of siblings, while the parent together with 1 randomly chosen offspring contributed as a pair in the parent-offspring analysis.

We identified spousal pairs by means of information from the Danish Central Person Registry, and included both married couples and civil partnerships. Each spouse was required to have an LDL-C measurement before and after statin initiation as described above, and both statin initiation dates had to fall within the time period of the marriage. An individual was allowed to be included in the analysis of both first-degree relatives and spousal pairs (126 individuals).

Statistical Analysis

The sources of variation in attained LDL-C level were investigated by a mixed-effect model, with the logarithm of the LDL-C level as outcome, using the SAS procedure PROC MIXED. More detailed description of the model, data structure, the SAS code used, and evaluation of model assumptions are available in Data S1, Table S1, and Figures S1, S2. Each individual contributed 2 observations: 1 pre-statin and 1 on-statin LDL-C level. We decomposed the variation into 3 sources of variance: variance in pre-statin LDL-C level, variance in statin response, and residual variance. Moreover, we distinguished between individual and familial variance, yielding 5 final variance components: inter-family and inter-individual variance in pre-statin LDL-C level, inter-family and inter-individual variance in statin response, and intra-individual (residual) variance. The inter-family variance components represent factors common to family members at the 2 measurement times, the inter-individual variance components represent factors that are not included in the inter-family variance components and are constant for the individual at the 2 measurement times and the intra-individual variance component represents individual factors that are different at the 2 measurement times and measurement errors. Accordingly, the model included 1 fixed and 2 random intercepts (inter-family and inter-individual), and 1 fixed and 2 random slopes (inter-family and inter-individual). The intercept represents the pre-statin LDL-C level, while the slope represents the statin response (ie, the change in LDL-C after statin initiation). Adjustment was made for age (2-year intervals), sex, period (1-year intervals), region of residence, indication for statin treatment (established cardiovascular disease; risk factors for cardiovascular disease; or no risk factors for cardiovascular disease [cf. Table S2 for definitions]), education, disposable household income (deciles), and the interaction between these covariates and a binary variable indicating treatment status (if not specified, the variables are categorized as in Table 1). Moreover, the model was adjusted for type and dose of the initial statin prescription; in addition to the 7 levels indicating the initial dose (simvastatin 10, 20, and 40 mg and atorvastatin 10, 20, 40, and 80 mg), this variable had an eighth level used for all the pretreatment observations. The relative importance of the different variance components was

Table 1. Characteristics of the Cohorts of First-Degree Relatives and Spousal Pairs

	Cohort	
	First-degree relatives	Spousal pairs
Number of individuals	5148	3198
Number of families	2541	1599
Characteristics at statin initiation		
Age, median (IQR), y	55 (48, 65)	64 (57, 69)
Female sex	2604 (50.6%)	1595 (49.9%)
Period		
2008–2014	1916 (37.2%)	1334 (41.7%)
2015–2016	1886 (36.6%)	1094 (34.2%)
2017–2018	1346 (26.1%)	770 (24.1%)
Region of residence		
Capital region	2790 (54.2%)	2005 (62.7%)
Zealand	661 (12.8%)	302 (9.4%)
Southern Denmark	871 (16.9%)	443 (13.9%)
Northern Denmark	826 (16.0%)	448 (14.0%)
Education, y*		
<10 y	1632 (31.7%)	869 (27.2%)
10–12 y	2294 (44.6%)	1488 (46.5%)
13–15 y	881 (17.1%)	598 (18.7%)
≥15 y	256 (5.0%)	181 (5.7%)
Disposable household income†		
1 quintile	582 (11.3%)	267 (8.3%)
2 quintile	973 (18.9%)	478 (14.9%)
3 quintile	1019 (19.8%)	592 (18.5%)
4 quintile	1068 (20.7%)	800 (25.0%)
5 quintile	1277 (24.8%)	1036 (32.4%)
Indication for statin treatment		
Established CVD	1938 (37.6%)	1172 (36.6%)
Other risk factors for CVD‡	1419 (27.6%)	1028 (32.1%)
No registered risk factors for CVD	1791 (34.8%)	998 (31.2%)
Initial statin type and dose		
Simvastatin 10 mg	147 (2.9%)	124 (3.9%)
Simvastatin 20 mg	746 (14.5%)	522 (16.3%)
Simvastatin 40 mg	1977 (38.4%)	1246 (39.0%)
Atorvastatin 10 mg	265 (5.1%)	181 (5.7%)
Atorvastatin 20 mg	572 (11.1%)	385 (12.0%)
Atorvastatin 40 mg	830 (16.1%)	469 (14.7%)
Atorvastatin 80 mg	611 (11.9%)	271 (8.5%)

CVD indicates cardiovascular disease; and IQR, interquartile range.

*147 individuals with missing information for education were imputed using mode imputation as 10 to 12 years.

†254 individuals with missing information for disposable household income were imputed as 10th decile in adjustment, consequently here as 5th quintile (mode imputation).

‡Other risk factors for CVD includes diabetes, hypertension, chronic kidney disease, and familial hypercholesterolemia (cf. Table S2 for definitions).

evaluated expressing each component as a percentage of the sum of all the variance components, hereafter referred to as total variance. Ninety-five percent CI for percentages of explained variance and *P* values were calculated from the estimated covariance matrix for the variance components using the SAS procedure PROC SIMNORMAL. The number of simulations was set to 2 000 000 to provide stable results to the fourth decimal place across different simulations.

The analyses were repeated including only adjustment for type and dose of the initial statin prescription, in order to assess the effect of adjustment.

We also performed several sensitivity analyses. First, we excluded individuals (and consequently their families) who did not refill their statin prescription within 24 weeks of filling the first prescription (to evaluate the effect of noncompliance). Second, we further excluded individuals (and their families) where the number of pills of the first statin prescription covered <75% or >150% of the days to refill, to eliminate low adherence, pill splitting, or use of several pills per day. Third, we identified cohort members who had their pre-statin LDL-C measured during an acute inpatient hospitalization, and performed an extra adjustment to control for possible transient effects on the LDL-C level from acute conditions. Fourth, to evaluate whether the time interval between statin prescription and the on-statin LDL-C level affected the results, we repeated the analysis after excluding individuals (and their families) with on-statin LDL-C measured <6 weeks after statin initiation. Lastly, to evaluate the robustness of the model chosen, we performed several sensitivity analyses using alternative models, as described in Data S1.

RESULTS

Cohort Description

Among 89 006 individuals who initiated simvastatin or atorvastatin and had both a pre-statin and an on-statin LDL-C measurement, we identified 2541 families of first-degree relatives and 1599 spousal pairs, for a total of 8220 individuals. Among these, there were 3198 spouses and 5148 first-degree relatives (126 individuals were included both as a spouse and as a first-degree relative). Among first-degree relatives, 2075 individuals contributed to the analysis of siblings and 3116 individuals contributed to the analysis of parent-offspring (43 individuals contributed to both analyses).

The median age at statin initiation was 55 years among first-degree relatives (interquartile range, 48–65), and 64 years among spouses (interquartile range, 57–69). There were minor differences between these groups in region of residence and disposable household income (Table 1, characteristics for the cohort of siblings and parent-offspring are shown in Table S3). Simvastatin 40 mg was the most commonly prescribed initial statin and strength in both groups. The pre-statin LDL-C levels were similar among first-degree relatives

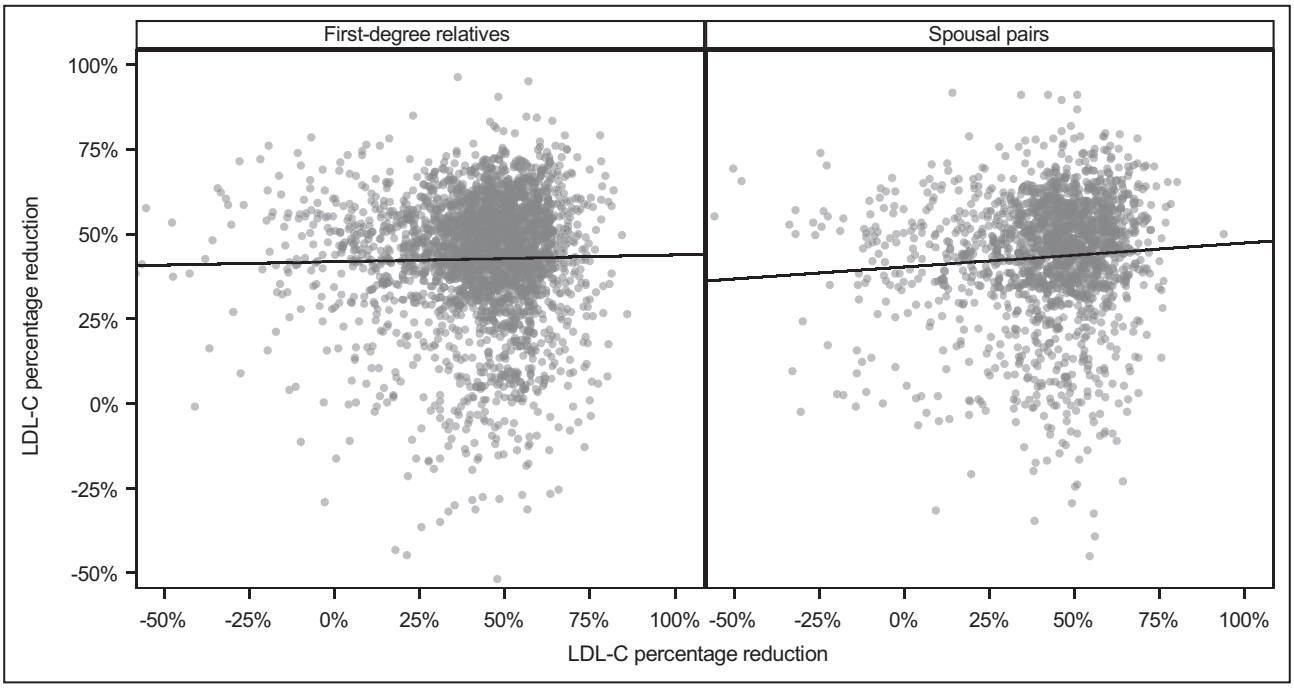

Figure 1. LDL-C percentage reduction and its correlation among pairs of first-degree relatives and spousal pairs.
The assignment of pairs to the horizontal vs the vertical axis was done randomly. The Pearson correlation was 0.02 among pairs of first-degree relatives and 0.08 among spousal pairs. LDL-C indicates low-density lipoprotein cholesterol.

(4.05 mmol/L) and spousal pairs (3.94 mmol/L), as were the on-statin LDL-C levels (2.26 mmol/L among first-degree relatives, and 2.21 mmol/L among spouses), and the percentage reductions in LDL-C (42.6% among first-degree relatives, and 42.4% among spouses). There were modest correlations in the percentage reduction in LDL-C levels (Figure 1) among both first-degree relatives (Pearson correlation coefficient=0.02) and spouses (Pearson correlation coefficient=0.08).

Decomposition of the Variation in Attained LDL-C

We first log-transformed all LDL-C levels to reduce skewness in the data, and then identified the proportion of variance, expressed as percentage of the total variance, explained by different factors. Among first-degree relatives, the crude total variance was 0.159, of which 9.8% was explained by dose and type of the initial statin and 6.1% by the other baseline covariates included in the model (similar proportions were observed in the cohort of spousal pairs), leaving an unexplained variance of 0.132 that was further decomposed into the 5 variance components (inter-family and inter-individual variance in pre-statin LDL-C level, inter-family and inter-individual variance in statin response, and intra-individual variance). Of this total variance, the inter-family variance in pre-statin LDL-C level constituted 7.4% (CI, 5.5–9.3), while the inter-individual variance in pre-statin LDL-C level was 29.7% (CI, 27.5–32.0). The corresponding figures for the inter-family and inter-individual variance in LDL-C response to statin initiation were 3.3% (CI, 0.5–6.2) and 45.6% (CI, 42.0–49.2), respectively (Table 2 and Figure 2). We found similar levels of inter-individual and inter-family variance in statin response in the analyses of siblings and parent-offspring pairs (Table S4).

The inter-family variance in pre-statin LDL-C level for spouses was lower (2.7% [CI, 0.5– 4.9]) than it was for first-degree relatives (7.4% [CI, 5.5–9.3], *P* value for difference=0.001). In contrast, despite their genetic unrelatedness, spousal pairs showed a higher inter-family variance in statin response (6.0% [CI, 2.2–9.7]) than first-degree relatives (3.3% [CI, 0.5– 6.2]), although this difference was not significant (*P*=0.27).

When the statistical adjustment was limited to type and dose of the initial statin, we found a greater total (unexplained) variance, which stemmed mostly from an increase in the estimated variance components of pre-statin LDL-C level, while the estimated variance components of statin response remained unchanged. This pattern was consistent across the different cohorts (Table S5).

In the sensitivity analysis that excluded individuals who did not refill their statin prescription (along with their family members), all results were largely unchanged, with the notable exception that the inter-individual variance in LDL-C response to

Table 2. Variance Components for LDL-C Levels and Statin Response in the Cohorts of First-Degree Relatives and Spousal Pairs

	First-degree relatives		Spousal pairs		
	Variance component* (95% CI)	Percentage of the total variance (95% CI)	Variance component* (95% CI)	Percentage of the total variance (95% CI)	*P* value†
Variance in pre-statin LDL-C level					
Inter-family variance	0.010 (0.008– 0.013)	7.4% (5.5%–9.3%)	0.003 (0.002– 0.010)	2.7% (0.5%–4.9%)	0.001
Inter-individual‡ variance	0.039 (0.036– 0.043)	29.7% (27.5%– 32.0%)	0.041 (0.037– 0.046)	31.5% (28.7%– 34.4%)	0.34
Variance in statin response					
Inter-family variance	0.004 (0.002–0.013)	3.3% (0.5%–6.2%)	0.008 (0.005–0.017)	6.0% (2.2%–9.7%)	0.27
Inter-individual‡ variance	0.060 (0.055– 0.067)	45.6% (42.0%– 49.2%)	0.062 (0.056– 0.071)	47.6% (43.1%– 52.0%)	0.50
Residual variance					
Intra-individual variance	0.018 (0.016– 0.021)	13.9% (11.9%– 16.0%)	0.016 (0.014– 0.019)	12.2% (9.8%–14.8%)	0.32
Total variance	0.132	100%	0.131	100%	

LDL-C indicates low-density lipoprotein cholesterol.
*The model was adjusted for age, sex, period, region of residence, indication for statin treatment, education, disposable household income, and type and dose of the initial statin prescription.
†*P* value for difference between estimates for first-degree relatives and spousal pairs.
‡Inter-individual (but within-family) variance.

statin therapy was lower than in the main analysis (Table S6). The estimate of inter-individual variance in statin response decreased further when we further excluded individuals (and their families) who did not have a refill pattern consistent with the use of 1 pill per day, while the other estimates were still unchanged (Table S7).

Results were unaffected by adjustment for pre-statin LDL-C level measured during an acute inpatient contact (Table S8). Similarly, the exclusion of individuals (and their families) with on-statin LDL-C levels measured <6 weeks after statin initiation produced results compatible with the main analysis, considering the reduced sample size in this sensitivity analysis (Table S9).

Excluding individuals with very low or very high levels of pre-statin LDL-C or on-statin LDL-C (along with their families) (ie, observations that could have had a high influence on the results), we observed minimal differences in the estimates of inter-family variance components (Table S10).

As an alternative approach, we modeled only statin response, using the difference between the logarithm of the on-statin LDL-C level and the pre-statin LDL-C level as outcome. The estimates of the inter-family variance in statin response for both cohorts were very similar to those from the main analysis. Other alternative approaches were evaluated and the results are shown in Table S11.

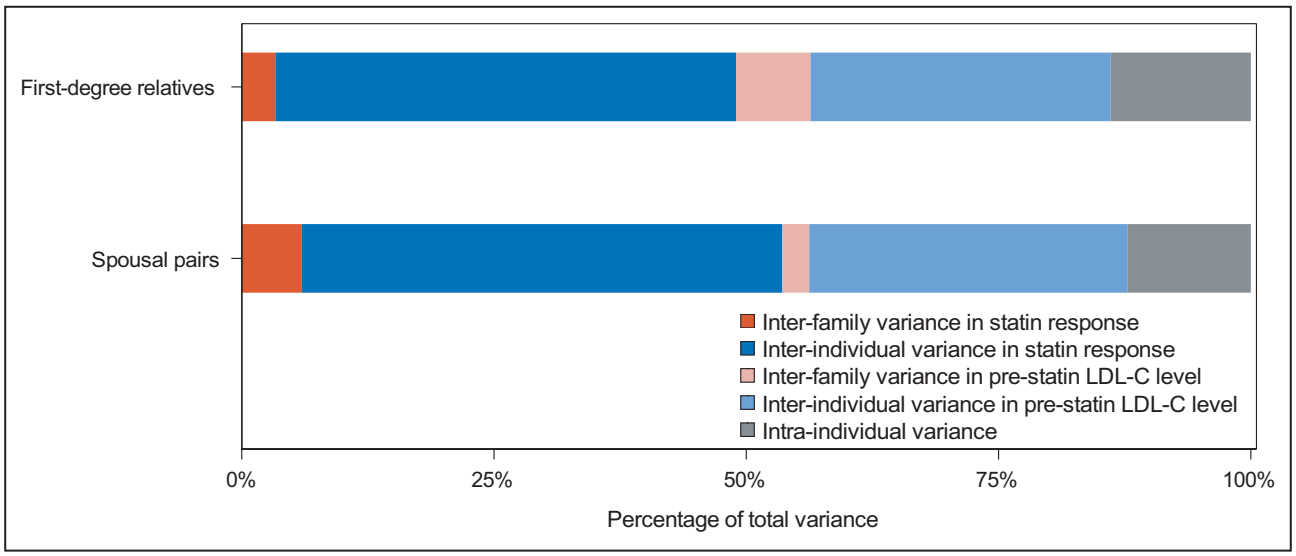

Figure 2. Variance components for LDL-C levels and statin response in the cohorts of first-degree relatives and spousal pairs.
LDL-C indicates low-density lipoprotein cholesterol.

Finally, for comparison with previous studies, we considered only the pre-statin part of the model and disregarded the variance components for statin response and found that 14.9% of the total variance in pre-statin LDL-C could be explained by factors common among members of the same family of first-degree relatives, while the corresponding value was 5.0% among spouses.

DISCUSSION

It is well known that there is a large variation in attained LDL-C level after initiation of statin treatment.[3–5] In this nationwide register study, we found that individual factors accounted for most of the variance in response to statin treatment, in both first-degree relatives and spouses. The amount of variance in response to statin treatment that could be attributed to factors shared among first-degree relatives and spouses was modest and very similar. In contrast, the shared variance in pre-statin LDL-C levels was significantly higher for first-degree relatives compared with spouses. This suggests that shared genetic factors may be more important in determining pre-statin LDL-C levels, whereas shared environmental factors (eg, shared behaviors) may be more important in affecting response to statin treatment.

Familial resemblance of LDL-C levels before any initiation of statin treatment has been investigated in several studies, indicating a significant genetic contribution to the attained level in the individual.[7] We found that factors common to members of the same family accounted for ≈15% of the variation in pre-statin LDL-C level. Our estimate should be interpreted with caution because it is based on a population selected for statin use. However, it is not very different from that found based on genomewide association studies data among non-Hispanic White members of Kaiser Permanente in the United States. Here, the overall variance explained by genomewide significant loci ranged from 14% to 20% in non-Hispanic White individuals.[14]

Only 1 study has previously investigated familial resemblance of the LDL-C level in response to statin treatment. Based on the Kaiser Permanente GERA cohort, Oni-Orisan et al observed a modest heritability of 12% in a smaller sample of 1036 first-degree relatives.[5] However, the degree of heritability observed by Oni-Orisan et al had a high uncertainty, including the observation of no heritability. Adding to the uncertainty of the heritability estimate by Oni-Orisan et al, it assumed there is no effect of common familial environment, which seems unlikely, since first-degree relatives may still share tradition and behaviors because of their common origin, even though they are no longer in the same household as adults. Therefore, the inter-family variance in statin response among first-degree relatives can be interpreted as an upper bound for the influence of shared genetics, while the same figure among spouses represents the influence of cohabitation (eg, nutrition, leisure, and sleep). In particular, our observation of spousal resemblance indicates that common familial environment contributes at least as much as common genetics to LDL-C levels on statin therapy. The limited influence of genetics in the LDL-C response to statin treatment observed in our study is in line with findings of genomewide association studies where only a small number of variants often with a rather modest effect have been found to be associated with LDL-C response to statin therapy.[15] The proportion of variation in statin responsiveness attributable to shared environment and to individual factors suggests that the response of LDL-C to statin treatment could be improved by identifying factors limiting the response and adjusting management accordingly.

A major strength of this study is the inclusion of a large population of first-degree relatives, allowing for the distinction between siblings and parent-offspring pairs, and the inclusion of genetic unrelated relatives (spouses) allowing us to evaluate the effect of cohabitation. Moreover, we performed several sensitivity and extra analyses to evaluate the assumptions underlying the model (eg, model definition, timing of LDL-C measurements, definition of statin use), and found very similar results in all of them, confirming the robustness of our results. However, the study also had some limitations. First, information on statin initiation relied on filled prescriptions, which does not guarantee that the medication was actually taken. However, when we restricted the analysis to individuals who refilled their statin prescription, the estimated inter-family variance was similar. Second, the design of the study required the presence of 2 registered LDL-C measurements, 1 before and 1 after statin initiation. Moreover, it was also required to have a relative in the same population. The statin response might affect the existence of a measurement after statin initiation if the drug was not used; however, this does not seem to affect the estimated inter-family variance. Lastly, the Danish population consists mainly of White individuals; therefore, the results of this study may not be generalizable to other populations.

CONCLUSIONS

In conclusion, we found that individual factors affecting statin response accounted for most of the total variation in both first-degree relatives and spouses, while factors affecting statin response common within spouses and first-degree relatives accounted for a similar, but modest share. These results suggest that shared familial environment and shared genetics have modest effects on the response to statins.

ARTICLE INFORMATION

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G. Corn had full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

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Disclosures

None.

Supplemental Material

Data S1
Tables S1–S11
Figures S1–S2
Reference 16

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Supplemental Material

Data S1.

Supplemental Methods

Description of the model

We decomposed the variation in attained low-density lipoprotein cholesterol (LDL-C) level by means of a mixed-effect model including one fixed and two random intercepts (inter-family and inter-individual), and one fixed and two random slopes (inter-family and inter-individual), yielding a total of five variance components. Let $i = 1,2, \dots, n$ indicate the family, $j = 1,2$ the individual within the family and $k = 1,2$ the observation within each individual, then the model can be expressed with the equation:

$$y_{ijk} = \underbrace{x_{ij}^T\beta + u_i^0 + v_j^0}_{intercept} + \underbrace{\left(x_{ij}^T\gamma + u_i^1 + v_j^1\right) * time}_{slope} + \varepsilon_{ijk}$$

where

- y_{ijk} is the logarithm of the LDL-C level for the observation k (pre-statin LDL-C if $k = 1$, on-statin LDL-C if $k = 2$), of the individual j, of the family i;
- the variable $time$ assumes the value 0 when $k = 1$, i.e. we are considering a pre-statin LDL-C level, and 1 when $k = 2$, i.e. we are considering an on-statin LDL-C level;
- $x_{ij}^T\beta$ and $x_{ij}^T\gamma * time$ represent the adjusting terms for the intercept and the slope respectively;
- u_i^0 is a random effect normally distributed according to $\mathcal{N}\left(0, \sigma_{0,fam}^2\right)$, and $\sigma_{0,fam}^2$ represents the inter-family variance in pre-statin LDL-C level;
- u_i^1 is a random effect normally distributed according to $\mathcal{N}\left(0, \sigma_{1,fam}^2\right)$, and $\sigma_{1,fam}^2$ represents the inter-individual variance in statin response;
- v_j^0 is a random effect normally distributed according to $\mathcal{N}\left(0, \sigma_{0,id}^2\right)$, and $\sigma_{0,id}^2$ represents the inter-individual variance in pre-statin LDL-C level;
- v_j^1 is a random effect normally distributed according to $\mathcal{N}\left(0, \sigma_{1,id}^2\right)$, and $\sigma_{1,id}^2$ represents the inter-individual variance in statin response;
- ε_{ijk} represents the residuals, which are normally distributed according to $\mathcal{N}(0, \sigma_{res}^2)$. σ_{res}^2 is the intra-individual (or residual) variance.

Data structure and SAS code

This section presents the data structure and SAS code used in the article to estimate the variance components. For simplicity, we have included only age and sex as adjustment variables in the following description.

The following table includes example data for four individuals grouped in two families.

FamId	PersonId	PreLDL	OnLDL	Time	Y	Treatment	AdjAge	AdjSex
1	1	4.0	2.5	0	1.39	No_statin	56-57	M
1	1	4.0	2.5	1	0.92	Simva40	56-57	M
1	2	3.7	2.6	0	1.31	No_statin	62-63	M
1	2	3.7	2.6	1	0.96	Atorva20	62-63	M
2	1	2.8	2.0	0	1.03	No_statin	58-59	F
2	1	2.8	2.0	1	0.69	Simva20	58-59	F
2	2	3.2	1.8	0	1.16	No_statin	72-73	M
2	2	3.2	1.8	1	0.59	Atorva40	72-73	M

The data were analyzed in SAS using the procedure PROC MIXED as described below.

```
data VarCompAnalysis;
input FamId PersonId PreLDL OnLDL Time Y Treatment $10. AdjAge $6. AdjSex $1.;
datalines;
1 1 4.0 2.5 0 1.39 No_statin 56-57 M
1 1 4.0 2.5 1 0.92 Simva40   56-57 M
1 2 3.7 2.6 0 1.31 No_statin 62-63 M
1 2 3.7 2.6 1 0.96 Atorva20  62-63 M
2 1 2.8 2.0 0 1.03 No_statin 58-59 F
2 1 2.8 2.0 1 0.69 Simva20   58-59 F
2 2 3.2 1.8 0 1.16 No_statin 72-73 M
2 2 3.2 1.8 1 0.59 Atorva40  72-73 M
…
;
run;



proc mixed data= VarCompAnalysis noclprint cl covtest method=ml asycov;
           class FamId PersonId Treatment AdjAge AdjSex;
           model Y = Treatment AdjAge AdjSex AdjAge*Time AdjSex*Time
                     / solution;
           random Intercept time / sub=FamiId type=vc;
           random Intercept time / sub=PersonId(FamId) type=vc;
run;
```


Evaluation of model assumptions

As described in the previous section the model assumes normality and homoscedasticity of the random effects. To evaluate how this assumption, or the lack of it, influenced our results, we performed a visual check of the normality and homoscedasticity of the residuals using a conditional standardized residuals vs. fitted values plot and a quantile-quantile plot (QQ-plot) for the conditional standardized residuals (figure S1 and S2). These plots revealed that the assumptions of normality and homoscedasticity of the residuals are generally true, except for few observations. In addition, we performed a bootstrap analysis resampling the families with replacement. We sampled 2541 families of first-degree relatives and 1599 spousal pairs one

hundred times and estimated variance components, and percentages of variance explained as done in the main models. Thereafter, we calculated the summary estimates and confidence intervals by means of medians and 2.5- and 97.5-percentiles. The results of this analysis are included in table S1 and show very similar results to the main analysis.

Evaluation of the robustness of the variance components estimates

To test the robustness of the model chosen for the main analysis in estimating inter-family variance components, we performed several sensitivity analyses using alternative models. The following sections describes these models in more detail.

Covariance between the inter-family random effects

The main model is based on the assumption of random effects being independent of each other. The data available allow us to identify one covariance, in particular the covariance between the inter-family random intercept and the inter-family random statin response. We, therefore, repeated the main analysis adding this covariance term to the model.

Modelling only statin response

In this alternative model, we used the difference between the logarithm of the pre-statin and the on-statin LDL-C levels as outcome. Consequently, each individual contributed with one observation to the model and therefore it was only possible to distinguish between two variance components: the inter-family variance in statin response and the intra-individual (residual) variance that, here, includes the inter-individual variance in statin response and two times the residual variance from the main model. Adjustment was made in the same way as in the main model in the intercept part.

Modelling only the untreated LDL-C level

The main model is based on the specific population of statin initiators having two registered LDL-C measurements, one before and one after statin initiation. To have an alternative and potentially more accurate estimate of the inter-family variance in untreated LDL-C level, we extended our population by not requiring statin treatment and an on-statin LDL-C measurement. In particular, we identified all adults (≥18 years old) resident in Denmark (known address) with a registered untreated LDL-C measurement, i.e. the individual had to be naïve to all lipid-lowering drugs at the time of the laboratory test (n=1,682,420). If more measurements were available for one person, then one was chosen randomly. In this cohort, we identified families of first-degree relatives and spousal pairs as described in the methods section for the main analyses (940,959 first-degree relatives and 440,634 spouses). The model included the logarithm of the untreated LDL-C level as outcome and one fixed and one random intercept, allowing the identification of the inter-family variance in untreated LDL-C level and the intra-individual (residual) variance. The model was adjusted for age, sex, period, region of residence, education and disposable household income. Due to computational issues, we randomly sampled one tenth of the families/pairs available (the analysis included 94,193 first-degree relatives and 44,064 spouses).

Results

Results are presented in table S11. In the first two alternative models, where the population included was the same as in the main model, we observed results very similar to the main model. In the third alternative model, where we extended the study population, we found an inter-family variance component of untreated LDL-C level twice that from the main analysis for first-degree relatives, while the estimate was similar for spouses. The main analysis is based on individuals that initiate statin, while this is not the case for all individuals used in the alternative model. Thus, the finding may indicate that statin initiation also is related to factors shared by first-degree relatives, i.e. genetics or shared behaviors not shared to the same

degree by spouses. Moreover, the difference observed in the third model underlines the need for caution in generalizing the results from this study to populations other than statin initiators.

However, the results from the first two alternative models demonstrate the stability of the chosen model when analyzing the variability in LDL-C level among statin initiators, reinforcing our conclusion of no major influence of genetic constitution on statin response.

Table S1. Variance components for LDL-C levels and statin response in cohorts of first-degree relatives and spousal pairs (results from bootstrap analysis[*]).

	First-degree relatives		Spousal pairs	
	Variance component[†] (95% CI)	Percentage of the total variance (95% CI)	Variance component[†] (95% CI)	Percentage of the total variance (95% CI)
Variance in pre-statin LDL-C level				
Inter-family variance	0.010(0.007,0.012)	7.4%(5.6%,9.3%)	0.003(0.001,0.006)	2.6%(0.4%,4.8%)
Inter-individual[‡] variance	0.039(0.035,0.043)	30.0%(27.8%,32.1%)	0.041(0.036,0.048)	31.7%(28.1%,36.3%)
Variance in statin response				
Inter-family variance	0.005(0.001,0.009)	3.8%(0.8%,6.4%)	0.008(0.003,0.012)	5.9%(2.6%,9.3%)
Inter-individual[‡] variance	0.059(0.051,0.067)	45.0%(40.7%,49.1%)	0.063(0.045,0.077)	48.6%(35.1%,56.5%)
Residual variance				
Intra-individual variance	0.018(0.015,0.021)	13.6%(10.6%,16.8%)	0.015(0.009,0.025)	11.7%(7.0%,19.6%)

CI Confidence Interval

[*] The bootstrap analysis was performed resampling the families with replacement. We sampled 2541 families of first-degree relatives and 1599 spousal pairs one hundred times and estimated variance components and percentages of variance explained as done in the main models. Thereafter we calculated the summary estimates and confidence intervals by means of medians and 2.5- and 97.5-percentiles.

[†] The model was adjusted for age, sex, period, region of residence, indication for statin treatment, education, disposable household income, type and dose of the initial statin prescription.

[‡] Inter-individual (but within-family) variance.

Table S2. Codes used in the definition of indication for treatment.

The hierarchical algorithm was previously introduced by Kildemoes et *al.* [16] and is presented here in a revised form. Individuals were classified in one of the three exclusive indication groups according to the diagnosis-, surgery- and drug codes in the table below. If individuals fulfilled the criteria for more than one category, they were classified according to the highest one.

Indication	Diagnosis codes (ICD8 and ICD10)	Surgery codes (NOMESCO Classification of Surgical Procedures)	Drug codes (ATC codes)
Individuals with established CVD			
Myocardial infarction	410.00-410.99 I21-I23, I24.1, I25.2		
Ischemic Heart Disease	411.00-414.99 I20, I24-I25 (ex. I24.1, I25.2)	KFNA-KFNG	≥2 C01D, C01d+B01A (within 6 months)
Stroke	432-437 I63-I66, I69.3-I694 G45-G46		
Peripheral arterial disease	440-441, 444.00-444.49 I70-I71, I73.9- I74.9, K55.0	KPE, KPF (only KPxE, KPxF, KPxH, KPxP, KPxQ)	
Potential atherosclerotic conditions	400-404 (ex. 401.99), 427, 424, I11-I15, I34-37, I44-I50	KPCG10, KPDG10, KPDG21-24 KPA, KPB, KPC, KPD (only KPxE, KPxF, KPxH, KPxP, KPxQ)	C01A, C01B, C03C, B01AC(-04, -06, -07, -30) (≥ 2 filled prescriptions in 6 months)
Individuals <u>without</u> established CVD, but with other cardiovascular risk factors			
Diabetes	249.00-250.99 E10-E11		A10 (≥ 2 filled prescriptions in 6 months)
Chronic renal insufficiency	403.99, 404.99, 581-584, 590.09 E10.2, E11.2, E13.2, E14.2, I12-I13, N03-N04, N07-N08, N11, N18, Z940, Z99.2 (874.40, 943.00, 943.40, Z49.1, Z49.2 at least 12 times and for at least 90 days)	574.8-574.9 KKAS0-KKAS2 BJFD2 or BJFD at least 12 times and for at least 90 days	
Primary hypertension	401.99 I10		C02, C07, C08, C09, C03B, C03A (≥ 2 filled prescriptions in 6 months)
Familial hypercholesterolemia	272.00 E78.0B		

ICD International Classification of Disease; ATC Anatomical Therapeutic Chemical Classification; CVD Cardiovascular disease.

Table S3. Characteristics of the cohorts of siblings and parent-offspring pairs.

	Cohort	
	Siblings	**Parent-offspring pairs**
Number of individuals	2075	3116
Number of families	1026	1558
Characteristics at statin initiation		
Age [median(IQR)]	53(49, 58)	58(48, 72)
Female sex	966(46.6%)	1660(53.3%)
Period		
2008-2014	694(33.4%)	1239(39.8%)
2015-2016	779(37.5%)	1122(36.0%)
2017-2018	602(29.0%)	755(24.2%)
Region of residence		
Capital region	1018(49.1%)	1799(57.7%)
Zealand	287(13.8%)	378(12.1%)
Southern Denmark	410(19.8%)	465(14.9%)
Northern Denmark	360(17.3%)	474(15.2%)
Education (years)*		
<10 years	605(29.2%)	1033(33.2%)
10-12 years	944(45.5%)	1373(44.1%)
13-15 years	374(18.0%)	515(16.5%)
≥15 years	115(5.5%)	147(4.7%)
Disposable household income†		
1 quintile	170(8.2%)	415(13.3%)
2 quintile	282(13.6%)	697(22.4%)
3 quintile	419(20.2%)	603(19.4%)
4 quintile	479(23.1%)	604(19.4%)
5 quintile	632(30.5%)	658(21.1%)
Indication for statin treatment		
Established CVD	686(33.1%)	1265(40.6%)
Other risk factors for CVD‡	593(28.6%)	832(26.7%)
No registered risk factors for CVD	796(38.4%)	1019(32.7%)
Initial statin type and dose		
Simvastatin 10 mg	55(2.7%)	92(3.0%)
Simvastatin 20 mg	303(14.6%)	445(14.3%)
Simvastatin 40 mg	735(35.4%)	1268(40.7%)
Atorvastatin 10 mg	110(5.3%)	157(5.0%)
Atorvastatin 20 mg	253(12.2%)	323(10.4%)
Atorvastatin 40 mg	348(16.8%)	487(15.6%)
Atorvastatin 80 mg	271(13.1%)	344(11.0%)

IQR, interquartile range; CVD cardiovascular disease

* 85 individuals with missing information for education were imputed using mode imputation as 10-12 years.

† 232 individuals with missing information for disposable household income were imputed as 10th decile in adjustment, consequently here as 5th quintile (mode imputation).

‡ Other risk factors for CVD includes diabetes, hypertension, chronic kidney disease and familial hypercholesterolemia (cf. table S2 for definitions).

Table S4. Variance components for LDL-C levels and statin response in the cohorts of siblings and parent-offspring pairs.

	Siblings		Parent-offspring pairs		p-value[†]
	Variance component[*] (95% CI)	Percentage of the total variance (95% CI)	Variance component[*] (95% CI)	Percentage of the total variance (95% CI)	
Variance in pre-statin LDL-C level					
Inter-family variance	0.010(0.007,0.015)	7.6%(4.8%,10.5%)	0.010(0.007,0.014)	7.6%(5.0%,10.2%)	*0.99*
Inter-individual[‡] variance	0.037(0.033,0.043)	28.3%(25.0%,31.6%)	0.039(0.035,0.044)	30.5%(27.4%,33.5%)	*0.35*
Variance in statin response					
Inter-family variance	0.007(0.003,0.021)	5.1%(0.6%,9.6%)	0.004(0.001,0.026)	2.8%(0.0%,6.5%)	*0.43*
Inter-individual[‡] variance	0.063(0.055,0.074)	48.4%(42.8%,53.8%)	0.056(0.049,0.064)	43.2%(38.3%,47.8%)	*0.16*
Residual variance					
Intra-individual variance	0.014(0.011,0.018)	10.6%(7.8%,13.6%)	0.021(0.018,0.024)	16.0%(13.3%,18.9%)	*0.009*
Total variance	0.131	100%	0.129	100%	

CI Confidence Interval

[*] The model was adjusted for age, sex, period, region of residence, indication for statin treatment, education, disposable household income and type and dose of the initial statin prescription.

[†] P-value for difference between estimates for siblings and parent-offspring pairs.

[‡] Inter-individual (but within-family) variance.

Table S5. Variance components for LDL-C levels and statin response in cohorts of first-degree relatives and spousal pairs (unadjusted analysis).

	First-degree relatives		Spousal pairs		p-value[†]
	Variance component[*] (95% CI)	Percentage of the total variance (95% CI)	Variance component[*] (95% CI)	Percentage of the total variance (95% CI)	
Variance in pre-statin LDL-C level					
Inter-family variance	0.014(0.011,0.017)	9.5%(7.5%,11.6%)	0.006(0.003,0.012)	3.8%(1.4%,6.2%)	*0.0004*
Inter-individual[‡] variance	0.046(0.042,0.050)	32.0%(29.7%,34.2%)	0.051(0.046,0.056)	35.1%(32.1%,38.0%)	*0.11*
Variance in statin response					
Inter-family variance	0.004(0.002,0.013)	2.9%(0.3%,5.6%)	0.010(0.006,0.018)	6.7%(3.3%,10.2%)	*0.09*
Inter-individual[‡] variance	0.060(0.054,0.067)	41.9%(38.3%,45.4%)	0.061(0.054,0.070)	42.3%(37.9%,46.6%)	*0.88*
Residual variance					
Intra-individual variance	0.020(0.017,0.022)	13.7%(11.6%,15.8%)	0.017(0.015,0.021)	12.1%(9.6%,14.7%)	*0.34*
Total variance	0.143	100%	0.145	100%	

CI Confidence Interval

[*] The model was adjusted for type and dose of the initial statin prescription.

[†] P-value for difference between estimates for first-degree relatives and spousal pairs.

[‡] Inter-individual (but within-family) variance.

Table S6. Variance components for LDL-C levels and statin response in the cohorts of first-degree relatives and spousal pairs refilling their statin prescription[*].

	First-degree relatives		Spousal pairs		p-value[‡]
	Variance component[†] (95% CI)	Percentage of the total variance (95% CI)	Variance component[†] (95% CI)	Percentage of the total variance (95% CI)	
Variance in pre-statin LDL-C level					
Inter-family variance	0.009(0.007,0.013)	8.1%(5.5%,10.6%)	0.003(0.001,0.015)	2.3%(0.0%,5.1%)	*0.003*
Inter-individual[§] variance	0.038(0.035,0.042)	34.3%(31.4%,37.2%)	0.043(0.038,0.048)	36.4%(32.9%,40.0%)	*0.37*
Variance in statin response					
Inter-family variance	0.003(0.001,0.015)	2.9%(0.0%,6.0%)	0.006(0.003,0.017)	5.0%(0.9%,9.1%)	*0.42*
Inter-individual[§] variance	0.042(0.037,0.049)	37.9%(33.4%,42.2%)	0.053(0.046,0.061)	44.9%(39.6%,50.0%)	*0.04*
Residual variance					
Intra-individual variance	0.019(0.017,0.022)	16.9%(14.3%,19.6%)	0.013(0.011,0.017)	11.3%(8.6%,14.3%)	*0.01*
Total variance	0.112		0.118		

CI Confidence Interval

[*] Only families where all members refilled their statin prescription within 24 weeks after the first prescription were included in the analysis (3743 first-degree relatives and 2352 spouses)

[†] The model was adjusted for age, sex, period, region of residence, indication for statin treatment, education, disposable household income and type and dose of the initial statin prescription.

[‡] P-value for difference between estimates for first-degree relatives and spousal pairs.

[§] Inter-individual (but within-family) variance.

Table S7. Variance components for LDL-C levels and statin response in the cohorts of first-degree relatives and spousal pairs with a refilling pattern consistent with the use of one pill per day[*].

	First-degree relatives		Spousal pairs		p-value[‡]
	Variance component[†] (95% CI)	Percentage of the total variance (95% CI)	Variance component[†] (95% CI)	Percentage of the total variance (95% CI)	
Variance in pre-statin LDL-C level					
Inter-family variance	0.007(0.005,0.013)	7.5%(4.0%,11.0%)	0.003(0.001,0.018)	3.4%(0.0%,7.3%)	*0.12*
Inter-individual[§] variance	0.038(0.034,0.043)	38.7%(34.7%,42.6%)	0.038(0.032,0.044)	36.8%(32.0%,41.7%)	*0.56*
Variance in statin response					
Inter-family variance	0.002(0.000,0.211)	1.7%(0.0%,5.6%)	0.006(0.003,0.021)	5.6%(0.2%,11.0%)	*0.25*
Inter-individual[§] variance	0.034(0.028,0.042)	33.9%(28.0%,39.4%)	0.038(0.031,0.048)	37.3%(30.0%,44.3%)	*0.45*
Residual variance					
Intra-individual variance	0.018(0.016,0.021)	18.3%(14.9%,21.9%)	0.017(0.014,0.021)	16.8%(12.8%,21.3%)	*0.61*
Total variance	0.100	100%	0.102	100%	

CI Confidence Interval

[*] Only families where all members refilled their statin prescription within 24 weeks after the first prescription and where the number of pills of the first statin prescription covered between 75% and 150% of the days to refill, indicating the use of one pill per day, were included in the analysis (2338 first-degree relatives and 1484 spouses).

[†] The model was adjusted for age, sex, period, region of residence, indication for statin treatment, education, disposable household income and type and dose of the initial statin prescription.

[‡] P-value for difference between estimates for first-degree relatives and spousal pairs.

[§] Inter-individual (but within-family) variance.

Table S8. Variance components for LDL-C levels and statin response in cohorts of first-degree relatives and spousal pairs (extra adjustment).

	First-degree relatives		Spousal pairs		p-value[†]
	Variance component[*] (95% CI)	Percentage of the total variance (95% CI)	Variance component[*] (95% CI)	Percentage of the total variance (95% CI)	
Variance in pre-statin LDL-C level					
Inter-family variance	0.009(0.007,0.012)	7.2%(5.3%,9.1%)	0.003(0.002,0.010)	2.6%(0.4%,4.8%)	*0.002*
Inter-individual[‡] variance	0.039(0.036,0.042)	29.7%(27.4%,31.9%)	0.041(0.037,0.045)	31.2%(28.3%,34.0%)	*0.41*
Variance in statin response					
Inter-family variance	0.004(0.002,0.013)	3.4%(0.5%,6.2%)	0.008(0.005,0.017)	6.0%(2.2%,9.7%)	*0.27*
Inter-individual[‡] variance	0.060(0.054,0.066)	45.7%(42.1%,49.3%)	0.063(0.056,0.071)	48.3%(43.8%,52.7%)	*0.38*
Residual variance					
Intra-individual variance	0.018(0.016,0.021)	14.0%(12.0%,16.1%)	0.016(0.013,0.019)	11.9%(9.6%,14.5%)	*0.20*
Total variance	0.131	100%	0.131	100%	

CI Confidence Interval

[*] The model was adjusted for age, sex, period, region of residence, indication for statin treatment, education, disposable household income, type and dose of the initial statin prescription and whether the pre-statin LDL-C level was measured during an acute inpatient contact.

[†] P-value for difference between estimates for first-degree relatives and spousal pairs.

[‡] Inter-individual (but within-family) variance.

Table S9. Variance components for LDL-C levels and statin response in the cohorts of first-degree relatives and spousal pairs excluding on-statin LDL-C measurements <6 weeks from statin initiation[*].

	First-degree relatives		Spousal pairs		p-value[‡]
	Variance component[†] (95% CI)	Percentage of the total variance (95% CI)	Variance component[†] (95% CI)	Percentage of the total variance (95% CI)	
Variance in pre-statin LDL-C level					
Inter-family variance	0.007(0.005,0.012)	5.4%(2.8%,7.9%)	0.003(0.001,0.015)	2.7%(0.0%,5.6%)	*0.18*
Inter-individual[§] variance	0.038(0.034,0.043)	29.0%(25.9%,32.2%)	0.035(0.031,0.041)	28.1%(24.3%,31.9%)	*0.70*
Variance in statin response					
Inter-family variance	0.007(0.004,0.019)	5.4%(1.2%,9.7%)	0.010(0.006,0.023)	8.2%(2.7%,13.6%)	*0.44*
Inter-individual[§] variance	0.061(0.053,0.071)	46.9%(41.7%,52.0%)	0.061(0.052,0.072)	48.5%(42.1%,54.6%)	*0.71*
Residual variance					
Intra-individual variance	0.017(0.015,0.021)	13.2%(10.5%,16.2%)	0.016(0.013,0.020)	12.6%(9.3%,16.2%)	*0.77*
Total variance	0.130	100%	0.126	100%	

CI Confidence Interval

[*] Only families where all members had their on-statin LDL-C measured 6 weeks or more after statin initiation were included in the analysis (2534 first-degree relatives and 1666 spouses).

[†] The model was adjusted for age, sex, period, region of residence, indication for statin treatment, education, disposable household income and type and dose of the initial statin prescription.

[‡] P-value for difference between estimates for first-degree relatives and spousal pairs.

[§] Inter-individual (but within-family) variance.

Table S10- Variance components for LDL-C levels and statin response in the cohorts of first-degree relatives and spousal pairs after exclusion of individuals with very low or very high levels of pre-statin LDL-C or on-statin LDL-C[*,†].

	First-degree relatives		Spousal pairs		p-value[§]
	Variance component[‡] (95% CI)	Percentage of the total variance (95% CI)	Variance component[‡] (95% CI)	Percentage of the total variance (95% CI)	
Variance in pre-statin LDL-C level					
Inter-family variance	0.009(0.007,0.012)	6.6%(4.7%,8.4%)	0.004(0.002,0.009)	2.8%(0.8%,4.8%)	*0.007*
Inter-individual[‖] variance	0.039(0.036,0.042)	29.9%(27.6%,32.1%)	0.039(0.035,0.043)	29.8%(27.1%,32.4%)	*0.95*
Variance in statin response					
Inter-family variance	0.004(0.002,0.013)	3.3%(0.5%,6.2%)	0.007(0.004,0.016)	5.6%(1.9%,9.3%)	*0.34*
Inter-individual[‖] variance	0.060(0.055,0.067)	46.6%(42.9%,50.1%)	0.068(0.062,0.076)	52.6%(48.2%,56.9%)	*0.04*
Residual variance					
Intra-individual variance	0.018(0.016,0.020)	13.6%(11.7%,15.7%)	0.012(0.010,0.015)	9.2%(7.0%,11.5%)	*0.004*
Total variance	0.130	100%	0.130	100%	

CI Confidence Interval

[*] In this analysis, individuals with very low or very high values for pre-statin LDL-C or on-statin LDL-C were excluded together with their families (11 first-degree relatives [0.41%] and 8 spouses [0.25%] were excluded in total). Extreme values were defined as values below the 0.05% quantile or above the 99.5% quantile of the distribution of pre-statin LDL (on-statin LDL-C respectively) in the total population of statin initiators having both LDL-C measurements.

[†] Overall, we observed minimal differences in the inter-family variance components compared with the result of the main analysis (Table 2). More specifically, among first-degree relatives, the decomposition of the variance in attained LDL-C was virtually unaltered, while, among spouses, the inter-individual variance in statin response accounted for a bigger share compared to the main analysis (Table 2), at the expense of the inter-individual variability in pre-statin LDL-C and the residual variance.

[‡] The model was adjusted for age, sex, period, region of residence, indication for statin treatment, education, disposable household income and type and dose of the initial statin prescription.

[§] P-value for difference between estimates for first-degree relatives and spousal pairs.

[‖] Inter-individual (but within-family) variance.

Table S11. Comparison of inter-family variance components estimated in the main model and in alternative models.

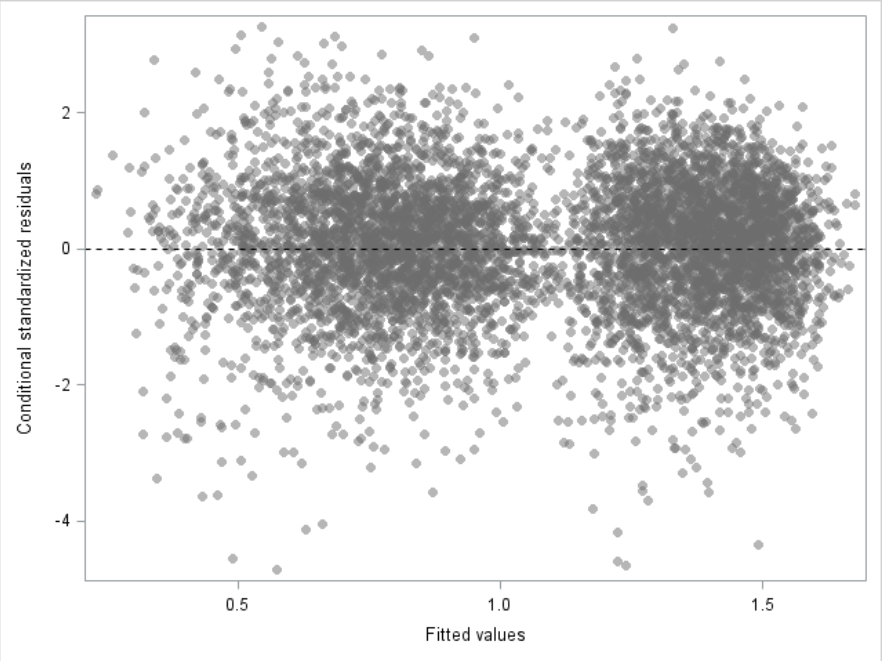
For further description of the alternative models, see Data S1.

	Main model (see Table 2)	Alternative model
Alternative model: model with covariance between the inter-family random intercept and the inter-family random statin response		
First-degree relatives		
Inter-family variance in statin response	0.004(0.002,0.013)	0.003(0.001,0.020)
% of total variance	3.3%(0.5%,6.2%)	2.4%(0.0%,5.4%)
Inter-family variance in pre-statin LDL-C level	0.010(0.008,0.013)	0.009(0.007,0.012)
% of total variance	7.4%(5.5%,9.3%)	6.9%(4.9%,8.9%)
Spousal pairs		
Inter-family variance in statin response	0.008(0.005,0.017)	0.008(0.005,0.017)
% of total variance	6.0%(2.2%,9.7%)	6.1%(2.3%,9.9%)
Inter-family variance in pre-statin LDL-C level	0.003(0.002,0.010)	0.004(0.002,0.010)
% of total variance	2.7%(0.5%,4.9%)	2.8%(0.5%,5.0%)
Alternative model: model including only statin response		
First-degree relatives		
Inter-family variance in statin response	0.004(0.002,0.013)	0.003(0.001,0.020)
% of total variance	3.3%(0.5%,6.2%)	-*
Spousal pairs		
Inter-family variance in statin response	0.008(0.005,0.017)	0.008(0.005,0.017)
% of total variance	6.0%(2.2%,9.7%)	-†
Alternative model: model including only the untreated LDL-C level‡		
First-degree relatives		
Inter-family variance in untreated LDL-C level	0.010(0.008,0.013)	0.020(0.019,0.021)
% of total variance	7.4%(5.5%,9.3%)	-§
Spousal pairs		
Inter-family variance in untreated LDL-C level	0.003(0.002,0.010)	0.002(0.001,0.005)
% of total variance	2.7%(0.5%,4.9%)	-‖

*,†,§,‖ Since the alternative model does not contain all the variance components included in the main model, it is meaningless to compare the percentages from the two models because of the different denominator. We, instead, calculated an approximate percentage of the total variance by substituting in the main model the estimate of the inter-family variance with the corresponding estimate obtained from the alternative model. By use of this approach, we obtained: 2.3% (*), 6.2% (†), 14.2% (§) and 1.8% (‖).

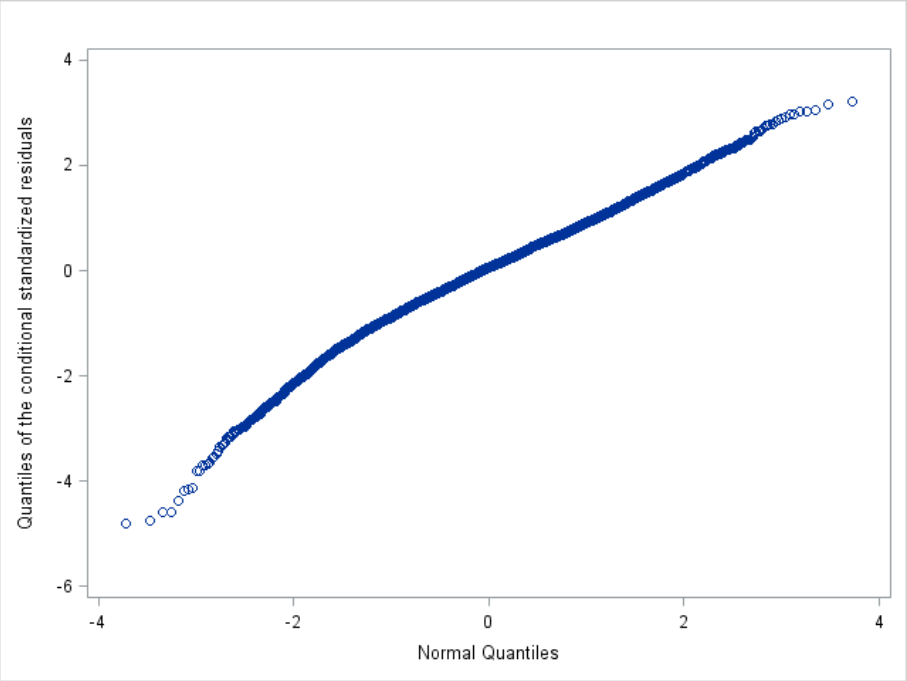
‡ This analysis included 94,193 first-degree relatives and 44,064 spouses

Figure S1. Conditional standardized residuals vs. fitted values plot.



The figure shows the conditional standardized residuals plotted against the fitted values from the model. Nine observations are not included in this graph because of conditional standardized residuals <-5.

Figure S2. QQplot for the conditional standardized residuals.



The figure shows the quantiles of conditional standardized residuals plotted against the quantiles of the normal distribution. Nine observations are not included in this graph because of conditional standardized residuals <-5.