# Influence of *PPARA, RXRA, NR1I2* and *NR1I3* gene polymorphisms on the lipid-lowering efficacy and safety of statin therapy

*Influência de polimorfismos nos genes* PPARA, RXRA, NR1I2 *e* NR1I3 *na eficácia hipolipemiante e na segurança da terapia com estatinas*

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## ABSTRACT

**Objective:** The aim of the present study was investigate the association between six genetic variants in the nuclear receptor genes *PPARA*, *RXRA*, *NR1I2* and *NR1I3* and the lipid-lowering efficacy and safety of statin therapy. **Subjects and methods:** The study was carried out on 240 Brazilian hypercholesterolemic patients on simvastatin and atorvastatin therapy. The polymorphisms were analyzed by PCR-based methods. **Results:** The *NR1I3* rs2307424 genotype distribution was different between subjects with and without adverse drug reactions. Among subjects in the ADR group, no T/T homozygotes were observed for this polymorphism, while in the non-ADR group the frequency of this genotype was 19.4% (P = 0.007, after multiple testing corrections P = 0.042). **Conclusion:** The polymorphisms investigated in *PPARA* (rs1800206), *RXRA* (rs11381416), and *NR1I2* (rs1523130) did not influence the lipid-lowering efficacy and safety of statin. Our results show the possible influence of *NR1I3* genetic variant on the safety of statin. Arq Bras Endocrinol Metab. 2013;57(7):513-9

**Keywords**

Pharmacogenetics; hydroxymethylglutaryl-CoA reductase inhibitors; nuclear receptors; single nucleotide polymorphisms

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## RESUMO

**Objetivo:** O objetivo deste estudo foi investigar a associação de seis variantes genéticas nos genes de receptores nucleares *PPARA*, *RXRA*, *NR1I2* e *NR1I3* na eficácia hipolipemiante e na segurança da terapia com estatinas. **Sujeitos e métodos:** O estudo foi realizado com 240 pacientes hipercolesterolêmicos em terapia com sinvastina e atorvastatina. Os polimorfismos foram analisados por meio de métodos baseados em PCR. **Resultados:** A distribuição da frequência genotípica do polimorfismo *NR1I3* rs2307424 foi diferente entre os pacientes com e sem efeito adverso à medicação; entre os sujeitos do grupo com efeitos adversos, nenhum homozigoto T/T foi observado, enquanto no grupo de indivíduos sem efeitos adversos a frequência desse genótipo foi 19,4% (P = 0,007, após correção para múltiplos testes P = 0,042). **Conclusão:** Os polimorfismos investigados nos genes *PPARA* (rs1800206), *RXRA* (rs11381416) e *NR1I2* (rs1523130) não foram associados com eficácia hipolipemiante e segurança da terapia com estatinas. Nossos resultados mostram uma possível influência de variantes do gene *NR1I3* (rs2307424) no desenvolvimento de efeitos adversos à terapia com estatinas. Arq Bras Endocrinol Metab. 2013;57(7):513-9

**Descritores**

Farmacogenética; inibidores de hidroximetilglutaril-CoA redutases; receptores nucleares; polimorfismo de nucleotídeo único

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Received on Sept/11/2012
Accepted on Mar/5/2013

## INTRODUCTION

In Brazil, as in most of the world, cardiovascular disease (CVD) is the leading cause of death (1). Among the classical risk factors for CVD, dyslipidemia is considered important, and lipid-lowering therapy is the central approach in the primary and secondary prevention of CVD (2). The 3-hydroxy-3-methylglutaryl-coenzyme A (HMG-CoA) reductase inhibitors (statins) function as competitive inhibitors of the rate-limiting enzyme in the cholesterol biosynthesis pathway and are the most prescribed drugs for dyslipidemia treatment and CVD prevention worldwide (3). Although statins

are usually highly effective and generally well tolerated, interindividual variation have been seen in relation to lipid-lowering efficacy and adverse effect occurrence. Just as genetic variability influence plasma lipid and lipoprotein levels, gene polymorphisms are also related with drug-response differences and may account for 15%-30% of this variability (4).

Nuclear receptors are a superfamily of more than 50 binding-activated transcription factors directly involved in gene expression control in different metabolic pathways and in response to a wide range of developmental, physiological, and environmental stimuli (5). Peroxisome proliferator-activated receptor alpha (PPARα or NR1C1) regulates a variety of target genes involved in lipid and glucose metabolism, inflammatory response, and energy homeostasis (6). To bind to DNA and regulate transcription of target genes, this nuclear receptor requires heterodimerization with the retinoid X receptor alpha (RXRα or NR2B1), another member of nuclear receptor superfamily. RXRα influences a variety pathways because of its ability to activate transcription of target genes as a homodimer or as an obligate partner of other nuclear receptors, such as pregnane X receptor (PXR or NR1I2) and constitutive androstane receptor (CAR or NR1I3) (5,7). PXR and CAR were originally identified as xenosensors that regulate the expression of drug-metabolizing enzymes/transporters (Phase I, II and III); however, recent studies have shown that they also affect other metabolic pathways, such as lipid homeostasis and metabolism (8,9).

Considering the influence of nuclear receptors on lipid metabolism and on the recognition/metabolism of xenobiotic compounds, and that few studies addressed the influence of genetic variants in the genes encoding PPARα, RXRα, PXR, and CAR, the aim of the present study was to evaluate the association between *PPARA* rs1800206, *RXRA* rs11381416, *NR1I2* rs1523130 and rs2472677, and *NR1I3* rs2307424 and rs2501873 polymorphisms and the lipid-lowering efficacy and safety of simvastatin and atorvastatin in a Brazilian population of European ancestry.

## MATERIALS AND METHODS

### Study subjects and protocol

Two hundred forty Brazilian hypercholesterolemic patients of European descent from a cardiovascular clinic in southern Brazil were investigated in a cohort study according to simvastatin or atorvastatin treatment. Exclusion criteria were: unstable or uncontrolled clinically diseases, uncontrolled hypothyroidism, and impaired hepatic or renal function. None of the patients were on previous therapy with statins or other lipid-lowering drugs. The statin therapy used (simvastatin or atorvastatin) and the doses administrated were determined by the physician according to the clinical characteristics of each patient.

Two hundred forty patients were analyzed for the lipid-lowering efficacy of the therapy. They had their lipid and lipoprotein concentrations measured at baseline and after approximately 6 months (5.97 ± 2.44 months) of treatment. Therefore, ninety-eight patients who remained on same dosage and statin therapy for more than a year (37.70 ± 23.12 months, minimum: 12 and maximum: 131 months) without presenting adverse drug reaction (ADR) were named the non-ADR group. Their genotypes and allele frequencies were compared with those of the 30 patients who developed adverse drug reactions (ADR group), initial time mean of adverse manifestation in ADR group was 6.83 ± 6.42 months (minimum: 1 and maximum 24 months). Adverse drug reaction were considered one or more events of myalgia concomitant with simvastatin/atorvastatin treatment, which was defined as proximal or diffuse muscle pain, tenderness and/or weakness, or both pain and weakness, with normal or increased serum creatine phosphokinase (CPK) levels, and alteration of hepatic function (ascertained by elevated serum levels of hepatic aminotransferases) (10,11). Patients were also screened and not included in ADR-group if other unrelated conditions that may contribute independently to muscle aches, as arthritis, viral myalgia, exercise-induced myalgia and fibromyalgia, or alteration of hepatic function were present. This protocol was approved by the Ethics Committee of Universidade Federal do Rio Grande do Sul, Universidade Federal de Ciências da Saúde de Porto Alegre, and Centro Universitário Metodista do IPA. A written informed consent form was signed by every subject included in the study.

### Biochemical analyses

Blood samples were collected from individuals after a 12-hour fast. Total cholesterol (TC), high-density lipoprotein cholesterol (HDL-C), triglyceride (TG), and glucose levels were determined by conventional enzymatic methods. Low-density lipoprotein cholesterol

(LDL-C) was calculated according to (12), when plasma triglycerides were below than 4.52 mmol/L. Glucose was measured by conventional methods.

### DNA analyses

Genomic DNA was isolated from peripheral blood leukocytes by a standard salting-out procedure (13). The polymorphisms c.484C>G (rs1800206; NM/NP_001001928.1; p.Leu162Val) of the *PPARA* gene and the deletion/insertion polymorphism -/A (rs11381416) of the *RXRA* gene were determined using PCR and restriction mapping (PCR-RFLP), as previously described (14,15).

The polymorphism c.-1663T>C (rs1523130; NM_003889.3) of *NR1I2* was determined using an in-house-designed PCR-RFLP. The amplification reactions used the following primers: forward, 5'-GTCATGAGGATATTGGACCG-3', and reverse, 5'-TAGCCATGGCCTTCTGATCT-3'. The polymorphism c.-22-7659T>C (rs2472677; NM_003889.3) of *NR1I2* and c.540C>T (rs2307424; NM_001077469.1; NP_001070937.1: p. Pro180Pro) and c.238+1099A>G (rs2501873; NM_001077469.1) of *NR1I3* were determined by allelic discrimination with TaqMan 5'-nuclease assays. Genotyping for rs2472677 (ID: C_26079845_10), rs2307424 (ID: C_25746794_20), and rs2501873 (ID: C_16033320_10) polymorphisms were performed with validated TaqMan genotyping assays (Real Time PCR, Applied Biosystems, California, USA).

For analytical adjustment purposes, the total sample was genotyped for *APOE* allele variants (E*2, E*3 and E*4) as previously described (16).

### Statistical analyses

Continuous variables were expressed as means ± standard deviations. TG levels were log-transformed before analyses because of their skewed distribution, although non-transformed values are shown in the Results.

Allele frequencies were estimated by gene counting. The agreement of genotype frequencies with the Hardy-Weinberg equilibrium expectations was tested using chi-square test. GraphPad InStat version 2.04a (GraphPad Software, San Diego, California, USA) was used to compare allele and genotype frequencies among groups by Fisher's exact test and the chi-square test, respectively. When appropriate, chi-square tests as described by Roff and Bentzen (17) (CHITEST.EXE software) were performed. Haplotype frequencies and linkage disequilibrium were estimated with Multiple Locus Haplotype Analysis version 2.0 (18,19) and ARLEQUIN software version 3.1 (20). Dmax (D theoretical maximum) and D' (the relative magnitude of D as compared with its theoretical maximum, calculated as D/Dmax) values were calculated as described by Lewontin (21).

Because of lower homozygous genotype frequencies, rs1800206 genotypes of the *PPARA* gene were grouped as C allele homozygotes (C/C) and G allele carriers (C/G and G/G), and rs11381416 genotypes of the *RXRA* gene were grouped as homozygous without the A insertion (–/–) and carriers of the A insertion (–/A and A/A). Considering the possible difference in the lipid-lowering efficacy of simvastatin and atorvastatin therapy in different doses, we created a standardized statin dosage variable transforming the daily doses of simvastatin to equivalent doses of atorvastatin by using the dose equivalence ratio 2:1 for simvastatin:atorvastatin, based on published side-by-side comparisons Jones and cols. (22) also used by Kivistö and cols. (23).

To determine the association of the genotypes and diplotypes with baseline lipid levels or response to statin treatment (mean percentage changes in plasma lipid levels), means of each variable were compared with a General Linear Model using the type III sums of squares. Age, gender, smoking status, standardized statin dosage, treatment period (months), baseline lipid levels, and dummy variables for the presence of E*2 and E*4 *APOE* alleles (*APOE* dummy variables) were included in each model as covariates for the lipid-lowering response. For baseline lipid level association analyses, age, gender, smoking status, and *APOE* dummy variables were used as covariates. Patients with E*2/E*4 genotype were excluded from analyses. Pairwise comparisons among genotypes were performed by least significant difference with no adjustments. Statistical analysis was performed using SPSS 16.0 for Windows®. The Benjamini and Hochberg false discovery rate procedure was performed for multiple testing correction (24), corrected P values were inserted in the tables and the text when significant P were detected in analyses.

## RESULTS

### Characteristics of the study population

The characteristics of the total sample and, separately, of samples analyzed for lipid-lowering response and the

ADR and non-ADR groups, are presented in table 1. The total sample comprised 240 patients aged between 25 and 88 years (62.23 ± 10.68 years), 68.2% were females. When the groups were compared, only the frequencies of CVD and the use of calcium channel blocker as concomitant therapy showed significant differences. None of the other characteristics differ between groups.

The genotype frequencies observed did not show statistically significant differences compared with those expected under the Hardy-Weinberg equilibrium. Linkage disequilibrium was not detected between the polymorphisms of *NR1I2* and *NR1I3* (D' = 0.119, P = 0.088 and D' = 0.109, P = 0.108, respectively).

### Association between *PPARA*, *RXRA*, *NR1I2* and *NR1I3* polymorphisms and statin efficacy and safety

Table 2 shows the mean percent modifications of lipid and lipoprotein levels in all patients investigated for lipid-lowering response and according to polymorphisms genotypes. Overall, statin therapy significantly reduced the plasma levels of TC (-26.36%, P < 0.001), LDL-C (-36.44%, P < 0.001) and TG (-10.18%, P < 0.001), whereas the increase in HDL-C did not reach statistical significance (3.96%, P = 0.321).

**Table 1.** Main characteristics of patients

| Characteristics | Lipid-lowering response | Non-ADR-group | ADR-group |
|---|---|---|---|
| No. | 240 | 98 | 30 |
| Age (y) | 62.07 ± 10.7 | 65.17 ± 9.54 | 61.77 ± 8.7 |
| Sex (% male) | 31.7 | 35.7 | 30 |
| Smoking | | | |
| Never (%) | 80.8 | 73.2 | 80 |
| Past (%) | 10.4 | 15.5 | 13.3 |
| Current (%) | 8.8 | 11.3 | 6.7 |
| CVD (%)[1] | 34.3 | 53.1 | 33.3 |
| Hypertension (%) | 72.1 | 80.6 | 70 |
| Diabetes (%) | 19.3 | 27.6 | 10 |
| Glucose (mmol/L) | 5.54 ±1.37 | 5.74 ± 1.72 | 5.42 ± 0.81 |
| Concomitant therapies | | | |
| ACE inhibitor (%) | 22.1 | 28.6 | 23.3 |
| β-Blocker (%) | 35.8 | 49.5 | 40 |
| Calcium channel blocker (%)[2] | 16.7 | 28.6 | 6.7 |
| Diuretics (%) | 41.7 | 52.0 | 43.3 |

Values for age and glucose levels are expressed as mean ± SD. [1] Lipid-lowering response x Non-ADR group, P = 0.002. [2] Lipid-lowering response x Non-ADR group, P = 0.017; ADR group x Non-ADR group, P = 0.013. ADR: adverse drug reaction; CVD: previous coronary heart disease; ACE: angiotensin-converting enzyme.

When a recessive model was tested for *NR1I3* rs2501873 polymorphism, a greater reduction of LDL-C was observed in G allele carriers than in A/A homozygotes (-37.63 ± 16.79% *versus* -29.82 ± 21.66%; P = 0.026, after multiple testing corrections P = 0.156). Significant difference in lipid and lipoprotein reduction was not observed among the other polymorphism genotypes.

As shown in table 3, a significant difference in frequencies distribution was observed for *NR1I3* rs2307424 polymorphism between subjects with or without ADR (P = 0.007, after multiple testing corrections P = 0.042). Among subjects in the ADR group, no T/T homozygotes were observed, while in non-ADR group, the frequency of this genotype was 19.4%. No other polymorphisms were significantly associated with ADR.

## DISCUSSION

In this study, we examined the association between *PPARA* rs1800206, *RXRA* rs11381416, *NR1I2* rs1523130 and rs2472677, and *NR1I3* rs2307424 and rs2501873 polymorphisms with simvastatin and atorvastatin response, considering both lipid-lowering efficacy and adverse effect occurrence. Our major findings were the association of *NR1I3* gene polymorphisms with statin safety. As described previously, PXR (*NR1I2*) and CAR (*NR1I3*) were originally identified as xenosensors that regulate the expression of drug-metabolizing enzymes/transporters (Phase I, II and III). These include the phase I enzymes cytochrome P450 (CYP) 2B6, CYP2B9, CYP2C8, CYP2C9, CYP3A4 and CYP3A7, the phase II enzymes the glutathione--*S*-transferases (GSTs), UDP-glucuronosyltransferases (UGTs), and sulfotransferases (SULTs); and the transporters, multidrug resistance protein 1 (MDR1), MDR2, multidrug resistance-associated protein 2 (MRP2), and the organic anion transporter polypeptide 2 (OATP2) (25). Simvastatin and atorvastatin are mainly metabolized by CYP3A4 and transported by MDR1, and therefore polymorphisms in these genes might affect its metabolism/transport and efficacy/safety of treatment. To the best of our knowledge, no study available showed the association of these polymorphisms with statins pharmacokinetics.

Despite the recognized importance of CAR in xenobiotic/endobiotic metabolism, conjugation, and transport, not much is known about the genetic varia-

**Table 2.** Baseline and mean percent change of lipid and lipoprotein levels after statin treatment

| | N | TC (mmol/L) | LDL-C (mmol/L) | HDL-C (mmol/L) | TG (mmol/L) |
|---|---|---|---|---|---|
| Baseline | 240 | 6.56 ± 1.06 | 4.46 ± 0.93 | 1.30 ± 0.31 | 1.85 ± 0.94 |
| Treatment | 240 | 4.78 ± 0.87 | 2.76 ± 0.72 | 1.31 ± 0.35 | 1.53 ± 0.69 |
| P | | < 0.0001 | < 0.0001 | 0.321 | < 0.0001 |
| % Change Overall | 240 | -26.36 ± 12.96 | -36.44 ± 17.68 | 3.96 ± 24.9 | -10.18 ± 33.14 |
| *PPARA* rs1800206 | | | | | |
| C/C | 206 | -25.97 ± 13.18 | | 4.60 ± 25.47 | -9.99 ± 34.22 |
| C/G or G/G | 30 | -28.34 ± 11.99 | -38.43 ± 15.05 | 1.48 ± 22.05 | -10.34 ± 25.83 |
| P | | 0.357 | 0.623 | 0.361 | 0.880 |
| *RXRA* rs11381416 | | | | | |
| _/_ | 195 | -26.13 ± 12.91 | -36.42 ± 17.51 | 4.97 ± 25.5 | -10.31 ± 32.57 |
| _/A or A/A | 39 | -28.16 ± 12.48 | -38.66 ± 15.55 | 0.24 ± 23.17 | -7.40 ± 36.90 |
| P | | 0.865 | 0.994 | 0.597 | 0.739 |
| *NR1I2* rs1523130 | | | | | |
| T/T | 27 | -23.13 ± 11.66 | -34.07 ± 16.47 | 0.94 ± 18.07 | -4.37 ± 37.74 |
| T/C | 126 | -27.39 ± 13.54 | -37.37 ± 18.11 | 4.23 ± 27.37 | -10.41 ± 32.70 |
| C/C | 79 | -25.49 ± 12.57 | -35.85 ± 17.46 | 5.20 ± 23.87 | -10.39 ± 33.09 |
| P | | 0.290 | 0.485 | 0.407 | 0.612 |
| *NR1I2* rs2472677 | | | | | |
| C/C | 33 | -25.90 ± 14.71 | -36.17 ± 19.24 | 0.46 ± 18.30 | -8.58 ± 39.90 |
| C/T | 120 | -26.26 ± 13.83 | -35.86 ± 18.67 | 2.44 ± 23.48 | -7.78 ± 33.33 |
| T/T | 83 | -26.43 ± 11.19 | -37.39 ± 15.97 | 8.05 ± 28.78 | -13.77 ± 30.15 |
| P | | 0.935 | 0.742 | 0.170 | 0.442 |
| *NR1I3* rs2307424 | | | | | |
| T/T | 31 | -26.80 ± 11.41 | -36.03 ± 16.2 | 4.70 ± 28.14 | -8.38 ± 36.29 |
| T/C | 89 | -28.15 ± 13.07 | -37.87 ± 17.46 | -0.33 ± 18.82 | -13.22 ± 32.67 |
| C/C | 116 | -24.69 ± 13.32 | -35.48 ± 18.48 | 7.45 ± 27.74 | -8.11 ± 32.86 |
| P | | 0.436 | 0.940 | 0.255 | 0.666 |
| *NRI13* rs2501873 | | | | | |
| G/G | 79 | -27.38 ± 13.52 | -37.73 ± 16.92 | 4.78 ± 22.53 | -9.01 ± 36.66 |
| G/A | 120 | -26.42 ± 12.49 | -37.57 ± 16.79 | 2.27 ± 24.73 | -7.64 ± 32.57 |
| A/A | 37 | -23.41 ± 13.65 | -29.82 ± 21.66 | 9.20 ± 30.39 | -20.03 ± 26.10 |
| P | | 0.436 | 0.084[1] | 0.563 | 0.530 |

Covariates included in the model are: age, gender, smoking status, standardized statin dosage, treatment period (months), baseline lipid levels, and *APOE* dummy variables; unadjusted and untransformed values are expressed as mean ± SD; [1] G carriers *versus* A/A for *NR1I3* rs2501873, P = 0.026, corrected P = 0.156 (-37.63 ± 16.79 *versus* -29.82 ± 21.66). TC: total cholesterol; LDL-C: low-density lipoprotein cholesterol; HDL-C: high-density lipoprotein cholesterol; TG: triglycerides.

tion of this gene. In our study, we primarily detected the association of rs2501873, a transition substitution A>G in intron 3 of the gene with statin treatment response, although after multiple testing correction p-values did not remain statistically significant. The *NR1I3* rs2307424 polymorphism, another polymorphism in same gene, is a transition substitution C>T at codon 180 of exon 5, corresponding to a silent mutation, proline/proline (P180P), in the ligand-binding domain. This polymorphism there is located in a region of the gene that encodes a multifunctional domain of the protein. This domain mediates, among other actions, the dimerization with RXRα, interaction with co-activator proteins, nuclear localization, and transactivation functions, as reviewed by (26). To our knowledge, the functionality of these polymorphisms has not been tested, but our results encourage a functional study of them. In present study, no patient in the ADR group was T/T homozygote at *NR1I3* rs2307424, and the genotype distribution of this variant was different between the ADR group and the controls.

Advanced age, female gender, multisystemic diseases, and multiple and/or concomitant medications have been described as risk factors for statin ADRs (27). In our study, a comparison of clinical characteristics between ADR and non-ADR groups (Table 1) showed that these risk factors do not appear to confound the genetic influence of the *NR1I3* rs2307424 polymorphism, because frequencies in the groups were similar. With exception of calcium channel blocker use, that was more frequent in non-ADR group than in ADR patients, however, these drugs are classical CYP3A4 and glycoprotein P inhibitors, and might increase risk of adverse drug reaction (28-30).

**Table 3.** Genotype and allele frequencies of the polymorphisms in non-ADR group and patients that developed adverse drug reaction (ADR group)

| Polymorphism | Genotypes | | | Alleles | |
|---|---|---|---|---|---|
| *PPARA* rs1800206 | C/C | C/G or G/G | | C | G |
| non-ADR group | 85 (86.7%) | 13 (13.3%) | | 92.86% | 7.14% |
| ADR group | 29 (96.7%) | 01 (3.3%) | | 98.33% | 1.66% |
| P | 0.111 | | | 0.204 | |
| *RXRA* rs11381416 | _/_ | _/A or A/A | | _ | A |
| non-ADR group | 80 (81.6%) | 18 (18.4%) | | 89.80% | 10.20% |
| ADR group | 25 (83.3%) | 05 (16.7%) | | 91.67% | 8.33% |
| P | 1.00 | | | 0.807 | |
| *NR1I2* rs1523130 | T/T | T/C | C/C | T | C |
| non-ADR group | 10 (10.9%) | 44 (47.8%) | 38 (41.3%) | 34.78% | 65.22% |
| ADR-group | 06 (20%) | 15 (50%) | 09 (30%) | 45.0% | 55.0% |
| P | 0.331 | | | 0.169 | |
| *NR1I2* rs2472677 | C/C | C/T | T/T | C | T |
| non-ADR group | 17 (17.3%) | 46 (46.9%) | 35 (35.7%) | 40.82% | 59.18% |
| ADR group | 09 (30.0%) | 13 (43.3%) | 08 (26.7%) | 51.67% | 48.33% |
| P | 0.296 | | | 0.180 | |
| *NR1I3* rs2307424 | T/T | T/C | C/C | T | C |
| non-ADR group | 19 (19.4%) | 38 (38.8%) | 41 (41.8%) | 38.78% | 61.22% |
| ADR group | 0 (0%) | 19 (63.3%) | 11 (36.7%) | 31.67% | 68.33% |
| P | 0.007[1,2] | | | 0.361 | |
| *NR1I3* rs2501873 | G/G | G/A | A/A | G | A |
| non-ADR group | 38 (38.8%) | 44 (44.9%) | 16 (16.3%) | 61.22% | 38.78% |
| ADR group | 08 (26.7%) | 14 (46.7%) | 08 (26.7%) | 50.0% | 50.0% |
| P | 0.366 | | | 0.136 | |

Values are expressed as frequency (percentage). [1] P = 0.007 ± 0.005 (±2 standard error), calculated using the Roff and Bentzen (1989) method. [2] Corrected P = 0.042. ADR: adverse drug reaction.

To the best of our knowledge, only one previous study (31) investigated the association of *PPARA* rs1800206 polymorphism with the lipid-lowering efficacy of statins; they found no association between *PPARA* rs1800206 genotypes and lipid baseline levels or lipid-lowering response to fluvastatin. Our results, taken together with this study, indicate lack of association this polymorphism with in lipid-lowering response to statins. In our study, *NR1I2* rs1523130 and rs2472677 polymorphisms did not show any influence on the lipid-lowering response or safety of simvastatin or atorvastatin treatment. Although *NR1I2* rs1523130 T allele has been associated with higher hepatic basal CYP3A4 activity, reduced hepatic induction of CYP3A4, lower intestinal levels of CYP3A4 and PXR mRNA, and lower promoter activity (32). No others studies investigated the association of this variant with statin pharmacogenetics.

There were some limitations to our study. First, our investigation addressed the effect of only one or two polymorphisms per gene, and it only took into account the *APOE* genotypes as the covariates between all polymorphisms previously related with the variables analyzed. Second, although we created a standardized statin dosage variable to equate the differences between treatment efficacies. Third, as previously discussed, we cannot exclude the possibility of a statistical type II error due the small sample size analyzed for adverse effects. Despite these limitations, and although our findings require confirmation in larger and in different populations, they suggest candidate polymorphisms for association with statin pharmacogenetics.

In summary, our study demonstrates that, in a Brazilian population of European descent, *NR1I3* rs2501873 polymorphism might modify the lipid-lowering response to statins, and *NR1I3* rs2307424 polymorphism may influence adverse reactions to simvastatin and atorvastatin, and should be considered genetic contributors to interindividual differences. To our knowledge, this study is the first to describe these associations; therefore, additional studies are warranted to confirm them. Pharmacogenetic studies are an opportunity to discover safer and more efficient pharmacotherapies, and although our study does not intend to explain all the variability determined by genetic variants, it is an effort to help in this area of research.

Acknowledgments: the authors would like to thank the staff of Centro de Diagnóstico Cardiológico for their assistance. The authors are also thankful for the financial support provided by Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq, Brazil), Institutos do Milênio (CNPq, Brazil), Programa de Apoio a Núcleos de Excelência (Pronex, Brazil), Fundação de Amparo à Pesquisa do Estado do Rio Grande do Sul (Fapergs, Brazil), Programa de Bolsas Reuni/UFCSPA, and Proap-Capes.

Disclosure: no potential conflict of interest relevant to this article was reported.

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