Genetics and Statins in the Progression for Age-Related Macular Degeneration

Genética e Toma de Estatinas na Progressão da Degenerescência Macular da Idade

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ABSTRACT

INTRODUCTION: Age-related macular degeneration (AMD) is a multifactorial disease with unclear pathophysiology. One of the pathways identified is lipid metabolism. Drusen, AMD hallmark, are lipid-rich deposits. Additionally, genes participating in lipid homeostasis are associated with the disease. Statins have been studied as possible protective drugs against AMD, though inconclusively.

We aim to explore statin's role in AMD progression, considering AMD genetic risk, in the Coimbra Eye Study.

MATERIAL AND METHODS: Six hundred eighty-three subjects (1321 eyes): 112 subjects (168 eyes) progressed over 6.5 years, and 561 subjects (1153 eyes) did not. Progressors were subjects who progressed from grade 0, 1a or 1b to grades 2,3 or 4 between visits (Rotterdam Classification). Only subjects taking statins between visits were considered. A Genetic Risk Score was calculated using 52 SNPs. Strength of statins was defined as high, medium, and low, according to INFARMED guidelines. To investigate the association between statins and AMD progression, an extended Cox regression analysis was used, adjusted for baseline-age, -sex, -body mass index, -smoking, -diabetes, and -arterial hypertension. Statin use was corrected by the genetic risk score. Correlation between eyes was considered.

RESULTS: The use of statins was significantly associated with a decreased risk in AMD progression (HR=0.509, 95% CI 0.302-0.860, p=0.012), when corrected for age, sex, body mass index, smoking, diabetes and arterial hypertension. This association was maintained for medium- and high-dose statins (HR=0.482, 95% CI 0.285- 0.814, p= 0.006), but not for low-dose statins (HR=1.321, 95% CI 0.159-10.995, p= 0.797), corrected for the same variables. After controlling for the Genetic Risk Score, statins remained significantly associated with a risk reduction for AMD progression (HR=0.516, 95% CI 0.293-0.91, p= 0.022).

CONCLUSION: The use of statins suggested a protection against AMD progression after controlling for age, sex, obesity, smoking habits, diabetes and arterial hypertension, highlighting lipids involvement in AMD pathophysiology. When considering AMD genetic risk, statins maintained its association with a decreased risk of progression. Our results indicate a benefit of these drugs in the delay of the disease course, even weighting the AMD genetic profile, contributing to a possible therapeutic strategy.

KEYWORDS: Disease Progression; Genetic Predisposition to Disease; Hydroxymethylglutaryl-CoA Reductase Inhibitors; Macular Degeneration/genetics.

RESUMO

INTRODUÇÃO: A degenerescência macular da idade (DMI) é uma doença multifactorial cuja fisiopatologia permanece inconclusiva. O metabolismo lipídico é um dos mecanismos identificados. Os drusen, característicos da doença, são ricos em lípidos. Adicionalmente, estão associados à doença genes da homeostase lipídica. As estatinas são fármacos estudados como alvo terapêutico, embora sem resultados conclusivos.

O nosso objectivo é avaliar se as estatinas são protectoras da progressão da DMI, considerando o risco genético para a doença.

MATERIAL E MÉTODOS: Seiscentos oitenta três participantes (1321 olhos): 112 progressores ao longo de 6,5 anos (168 olhos) e 561 não progresores (1153 olhos). Os progressores progrediram do nível 0, 1a ou 1b para os níveis 2,3 e 4 entre visitas do Coimbra Eye Study (Classificação de Roterdão). Foram apenas considerados os participantes que tomaram estatinas entre as visitas. O risco genético foi calculado com os 52 SNPs. Definiu-se a potência das estatinas de acordo com guidelines do INFARMED. Calculou-se a associação entre as estatinas e a progressão da DMI usando uma extensão do modelo de regressão de COX, ajustado à baseline para a idade, sexo, IMC, tabaco, diabetes e pressão arterial. A toma de estatinas foi corrigida pelo risco genético. Considerou-se a correlação entre olhos.

RESULTADOS: A associação entre as estatinas e risco diminuído de progressão da DMI foi estatisticamente significativa (HR=0,509, 95% CI 0,302-0,860, *p*=0,012), ajustada à baseline para a idade, sexo, IMC, tabaco, diabetes e pressão arterial. Esta associação manteve-se para as estatinas de alta e média potência (HR=0,482, 95% CI 0,285-0,814, *p*=0,006), mas não para as de baixa potência (HR=1,321, 95% CI 0,159-10,995, *p*=0,797). A associação à diminuição do risco de progressão manteve-se significativa depois de ajustada ao risco genético para a DMI (HR=0,516, 95% CI 0,293-0,91, *p*=0,022).

CONCLUSÃO: A toma de estatinas sugeriu uma protecção contra a progressão da DMI, mesmo considerando o risco genético para a doença, evidenciando o envolvimento lipídico na sua fisiopatologia. Estes resultados indicam que as estatinas podem ser benéficas na gestão da doença, mesmo ponderando o perfil genético da DMI, contribuindo para uma possível estratégia terapêutica.

PALAVRAS-CHAVE: Degenerescência Macular/genética; Inibidores da Hidroximetilglutaril-CoA Redutase; Predisposição Genética para a Doença; Progressão da Doença.

INTRODUCTION

Age-related macular degeneration (AMD) is a disease that affects the macula, leading to irreversible vision loss. It is the main cause of adult blindness in people over 55 years old in developed countries.[1] AMD prevalence is estimated in 8%,[2] but it is expected to achieve 288 million by 2040.[3]

Age-related macular degeneration is a multifactorial disease, with a complex aetiology and with several pathways and mechanisms that may contribute for its pathophysiology.[1] The lipid metabolism pathway stands out as one, for different reasons: drusen, the main characteristic of the disease, are rich in lipids. Also, high serum cholesterol levels have been associated with AMD in several studies.[4] Lipid homeostasis genes are independently associated with AMD,[5] which has been corroborated by metabolomic evidence.[6,7] As so, statins arise as a possible drug option for AMD management and several studies have reported

their use in the protection of AMD onset or progression, with conflicting results.[8–11] Additionally, no study has yet addressed the question on how statins and AMD genetic risk may interact within themselves on the outcome on AMD progression. With this study, we will assess the risk for AMD progression associated with the use of systemic concomitant statins in the Coimbra Eye Study (CES), considering the genetic risk for AMD.

MATERIAL AND METHODS

THE COIMBRA EYE STUDY

The Coimbra Eye Study comprises different studies: The Epidemiological Study of the Prevalence of Age-Related Macular Degeneration in Portugal (NCT01298674) was performed in two cohorts (Mira and Lousã); The Lifestyle and Food Habits Questionnaire in the Portuguese Population Aged 55 or More (NCT01715870) performed in two cohorts (Mira and Lousã) and the Five-year Incidence of Age-related Macular Degeneration in the Central Region of Portugal Study (NCT02748824) performed in Mira Cohort. This manuscript refers to the Epidemiological Study of the Prevalence of Age-Related Macular Degeneration in Portugal (NCT01298674) and to the Five-year Incidence of Age-related Macular Degeneration in the Central Region of Portugal Study (NCT02748824), in the Mira cohort. These are epidemiological and cross-sectional studies. The baseline visit was considered the visit of The Epidemiological Study of the Prevalence of Age-Related Macular Degeneration in Portugal (NCT01298674) and the follow-up visit was considered the visit of the Five-year Incidence of Age-related Macular Degeneration in the Central Region of Portugal Study (NCT02748824), 6.5 years later.

Different ophthalmological exams were performed: best-corrected visual acuity with Early Treatment Diabetic Retinopathy Study (ETDRS) charts, colour fundus photography (CFP) (Topcon® fundus camera, TRC-NW8; Topcon Corp., Tokyo, Japan), Spectral Domain Optical Coherence Tomography (SD-OCT), fundus autofluorescence (FAF), and Infra-Red (IR) imaging (Spectralis HRA+OCT Heidelberg Engineering, Heidelberg, Germany). Patients were assessed by an ophthalmologist and blood was drawn for genetic analysis. Medical history was recorded by a study nurse.

The tenets of the Declaration of Helsinki and International Conference on Harmonization - Good Clinical Practice Guidelines were followed. The studies obtained AIBILI's Ethics Committee approval. Patients signed the informed consent before undergoing any study procedures and were informed of possible consequences.

Details of these studies can be found elsewhere.[12–14]

CONCOMITANT MEDICATION

Patients were asked to bring the concomitant medication they were taking at the day of the visit. In case of oblivion, the medication was checked in the patients file of the Primary Health Care Unit. Each medication was managed as Anatomical Therapeutic Chemical (ATC) codes.

Lipid modifying agents, plain: C10AB - Fibrates; C10AC – Bile acid sequestrants; C10AD – Nicotinic acid and derivatives; C10AX - Other lipid modifying agents; C10AA - HMG CoA reductase inhibitors; C10BA - Combinations of various lipid modifying agents.

Subjects who were already taking statins before the first study visit were excluded from this analysis. Only those who started taking statins within the two visits were considered, in order not to consider a possible effect of statins before the progression of AMD.

The strength of statins was defined as high, medium, and low, according to the Portuguese National Authority of Medicines (INFARMED).[15] High-strength statins include atorvastatin 40-80 mg and rosuvastatin 20-40 mg. Medium-strength statins include atorvastatin 10-20 mg, rosuvastatin 5-10 mg, simvastatin 20-40 mg, pravastatin 20-80 mg, fluvastatin 40 mg and pitavastatin 2-4 mg. Low-strength statins include simvastatin 10 mg, pravastatin 10 mg, fluvastatin 20-40 mg and pitavastatin 1 mg.

GENOTYPING AND GENETIC RISK SCORE (GRS)

Genotyping and calculation of the genetic risk score of participants was performed according to what is published elsewhere.[16] In brief, genotyping was performed within EYE-RISK project of the E3 - The European Eye Epidemiology Consortium. A single-molecule molecular inversion probe and next generation sequencing were used to target single nucleotide polymorphisms (SNPs) and coding and splice-site regions of 10 AMD-related genes (*ARMS2*, *C3*, *C9*, *CD46*, *CFB*, *CFH*, *CFI*, *HTRA1*, *TIMP3* and *SLC16A8*) and 3 genes associated with inherited macular dystrophies (*ABCA4*, *CTNNA1*, and *PRPH2*). Ten SNPs were genotyped by KASP genotyping assays to ensure a full genotyping of the 52 variants identified by the International AMD Genomics Consortium (IAMDGC).[17] Sixty-nine SNPs were successfully genotyped.

The GRS was calculated for each participant individually, considering the number of minor alleles of variant *i*, N_i and their effect size β_i (natural logarithm of the fully conditioned odds ratio of the minor allele of variant *i*) determined by the genome-wide association study (GWAS) of the IAMDGC, according to the formula: $GRS = \sum_{i=1}^{52} (N_i \beta_i)$ In order to have a GRS value calculated, only participants with the five major risk variants genotyped (*CFH* rs570618, *CFH* rs10922109, *C2/CFB/SKIV2L* rs429608, *ARMS2/HTRA1* rs3750846 and *C3* rs2230199) were considered. Otherwise, GRS was considered null. No data imputation was performed.

GRADING OF IMAGES

The grading of images was based on the eye with the most severe phenotype. Graders were ophthalmologists by CORC (Coimbra Ophthalmology Reading Centre, AIBILI, Portugal.

The Rotterdam Classification was used: presence of AMD was considered 2a (soft, indistinct or reticular

drusen) and 2b (soft, distinct drusen with pigmentary irregularities), 3 (soft, indistinct drusen with pigmentary irregularities) and 4 (geographic atrophy or choroidal neovascularization) and no AMD were participants graded as 0 (no AMD or drusen <63 μm), and stages 1a and 1b (soft, distinct drusen or pigmentary irregularities).[18,19]

DEFINITION OF PROGRESSORS AND NON-PROGRESSORS

Participants with AMD in the Epidemiological Study of the Prevalence of Age-Related Macular Degeneration in Portugal (NCT01298674) were excluded from this analysis, because they were not considered at risk for progression. Only participants graded with no AMD at this baseline study and who presented AMD at the 6.5-year visit were considered for analysis. This is to say that progressors were defined as not having AMD at baseline (stages 0 or 1) and having AMD at the follow-up visit (stages 2,3 or 4). Non-progressors were defined as participants who did not have AMD (stages 0 or 1) at both visits.

STATISTICAL ANALYSIS

Data normality was tested using the Shapiro-Wilk test and visually verified with histogram. Continuous variables were presented as mean ± standard deviation and categorical variables were presented as frequency (N), and percentage (%).

Differences in variables of interest between groups (progressors and non-progressors and participants taking and not taking lipid lowering drugs) were analysed with Mann-Whitney U-test for continuous variables not following a normal distribution and a Pearson's chi-squared test (or Fisher's exact test) for categorical variables.

To investigate whether statins intake is associated with AMD progression, we used a time to event (or survival analysis), which takes into account the effect of censored observations and is widely used in ophthalmic research. An extended Cox regression model was tested, with statins intake as a time dependent covariate. The model was adjusted for potential confounding known to influence AMD, including age, gender, body mass index (BMI), smoking status, diabetes and arterial hypertension assessed at the baseline and tested for the cox proportional hazard assumption, for influential data and for nonlinearity in the relationship between the log hazard and the covariates. The correlation between the two eyes for the same patient was taken into account using a robust (sandwich) standard error.

ETHICAL CONSIDERATIONS

Coimbra Eye Study complied with the tenets of the Declaration of Helsinki, the International Conference on Harmonization - Good Clinical Practice Guidelines. The studies obtained AIBILI's Ethics Committee approval. Patients signed the informed consent with study procedures and possible consequences explanation. Participant's rights to physical and mental integrity as well as to privacy and protection of their data were safeguarded in accordance with the Regulation (EU) 2016/679 of the European Parliament and of the Council of 27 April 2016 – General Data Protection Regulation (GDPR) and applicable national law.

RESULTS

Six hundred eighty-three subjects were eligible for the analysis, with a mean age of 65.6 ± 6.8 years at baseline and 55.2 % were females. The mean follow-up time was 6.3 ± 0.6 years and 170 subjects started taking statins between the baseline and the follow-up visit.

Baseline results between progressors and non-progressors are presented in Table 1. A total of 561 participants did not progress between the two studies and 122 participants did. This corresponds to 1321 eyes analysed: 168 progressors and 1153 not progressors. Age (p<0.001), smoking (p=0.034), BMI (p=0.003), years of follow up (p=0.018), oral antidiabetic medication (p=0.021) and use of statins (p=0.009) were significantly associated with AMD progression. In the group of non-progressors, the most used statin was atorvastatin (50.3%), followed by simvastatin (28.5%). In the group of progressors, these statins were as well the most prescribed, corresponding to 36.8% each.

Within the groups of progressors and non-progressors, we further analysed differences between participants taking lipid-lowering drugs and those not taking lipid-lowering drugs. Results are shown in Table 1.

The association of the use of statins and AMD progression is represented in Table 2. When adjusted for age, gender, BMI, smoking, diabetes and arterial hypertension at baseline, statins reduced the risk for AMD progression in 50% (HR=0.509, 95%CI 0.302-0.860, p=0.012), using an extended Cox regression analysis, with use of statins as exposure (binary variable) and time dependent.

We then separated statins by strength (Table 3). Low strength statins were not significantly associated with AMD progression (p=0.797). However, medium- and high-strength statins maintained the association with AMD progression, presenting the same risk reduction of about 50% (HR=0.482, 95% CI 0.285-0.814, p=0.006), adjusting for the same variables.

Finally, we included the GRS in the model, in order to assess how the genetic risk for AMD could interfere with the protection of statins, maintaining the model adjusted for age, gender, BMI, smoking, diabetes and arterial hypertension (Table 4). The use of statins was significantly associated with progression of AMD, with a reduction in the risk for progression of about 50% (HR=0.516, 95% CI 0.293, 0.91, p= 0.022). For each increasing unit in the GRS, the risk for progression increased 1.29 (HR=1.29, 95% CI 1.068-1.559, p= 0.008).

DISCUSSION

Age-related macular degeneration is a multifactorial disease. Both genetics and environment play a role in its aetiology, meaning that different risk factors contribute for its pathophysiology. This translates into different disease

Table 1. Baseline characteristics and use of concomitant medication during the period between the baseline and the follow-up visit.

	Non-progressor (N=561)			Progressor (N=122)			*p*-value
	No lipid-lowering drugs (N=388)	On lipid-lowering drugs (N=173)	*p*-value	No lipid-lowering drugs (N=100)	On lipid-lowering drugs (N=22)	*p*-value	
Gender, n (%)			0.5			0.7	0.9[a]
Female	210 (54.1%)	99 (57.2%)		55 (55.0%)	13 (59.1%)		
Male	178 (45.9%)	74 (42.8%)		45 (45.0%)	9 (40.9%)		
Age at baseline, Mean (SD)	65.3 (6.9)	64.2 (5.6)	0.2	68 (7.6)	69.7 (6.6)	0.3	**<0.001**[b]
Smoking, n (%)			0.6			> 0.9	**0.034**[a]
Non-smoker	363 (93.6%)	164 (94.8%)		88 (88.0%)	20 (90.9%)		
Smoker/Ex-smoker	25 (6.4%)	9 (5.2%)		12 (12.0%)	2 (9.1%)		
Body Mass Index, Mean (SD)	27.7 (3.9)	28.4 (3.6)	**0.033**	26.9 (3.4)	27.1 (3.4)	0.9	**0.003**[b]
Diabetes, n (%)	48 (12.4%)	19 (11.0%)	0.6	9 (9.0%)	5 (22.7%)	0.13	0.9[a]
Years of follow-up, Mean (SD)	6.3 (0.6)	6.5 (0.5)	0.091	6.2 (0.8)	6.3 (0.7)	0.6	**0.018**[b]
On insulin, n (%)	9 (2.3%)	5 (2.9%)	0.8	2 (2.0%)	1 (4.5%)	0.5	>0.9[a]
On oral antidiabetic medication, n (%)	55 (14.2%)	43 (24.9%)	**0.002**	7 (7.0%)	4 (18.2%)	0.11	**0.021**[a]
On antihypertensive medication, n (%)	154 (39.7%)	95 (54.9%)	**<0.001**	42 (42.0%)	13 (59.1%)	0.14	0.9[a]
Exposure to statins, n (%)	151 (26.9%)			19 (15.6%)			**0.009**[a]
Exposure to statins by drug, n (%)							0.7[a]
	Non-progressor (N=561)			**Progressor (N=122)**			***p*-value**
atorvastatin	76 (50.3%)			7 (36.8%)			
fluvastatin	1 (0.7%)			0 (0.0%)			
pitavastatin	7 (4.6%)			1 (5.3%)			
pravastatin	9 (6.0%)			2 (10.5%)			
rosuvastatin	15 (9.9%)			2 (10.5%)			
simvastatin	43 (28.5%)			7 (36.8%)			
Exposure to Statins by strength, n (%)							>0.9[a]
Low	10 (6.6%)			1 (5.3%)			
Medium/High	141 (93.4%)			18 (94.7%)			

[a] Pearson's Chi-squared test, Fisher's exact test
[b] Mann-Whitney U-test

Table 2. Extended Cox regression analysis, with use of statins as exposure (binary variable), time-dependent.

Variables	Coef. (,β.)	td. Err. (,*se*.(,β.))	,*HR*.(,*e*-,β..)	95% CI	*p*-value (Wald test)
Use of statins					
Yes	-0.675	0.224	0.509	[0.302, 0.860]	**0.012**
Age at baseline	0.062	0.011	1.064	[1.036, 1.094]	**<0.001**
Gender					
Male	-0.108	0.171	0.898	[0.598, 1.347]	0.602
Body Mass Index at baseline	-0.042	0.022	0.959	[0.917, 1.004]	0.073
Smoking at baseline					
Smokers	0.588	0.253	1.801	[1.073, 3.023]	0.026
Diabetes at baseline					
Yes	0.051	0.247	1.052	[0.593, 1.866]	0.863
Arterial hypertension at baseline					
Yes	0.321	0.162	1.378	[0.932, 2.040]	0.108

Coef = coefficient; CI = confident interval; HR = hazard ratio; Std.Err. = standard error.

pathways, like the complement system, oxidative stress, lipid metabolism, extracellular matrix remodeling and angiogenesis.[1] Of these, at the therapeutic level, only two are targeted for AMD treatment: the angiogenesis pathway for neovascular AMD and, very recently, the complement system for geographic atrophy. Also, antioxidants have shown

Table 3. Extended Cox regression analysis, with exposure to statins by strength, time-dependent.

Variables	Coef. (,β.)	td. Err. (,*se.*(,β.))	,*HR.*(,*e*-,β..)	95% CI	*p*-value (Wald test)
Statins exposure by strength, Low	0.278	0.718	1.321	[0.159, 10.995]	0.797
Statins exposure by strength, Medium/High	-0.73	0.232	0.482	[0.285, 0.814]	**0.006**
Age at baseline	0.063	0.011	1.065	[1.036, 1.095]	**<0.001**
Gender					
Male	-0.102	0.171	0.903	[0.603, 1.353]	0.621
Body Mass Index at baseline	-0.042	0.022	0.959	[0.916, 1.004]	0.072
Smoking at baseline					
Smokers	0.573	0.253	1.774	[1.052, 2.991]	0.032
Diabetes at baseline					
Yes	0.059	0.247	1.061	[0.598, 1.884]	0.839
Arterial hypertension at baseline					
Yes	0.322	0.162	1.38	[0.933, 2.043]	0.107

Coef = coefficient; CI = confident interval; HR = hazard ratio; Std.Err. = standard error.

Table 4. Extended Cox regression analysis, with interaction between use of statins (binary variable) and Genetic Risk Score, time-dependent.

Variables	Coef. (,β.)	td. Err. (,*se.*(,β.))	,*HR.*(,*e*-,β..)	95% CI	*p*-value (Wald test)
Use of statins					
Yes	-0.662	0.235	0.516	[0.293, 0.91]	**0.022**
Genetic Risk Score	0.255	0.074	1.29	[1.068, 1.559]	**0.008**
Age at baseline	0.056	0.012	1.058	[1.028, 1.089]	**<0.001**
Gender					
Male	-0.095	0.18	0.91	[0.591, 1.4]	0.667
Body Mass Index at baseline	-0.039	0.023	0.961	[0.916, 1.009]	0.111
Smoking at baseline					
Smokers	0.479	0.274	1.614	[0.905, 2.879]	0.105
Diabetes at baseline					
Yes	-0.208	0.275	0.812	[0.42, 1.572]	0.537
Arterial hypertension at baseline					
Yes	0.277	0.168	1.32	[0.877, 1.986]	0.184

Coef = coefficient; CI = confident interval; HR = hazard ratio; Std.Err. = standard error.

benefits only in the intermediate and late stages of AMD. Hence, the lipid metabolism pathway remains untargeted, but biologically plausible.

Statins are drugs that have long been considered for the treatment of AMD. Their approved therapeutic indication is lowering cholesterol levels through the inhibition of hydroxymethylglutaryl-coenzyme A (HMG-CoA) reductase. Nonetheless, statins present pleiotropic effects, probably since the inhibition of HMG-CoA reductase inhibits the synthesis of mevalonate, a precursor of not only endogenous cholesterol, but also of other compounds.[20]

Some studies have tried to assess the probable benefit of the use of statins in AMD. Results have been inconclusive so far, as some studies report protection against AMD or AMD progression and others do not, probably due to heterogenicity in duration, design and controlled variables. The only clinical trial up to now, with simvastatin 40 mg,[21] published a 2-fold decrease in the risk of AMD progression in the statin group of patients, but a later analysis reported that protection was shown only in patients with intermediate bilateral AMD. To solve this problem, meta-analyses have tried to answer this question and so far statins are not considered as protective.[22–25]

Our results contradict this conclusion, as they show, in a survival analysis, that statins are protective against AMD progression. Our model is corrected for age, gender, BMI, smoking, diabetes and arterial hypertension. Age and smoking are risk factors well associated with the disease,[26]

so their confounding effects could not be ignored. We also adjusted the model for gender and BMI. Despite not being acknowledged as completely established risk factors, some studies state that female gender and obesity may be considered risk factors for AMD.[27–29] Additionally, we corrected the model for diabetes and arterial hypertension, which are co-morbidities that need to be considered in order to assess the role of statins, since they frequently course along with hypercholesteremia. Also, AMD and cardiovascular disease share common risk factors.[30,31]

When separated by strength, low-dose statins lost the statistical association, suggesting that the potency of these drugs may be associated with the protection against AMD progression. To the best of our knowledge, there are no studies that analyse low- and high-strength statins, so this is a point to build upon in terms of research. When GRS was included in the model, statins remained associated with AMD progression, reducing the risk in about 50%. This is probably the most important result, as GRS is calculated quite comprehensively, by weighting the SNPs that are associated with AMD, the number of alleles and their effect size. Still, the risk presented significantly reduced in those participants taking statins. We must highlight that the intake of statins was only considered in between the visits of the two studies, so no confounding effect could have arisen from previous intake time. Additionally, our model is time dependent, meaning that the duration of statin treatment was considered in assessing risk reduction. The results are quite interesting, in our point of view, since we must acknowledge that our sample of participants who progressed and who took statins within visits is rather small (n=19). Also, we did not adjust the model for lipid profile because we had scarce information on this. Most of patients did not provide this information, so it was not possible. Should it have been, this would be important information, as hypercholesterolemia has been associated with AMD[30,32] and a low-fat diet is also an important protective factor for AMD.[33,34] The evidence of AMD and lipid association relies on this evidence, along with the fact that drusen are largely composed of lipids. Hence, knowing baseline cholesterol levels would help in the understanding of the mechanism against AMD protection.

In conclusion, our results suggest that statins may be protective against AMD progression, reducing the risk to half, and that this may be a topic worthy of revisiting through a well-designed clinical trial with a large sample to assess the intervention causality.

PREVIOUS PRESENTATIONS

This work was presented at EURETINA 2023 (oral presentation).

CONTRIBUTORSHIP STATEMENT / DECLARAÇÃO DE CONTRIBUIÇÃO

MLC: Responsible for creating the manuscript.

RC: Responsible for statistical analysis.

JBM, JCV, RS, PB: Contributed with their expertise to the manuscript conclusion.

RS: Principal Investigator of the CES.

All authors approved the final version to be published.

MLC: Responsável pela elaboração do manuscrito.

RC: Responsável pela análise estatística.

JBM, JCV, RS, PB: Contribuíram com a sua experiência para a conclusão do manuscrito.

RS: Investigador Principal do CES.

Todos os autores aprovaram a versão final a ser publicada.

RESPONSABILIDADES ÉTICAS

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