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Direct-to-Consumer Personal Genome Testing for Age-related Macular Degeneration

Running title: Commercial prediction of AMD

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ABSTRACT

Purpose: Genetic testing may be the next step in clinical medicine for a more personalized approach in determining risk of disease. Direct-to-consumer (DTC) personal genome tests may fulfill this role. We explored the practicability and predictive value of DTC-tests from four companies (23andMe, deCODEme, Easy DNA, Genetic testing laboratories) for age-related macular degeneration (AMD).

Methods: Body specimens of three individuals were collected and sent to four companies for DNA genotyping and disease risk estimation. In addition, DNA was also genotyped using Illumina HumanOmniExpress 12v1 array in the Rotterdam Study laboratory, and risk estimates of AMD were calculated using the validated prediction model from the population-based Three Continent AMD Consortium.

Results: Genotyped results of the four DTC-tests matched genotyping performed by the Rotterdam Study laboratory. The estimated risks provided by the companies varied considerably in the tested individuals, from a 1.6-fold difference for overall relative risk to an up to 12-fold difference for lifetime risk. The lifetime risks for the individuals ranged from 1.4-16.1% in the DTC-tests, while they varied from 0.5-4.2% in the validated prediction model. Most important reasons for the differences in risks were the testing of only a limited set of genetic markers, the choice of the reference population, and the methodology applied for risk calculation.

Conclusion: Direct-to-consumer personal genome tests are not suitable for clinical application as yet. More comprehensive genetic testing and inclusion of environmental risk factors may improve risk prediction of AMD.

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INTRODUCTION

Genetic studies of age-related macular degeneration (AMD) have elucidated a major proportion of its genetic background. Currently, genome-wide studies (GWAS) have identified associations with >30 genetic loci for this disease, explaining a large part of the heritability of AMD[1, 2]. Subsequently, these genomic findings have been incorporated into prediction models, many of which provide a >80% discriminative accuracy for late AMD[3-22]. This high predictive ability makes AMD particularly suitable for genetic testing, which may be the next step to a more personalized approach in clinical medicine.

Direct-to-consumer (DTC) personal genome tests had been made available for consumers and thousands have purchased these tests via the internet to determine a personal disease risk. Recently, methods of three DTC-tests have been examined and compared for several diseases[23]. AMD was the disease for which each test obtained the best predictive ability. Several companies offered genetic tests for AMD and implementation of these tests in the clinic could help identify individuals at risk of developing the disease to apply risk dependent patient care and surveillance strategies. Therefore, the accuracy of the risk estimates will be a great concern, and will determine whether such tests will be meaningful in the clinic.

In this study, we evaluated the results of AMD prediction tests provided by four major companies. We sent bio-samples from three individuals to these companies to test proof of principle, and reviewed the sampling process, the type of analysis, the genotyping, and the risk information. In addition, we compared results to a validated prediction model based on population studies.

METHODS

Experimental design

Evaluation of test methodology

Study participants

Three investigators (GB, JV, CK) agreed to voluntarily participate in the study, and signed informed consent.

DTC-tests for AMD

We searched for internet-based DTC-tests for AMD using a web search engine and the word groups "genetic testing for age-related macular degeneration", "genetic prediction of age-related macular degeneration", and "genetic tests for age-related macular degeneration". Only companies available for European citizens and testing more than one single nucleotide polymorphism (SNP) were eligible, and of these, four companies were selected; i.e., 23andMe, deCODEme, Easy-DNA, The Genetic Testing Laboratories, Inc.

23andMe

https://www.23andme.com/

This privately-held American company was founded in 2006 with the intention to empower individuals in accessing their own genetic information and to stimulate a way into more personalized medicine. One can order a single 'spit' kit for $99 (shipping costs $14.95 - $118.95) from the website on internet, and a sample collection kit will be sent by mail with instructions how to provide a saliva sample and details for returning the sample. An assisted collection kit for persons having trouble to spit can be ordered together with the DTC-kit for an additional $25, requiring only half the amount of saliva. The returned saliva sample will arrive at the contracted LabCorp's Clinical Laboratory Improvement Amendments (CLIA) certified laboratory, where DNA will be isolated from cells in the saliva and processed on an Illumina® HumanOmniExpress array customized by 23andMe (>1 million SNPs, call rate above 98%). These SNPs

provide information about traits, carrier status, and risks for over hundred diseases, including AMD. The risk for developing AMD is estimated based on the risk in the reference population and an overall relative risk (RR) representing risks of five SNPs: *CFH* rs1061147; *C2* rs547154; *LOC387715/ARMS2* rs3750847; *C3* rs2230199; *TIMP3* rs9621532 [11, 24-36]. AMD risk in the reference population differed for males and females and was 6.5 and 7% respectively. Methods of risk calculation have been described in a white paper[37], accessible after login to the 23andMe website. No health reports including risk prediction and carrier status are currently provided for new customers.

DeCODE

http://www.decodeme.com/

DeCODE was founded in 1996 and the headquarters are located in Reykjavik, Iceland. This company developed the deCODEme test, which provide results for 47 conditions and traits. Unfortunately, new tests are no longer offered by the company. Costs were $1100 per test, with no extra costs for shipping. After purchasing the test from the internet, a buccal swab kit will was sent in the mail with instructions how to collect and return the sample. The samples were processed at a CLIA certified lab, the deCODE laboratory in Reykjavik, for DNA isolation. Genotyping was performed on an Illumina Human 1M Beadchip which determines >1 million SNPs. Validation occurred by bi-directional Sanger sequencing and independent SNP genotyping platforms.

A overall RR for developing AMD was calculated based on six risk variants: *ARMS2/HTRA1* rs3750847, *C2/FB* rs9332739 and rs547154, *C3* rs230199 and, *CFH* rs1061147 and rs1329428 [27, 38]. Subsequently, for the tested individual a lifetime risk was calculated based on the overall relative risk and the AMD risk in the reference population, which was set at 8%. A white paper[39] describing the risk calculation is available after login to the deCODEme website.

Easy-DNA

http://www.easy-dna.com / http://www.easydna.co.uk / http://www.easydna.eu

Easy-DNA is an international company which provides a genetic DNA predisposition test on 25 conditions and diseases. This test can be purchased from the internet for €299/$299/£299 including shipping costs. A kit will be sent by mail for collection of a blood sample, and includes submission forms, instructions for collecting the blood sample from a punctured finger, the sample collection kit and a self-addressed envelope. This company does not provide information on the genotyping method, but states that results are provided for *CFH* rs1061170 and *C2* rs800292 [40, 41]. Risk estimates are presented as lifetime and overall RR of AMD. Risk of AMD in the reference population was set at 8%. Methods for risk calculation was not provided by the company.

The Genetic Testing Laboratories, Inc (GTL)

http://www.gtldna.com/predisposition.html

This company provides a DNA predisposition test which will reveal the genetic and environmental predisposition for 25 diseases and conditions including AMD. The DNA predisposition test costs $285 with additional costs of $45 for shipping outside the Contiguous United States. After purchasing the kit from the internet, it will be sent to your own physician or a professional collector agency appointed by GTL to collect the sample, which can be a bucal or a blood sample. The sample will be processed by a CLIA accredited laboratory. As for Easy-DNA, this company also is unclear on genotyping method, but states that results are provided for *CFH* rs1061170 and *C2* rs800292 [40, 41]. Lifetime and overall RR are provided for each tested person. Risk of AMD in the reference population was set at 8%.The risk calculation method of this company was not available for consumers or professionals.

We followed each company's instructions for the collection of bio-samples used for DNA extraction. We sent the samples to the various laboratories associated with the companies, and awaited the results.

Genotyping in Rotterdam

Genotyping for the three individuals was also performed at the Rotterdam Study Laboratory: Genetic Laboratory of Internal Medicine at the Erasmus Medical Center in Rotterdam, the Netherlands. Genomic DNA was extracted from peripheral leukocytes and all participants were genotyped using the Illumina HumanOmniExpress 12v1_J microarray. Call rate for the genotyping was >97.5%.We imputed genotype data to Hapmap 3 release 2 and 1000 genomes phase I V3.

Assessment of covariates

The covariates age, length, weight, smoking status, and family history regarding AMD were obtained by interview. Body mass index (BMI) was calculated by dividing weight (kg) by the height squared (m^2). AMD phenotype was evaluated by standard ophthalmologic examination including fundus photography (Topcon TRC-50EX fundus camera, Topcon Optical Co, Tokyo, Japan and Sony DXC-950P digital camera, Sony Corporation, Tokyo, Japan) after pharmacological mydriasis. Images were graded according to the Wisconsin Age-Related Maculopathy Grading[2] and the modified international classification system[42] by graders from the Rotterdam Study.

Risk score Three Continent AMD Consortium prediction model and DTC-tests

The Three Continent AMD Consortium (3CC) developed a validated prediction model including a total risk score based on 31 variables; 26 genetic variants associated with AMD, age, sex, smoking, BMI, and AMD phenotype. The prediction model had 87% discriminative accuracy for incident late AMD [22]. For each individual in this study this summary risk score was calculated. Based on the risk score, lifetime risks could be assessed for each individual.

Ancestry assessment

Ancestry of the three individuals was determined using multi-dimensional scaling (MDS) protocol from ENIGMA [43] using Hapmap 3 release 2 as the reference.

Statistical analysis

Test results included predicted risks for several diseases from four companies. For the purpose of this study, we only evaluated the predicted risks for AMD. 23andMe provided odds ratios (OR) and the other companies relative risks (RR) per SNP per genotype, but all were adjusted for the average risk of the SNP in the population, and will be referred to as OR and RR, respectively. Genotype frequency, risks per genotype, overall RR, lifetime population risk and lifetime risk of the tested individual were obtained from the test results.

Minor allele frequencies were not provided by the companies, but calculated using the formula:

$$p+q = 1$$

With p representing the major allele and q the minor allele. For the different genotypes, frequencies could be calculated after applying this information; homozygous for major alleles = p^2, heterozygous = $2pq$ and homozygous for minor alleles = q^2.

All analyses were performed using SPSS version 20.0 (SPSS INC, Chicago, Illinois) except for the MDS-analysis which was performed using R software.

RESULTS

Demographic characteristics of the three study subjects are provided in **table 1**. All three were younger than the average age of AMD onset, and none had any features of AMD, as determined by grading of fundus photographs. One had a history of smoking, and one had a positive family history for late AMD. All three were Caucasian and had northern/western European ancestry (**Supplementary figure 1**).

DTC-tests

Details of the DTC-tests are given in **table 2**. Tests differed considerably in price, the most costly being 11x more expensive than the cheapest test. Sampling methods varied from saliva, buccal swap to blood from a finger prick. One participant particularly had difficulty to deliver the saliva specimen of 2.5 ml for 23andMe, which required ~1 hour of sampling time. Genetic Testing Laboratories (GTL) required for all participants and Easy-DNA only for US-residents a physician or another health professional assigned by the company to collect the blood sample and only the collectors obtained the test results. However, the forms for requesting the test from GTL were open access. Delivery time for test results ranged from 2-4 weeks for most tests; results from one Easy-DNA test were delayed up to 8 weeks without notice or explanation.

In contrast to the statement of Easy-DNA and GTL, the SNP rs800292 is located in the *CFH* gene, not in *C2* (**table 3**). Thus, these two companies only tested risk variants in *CFH.* DeCODEme and 23andMe covered 4 and 5 AMD loci, respectively. The tested SNPs varied among tests, however, there was considerable overlap. Individual genotypes at these SNP locations are shown in **table 3**. Risk-increasing as well as risk-decreasing variants were present in all three individuals. The effect estimates of these variants showed the largest range in individual 2, in particular for the risks predicted by 23andMe and deCODEme. The lifetime AMD population risk used by the companies varied from 6.5-8%, and varied for gender in the 23andMe calculations. For 23andMe and deCODEme the ancestry of the reference populations was European, for GTL and Easy-DNA this was European Tuscan. Only for individual 1 the Easy-DNA test listed European ancestry as the reference population. Genotypes identified by the DTC-tests were identical to those determined at the Rotterdam Study laboratory in all three individuals.

The inter-test variability of the overall relative and life-time risks was large in all three individuals , but most profoundly in individual 3 (**table 3**). For this person, these risks were lower and higher than the population risk, depending on the test. Lifetime risks between lowest and highest estimate differed by factor 1.7, 1.6, and 11.5 for individuals 1, 2, and 3, respectively.

Risk prediction based on Three Continent AMD Consortium

The prediction model developed by the population-based Three Continent AMD Consortium (3CC) consists of 31 variables which were represented in a total risk score indicating the risk of developing late AMD[22]. For each individual the total risk score was calculated (**table 4**) and used to assess lifetime risks. Lifetime population risk for developing late AMD was 17.4% at life expectancy of 90 years in the 3CC cohort. Lifetime risks for all three individuals were also calculated using the 3CC risk score, and were 4.2%, 0.5%, and 0.5% respectively (**table 4**). Although the population risk in the 3CC cohort was much higher than for the DTC-tests, lifetime risks for the three individuals were considerably lower than the lifetime risks provided by the companies (4.9-8.6; 4.0-6.5; 1.4-16.1, **table 3**).

DISCUSSION

Until recently, anyone could order a DTC-test and get a personal risk estimate for common diseases. Interpretation of the test results and evaluation of their validity has been difficult, even for professionals. Our study shows that predicted risks of AMD vary considerably among DTC-tests, and none may represent the true disease risk.

We examined four DTC-tests in three individuals, and compared test results to predicted risks from a validated model developed in the large population-based Three Continent AMD Consortium (3CC)[22]. Predicted risks varied widely within each individual, and differences between highest and lowest estimates for lifetime risk were up to 12-fold. Within the same person, overall relative risks could be increased as well as decreased, depending on which test was used. All tests provided higher estimates for lifetime risk than the 3CC model. Several key points explain these differences.

First, the DTC-tests genotyped only 2-6 SNPs to calculate the risk of AMD. These risks were often based on case-control studies instead of population-based studies which often comprise lower risks[22]. Recent reports show that >30 loci have been associated by GWAS studies[1, 2]. Not testing a comprehensive set of SNPs may lead to imbalance of harmful and protective SNPs, and provide a very different overall risk estimate. For example, individual 2 had several important risk-increasing as well as risk-decreasing variants (**table 4**), and not testing these hampered accurate risk profiling (**table 3**). This was also acknowledged for the population at large; inclusion of an extended set of variants increased risk prediction in three population-based studies.[22]. We expect that even more common and rare variants will be identified for AMD in the near future, and inclusion of these variants will further refine personalized risk prediction.

Second, the lifetime population risk and reference population differed among the DTC-tests. The lifetime population risk used by 23andMe was lower than that used by the other companies, and differed for men and women. Which population had been used as reference for the calculation of the lifetime AMD population risk was not specified by any of the companies. They were all lower than the lifetime population risk estimate in 3CC (6.5-8% versus 17.4%, respectively). Lifetime population risks were

based on life expectancy of 79 years for 23andMe and 90 years for 3CC. No information was provided on life expectancy by the other companies. The average life expectancy is currently above 80 years in western Europe and 79 years in the United States [44]. Life expectancy increases once a certain age has been reached: for instance, persons who reached the age of 80 years during 2008-2010 in France still had an average life expectancy of 8.3 years for men and 10.6 years for women [45]. In these persons, a life expectancy of 90 years is not unrealistic. Ancestry also influences the risk estimates. All companies asked the applicant for their ethnicity and used questionnaire data for analysis. However, calculation of ancestry is more accurate using multi-dimensional scaling (MDS) analysis with genotype data. In GTL and Easy DNA, all results were based on European Tuscan ancestry, although European ethnicity was stated by the individuals at application. MDS analysis with genotype data from all three individuals confirmed their northern/western European ancestry comparable with their appearance (**supplementary figure 1**). Why a Tuscan ancestry was chosen for these individuals is unclear and incorrect. The choice of two different ancestries (European Tuscan and European) in one individual (**table 3**) in these tests is presumably an unintended error.

The conversion to a different ancestry can lead to an alteration of the risk, since the frequency of genotypes may differ among ethnicities. The minor allele frequency (MAF) for the *CFH* rs1061170 variant in the Easy-DNA and GTL tests was set at 17% for those with Tuscan ancestry. MAF for this variant varies among ethnicities: ~36% in Europeans and Africans, ~17% in Latinos/Hispanics and ~10-15% in Asians[46]. Tuscans cluster more closely with northern/western Europeans than with Latinos/Hispanics (**supplementary figure 1**), and literature indicates that the actual MAF of the *CFH* rs1061170 variant in an Italian population is also 36%[47]. Therefore, these companies should have used a MAF of 36% rather than 17% for European Tuscans. Not using the correct MAF resulted in higher risks since all risks per SNP have been adjusted for the average risk of the SNP in the population, which can be calculated using the risk per genotype and genotype frequency. This effect is particularly visible in the risks for individual 1 (**table 3**); risks provided by Easy-DNA used the European ancestry as reference population and a MAF of 36% resulting in an RR of 1.26, while GTL used the European Tuscan ancestry with a MAF of 17% resulting in a higher RR of 1.60. For carriers of the *CFH* rs1061170 CC-genotype this difference in risk will be even more extreme. In summary, an incorrect reference population was assigned to the three individuals and to this reference population (Tuscans) an incorrect MAF for the *CFH* rs1061170 SNP was

assigned. In this particular case the largest effect on risk prediction of AMD was the incorrect assigned MAF. This most likely influenced the risk prediction for the other diseases predicted by the companies as well.

Third, there were mistakes in assignment of an AMD risk variant. Easy-DNA and GTL stated that the tested SNP rs800292 was located in the *C2* gene, when in fact this particular rs-number is located in the *CFH* gene[48]. Apart from the incorrect gene, the direction of the risk for this variant was opposite of that reported in 3CC[22]; in the tests from Easy-DNA and GTL the T allele was set as the risk variant, increasing the risk of AMD, while in 3CC this allele decreased the risk of AMD.

Fourth, the DTC-tests lacked inclusion of non-genetic risk factors. Only 23andMe took age and gender into account in their risk calculation. Age is the most important non-genetic factor associated with AMD known to date, and it is therefore prudent to incorporate this factor in risk predictions of AMD [49]. None of the companies included environmental factors in their risk prediction. We recommend inclusion of smoking since this factor is an important environmental risk factor for AMD[50], which also shows interaction with genetic risk variants[40]. Inclusion of non-genetic risk factors can improve the predictive ability of the test[22].

Lastly, the companies applied different methods for their risk calculation. A recent study examined and compared the methods from three DTC-tests (23andMe, deCODEme and Navigenics) for several diseases including AMD[23]. The authors showed that the formulas used by deCODEme can lead to a predicted risk exceeding 100% in high risk cases. The formulas used by 23andMe followed the Bayes' theorem preventing risks to exceed 100%, leading to more realistic risk estimates. Unfortunately, methods for risk calculation were not provided by Easy-DNA or GTL, and could therefore not be evaluated.

Recently, many companies stopped offering DTC-tests. Several issues played a role. First, the Food and Drug Administration (FDA) questioned the evidence of the safety and efficacy of these prediction tests[51]. Second, it was unclear what actions the individual will take when made aware of his/her genetic profile. Third, health care professionals lacked guidelines for counselling and patient management after genetic

profiling. Do these issues apply to DTC-tests for AMD? Our study encountered no genotyping errors. Nevertheless, predictions were inaccurate based on methodology. It is indeed unclear what an individual should do when diagnosed with a high genetic risk of AMD, and what a clinician should advise such patients. Cessation of smoking and lowering BMI is advice which applies to all persons. However, it is likely that individuals who have been made aware of a high genetic risk after testing will be more motivated to make drastic life style changes than persons who are ignorant.

Although genetic testing for prediction of disease risk is the next step to personalized medicine, the current state of the art is that most DTC-tests are accurate at genotyping, but not at risk prediction. Improvement can be achieved by incorporation of a more comprehensive set of genetic markers with population-based risks. Inclusion of non-genetic risk factors, a more adequate choice of the reference population, and implementation of valid methodology for risk calculation will further improve these tests. Only then will these genetic tests become suitable for clinical practice.

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Authors' contribution:

Concept and design: AH, CCWK, CMvD, JRV **Acquisition of data:** CCWK, GHSB ,JRV **Analysis and interpretation**: GHSB, CCWK, NA **Drafting article:** GHSB, CCWK **Critically revising article**: AH, CMvD, NA, JRV **Funding:** AH, GHSB, CCWK, JRV

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Table 1: Discriptives of the participants

Variable	*Individual 1*	*Individual 2*	*Individual 3*
Age (yrs)	45	29	51
Sexe	Female	Female	Male
Ethnicity	Caucasian	Caucasian	Caucasian
Ancestry	Northern/western European	Northern/western European	Northern/western European
BMI (kg/m²)	22.7	20.2	24.3
Smoking	never	never	past
AMD phenotype	none	none	none
Family history of AMD	grandmother	none	none

Abbreviations: AMD = age-related macular degeneration,
BMI = body mass index, yrs = years

Table 2; Overview genetic testing companies

Company name	Website	Costs per kit	DNA source	Easy to collect?	Additional notes
23andMe	https://www.23andme.com	$99 / € 74	saliva	Difficult in one participant	Streetaddress is needed to deliver DTC-test
deCODEme*	https://www.decodeme.com	$1100 / € 821	bucal	yes	-
Easy-DNA	http://www.easygenetictest.com	$299 / € 299	blood	yes	For US residents: Sample needs to be collected by physiscian or professional collector
The Genetic Testing Laboratories	http://www.gtldna.com/	$285 / € 213	blood	yes	Sample needs to be collected by physiscian or professional collector

* deCODEme do not offer any new testing possibilities

Table 3; Risks of the tested variants, overall risk and lifetime risk per company for each individual

AMD		*Individual 1*								*Individual 2*								*Individual 3*							
		23andMe		**deCODEme**		**Easy-DNA***		**GTL**		**23andMe**		**deCODEme**		**Easy-DNA**		**GTL**		**23andMe**		**deCODEme**		**Easy-DNA**		**GTL**	
Gene	**SNP number**	genotype	OR	genotype	RR	genotype	RR	genotype	RR	genotype	OR	genotype	RR	genotype	RR	genotype	RR	genotype	OR	genotype	RR	genotype	RR	genotype	RR
CFH	rs1061147	AC	0.97	AC	1.56[‖]					CC	0.34	CC	0.21[‖]					AC	0.97	AC	1.56[‖]				
CFH	rs1329428			GG								AA								GG					
CFH	rs1061170					CT	1.26	CT	1.60					TT	0.64	TT	0.64					CT	1.60	CT	1.60
CFH[†]	rs800292					CC	0.67	CC	0.63					CT	1.26	CT	1.26					CT	1.26	CT	1.26
C2	rs547154	GG	1.07	CC	1.10					GG	1.07	CC	1.10					GT	0.57	AC	0.58				
C2	rs9332739			GG	1.06							GG	1.06							GG	1.06				
LOC387715/ARMS2	rs3750847	CC	0.47	GG	0.46					CT	1.63	AG	1.59					CC	0.47	GG	0.46				
C3	rs2230199	CG	1.37	CG	1.29					CG	1.37	CG	1.29					GG	0.79	CC	0.76				
TIMP3	rs9621532	AA	1.02							AA	1.02							AA	1.02						

	23andMe	**deCODEme**	**Easy-DNA***	**GTL**	**23andMe**	**deCODEme**	**Easy-DNA**	**GTL**	**23andMe**	**deCODEme**	**Easy-DNA**	**GTL**
Overall RR[‡]	0.70	1.01	0.85	1.00	0.70	0.50	0.81	0.81	0.22	0.34	2.01	2.01
Lifetime population risk (%)	7.0	8.0	8.0	8.0	7.0	8.0	8.0	8.0	6.5	8.0	8.0	8.0
Lifetime risk[§] (%)	4.9	8.6	6.8	8.1	5.9	4.0	6.5	6.5	1.4	2.7	16.1	16.1

abbreviations: AMD = age-related macular degeneration, GTL = The Genetic Testing Laboratories, OR = Odds Ratio, RR = Relative Risk

* Reference population for individual 1 was set to European and differed from individual 2 and 3 for the Easy-DNA test which was set to European (Tuscans)

† Easy-DNA and GTL referred to this SNP as though it was located within the *C2* Gene

‡ The overall RR provided by each company is based on all the tested genetic variants.

§ The lifetime risk is calculated multiplying the overall RR with the population risk

‖ RR based on haplotype rs1061147 and rs1329428 in the *CFH* gene

Table 4; Risk estimates from the Three Continent AMD consortium prediction model

Variable	**Code**	**Risk per code**	*Individual 1*	*Individual 2*	*Individual 3*
ARMS2 rs10490924	GG=0 / GT=1 / TT=2	0 / 0.779 / 1.720	0	0.779	0
ADAMTS9 rs6795735	CC=0 / TC=1 / TT=2	0 / 0.130 / 0.424	0	0.424	0.424
SLC16A8 rs8135665	CC=0 / TC=1 / TT=2	0 / 0.313 / 0.648	0.313	0	0.313
Sexe	M=0 / F=1	0 / 0.320	0.320	0.320	0
CETP rs3764261	CC=0 / CA=1 / AA=2	0 / 0.215 / 0.478	0.215	0	0
CFH rs1061170	TT=0 / TC=1 / CC=2	0 / 0.175 / 0.278	0.175	0	0.175
Smoking	Never=0 / Past=1 / Current=2	0 / 0.164 / 0.651	0	0	0.164
MYRIP rs2679798	AA=0 / AG=1 / GG=2	0 / 0.059 / 0.156	0.059	0.156	0
VEGFA rs943080	CC=0 / TC=1 / TT=2	0 / 0 / 0.098	0	0	0.098
TNFRSF10A rs13278062	TT=0 / TG=1 / GG=2	0 / 0.093 / 0.196	0.093	0	0
TGBR1 rs334353	TT=0 / TG=1 / GG=2	0 / 0.039 / -0.336	0.039	0.039	0
IER3/DDR1 rs3130783	AA=0 / AG=1 / GG=2	0 / 0.029 / 0.166	0	0.029	0.029
SKIV2L rs429608	GG=0 / GA=1 / AA=2	0 / 0.027 / 0.590	0	0	0.027
Age (yrs)	=<65=0 / 65-75=1 / 75+=2	0 / 1.558 / 2.433	0	0	0
AMD baseline grade	Level 10=0 / Level 20=1 / Level 30=2 / Level 40=3	0 / 1.458 / 2.560 / 3.398	0	0	0
BMI (kg/m^2)	=<25=0 / 25+=1	0 / 0.007	0	0	0
C2/CFB rs4151667	TT=0 / TA or AA=1	0 / -1.245	0	0	0
B3GALTL rs9542236	TT=0 / TC=1 / CC=2	0 / -0.231 / -0.169	0	0	0
LIPC rs12912415	AA=0 / AG or GG=1	0 / -0.098	0	0	0
COL8A1 rs13081855	GG=0 / GT=1 / TT=2	0 / 0.223 / 0.890	0	0	0
TIMP3 rs5749482	GG=0 / GC or CC=1	0 / -0.357	0	0	0
C3 rs2230199	CC=0 / GC=1 / GG=2	0 / -0.033 / 0.755	-0.033	-0.033	0
ABCA1 rs1883025	CC=0 / TC=1 / TT=2	0 / -0.046 / 0.076	-0.046	-0.046	0
LPL rs256	CC=0 / TC or TT=1	0 / -0.048	0	-0.048	-0.048
CFI rs10033900	CC=0 / TC=1 / TT=2	0 / -0.070 / -0.223	0	-0.070	-0.070
C3 rs433594	GG=0 / GA=1 / AA=2	0 / -0.110 / -0.591	-0.110	-0.110	0
FRK/COL10A1 rs3812111	TT=0 / TA=1 / AA=2	0 / -0.278 / -0.118	0	0	-0.118
RAD51B rs8017304	AA=0 / AG=1 / GG=2	0 / -0.414 / -0.138	0	0	-0.414
C2/CFB rs641153	GG=0 / GA or AA=1	0 / -0.592	0	0	-0.592
CFH rs800292	GG=0 / GA=1 / AA=2	0 / -0.899 / -1.614	0	-0.899	-0.899
CFH rs12144939	GG=0 / GT=1 / TT=2	0 / -0.947 / -1.195	0	-0.947	0
Total risk score			**1.025**	**-0.406**	**-0.911**
Lifetime risk (%)			**4.2**	**0.5**	**0.5**

Abbreviations: AMD = age-related macular degeneration; BMI = body mass index; F = female; M= male; yrs = years

LEGEND

Supplementary figure 1. Ancestry of tested individuals

Abbreviations: CEU = Utah residents with ancestry from northern and western Europe; CHB = Han Chinese in Beijing, China; YRI = Yoruba in Ibadan, Nigeria; TSI = Tuscans in Italy; JPT = Japanese in Tokyo, Japan; CHD = Chinese in Metropolitan Denver, Colorado; MEX = persons with Mexican ancestry in Los Angeles, California; GIH = Gujarati Indians in Houston, Texas; ASW = persons with African ancestry in Southwest USA; LWK = Luhya in Webuye, Kenya; MKK = Maasai in Kinyawa, Kenya.

Genetic markers from the three tested individuals were compared with those from 11 populations. Each square represents a persons and every person was assigned two dimensions in de MDS analysis based on their genome and plotted according to these two dimensions; on the x-axis dimension 1 and on the y-axis dimension 2. Every population has their unique color: CEU = light blue, CHB = turquoise, YRI = yellow, TSI = green, JPT = purple, CHD = orange, MEX = grey, GIH = black, ASW = olive-green, LWKK = magenta, MKK = blue. The tested individuals are visible as red squares and cluster together with persons in the CEU sample

Supplementary figure 1