European Journal of Epidemiology

Ophthalmic epidemiology in Europe: the “European Eye Epidemiology” (E3) consortium

Cécile Delcourt1,2, Jean-François Korobelnik1,2,3, Gabriëlle H.S Buitendijk4, Paul J Foster5, Christopher J Hammond6, Stefano Piermarocchi7, Tunde Peto5, Nomdo Jansonius8, Alireza Mirshahi9, Ruth E Hogg10, Lionel Bretillon11-13,  Fotis Topouzis14, Gabor Deak15, Jakob Grauslund16, Rebecca Broe16, Eric H Souied17, Catherine Creuzot-Garcher11-13,18, José Sahel19-24, Vincent Daien25-27, Terho Lehtimäki28,29, Hans-Werner Hense30, Elena Prokofyeva31-33, Konrad Oexle34, Jugnoo S Rahi5,35,36, Phillippa M Cumberland35,36, Steffen Schmitz-Valckenberg37, Sascha Fauser38, Geir Bertelsen39,40, Carel Hoyng41, Arthur Bergen42, Rufino Silva43-45, Sebastian Wolf46, Andrew Lotery47, Usha Chakravarthy48, Astrid Fletcher49, Caroline C.W. Klaver4, on behalf of the European Eye Epidemiology(E3) consortium.

1: Univ. Bordeaux, F-33000 Bordeaux,  France

2: INSERM, ISPED, Centre INSERM U897-Epidemiologie-Biostatistique, F-33000 Bordeaux, France

3: CHU de Bordeaux, Service d’Ophtalmologie, Bordeaux, F-33000, France

4: Dept Ophthalmology, Dept Epidemiology, Erasmus Medical Center, Rotterdam, the Netherlands

5: NIHR Biomedical Research Centre at Moorfields Eye Hospital and UCL Institute of Ophthalmology, London, UK, EC1V 2PD

6: Departments of Ophthalmology and Twin Research & Genetic Epidemiology, King’s College London, St Thomas’ Hospital, London, UK SE1 7EH

7: University of Padua, Dept. of Ophthalmology, Padua, Italy

8: NIHR Biomedical Research Centre at Moorfields Eye Hospital NHS Foundation Trust and UCL Institute of Ophthalmology

9: Dept. Ophthalmology, University Medical Center, Mainz, Germany

10: Centre for Experimental Medicine, Queen’s University Belfast, Grosvenor Road, Belfast, Northern Ireland

11: INRA, UMR1324 Centre des Sciences du Goût et de l’Alimentation, F-21000 Dijon, France

12: CNRS, UMR6265 Centre des Sciences du Goût et de l’Alimentation, F-21000 Dijon, France

13: Université de Bourgogne, Centre des Sciences du Goût et de l’Alimentation, F-21000 Dijon, France

14: Laboratory of Research and Clinical Applications in Ophthalmology, Department of Ophthalmology, Aristotle University of Thessaloniki, AHEPA Hospital, Thessaloniki, Greece

15: Vienna Reading Center, Department of Ophthalmology, Medical University of Vienna, Vienna, Austria

16: Institute of Clinical Research, University of Southern Denmark, and Department of Ophthalmology, Odense University Hospital, Odense, DK-5000, Denmark

17: Service d’ophtalmologie, Centre Hopsitalier Intercommunal de Creteil, CRC, CRB, Universite Paris Est, Creteil, France

18: CHU, Department of Ophthalmology, F-21000 Dijon, France

19: Institut de la Vision, UPMC Univ Paris 06, UMR\_S 968, Paris, F-75012, France;
20: INSERM, U968, Paris, F-75012, France;
21: CNRS, UMR\_7210, Paris, F-75012, France;
22: Centre Hospitalier National d’Ophtalmologie des Quinze-Vingts, INSERM-DHOS CIC 1423, Paris, F-75012, France;
23: Fondation Ophtalmologique Rothschild, Paris, F-75019 Paris, France;
24: Institute of Ophthalmology-University College London, London EC1V 9EL, United Kingdom

25: Inserm, U1061, Montpellier, F-34093 France

26: Univ Montpellier 1, Montpellier, F-34000 France

27: Department of Ophthalmology, Gui De Chauliac Hospital, Montpellier, F-34000, France

28: Department of Clinical Chemistry, Fimlab Laboratories, Tampere, Finland.

29: University of Tampere School of Medicine, Tampere, Finland.

30: Clinical Epidemiology, Institute of Epidemiology and Social Medicine, University Münster, D 48129 Münster, Germany.

31: Inserm U1018, Centre for Research in Epidemiology& Population Health (CESP), Epidemiology of occupational and social determinants of health, Villejuif, France

32: Université de Versailles Saint-Quentin, Villejuif, France

33: Northern State Medical University, Arkhangelsk, Russia

34: Institute of Human Genetics, Technische Universität München, 81675 Munich, Germany

35: Population, Policy and Practice Programme, UCL Institute of Child Health, London, UK WC1N 1EH

36: Ulverscroft Vision Research Group, UCL Institute of Child Health, London, UK WC1N 1EH

37: GRADE Reading Center, Department of Ophthalmology, University of Bonn, 53127 Bonn, Germany

38: University Hospital Cologne, Center of Ophthalmology, 50924 Cologne, Germany

39: Department of Ophthalmology, University Hospital of North Norway, Tromsø, Norway.

40: Department of Community Medicine, Faculty of Health Sciences, University of Tromsø, Tromsø, Norway.

41: Radboud University Medical Center, Department of Ophthalmology, Nijmegen, The Netherlands

42: Academic Medical Centre, Amsterdam, The Netherlands

43: Department of Ophthalmology, Centro Hospitalar e Universitário de Coimbra. Portugal (CHUC)

44: Faculty of Medicine, University of Coimbra. Portugal. (FMUC)

45: Association for Innovation and Biomedical Research on Light and Image, Coimbra, Portugal (AIBILI)

46: Department of Ophthalmology, Inselspital, University Hospital, University of Bern, Bern, Switzerland.

47: Clinical and Experimental Sciences, Faculty of Medicine, University of Southampton, England

48: Usha: Center for Experimental Medicine, Queen's University of Belfast, United Kingdom

49: Faculty of Epidemiology & Population Health, London School of Hygiene & Tropical Medicine, London, UK

Corresponding author : Cécile Delcourt, Inserm U897, Isped, Université de Bordeaux, 146 rue Léo Saignat, 33076 Bordeaux Cedex. Email: cecile.delcourt@isped.fr; tel: +33 5 57 57 11 91; fax: +33 5 57 57 14 86

**Electronic supplementary material**

**1958 British Birth cohort**

The 1958 British Birth is a prospective population-based cohort study that initially included 17,000 newborn children born during a single week in March 1958. Diverse and detailed biological, social, and lifestyle data have been collected in follow up surveys carried out periodically over time. [[1](#_ENREF_1)] Specifically, ophthalmic data have been collected at 7, 11, 16, and 44 years. During 2002 and 2003, an ophthalmic assessment was undertaken as part of a broad biomedical survey. It provided data on presenting and best visual acuity (3 metre Keeler crowded logMAR test), binocular near vision acuity (Keeler reduced Snellen reading test) and stereoacuity (Lang II stereo card). Additionally, around one quarter of the 9377 individuals still actively participating in the study were selected randomly and underwent non-cycloplegic autorefraction with an autorefractor (Retinomax 2, Nikon, Tokyo, Japan). Refractive data were available for 2267 subjects.  Participants were all of similar age at the time of the ophthalmic examination (44-45 years). Investigations on refractive error and visual function have been reported. [[2-4](#_ENREF_2)]The 1958 British Birth Cohort biomedical survey was funded by the Medical Research Council (grant G0000934, Health of the Public initiative, principal grant holders C Power and D Strachan). It was approved by the Institute of Child Health’s Research Ethics Committee and the South East Multi Centre Research Ethics Committee (ref: 01/1/44) and the Oversight Committee for the biomedical examination of the British 1958 British birth cohort. All subjects gave individual informed consent to participation.

**Alienor**

The Alienor (Antioxydants, Lipides Essentiels, Nutrition et maladies OculaiRes) Study is a population-based prospective study aiming at assessing the associations of age-related eye diseases (age-related macular degeneration (AMD), glaucoma, cataract, dry eye syndrome) with nutritional factors (in particular antioxidants, macular pigment and fatty acids), determined from plasma measurements and estimation of dietary intakes. It also takes into account other major determinants of eye diseases, including gene polymorphisms, environmental factors and vascular factors. The methods of this study have been published elsewhere.[[5](#_ENREF_5)]

Subjects of the Alienor Study were recruited from an ongoing population-based study on the vascular risk factors for dementia, the Three-City (3C) Study.[[6](#_ENREF_6)] The 3C Study included 9,294 subjects aged 65 years or more from three French Cities (Bordeaux, Dijon and Montpellier), among whom 2,104 were recruited in Bordeaux. They were initially recruited in 1999-2001 and followed-up about every two years since. The Alienor Study consists of eye examinations, which are proposed to all participants of the 3C cohort in Bordeaux since the third follow-up (2006-2008). Among the 1,450 participants re-examined between October 2006 and May 2008, 963 (66.4%) participated in the Alienor Study’s baseline eye examination. This research followed the tenets of the Declaration of Helsinki. Participants gave written consent for participation in the study. The design of this study has been approved by the Ethical Committee of Bordeaux (Comité de Protection des Personnes Sud-Ouest et Outre-Mer III) in May 2006.

Eye examinations included, for each eye, a recording of ophthalmological history, measures of visual acuity (ETDRS charts, Light House Low Vision, New York, NY) and refraction (Speedy K, Luneau, France), two 45° non mydriatic color retinal photographs (one centered on the macula, the other centred on the optic disc) (TRC NW6S, Topcon, Japan), measures of intraocular pressure (non contact tonometer (KT 800, Kowa, Japan) and central corneal thickness (Pachpen, Accutome Inc., Malvern Pa, USA) and break-up time test. In addition, from 2009, examinations with spectral-domain optical coherence tomography (Spectralis, Heidelberg Engineering, Heidelberg, Germany) and ultrawide field imaging (Optos Panoramic 200C, Optos plc, United Kingdom) were performed. AMD, other retinal diseases and glaucoma were classified using international classifications.

**AMRO-NL**

The AMRO-NL study is a hospital based study, consisting of 375 unrelated individuals with AMD, 200 unrelated individual with Primary open angle glaucoma (POAG) and 200 age-matched healthy control individuals without AMD and/or POAG. All subjects are Caucasian and recruited from the Netherlands Institute of Neuroscience (NIN) Amsterdam, The AMC Amsterdam, and Erasmus University Medical Centre Rotterdam, by newsletters, via patient organizations, and nursing home visits. Controls were at least 65 years old, and were usually unaffected spouses or non-related acquaintances of cases or individuals who attended the ophthalmology department for reasons other than retinal pathology. Clinical diagnosis including fundus grading (AMD) and IOP pressure as well as optic nerve head measurements (POAG) as well as clinical investigators (Prof dr de Jong, Prof dr Klaver) were exactly the same as in The Rotterdam study.

**Belfast case-control study of AMD**

This study was an add on extension study to the European Eye Study. This enrolled 205 participants through a community based sampling approach. Participants were aged over 65 years and gave a blood sample and underwent an ophthalmic and clinical examination and fundus photography. An additional 212 participants were recruited from the Belfast Macular clinic if they were aged over 50 and had a diagnosis of AMD on clinical examination.

**CARMA**

This study was conducted at two sites (Waterford Republic of Ireland and Belfast, Northern Ireland). A total of 403 participants were enrolled. To be eligible participants were required to have one eye with neovascular macular degeneration with a fellow eye free of late stage macular degeneration or if both eyes were free of late AMD at least one eye with 20 or more drusen. Visual acuity in at least one study eye was required to be 73 or more ETDRS letters. Participants were randomized to receive an oral daily antioxidant supplement or placebo. Participants were seen 6 monthly with clinical and ophthalmic examinations, fundus photography and blood sampling at every visit. AMD severity was graded based on the WARMSG definitions and participants were assigned to 6 mutually exclusive categories based on the Rotterdam staging system.

**CIC XV-XX**

The 794 enrolled AMD patients and controls are part of a larger cohort study initiated in our Centre since 2006 aiming at genotype phenotype correlation study and identification of predictive markers. This study was approved by CPP Ile de France V (Project n° 06693, EUDRACT 2006-A00347-44). Inclusion criteria of the AMD patients were women or men aged 55 or older with AMD changes in at least one eye. Exclusion criteria: other retinal disease (e.g. diabetic retinopathy, high myopia, or macular dystrophies). The subjects were included in the study after informed consent signature and underwent a complete ophthalmologic examination including Visual acuity measurement (ETDRS), fundus examination, Color Retinographies, SD-OCT (HRA, Spectralis, and Fundus AutoFluorescence imaging. A questionnaire about family history and personal medical history was completed. Genomic DNA was extracted from 10 mL blood leukocytes using the Illustra® kit according to the manufacturer protocol (GE Healthcare).

The patients with early AMD were followed twice a year and patients with advanced AMD and controls, yearly.

**Coimbra Eye Study**

 This was a cross-sectional, population-based study, including two Portuguese populations aged ≥ 55 years. A total population of 6,023 adults was recruited from two Portuguese primary health-care units in the central region of Portugal – one from a coastal town (n=3000) and another from an inland town (n=3023). Between August 2009 and April 2011, subjects were recruited from the primary health-care center of the coastal town (Mira) and between April 2012 and October 2013 from the health-care unit of the inland town (Lousã). Report 1 of the Coimbra Eye Study provided the first population-based data on prevalence of AMD in a Portuguese population (Mira) [[7](#_ENREF_7)]. In our second report we included additional data on prevalence and risk factors for AMD in Portugal, comparing two geographic different populations.

All participants underwent complete bilateral ophthalmological examination and two 35° non-simultaneous stereoscopic color fundus photographs were taken from fields 1M (centered on the optic disc), 2 (centered on the macula) and 3M (temporal to the macula), using a digital mydriatic Topocon® fundus camera (TRC-50EX; Topcon Corporation, Tokyo, Japan). Images were analyzed in a step-wise manner by a centralized reading centre (Coimbra Ophthalmology Reading Centre, CORC - AIBILI): the general analysis, aiming to identify major retinal pathology and a differential analysis for AMD lesions.

The main outcome measures consisted of age and gender adjusted prevalence of early and late of AMD. We also evaluated the potential risk factors that could explain AMD prevalence in our study population using logistic regression analysis. The International Classification and Grading System (ICGS) for ARM and AMD was the chosen classification [[8](#_ENREF_8)] and the signs of disease were stratified into 5 exclusive stages (ARM, stage 0 to 4) using the Rotterdam staging system [[9](#_ENREF_9), [10](#_ENREF_10)]. This AMD grading was supported by a software to visualize digital colour fundus images and to grade retinal lesions – Retmarker AMD Research (Critical Health, SA, Portugal). [[11](#_ENREF_11)]

Socio-demographic and past medical history data were collected in the study visit by interview and included: date of birth; gender; relatives with history of AMD; smoking habits and alcohol consumption; diagnosis of hypertension, diabetes and cardiovascular diseases. Height and weight were obtained using standardized techniques and equipment, to enable the calculation of body mass index (Kg/m2). Demographic and clinical characterstics were summarized using descriptive methods. Categorical variables were reported by frequencies and percentages and numerical variables with mean and SD. T-tests and chi-squares were used to assess differences in age, gender, race, familiar history of AMD, smoking, body mass index, hypertension, diabetes and age of menopause between the two populations. ANOVA, t-tests and chi-squares were used to evaluate the independence of early, late and no AMD, according to the relevant clinical and demographic covariates. Age and gender-specific prevalences were calculated for early and late stages of AMD. Multinomial logistic regression analyses were performed to assess the univariate association of AMD with age, gender, smoking, body mass index, hypertension, diabetes, age of menopause, by estimating the odds ratios (OR), followed by multivariate analysis for association of AMD with all significant risk factors.

**Coimbra Diabetic Retinopathy Study**

The DR screening program here described is conducted under the authority of Regional Health Administration in the Central Region of Portugal (ARSC), who is responsible of the coordination and logistics of the screening program involving the health units, family physicians, photographers/screeners and reading center. It covers the selection of patients and their call for screening, the implementation of the screening program itself, and the return of the results back to the health units, physicians and patients, ensuring as well the orientation of patients after the screening. Each geographic area has a portable non-mydriatic camera and a photographer/screener, and the screening site changes location to cover all geographic areas. The amount of time the camera stays at one health center depends on the number of patients for that location. The number of locations is such that the camera completes a “rotation” in 12 months, i.e., the camera should visit each health center every 12 months.

The diabetic patients, type 1 and 2,except those that had already received treatment for sight-threatening DR complications or unable to collaborate sufficiently with the screening procedures, are selected by their primary care unit and physician and summoned for a DR screening appointment within or near their residence area.

In the screening appointment the following procedures take place: (1) visual acuity testing, using a Snellen scale, measured with no correction or using the patient usual correction; (2) small questionnaire, including the visual acuity record, date of birth, gender, year of diagnosis of DM, and if on insulin treatment and since when; and (3) acquisition of two 45 degree non-mydriatic digital fundus images per eye / per patient, being the first image taken centered at the macula (field 2), and a second image centered on the nasal border of the optic disc (field 1) and then repeated the same procedure for the other eye. The photographers are requested to make a general quality assessment of the images and repeat them to achieve the best image possible, providing, however, a comment/reason for the cases with considerable low quality and with no improvement after repetition. These comments are customized as “myosis”, “cataract”, “corneal problems” and “bad patient collaboration”, and are included in the patient questionnaire data.

The program has been continuously running since 2001. However, in July 2011, the central reading center, Coimbra Ophthalmology Reading Center (CORC), introduced the Retmarker Screening technology (herein referred as RetmarkerSR; Retmarker SA, Coimbra, Portugal), reducing human grader burden, with the advantage of creating an auditable database of screening episodes [[12](#_ENREF_12)]. The RetmarkerSR solution is an automated software system capable of identifying signs of microvascular DR pathology. RetmarkerSR is a patented and class IIa CE marked medical device solution that analyses all image sets received and separates those with no signs of DR pathology or no evolution of DR compared to previous screening visits, from those with signs of DR pathology/evolution, requiring human grading. The analysis/grading is performed per eye, to evaluate the presence of referable DR (maculopathy and/or proliferative DR) in two different main steps: automated analysis - 1st assessment; human grading - 2nd assessment.

This second assessment is performed by trained and experienced non-ophthalmological graders, supervised by senior graders (ophthalmologists). Only image sets identified as having signs of DR pathology in the 1st assessment, are sent to human grading for classification of DR level.

Patients/eyes classified as R0 (without retinopathy) or RL (existence of microaneurysms, hemorrhages and/or exudates outside the central 1DD macular region) are not referred for an ophthalmological appointment and are scheduled to come back for screening one year later. Referable patients are those classified as non-proliferative DR with maculopathy (M) or proliferative DR (RP). Patients classified as M are referred as soon as possible, preferably within three months, for an ophthalmological appointment in the central hospital of their residence area. Patients classified as RP are considered urgent and should be examined within one month period in the ophthalmological department of the central hospital of their residence area.

**Creteil Study**

A total of 1080 French AMD patients and 406 controls were recruited in 4 French retinal Centres, at the department of Ophthalmology of Creteil in collaboration with CHU de Bordeaux, the Quinze-Vingts Hospital and the Centre of Imaging and Laser of Paris, between November 2005 and July 2007. Written informed consent was obtained, as required by the French bioethical legislation and local ethic committee (CCPPRB Henri Mondor), in agreement with the Declaration of Helsinki for research involving human subjects.

 Inclusion criteria of the AMD patients were (1) women or men aged 55 or older, and (2) with exudative AMD in at least one eye, (3) no association with other retinal disease (e.g. diabetic retinopathy, high myopia, or macular dystrophies). Patients underwent a complete ophthalmologic examination including best corrected visual acuity measurement, fundus examination, and retinal photographs. Fluorescein angiography (Topcon 50IA camera, Tokyo, Japan)- and if needed indocyanine green angiography (HRA, Heidelberg, Germany)- and Optical Coherence Tomography (Carl Zeiss Meditec, Inc) were performed. During the first visit AMD phenotypes in both eyes were analysed independently by each investigator (EHS and NL) prior to genetic testing according to color photographs and FA at presentation. When investigators disagreed on a particular clinical feature this patient was excluded from further analysis. A questionnaire about medical history was completed.

Controls were also recruited in the group of patients operated of cataract in our four centres. A total of 406 French women or men over 55 years with a normal fundus examination and a normal aspect of fundus photography were also recruited at the department of Ophthalmology of Creteil between 2002 and 2008. Information about their medical history including smoking was obtained.

Genomic DNA was extracted from 10 mL blood leukocytes using the Illustra® kit according to the manufacturer protocol (GE Healthcare). *CFH* Y402H, *ARMS2* rs10490924, HTRA1 rs11200638, rs2230199:C>G (C3:R102G) and T280M of *CX3CR1* SNPs were genotyped by quantitative PCR allelic discrimination using reagents and conditions from Custom Taqman SNP Genotyping Assays (Applera Corp., France), using ABI 7900HT (Applied Biosystems). 5% of the population was genotyped for quality control. 100% of these duplicates are concordant. The call rate of the result was controlled by Hardy Weinberg Test. All of the SNPs are in Hardy Weinberg equilibrium.

**Danish Cohort of Pediatric Diabetes 1987**

Every pediatric department in Denmark contributed with participants for a nationwide study on glycemic control in pediatric diabetes in 1987. It was estimated that 75% of all Danish children and adolescents below the age of 19 participated. This cohort was re-studied in 1995 where the main focus was microvascular complications. Yet another follow-up study was done in 2011. For E3 purposes we have included cross-sectional data from all 324 participants at the 1995-study and follow-up data from 2011 on 185 participants.

From both examinations bilateral retinal photographs are available on all participants, all graded for retinopathy with the ETDRS scale, and for the 2011 follow-up OCT-scans, BCVA, and history of laser treatment and vitrectomy. Furthermore data on retinal vessel analyzes are available from both examinations.

Extra-ocular data available from both examinations are age, sex, debut and duration of diabetes, BMI, BP, smoking history and relevant blood- and urine markers.

**Early Observational Markers Study**

This was a 3 centre European study undertaken in Belfast, Northern Ireland, Coimbra, Portugal and Milan, Italy. Each site enrolled 35 participants with neovascular macular degeneration in one eye. The fellow eye that was free of wet AMD was the designated study eye. Participants were seen 6 monthly with deep functional and morphological testing. Function included best corrected distance visual acuity, near acuity and reading speed, low luminance acuity and microperimetry. High resolution raster tomographic scans were acquired on the macula along with stereoscopic colour fundus photography, autofluorescence, red free and infra red reflectance images. Standardized fluorescein and indocyanine green angiography was also performed at every visit. The images have been graded for early AMD severity and for presence of geographic atrophy and for onset of neovascularization.

**Epic-Norfolk**

The European Prospective Investigation of Cancer (EPIC) is a 10 nation collaborative study which commenced in 1989. EPIC Norfolk enrolled 30,445men and women resident in the East Anglia region of England aged 40-79 years between 1993 – 97. The initial aim was to study dietary determinants of cancer, but the study was extended to identify other determinants of chronic disease. Visual health was included in the third health check (3HC) in which 8,623 people aged 48 – 92 years were examined. The cohort is 99.7% white. Ocular data include ocular history, measures of logMAR visual acuity, auto-refraction and keratometry (Humphrey 500), intraocular pressure and corneal biomechanics (Reichert ORA), axial biometry (Zeiss IOLMaster), retinal nerve fibre layer thickness using a Zeiss GDx-VCC, optic nerve head topography (Heidelberg HRT II), and fundus photography using a Topcon TRC-NW5S + Nikon D80 non-mydriatic camera. Images have been graded for AMD, diabetic retinopathy and glaucoma. Visual field testing (Zeiss HFA2 750i running 24-2 SITA threshold programme) was attempted in all those with features suggesting an increased risk of glaucoma (IOP > 24mmHg, abnormal GDx or HRT) and in 10% of those regarded as “normal”. Participants with abnormal features (VA > 0.34, IOP > 24mmHg, abnormal HRT or GDx, retinal abnormalities identified on photographs, visual field abnormalities, N = 1,703) were referred for examination by a senior ophthalmologist at the Norfolk and Norwich University Hospital. Additional measures include height, weight, blood pressure, hip and waist measures, cognitive function and banked plasma, serum and urine. Genotyping on Affymetrix UK Biobank 820K SNP array has recently been completed on 3HC participants. Extensive questionnaire data have been collected using HLEQ and EPAQ2, and generic EPIC questionnaires recording socio-demographics & economic status, medication, smoking and alcohol, leisure activities and falls. Detailed 5 day diet diaries have been completed. Further information from Paul Foster (p.foster@ucl.ac.uk).

**ERF**

The Erasmus Rucphen Family (ERF) study is a large, family-based study in a genetically isolated population located in the southwest of The Netherlands. This population was founded in the middle of the 18th century by fewer than 400 individuals and was isolated until the last few decades. An extensive genealogic database, including >80 000 individuals is available for this population. Genetic characterization of this population has been presented elsewhere. [[13](#_ENREF_13), [14](#_ENREF_14)] For the ERF study, 22 families were selected who had at least 6 children baptized in the community church between 1880 and 1900. All living descendants of the selected families and their spouses (18+ yrs) were invited to participate in the study (N=2755). The pedigree members were not selected on disease status. Data were collected between June 2002 and February 2005. Nonophthalmic examinations included anthropometric measurements, cardiovascular and endocrine assessments, neuropsychological tests, fasting blood samples, and interviews regarding medical history, medication, and putative risk factors. The ophthalmic examination comprised the assessment of best-corrected visual acuity, refraction, and intraocular pressure. Keratometry was determined by an automatic refractometer, and the eyes’ axial lengths by an intraocular lens calculator (IOL Master; Carl Zeiss Meditec, Inc., Dublin, CA). Scanning laser polarimetry was subsequently performed with the commercially available GDx VCC (Carl Zeiss Meditec, Inc.). In mydriasis, participants underwent fundus photography centered on the optic disc and macula (20° and 35°, TRC-50XT retinal camera; Topcon Medical Systems, Inc., Paramus, NJ) and confocal scanning laser ophthalmoscopy measurements (Heidelberg Retina Tomograph II; [HRT II] Heidelberg Engineering GmbH, Dossenheim, Germany). A total of 2940 subjects underwent ophthalmic examination. The Medical Ethics Committee of the Erasmus Medical Centre Rotterdam approved the study and informed consent was obtained from all participants.

**EUGENDA**

The EUGENDA (European Genetics Database) is a case-control study that investigates genetic and non-genetic factors in age-related macular degeneration (AMD).[[15](#_ENREF_15)]  The database includes DNA, plasma, serum samples, and data from a questionnaire with information on medical history, nutrition, and life-style.

Subjects have been recruited in Nijmegen (Netherlands) and Cologne (Germany). Currently, there are around 5000 participants in the database with about half of them controls.  The AMD grading is based on color fundus photography, SD-OCT, and fluoroscein angiography. The study was approved by the ethics committees in Cologne and Nijmegen.

**Eureye**

The EUREYE Study is an epidemiological study funded by the European Commission Vth Framework (QLK6-CT-1999-02094). Additional funding was provided by the Macular Disease Society of the UK, Thomas Pocklington Trust and the UK Medical Research Council

The main objectives of the EUREYE study are to (i) describe the prevalence of early and late age-related macular degeneration(AMD) in men and women aged 65 and over in the European setting (ii) to investigate the association of solar radiation with AMD (iii) to investigate the role of dietary factors especially antioxidants. The EUREYE study was specifically designed to exploit the diversity of European populations in their exposures to these potential factors. The participating centres in the EUREYE study span a latitude of 22°, from 60° north to 38° south, a nearly 3-fold gradient of UVR.

The EUREYE study is a multi-centre study in seven European countries following a common protocol. Participants (n=4753) were recruited from random sampling of the population aged over 65 years in the centres: Bergen (Norway), Tallinn (Estonia), Belfast (UK), Paris-Creteil (France), Verona (Italy), Thessaloniki (Greece), Alicante (Spain). Participants were interviewed by fieldworkers, underwent an ophthalmological examination including fundus photography and gave a blood sample for measurement of antioxidants and banking of DNA. Information collected at interview included education, smoking and alcohol use, brief medical history, lifetime history of residence, outdoor exposure and ocular protection, and a Food Frequency Questionnaire. AMD was assessed using digital Topcon fundus cameras (Topcon TRC-50EX, Topcon Corporation, Japan) and the settings were calibrated and standardised for all seven centres. The fundus images were graded at a independent reading centre (Erasmus University Rotterdam) using the International Classification System for Age Related Maculopathy.

**Fyns Diabetes Database**

Fyns Diabetes Database is a continuously updated database with diabetes-related data from patients in Funen County, Denmark. Data were collected from general physicians, diabetologists, and ophthalmologists. For E3-purposes, we have included cross-sectional data from 22,089 unique patients examined in 2003-2015.

Eye data from the database included bilateral levels of retinopathy and maculopathy, visual acuity, and history of laser treatment and vitrectomy. Extra-ocular data included age, gender, type and duration of diabetes, body mass index, smoking, blood pressure, and relevant blood markers (HbA1c, lipids, and renal clearance).

Mydriatic colour fundus photos (2-6 fields) were captured by the local ophthalmologist or at specialized hospital-based screening centers using different fundus cameras. Level of retinopathy was based on the international clinical diabetic retinopathy and diabetic macular edema disease severity scales as proposed by Wilkinson et al [[16](#_ENREF_16)].

**Generation R**

The Generation R Study is a population-based prospective birth-cohort study from fetal life until adulthood. The study is designed to identify early environmental and genetic causes leading to normal and abnormal growth, development and health from fetal life, childhood and young adulthood.[[17](#_ENREF_17)] Women who were living in the city of Rotterdam and pregnant during 2001-2005 were included in the study. In total, 9,778 pregnant women were enrolled. Data collection in children and their parents include questionnaires, interviews, detailed physical and ultrasound examinations, behavioural observations, Magnetic Resonance Imaging and biological samples. Biological samples were ascertained including blood, hair, faeces, nasal swabs, saliva and urine samples, and extensive data on DNA, RNA and microbiome is available. Children underwent a first physical examination at a research center at age 5; the ophthalmic examination included visual acuity, ocular biometry, fundus photographs and ophthalmologic history; those who had a history of ophthalmic care underwent a full ophthalmologic exam at a second step including cycloplegic refraction and orthoptic and ophthalmologic screening to diagnose strabismus and other ocular conditions. At 9 years of age children underwent a second physical examination; all children were examined for cycloplegic refraction, and OCT and MRI of the orbit was carried out.

**Guernsey AMD case cohort study and Southampton AMD/glaucoma case-control Study**

All participants were white, aged older than 55 years, and ascertained through the Southampton Eye Unit (UK) or research clinics undertaken (by AL) in Guernsey (UK) [[18](#_ENREF_18)]. Control patients were either spouses or partners of patients with disease or those who presented at eye clinics for an unrelated eye disease. An experienced retinal specialist examined all participants. Controls underwent a dilated retinal examination to exclude any clinical signs of age-related macular degeneration. Cases and controls were classified as having or not having disease on the basis of the AREDS classification system. Recruitment was approved by the Southampton and Southwest Hants local research ethics committee and followed the tenets of the Declaration of Helsinki. All participants provided informed written consent and underwent a detailed ophthalmic examination to confirm both positive and negative diagnoses.

**Gutenberg Health Study**

The GHS is an ongoing, prospective, interdisciplinary, single-center, population-based cohort study in the Rhine-Main Region in midwestern Germany with a total of 15,010 participants and follow-up after five years. The study sample is recruited from subjects aged between 35 and 74 years at the time of the exam. The sample was drawn randomly from local governmental registry offices and stratified by gender, residence (urban and rural) and decade of age. Exclusion criteria were insufficient knowledge of the German language to understand explanations and instructions, and physical or psychic inability to participate in the examinations in the study center. The main goals of the ophthalmological section are to assess the prevalence and incidence of ocular diseases and to explore risk factors, genetic determinants and associations with systemic diseases and conditions. The eye examination at baseline included a medical history, self-reported eye diseases, visual acuity, refractive errors, intraocular pressure, visual field, pachymetry, keratometry, fundus photography and tear sampling. The 5-year follow-up visit additionally encompassed optical coherence tomography, anterior segment imaging and optical biometry. The general examination included anthropometry; blood pressure measurement; carotid artery ultrasound; electrocardiogram; echocardiography; spirometry; cognitive tests; questionnaires; assessment of mental conditions; and DNA, RNA, blood and urine sampling. The GHS is the most extensive dataset of ophthalmic diseases and conditions and their risk factors in Germany and one of the largest cohorts worldwide.

**KORA**

KORA ("Kooperative Gesundheitsforschung in der Region Augsburg" which translates as “Cooperative Health Research in the Region of Augsburg”) is a population based study of adults randomly selected from 430,000 inhabitants living in Augsburg and 16 surrounding counties in Germany. The collection was done in 4 separate groups from 1984-2001 (S1-S4). All survey participants are residents of German nationality identified through the registration office. In the KORA S3 and S4 studies 4,856 and 4,261 subjects have been examined implying response rates of 75% and 67%, respectively. 3,006 subjects participated in a 10-year follow-up examination of S3 in 2004/05 (KORA F3), and 3080 of S4 in 2006/2008 (KORA F4). The age range of the participants was 25 to 74 years at recruitment. The study was approved by the local ethics committee. Written informed consent was obtained from all participants before enrollment in accordance with the Declaration of Helsinki.

Genome-wide genotyping using the Illumina 2.5M chip or the Illumina Omni Express chip was performed on a subset of individuals from the S3/F3. Samples with low call rate (<98%), sex-mismatch, exhibited excess heterozygosity rates or evidence for non caucasian anchestry were excluded. SNPs were excluded before imputation if they had a low a genotype call rate (<0.98), low minor allele frequency (<0.01) or Hardy-Weinberg P-value < 10−6. Phasing and imputation was performed with SHAPEIT v2 and IMPUTE v2.3.0 using the 1000g phase 1 integated reference panel. Only individuals of S3 had refraction assessment which was done by Retinomax analysis and eyeglass prescription inspection if available. Subjects with age-related macular degeneration, cataracts, retinitis pigmentosa, color blindness, other congenital eye problems, LASIK, artificial lenses, and other eye surgery were excluded.  Association analyses were  done with QUICKTEST version 0.95.The genomic control inflation factor was 1.016 (after filtering SNPs for maf > 1%, imputation quality info > 0.3).

**MARS**

The MARS Study is a longitudinal study designed to identify medical, environmental, and genetic factors with implications for the progression of ARM. From June 2001 to October 2003, we assembled a cohort of 1060 residents of the Muenster (Germany) region. Eligibility criteria for the baseline examination were described in detail previously. In brief, patients with ARM (drusen and/or retinal pigment epithelial [RPE] changes) in at least one eye, no or minimal lens opacity, thereby allowing good visualization of the retina, and age between 60 and 80 years were included into the study. Control subjects were volunteers, spouses, and companions of ARM patients who had no signs of ARM. Between November 2003 and August 2006, we re-examined 828 participants (85.5% of the initial cohort members eligible for re-examination).The median follow-up time was 2.6 years. Between 2007 and 2009, after a median time of further 4.8 years, we re-invited 403 participants of MARS II to a second follow up MARS III (net participation among eligible 72%). We replicated the baseline examination protocols during MARS II and III, additionally including the measurement of macular optical density (MPOD) by 2-wavelengths methods. The recruitment and research protocols were reviewed and approved by the Institutional Review Board of the University of Muenster, and written informed consent was obtained from all study participants, in compliance with the Declaration of Helsinki.

All subjects were interviewed by a trained interviewer using a standardized risk factor questionnaire. Detailed information was obtained about demographic characteristics, smoking history, lifestyle, medical history, and the current and past use of medications and vitamin supplements, in particular those containing L and/or Z. Physical examinations included measurements of height, weight, pulse rate, and blood pressure. Blood was drawn only at the baseline examination for biochemical and genetic analyses. Serum concentrations of L and Z were measured using standard methods at baseline and at MARS III.

**Montrachet**

Subjects of the MONTRACHET (Maculopathy Optic Nerve nuTRition neurovAsCular and HEarT diseases) study were recruited from an on going population-based study, the Three-City (3C) study, on the vascular risk factors for dementia [[19](#_ENREF_19)]. The 3C-Study was designed to examine the relationship between vascular diseases and dementia in 9,294 community-dwelling persons aged 65 years and over. The participants were selected from the electoral rolls and were only urban since they lived in 3 French cities, Bordeaux, Dijon and Montpellier. Eye examinations were proposed to 3C participants from 2009 in Dijon.

In Dijon, 4931 subjects participated to the first run of the 3C-Study in 1999. At the fifth run, a subgroup of participants was invited to participate to the Montrachet study. Written informed consent was obtained. We chose preferentially the participants having had an MRI (n = 1663) and we completed the recruitment with a random sample of 500 subjects without MRI. Therefore from October 22th, 2009 until March 31th, 2013, 900 volunteers with an MRI and 253 without an MRI were recruited in the Montrachet study. The participants of the Montrachet study represented 54.1% and 50.6% of the cohort still followed 10 years in Dijon after the initiation of the 3 C study, for those having had an MRI and those without MRI, respectively.

The Montrachet study is a population-based study designed to find associations between age-related eye diseases and neurologic and heart diseases in the elderly as a primary objective. The secondary objective was to report the prevalence of the main age-related eye diseases in the elderly as well as the influence of genetic and environmental risk factors. Fasting blood samples were drawn. Technicians conducted the eye examination in the Department of Ophthalmology, University Hospital Dijon, France. At the end of the eye examination participants were asked to fill at home a questionnaire about lifestyle, environment and nutrition (frequency food questionnaire). The following data were collected: ophthalmic history, visual acuity, refractive error, tonometry and pachymetry, slit-lamp examination and ocular surface evaluation, visual field, OCT imaging, macular pigment assessment, two 45° non mydriatic color retinal photographs, one centred on the macula and the one on the optic nerve head. AMD, other retinal diseases and glaucoma were classified using international classifications.

**MRC Older People Study**

The MRC Older People Study was a cluster randomised trial in general practice (family doctors) funded by the UK Medical Research Council and Departments of Health. The principal aim of the study was to evaluate different methods of health assessment and management of older people (aged 75+). The two main arms of the study were Universal or targeted assessment. Mortality, 2 year hospital admissions and nursing home admission and quality of life were the primary outcomes. A further aim of the study was to provide comprehensive data on the prevalence of health and social problems of older people and to investigate the relationship of these with health outcomes.

All people aged 75 and over on the GP lists were eligible for the study provided they were not resident in a nursing home or hospital and were not terminally ill. The study commenced in 1995 and 106 general practices (from the MRC General Practice Research Framework) and 33,000 patients aged over 75 years were recruited to the trial with response rates of 78%. In the Universal arm of the study, all participants (n=15,000) underwent an in-depth assessment by a study nurse, while in the Targeted arm, only participants with selected problems identified from a brief health check (n=1500) went on to have the detailed assessment. The detailed assessment included questionnaires: Mini-Mental State Examination for cognitive impairment, Geriatric Depression Scale, GHQ anxiety scale, Rose chest pain questionnaire for angina, MRC respiratory questionnaire, Activities of Daily Living, questions on incontinence and diabetes and a medical history including cardiovascular events. Lifestyle factors included current and past smoking behaviour, usual alcohol pattern and consumption of wine, beer and spirits and physical activity. Measurements included height, weight, mid upper arm circumference, demi span, waist and hip circumference, systolic and diastolic blood pressure, whispered voice test for hearing, Bailey Lovie Charts for visual acuity. Biological measurements included a urine dipstick for blood and protein, and a blood sample for a full biochemical screen. In 49 practices, the cause of visual impairment was assessed by medical record review of ophthalmologists’ diagnosis. Over 50 papers have been published including the main trial [[20](#_ENREF_20)] of which 8 papers are specifically on vision problems.

**MYST**

The MYopia STudy (MYST) is a high-myopia case-control study conducted from December 2009 to July 2012. High-myopic cases and emmetropic controls were recruited by eye care providers, opticians, and optometrists; by ophthalmologists from university hospitals (primarily from the Erasmus Medical Center, Leiden University Medical Center, and Nijmegen University Medical Center) and community hospitals (primarily from the Eye Hospital Rotterdam, the Focus Clinic Rotterdam, and the Amphia Hospital Breda); by public media outlets (www.myopiestudie.nl); and by door-to-door flyer distribution. High-myopic cases were defined as having refractive error ≤‒6 D, and emmetropic controls were defined as having refractive error ≥-1.5 D and ≤1.5 D. All participants were ≥25 years of age. The subjects were examined from 2010 through 2012. In total, 1057 participants (690 cases and 367 controls) were included in this study. Measurements in all studies were collected after receiving approval from the medical ethics committee of the Erasmus University Medical Center, and all participants provided written informed consent in accordance with the Declaration of Helsinki.

The MYST study protocol included a complete ophthalmological examination, a questionnaire, and peripheral blood sampling for genotyping. A non-cycloplegic measurement of refractive error and keratometry was performed for both eyes using a Topcon RM-A2000 autorefractor (Topcon Optical Company). BCVA with objective refraction was measured using standardized ETDRS protocols. [[21](#_ENREF_21)] Intraocular pressure was measured using Goldmann applanation tonometry. AL, corneal thickness, anterior chamber depth, and lens thickness were measured using a Lenstar LS900 (Laméris Ootech). For subjects with AL >30 mm, we used an A-scan ultrasound device (Pacscan, Sonomed Escalon) to measure AL, anterior chamber depth, and lens thickness. After dilating the pupils with tropicamide/phenylephrine 0.5/5%, we performed indirect ophthalmoscopy to quantify myopic degeneration of the retina (including the peripheral retina). Stereoscopic digital colour photographs (35°) were taken of the lens, the optic nerve head, and the macular area using a Topcon digital fundus camera (Topcon TRC 50EX; 0.44 megapixel, Topcon Optical Company) or a Sony DXC-950P digital camera (Sony Corporation, Minato, Japan). Fundus photographs were graded according to the META-PM grading protocol. [[22](#_ENREF_22)] We also performed fundus autofluorescence, infrared, and red-free measurements of the macular area of each eye (Heidelberg HRA-2, Heidelberg Engineering, Heidelberg, Germany). Optic discs were imaged using confocal laser scanning tomography (Heidelberg Retina Tomograph HRT II, Heidelberg Engineering). Optical coherence tomography was performed using Topcon 1000, 2000 (SD) and Topcon-DRI (SS) (Topcon Optical Company), and scans were made of both the optic nerve and the macula. The questionnaire included questions regarding the subject’s complete ocular and medical history, family history of myopia, education level, and near-work and outside activities during young childhood, adolescence, and adulthood.

**NICOLA**

The Northern Ireland Cohort for the Longitudinal Study of Ageing (NICOLA) study is a comprehensive, long-term study of adult development and ageing which started in December 2013 and consists of a random sample of men and women aged 50 years and above who will be representative of the Northern Ireland population (n=8500). Participants will take part in repeated waves of data collection every four years and will be followed up for a period of at least ten years. The main study comprises of two parts: firstly, a computer assisted personal interview (CAPI) which takes place in their own home, during which they provide information examining multiple aspects of their lives as they age including self-reported medical history; secondly, the participants will be invited to attend the Wellcome-Wolfson Clinical Research Facility (CRF) at Belfast City Hospital for a health assessment which includes the following assessments: *i) Anthropometry***:** Height, weight, waist and body composition, sitting, arm raise extension and finger snapping, timed get up and go, grip strength, walk speed and facial photograph; ii) *Respiratory*: Spirometer; iii) *Cardiovascular*: Blood pressure, pulse rate; iv) *Cognition*: Colour trails, Verbal fluency, Montreal Cognition Assessment, mini-mental state examination (MMSE); v) *Ophthalmology:* Visual acuity, auto refraction, corneal compensated intra-ocular pressure, colour fundus photographs, infra-red and quantitative auto fluorescent retinal images, wide field Optos colour images and Spectral Domain Optical Coherence Tomography(OCT) with Enhanced Depth Imaging (EDI) of the choroid; vi) *Sampling*: 50mls of blood. Urine. We expect approximately 50% of those who do the home interview to attend the health assessment (n=4,000).

**PAMDI**

The PAMDI (Prevalence of Age-related Macular Degeneration in Italy) Study is population-based cross-sectional study aiming at estimating the prevalence, risk factors of age-related macular degeneration (AMD) and vision-related quality of life in an Italian population and to analyze differences between urban and rural communities. The methods of this study have been published elsewhere.

Subjects aged 61 years or older were recruited from two communities in Northeast Italy: one living in an urban district of the city of Padua with a total of 2,495 inhabitants aged 61 years or older (representing 23.4% of the local population), and the other consisting of two municipalities in a rural area of Padua province, with a total of 3,189 inhabitants aged 61 or older (representing 21.8% of the local population). The two communities were representative of the general Italian population. The study was approved by the IRB of the University of Padua and performed in accordance with the Declaration of Helsinki.

The sample size calculated from the target population consisted of 885 subjects. After giving written informed consent, two questionnaires were self-administered: i) NEI 25-itemVisual Function Questionnaire and ii) Food Frequency Questionnaire. All subjects were also interviewed about past medical history, demographic features and life-style habits (e.g. smoking habit, alcohol consumption and sunlight exposure). Participants underwent a complete ophthalmological assessment, including 30°color fundus photographs of the posterior retina according to standard methods (ETDRS field 1M). The digital photographs were anonymized and sent to the Reading Centre (Moorfields Eye Hospital NHS Foundation Trust, London, UK) for grading. Raman Spectroscopy for measurement of xantofil pigments was also performed.

**POLA**

The Pathologies Oculaires Liées à l’Age (POLA) Study is a population-based study aimed at identifying the risk factors of age-related eye diseases. The methods of this study have been published elsewhere.[[23](#_ENREF_23)] For inclusion in the study, participants needed to be a resident of Sète (South of France) and aged 60 years and over. According to the 1990 population census, there were almost 12,000 eligible residents, of whom our objective was to recruit 3,000. The population was informed of the study through the local media. We also contacted 4,543 residents individually by mail and telephone, using the electoral roll. The baseline examinations took place in a mobile unit equipped with ophthalmologic devices. Between June 1995 and July 1997, 2,584 participants were recruited. This research followed the tenets of the Declaration of Helsinki. Participants gave written consent for participation in the study. The study was approved by the ethics committee of the University Hospital of Montpellier, France. A follow-up examination was performed in 1998-2000, in 1, 947 of the 2,436 survivors (79.9 %).

Eye examinations included, for each eye, a recording of ophthalmological history, measures of visual acuity and refraction, one 50° mydriatic color retinal photograph centered on the macula, assessment of lens opacities at slit lamp using LOCSIII and measures of intraocular pressure. AMD was graded according to the International Classification. Retinal vessel calibers were estimated using IVAN software.

**Rotterdam Study I/II/III**

The Rotterdam Studies are prospective cohort studies of people living in Ommoord, a district of the city of Rotterdam. This study investigates occurrence and risk factors of ophthalmic, cardiovascular, hepatic, neurological, psychiatric, dermatologic, oncological, respiratory and endocrine diseases in an elderly population. [[24](#_ENREF_24)] The ophthalmic part of the Rotterdam Study focusses on age-related macular degeneration, open angle glaucoma and myopia. In addition to risk analyses, potential biomarkers in various biosamples are investigated. [[24](#_ENREF_24)]

The Rotterdam Study consists of three cohorts of which the first started in 1990 and consisted of 7983 participants of 55 years and older (RS I, response rate of 78%). The second cohort started recruiting in 2000 and 3011 participants of 55 years and older were included (RS II, response rate of 67.3%). The third cohort also included people aged 45 years and older and consisted of 3932 participants (RS III, response rate 64.9%) starting from the year 2006. Follow-up of these cohorts took place about every 3-5 years. Participants underwent an extensive physical examination at a research center. The study was approved by the institutional review board (Medical Ethics Committee) of the Erasmus Medical Center and by the review board of the Netherlands Ministry of Health, Welfare and Sports, and participants gave written consent.

The eye examinations consisted of medical ophthalmic history, presenting and best corrected visual acuity, refractive error, intra ocular pressure, axial length, biometry, perimetry, and in depth imaging such as mydriatic color fundus photography (one centered on the macula 35° and one on the optic nerve head 20°), HRA, HRT and OCT.

**Southampton Liver Transplant**

We used a cross-sectional design to investigate whether donor or recipient *CFH* Y402H genotype was associated with AMD in LT patients [[25](#_ENREF_25)]. Patients were recruited between September 2009 and January 2011 from four liver transplant centres in the UK (London Kings College Hospital, London; Queen Elizabeth Hospital, Birmingham; Addenbrooke's Hospital, Cambridge; and University Hospital Southampton). Inclusion criteria included patients of Western European origin, aged ≥55 years, with a history of LT ≥5 years ago. Patients were retrospectively identified from preexisting hospital LT databases.

We determined AMD status by dilated fundus examination and digital photography (Topcon TRC 50DX camera, Topcon Corporation, Tokyo, Japan/Nidek NM-200D camera, Nidek Co. Ltd, Gamagori, Japan). Digital photographs were graded according to the Rotterdam grading system by 2 retinal fellows and any differences were arbitrated by a senior grader. We defined AMD as the presence of Rotterdam grades 1 through 4 in the worst eye). General medical history, smoking history, body mass index, and blood pressure were recorded. Recipient peripheral blood was taken for DNA genotyping and plasma complement component measurement. Donor age and sex information were obtained anonymously from each site. Donor tissue (when available) was obtained for DNA genotyping from samples stored at the time of liver transplant.

**IVAN Study**

The IVAN study is a randomized clinical trial aimed at comparing the efficacy and safety of ranibizumab and bevacizumab intravitreal injections to treat neovascular age-related macular degeneration. Adults ≥50 years old with previously untreated neovascular AMD in the study eye and best corrected visual acuity ≥25 letters on the Early Treatment Diabetic Retinopathy Study chart were eligible[[26](#_ENREF_26)]. Diagnosis was confirmed by fluorescein angiography. Participants without a subfoveal (within 200 μm) neovascular component were eligible if subretinal fluid or serous pigment epithelial detachment was subfoveal. To avoid including inactive or advanced disease, lesions comprising >50% fibrosis or blood were excluded. Only 1 eye from each participant was studied.

We recruited participants from 23 teaching and general hospitals in the United Kingdom (UK. A UK National Health Service (NHS) Research Ethics Committee gave approval (reference 07/NIR03/37). This trial is registered (ISRCTN92166560).

**Southampton nystagmus**

The study had the approval of the local and regional ethics committees and conformed to the tenets of the Declaration of Helsinki. Congenital idiopathic nystagmus patients underwent detailed clinical examination, including tests for logarithm of the minimum angle of resolution visual acuity, refraction, colour vision, intraocular pressure; anterior and posterior segment slitlamp examination, including iris transillumination testing in a darkened room; and orthoptic assessment [[27](#_ENREF_27), [28](#_ENREF_28)]. Some patients had detailed recordings of their nystagmus waveform performed using Skalar IRIS IR Light Eye Tracker equipment (Cambridge Research Systems Ltd, Rochester, England). Twenty-four eye movement recordings were completed for each patient. Binocular and uniocular saccades were recorded to calibrate amplitude measurements at ± 10° and ± 20° from fixation in the horizontal plain using a 1° red square target moving at 500-millisecond intervals.

**Southampton paediatric eye diseases**

Both adults and children were recruited from 5 ophthalmology clinics at Southampton General Hospital. Inclusion criteria was broad and related to the diagnosis of a genetic/inherited eye disease affecting the subject or a close family member before the age of 18 years. Patients/guardians were identified by their treating doctors and offered the opportunity to participate in a genetic study. Patients were grouped into one of 4 categories; predominantly anterior segment disease, predominantly posterior segment disease, predominantly ocular motility disorder or miscellaneous.

**Southampton POAG**

Primary open angle patients from a cohort of patients being recruited in Hampshire (UK) were included in this study. Patients were recruited following the tenets of the declaration of Helsinki, informed consent was obtained and the research was approved by the Southampton & South West Hampshire Research Ethics Committee. Patients were all diagnosed as POAG cases and further defined as normal tension glaucoma (NTG) if the average IOP over both eyes ≤21 mmHg, and high tension glaucoma (HTG) if otherwise. All showed visual field loss in at least one eye. A full description of the cohort is given [[29](#_ENREF_29)] [[30](#_ENREF_30)]. Further POAG cases were collected from Southampton, Portsmouth and additional sites on this study in Frimley, Cambridge, Torbay, Wolverhampton, Isle of Wight & Birmingham.

**Southampton Rod-cone Dystrophies**

Rod Cone Degeneration patients over the age of 18 were included in this study. Patients were recruited in Hampshire (UK) following the tenets of the declaration of Helsinki, informed consent was obtained and the research was approved by the Southampton & South West Hampshire Research Ethics Committee. Patients diagnosed with rod cone degeneration and were further defined as Retinitis Pigmentosa (RP), Cone Dystrophy (CD) Rod/Cone dystrophy (RCD), Cone/Rod dystrophy (CORD), Stargardt disease, Best Disease, Leber’s Congenital Amaurosis (LCA), Dominant Optic Atrophy or Other. Inheritance pattern & Best Corrected Visual Acuity (BCVA) were taken.

**Thessaloniki Eye Study**

The Thessaloniki Eye Study (TES), 2000-2005, is a cross-sectional, population-based, epidemiologic study of chronic eye diseases in the Greek population of Thessaloniki which is considered representative of the general population in the country. The initial recruitment frame consisted of 5,000 people 60 years of age or older, identified randomly from approximately 321,000 persons registered in the municipality registers. [[31](#_ENREF_31)] From the initial recruitment sample, 3,617 subjects were eligible and finally 2,554 participated in the study (participation rate 71%). [[32](#_ENREF_32)] The study was approved by the Aristotle University Hospital Ethics Committee and the University of California Los Angeles Human Subject Protection Committee. All study procedures adhered to the principles outlined in the Declaration of Helsinki for research involving human subjects and all participants gave written informed consent prior to their participation.

Subjects were invited to the study examination center for an extensive ophthalmologic screening examination. In an effort to increase participation rate and to minimize potential no-participation bias, a home visit eye examination was arranged for persons unable to visit the study examination center because of illness or major disability.

Visual acuity was measured with the Early Treatment of Diabetic Retinopathy Study (ETDRS) charts, and screening visual field (VF) examination (Humphrey Automated Field Analyzer II, Carl Zeiss Meditech, Dublin, CA) was performed for all participants with visual acuity of more than counting fingers. If the screening test was abnormal, a Full Threshold or Sita-Standard VF was performed. Intraocular pressure (IOP) was measured using a calibrated Goldmann applanation tonometer (Haag-Streit, Bern, Switzerland). Blood pressure (BP) was measured with an automated sphygmomanometer (model 705CP; OMRON Matsusaka Co Ltd, Matsusaka City, Japan) before instillation of mydriatic drops and after the participant was seated for 10 minutes. Somatometric data (height, weight) were also measured.

If the anterior chamber angle was open, dilation drops (5% phenylephrine and 0.5% tropicamide) were instilled. If the angle was potentially occludable, evaluation of the lens and fundus were performed without dilation, the participants were referred for laser peripheral iridotomy and dilated lens and fundus examination was completed afterwards. The presence of pseudoexfoliative material in the anterior chamber of the eye was also recorded. Fundus photos of the macula and disc (Topcon, Japan), optic disc imaging with the Heidelberg Retina Tomograph (HRT) and Heidelberg Retinal Flowmeter (HRF) images (Heidelberg Engineering, Heidelberg, Germany) were acquired. Finally, central corneal thickness (CCT) was measured using ultrasound pachymetry (A-scan, Quantel Medical, France) in a subset of subjects.

**Tromsø Eye Study**

Tromsø Eye Study (TES) is a substudy of the multipurpose Tromsø Study, a longitudinal population-based study started in 1974. The Tromsø Study and the cohort profile have been described elsewhere [[33](#_ENREF_33)]. A description of the large amount of variables collected is presented at <http://tromsoundersokelsen.uit.no/tromso/> Blood samples from each survey and DNA samples from the 3rd survey and onwards are stored in a biobank. The population is being followed up with registration of incident myocardial infarction, stroke, atrial fibrillation, diabetes and non-vertebral fractures. The study sample for the Tromsø Study is based upon the official population registry and all subjects were residents of the municipality of Tromsø. Ophthalmological data have been collected from the 5th Tromsø Study survey and onwards. Details on the study samples have been described elsewhere [[33](#_ENREF_33), [34](#_ENREF_34)].

In the 5th Tromsø Study survey (2001-02) a total of 8130 participants (30-89 years) attended (78.5% attendance rate). Visual acuity and refraction was collected on a subset of the participants. The 6th survey (2007-2008) consisted of two separate visits. The first visit included a questionnaire and physical examination comprising the measurement of blood pressure, height, weight and waist-to-hip ratio. Blood sampling, bone mineral density and pain threshold tests were also performed. A total of 12984 subjects (65.7% attendance rate), 30-87 years participated. Among the participants attending the first visit, 7958 participants were invited to the 2nd visit and 7307 participants (30-87 years) attended (91.8% attendance rate). Eye examinations were performed at the 2nd visit including ophthalmological history, five fields 45° color retinal photographs, spectral domain optical coherence tomography, visual acuity and refraction. In addition the 2nd visit comprised a second questionnaire, blood samples, cognitive tests, ultrasound of the carotid artery, 12-lead electrocardiogram, echocardiography, spirometry, and bone mineral densitometry. All images were graded for diabetic retinopathy and retinal vessel calibers were estimated using IVAN software. AMD was graded in participants 65 years and older according to the International Classification. The 7th survey started 2015 and aim to include all participants from the 6th survey and an additional random selection of residents 40 years and older. Data collected includes all variables from the 6th survey supplemented with intraocular pressure measurements.

The Tromsø Study and TES followed the tenets of the Declaration of Helsinki for research involving humans and were approved by the Regional Committee for Medical and Health Research Ethics. All participants gave an informed written consent.

**Twins UK**

The TwinsUK adult twin registry, based at St. Thomas' Hospital in London, compromises over 12,000 predominantly female Caucasian ancestry twins, from throughout the United Kingdom [[35](#_ENREF_35)], with mean age of 63 years (SD 13.9, range 18-90), of whom ~7,000 have attended for phenotyping. Twins largely volunteered unaware of the eye studies at the time of enrolment and gave fully informed consent under a protocol reviewed by the St. Thomas' Hospital Local Research Ethics Committee (EC04/015), which was performed in accordance with the Helsinki Declaration.

Various eye phenotypes have been collected on a subset of twins from 1998 to the current time. Refractive error was measured using non-cycloplegic autorefraction (ARM-10 autorefractor, Takagi Seiko, Japan) for over 6,000 participants. IOP was measured with a non-contact air-puff tonometer, the Ocular Response Analyser (ORA, Reichert®, Buffalo, NY) for 3,500 participants. The mean (Goldmann-equivalent) IOP was calculated from 4 readings (2 from each eye) for each participant; where quality indicators were poor or the two IOPs differed by more than 2mmHg, a third reading was taken. CCT was measured using an ultrasound pachymetry device provided with the ORA instrument. Retinal photographs of both eyes were obtained by one of two methods; a Kowa camera (Kowa-Europe, Dusseldorf, Germany) on a 30° width of field setting and developed on Ektachrome 64 film (Kodak, Rochester, NY) in 1,012 subjects, and subsequently using the Nidek AFC-210 non-mydriatic digital fundus camera (Nidek, Gamagori, Japan) for 2,500 twins. Macula images were graded initially according to International ARM Epidemiologic Study Group classification and latterly to the Rotterdam and modified AREDS classification systems. Retinal vessel calibres were graded using Ivan 1.1 software (University of Wisconsin). Disc and cup area was measured from stereo disc photographs (using the Nidek-3DX stereo camera, Gamagori, Japan; Polaroid film, Minnetonka, MN), with digitized images scanned from Polaroid images and StereoDx stereoscopic planimetric software (StereoDx) using a Z-screen (StereoGraphics Corp). Cortical cataract was graded on mydriatic retro-illumination cataract images using the Oxford Clinical Cataract Classification and Grading System. The amount of nuclear cataract was graded on Scheimpflug camera images using the pixel density at the centre of the nucleus (central nuclear dip score), available on 2,300 subjects over 50 years of age. Dry eye phenotypes were available from questionnaire data (~3,500 subjects), additional Ocular Surface Disease Index (OSDI) questionnaires, tear osmolarity, Schirmer’s values and tear breakup time (TBUT) on 600 participants. OCT imaging (Optovue) is currently being performed on twin subjects.

**Young Finns Study (YFS)**

The YFS cohort [[36](#_ENREF_36)] is a Finnish longitudinal population study sample on the evolution of cardiovascular risk factors from childhood to adulthood. The first cross-sectional study was conducted in the year 1980 in five different centers. It included 3,596 participants in the age groups of 3, 6, 9, 12, 15, and 18, who were randomly chosen from the national population register. After the baseline in 1980 these subjects have been re-examined in 1983 and 1986 as young individuals, and in 2001, 2007 and 2011-2012 as older individuals. The current study includes the 1571 participants who underwent retinal photography during the 2011–2012 and have the refractive error measurement data from both eyes. Refractive error was measured by using NIDEK AR-310AR autorefractor. Forty five degree digital retinal images centered on the macula of each eye were captured using a Canon nonmydriatic retinal camera (Canon CR6-45NM, USA) fitted with a Canon 10D digital SLR camera attachment. Images were read at a single reading center (Imperial College London) and photographer accreditation was performed prior to beginning the study. Imaging QC was conducted regularly throughout the study. One observer, blinded to subject data, undertook QC, including provision of feedback, and performed retinal grading. A semiautomated grading system was used to capture a range of retinal geometric parameters. Measured parameters included the (i) arteriolar diameters, (ii) arteriolar bifurcation angles, (iii) length/diameter ratios of arteriolar segments and arteriolar/venular diameter ratios (these parameters provide measures of arteriolar narrowing that are relatively unaffected by differences in optical refraction), (iv) arteriolar tortuosity (estimated as the actual length of the vessel divided by the straight line distance between bifurcations minus 1), and (v) arteriolar optimality ratio and optimality deviance. Optimality ratio is the ratio of sum of “daughter” arteriolar diameters divided by the “parent” arteriolar diameter corrected for asymmetry [[37](#_ENREF_37)]. YFS database is rich of different phenotypes having over 3000 clinical, anthropometric, laboratory, dietary, psychological, and socioeconomic parameters. YFS have genome-wide genetic data (GWAS) imputed to last 1000G reference and whole blood genome wide expression data (GWE from year 2011) and metabolomics data (from years 2001, 2007 and 2011). In addition, from subsamples, genome wide epigenetic measurements including methylome and microRNA measurements are available.

This study was carried out in accordance with the recommendations of the Declaration of Helsinki. All participants provided written informed consent and the study protocol was approved by the Ethics Committee.

References:

1. Power C, Elliott J. Cohort profile: 1958 British birth cohort (National Child Development Study). Int J Epidemiol 2006;35:34-41.

2. Rahi JS, Cumberland PM, Peckham CS. Myopia over the lifecourse: prevalence and early life influences in the 1958 British birth cohort. Ophthalmology 2011;118:797-804.

3. Rahi JS, Cumberland PM, Peckham CS. Visual impairment and vision-related quality of life in working-age adults: findings in the 1958 British birth cohort. Ophthalmology 2009;116:270-4.

4. Rahi JS, Cumberland PM, Peckham CS. Visual function in working-age adults: early life influences and associations with health and social outcomes. Ophthalmology 2009;116:1866-71.

5. Delcourt C, Korobelnik JF, Barberger-Gateau P, et al. Nutrition and Age-Related Eye Diseases: The ALIENOR (Antioxydants, Lipides Essentiels, Nutrition et Maladies Oculaires) Study. J Nutr Health Aging 2010;14:854-61.

6. Group CS. Vascular factors and risk of dementia: design of the Three-City Study and baseline characteristics of the study population. Neuroepidemiology 2003;22:316-25.

7. Cachulo Mda L, Lobo C, Figueira J, et al. Prevalence of Age-Related Macular Degeneration in Portugal: The Coimbra Eye Study - Report 1. Ophthalmologica 2015;233:119-27.

8. Bird AC, Bressler NM, Bressler SB, et al. An international classification and grading system for age-related maculopathy and age-related macular degeneration. The International ARM Epidemiological Study Group. Surv. Ophthalmol. 1995;39:367-74.

9. Augood CA, Vingerling JR, de Jong PT, et al. Prevalence of age-related maculopathy in older Europeans: the European Eye Study (EUREYE). Arch Ophthalmol 2006;124:529-35.

10. Van Leeuwen R, Klaver CC, Vingerling JR, Hofman A, De Jong PT. The risk and natural course of age-related maculopathy: follow-up at 6 1/2 years in the Rotterdam study. Arch. Ophthalmol. 2003;121:519-26.

11. Marques JP, Costa M, Melo P, et al. Ocular Risk Factors for Exudative AMD: A Novel Semiautomated Grading System. ISRN Ophthalmol 2013;2013:464218.

12. Ribeiro L, Oliveira CM, Neves C, et al. Screening for Diabetic Retinopathy in the Central Region of Portugal. Added Value of Automated 'Disease/No Disease' Grading. Ophthalmologica 2014.

13. Aulchenko YS, Heutink P, Mackay I, et al. Linkage disequilibrium in young genetically isolated Dutch population. Eur J Hum Genet 2004;12:527-34.

14. Pardo LM, MacKay I, Oostra B, van Duijn CM, Aulchenko YS. The effect of genetic drift in a young genetically isolated population. Ann Hum Genet 2005;69:288-95.

15. Fauser S, Smailhodzic D, Caramoy A, et al. Evaluation of serum lipid concentrations and genetic variants at high-density lipoprotein metabolism loci and TIMP3 in age-related macular degeneration. Invest Ophthalmol Vis Sci 2011;52:5525-8.

16. Wilkinson CP, Ferris FL, 3rd, Klein RE, et al. Proposed international clinical diabetic retinopathy and diabetic macular edema disease severity scales. Ophthalmology 2003;110:1677-82.

17. Kruithof CJ, Kooijman MN, van Duijn CM, et al. The Generation R Study: Biobank update 2015. Eur J Epidemiol 2014;29:911-27.

18. Ennis S, Jomary C, Mullins R, et al. Association between the SERPING1 gene and age-related macular degeneration: a two-stage case-control study. Lancet 2008;372:1828-34.

19. Vascular factors and risk of dementia: design of the Three-City Study and baseline characteristics of the study population. Neuroepidemiology 2003;22:316-25.

20. Fletcher AE, Price GM, Ng ES, et al. Population-based multidimensional assessment of older people in UK general practice: a cluster-randomised factorial trial. Lancet 2004;364:1667-77.

21. (ETDRS) ERGETDRS. Manual of Operations National Technical Information Service, Springfield, VA, 1985.

22. Ohno-Matsui K, Kawasaki R, Jonas JB, et al. International photographic classification and grading system for myopic maculopathy. Am J Ophthalmol 2015;159:877-83 e7.

23. Delcourt C, Diaz JL, Ponton-Sanchez A, Papoz L. Smoking and age-related macular degeneration. The POLA Study. Pathologies Oculaires Liees a l'Age. Arch Ophthalmol 1998;116:1031-5.

24. Hofman A, Darwish Murad S, van Duijn CM, et al. The Rotterdam Study: 2014 objectives and design update. Eur J Epidemiol 2013;28:889-926.

25. Khandhadia S, Hakobyan S, Heng LZ, et al. Age-related macular degeneration and modification of systemic complement factor H production through liver transplantation. Ophthalmology 2013;120:1612-8.

26. Chakravarthy U, Harding SP, Rogers CA, et al. Ranibizumab versus bevacizumab to treat neovascular age-related macular degeneration: one-year findings from the IVAN randomized trial. Ophthalmology 2012;119:1399-411.

27. Self JE, Shawkat F, Malpas CT, et al. Allelic variation of the FRMD7 gene in congenital idiopathic nystagmus. Arch Ophthalmol 2007;125:1255-63.

28. Self JE, Ennis S, Collins A, et al. Fine mapping of the X-linked recessive congenital idiopathic nystagmus locus at Xq24-q26.3. Mol Vis 2006;12:1211-6.

29. Ennis S, Gibson J, Griffiths H, et al. Prevalence of myocilin gene mutations in a novel UK cohort of POAG patients. Eye (Lond) 2010;24:328-33.

30. Gibson J, Griffiths H, De Salvo G, et al. Genome-wide association study of primary open angle glaucoma risk and quantitative traits. Mol Vis 2012;18:1083-92.

31. Topouzis F, Coleman AL, Harris A, et al. Association of blood pressure status with the optic disk structure in non-glaucoma subjects: the Thessaloniki eye study. Am J Ophthalmol 2006;142:60-7.

32. Topouzis F, Wilson MR, Harris A, et al. Prevalence of open-angle glaucoma in Greece: the Thessaloniki Eye Study. Am J Ophthalmol 2007;144:511-9.

33. Jacobsen BK, Eggen AE, Mathiesen EB, Wilsgaard T, Njolstad I. Cohort profile: the Tromso Study. Int J Epidemiol 2012;41:961-7.

34. Bertelsen G, Erke MG, von Hanno T, et al. The Tromso Eye Study: study design, methodology and results on visual acuity and refractive errors. Acta Ophthalmol 2013;91:635-42.

35. Moayyeri A, Hammond CJ, Hart DJ, Spector TD. The UK Adult Twin Registry (TwinsUK Resource). Twin Res Hum Genet 2013;16:144-9.

36. Raitakari OT, Juonala M, Ronnemaa T, et al. Cohort profile: the cardiovascular risk in Young Finns Study. Int J Epidemiol 2008;37:1220-6.

37. Hussain SM, Kahonen M, Raitakari OT, et al. Impact of fetal growth and preterm birth on the retinal microvasculature in mid-adulthood. Microcirculation 2015;22:285-93.