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DESCRIPTION OF PARTICIPATING STUDIES

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. 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. 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.

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). 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 and the signs of disease were stratified into 5 exclusive stages (ARM, stage 0 to 4) using the Rotterdam staging system. 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).

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 characteristics 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, familial 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.

Epic-Norfolk

The European Prospective Investigation of Cancer (EPIC) is a 10 nation collaborative study which commenced in 1989. EPIC Norfolk enrolled 30,445 men 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-NWSS + 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).
**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 an independent reading centre (Erasmus University Rotterdam) using the International Classification System for Age Related Maculopathy.

**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.

**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. 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 22nd, 2009 until March 31st, 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 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 \(^{12}\) of which 8 papers are specifically on vision problems.

**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-item Visual 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 xanthophyll pigments was also performed. Two papers have been published, regarding the epidemiologic results of the main study\(^ {13}\) and the associated risk factors \(^ {14}\).

**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.\(^ {15}\) 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.  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.

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, intraocular 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.

**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. From the initial recruitment sample, 3,617 subjects were eligible and finally 2,554 participated in the study (participation rate 71%). 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 non-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.19 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, venous thromboembolism, 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.19 20

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. Optical coherence tomography scans were graded for retinal thickness. 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.

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