Genetic correlations between diabetes and glaucoma: an analysis of continuous and dichotomous phenotypes

Vincent Laville[1], Jae H. Kang[2], Clara C. Cousins[3], Adriana I. Iglesias[4], Réka Nagy[5], Jessica N. Cooke Bailey[6,7], Robert P. Igo, Jr.[6], Yeunjoo E. Song[6,7], Daniel I. Chasman[8], William G. Christen[8], Peter Kraft[9,10], Bernard A. Rosner[2,10], Frank Hu[9,11], James F. Wilson[5,12], Puya Gharahkhani[13], Alex W. Hewitt[14,15], David A. Mackey[16], Pirro G. Hysi[17], Christopher J Hammond[17], Cornelia M. vanDuijn[4,18], Jonathan L. Haines[6,7], Veronique Vitart[5], John H. Fingert[19], Michael A. Hauser[20], Hugues Aschard[1,9], Janey L. Wiggs[3], Anthony P. Khawaja[21]§, Stuart MacGregor[13]§, Louis R. Pasquale[2,22]§, UK Biobank, International Glaucoma Genetics Consortium*, NEIGHBORHOOD Consortium*.

[1] Department of Computational Biology – USR 3756 CNRS), Institut Pasteur, Paris, France.
[2] Channing Division of Network Medicine, Brigham and Women's Hospital, Harvard Medical School, Boston, MA, USA.
[3] Department of Ophthalmology, Massachusetts Eye and Ear Infirmary, Harvard Medical School, Boston, MA, USA
[4] Departments of Ophthalmology and Clinical Genetics, Erasmus Medical Center, Rotterdam, The Netherlands.
[5] MRC Human Genetics Unit, Institute of Genetics and Molecular Medicine, University of Edinburgh, Western General Hospital Edinburgh
[6] Department of Population and Quantitative Health Sciences, Case Western Reserve University School of Medicine, Cleveland, OH, USA
[7] Institute for Computational Biology, Case Western Reserve University School of Medicine, Cleveland, OH, USA.
[8] Division of Preventive Medicine, Brigham and Women's Hospital, Boston, MA, USA.
[9] Department of Epidemiology, Harvard T. H. Chan School of Public Health, Harvard Medical School, Boston, MA, USA.
[10] Department of Biostatistics, Harvard T. H. Chan School of Public Health, Harvard Medical School, Boston, MA, USA.
[11] Department of Nutrition, Harvard T. H. Chan School of Public Health, Harvard Medical School, Boston, MA, USA.
[12] Centre for Global Health Research, Usher Institute for Population Health Sciences and Informatics, University of Edinburgh, Edinburgh, UK.
[13] Statistical Genetics, QIMR Berghofer Medical Research Institute, Brisbane, Australia.15[4] Centre for Eye Research Australia,
[14] Centre for Eye Research Australia, University of Melbourne, Royal Victorian Eye and Ear Hospital, East Melbourne, Australia.
[15] School of Medicine, Menzies Institute for Medical Research, University of Tasmania, Hobart, Tasmania, Australia.
[16] Lions Eye Institute, Centre for Ophthalmology and Visual Science, University of Western Australia, Perth, Western Australia, Australia.
[17] Department of Twin Research and Genetic Epidemiology, King's College London, UK.
[18] Nuffield Department of Population Health, University of Oxford, Oxford, UK.
[19] Department of Ophthalmology and Visual Science, University of Iowa, Iowa City, Iowa, USA
[20] Departments of Ophthalmology and Medicine, Duke University, Durham, NC, USA
[21] Department of Public Health and Primary Care, Institute of Public Health, University of Cambridge School of Clinical Medicine, Cambridge, UK.
[22] Icahn School of Medicine at Mount Sinai, Department of Ophthalmology, New York, NY, USA.
§ These authors contributed equally to the work.
* Members of the International Glaucoma Genetics Consortium and the NEIGHBORHOOD Consortium are listed in the supplementary note.

Corresponding author:

Louis R Pasquale, MD, FARVO; Icahn School of Medicine at Mt. Sinai; One Gustave L. Levy Place, Box 1183, New York, NY, USA 10029-6574; Email: Louis.Pasquale@mssm.edu; Voice: 212-241-6752; Fax: 212-241-5764

Running title: *Genetic correlations between diabetes and glaucoma*

Introduction:

Clarifying the relationship between diabetes mellitus and primary open-angle glaucoma (POAG) could help prioritize glaucoma detection efforts and focus glaucoma drug discovery. Studies show that patients with diabetes have higher intraocular pressure (IOP) than patients without diabetes and that increased fasting blood sugar (FBS) is associated with higher IOP.[1-6] However, the link between diabetes and IOP is complex as diabetes alters corneal hysteresis (CH) and corneal resistance factor (CRF), possibly confounding the true correlation between diabetes and IOP.[7, 8] The Ocular Response Analyzer (ORA) noncontact tonometer (NCT) generates both a Goldmann-correlated IOP (IOPg) and a cornea-compensated IOP (IOPcc), with the latter adjusting for corneal biomechanical properties. Among 110,573 participants in the UK Biobank where IOP was measured with the ORA NCT, self-reported diabetes was associated with higher IOPg but there was no significant difference in IOPcc between subjects with and without diabetes in multivariate analysis.[9] A meta-analysis of seven prospective cohort studies also shows that type 2 diabetes (T2D) is associated with increased risk of POAG;[10] however, this meta-analysis is not consistent with a study finding that POAG patients with T2D and no diabetic retinopathy had significantly slower rates of retinal nerve fiber layer thinning compared to POAG patients without T2D.[11]

Several other correlations between diabetes-related traits and IOP are notable. For example, there was a positive association between postprandial glucose level and IOP in patients with and without diabetes.[12, 13] Among non-obese individuals,[14] there was a positive relationship between insulin resistance and IOP.[15] Serum diabetes-related biomarkers positively associated with IOP include hemoglobin A1c (HbA1c),[16] high-density lipoprotein (HDL) and triglyceride (TG).[2] Several studies also showed a positive correlation between body mass index (BMI), a continuous trait positively linked to T2D,[17-19] and IOP.[4, 20] Currently, it is unclear if any of these diabetes-related traits translate into increased vulnerability to POAG.

Genetic analyses offer powerful tools to analyze relationships between various traits without confounding by reverse causality, measurement artifact or detection bias. One such tool is linkage disequilibrium (LD) score regression, which estimates the genetic correlation (r_g) between traits using genome-wide association study (GWAS) summary statistics.[21, 22] For example, Pickrell et al. reported strong genetic correlations between each of the following continuous diabetes-related traits and T2D using LD score regression: fasting blood sugar (FBS), TG, low-density lipoprotein (LDL), HDL and BMI.[23] For glaucoma-related traits, a strong genetic correlation between IOP measured with Goldmann applanation tonometry and POAG was reported using GWAS summary data from two large European-derived consortia.[24] Using LD score regression in a Japanese population, Shiga et al[25] found a positive genetic correlation between T2D and POAG (r_g=0.27; p=2.00E-04) but Kinai et al. found no significant correlations between various quantitative diabetes traits and POAG in the same population.[26] Another approach is to form panels of genome-wide significant markers for a trait and test them in relation to another trait of interest. In a multiethnic US population (n=69,685), 39 genome-wide significant diabetes alleles were not collectively associated with POAG (n=3,554 cases) after adjustment for T2D.[27]

A repository of existing GWAS summary statistics and an atlas of genetic cross-correlations can be found at LD Hub.[28] Given the preponderance of epidemiological evidence linking diabetes and glaucoma, we tested the hypothesis that there would be genetic correlations between diabetes- and glaucoma related traits. First, we used LD score regression to explore the

relations between quantitative glaucoma-related traits (IOP measured using various techniques in the International Glaucoma Genetics Consortium, as well as corneal-compensated IOP (IOPcc) and Goldmann-correlated IOP (IOPg) – both measured with the ORA in the UK BioBank study, central corneal thickness (CCT), CH, CRF, cup-disc ratio (CDR) and POAG) using existing GWAS summary statistics. Next, we performed LD score regression to assess the genetic correlation between diabetes-related traits (2-hour glucose, FBS, HbA1c, fasting insulin (FI), BMI, TG, LDL, HDL and T2D) and glaucoma-related traits. Finally, we compared our estimates of genetic correlations between selected diabetes quantitative traits and glaucoma quantitative traits to values derived from directly measured traits leveraging pedigree information in two Northern European island cohorts.

Methods:

The Institutional Review Board (IRB) of Partners Healthcare prospectively approved the genetic correlation analyses described in this work. The Icahn School of Medicine IRB has a reliance agreement with Partners to conduct this research. These analyses represent a retrospective study of publicly available summary genotype data. The island cohort studies described below were approved by the Scotland National Health Study.

Assembly of Genome-Wide Association Study Summary Statistics

We assembled publicly available GWAS summary statistics and outlined the traits, sample sizes, population characteristics, and trait heritability based on GWAS data for relevant studies in **Table 1**.[29-38] The GWAS summary data were accessed at http://jass.pasteur.fr/selectPhenotypes.html and at http://ldsc.broadinstitute.org. We used the European-derived subgroups of these studies. Details such as study demographics, detailed phenotype collection methods, adjustments for covariates, the genotyping platforms used and number of single nucleotide polymorphisms (SNPs) that passed quality control can be found in references listed in **Table 1**. The trait heritability based on classic twin studies and family studies as well as the methodology for determining these traits can also be found by referring to the appropriate references in **Table 1**. Heritability based on classic twin and family studies was high overall and upward of 0.95 for CCT[39] (**Table 1**). As expected, calculations of heritability for all these traits based on summary GWAS data were lower than values estimated from classic twin studies. Several hypotheses for the source of this 'missing heritability' have been proposed in the genetics literature.[40] In the studies of quantitative diabetes traits, efforts were taken to exclude patients with known diabetes. The studies of blood lipids and BMI contains patients with and without dyslipidemia – there was no concerted effort to exclude patients with diabetes. In the studies of IOP measured in various ways, measured CDR and in the studies of corneal biophysical properties, less than 1.5% of subjects were on treatment for glaucoma.

Genetic correlation between traits analyses

The methodology for estimating genetic correlation between traits using high throughput allelic markers has been previously described[21] and appears in the **Appendix**. We provide an overview of the method here. The genetic correlation $r_{g,}$ measures of the covariance between the genetic components of two traits scaled by their respective heritability. It ranges between -1

and +1, although occasional out-of-bounds-estimates arise due to estimation error.[41, 42] Negative r_g between trait pairs mean that alleles that are positively associated with phenotype 1 are negatively associated phenotype 2. Positive r_g between trait pairs mean that there are common alleles positively associated between both traits. An absolute value of $r_g \geq 0.5$ can be considered as strong while an absolute $r_g \leq 0.12$ can be regarded as weak. P-values < 6.9E-04 associated with r_g were considered as significant to correct for the multiple comparisons made (9 diabetes traits x 8 glaucoma traits). Power calculations[41] for all possible bivariate analyses are provided in **Supplemental Table 1**.

The Orkney and Shetlandic Cohorts: Pedigrees with measured intraocular pressure, central corneal thickness and serum diabetes-related biomarkers.

The Orkney Complex Disease Study (ORCADES) is a family-based, cross-sectional study that seeks to identify genetic factors influencing cardiovascular and other disease risk in the isolated archipelago of the Orkney Isles in northern Scotland.[43] In total, 2078 participants aged 16-100 years were recruited between 2005 and 2011, most having three or four grandparents from Orkney, the remainder with two Orcadian grandparents.

The Viking Health Study (VIKING) is a family-based, cross-sectional study that aims to identify genetic factors influencing cardiovascular and other disease risk in the population isolate of the Shetland Islands in northern Scotland. In total, 2105 participants were recruited between 2013 and 2015, each having at least three grandparents from Shetland.

Genetic diversity in both the ORCADES and VIKING populations is less than mainland Scotland, consistent with high levels of endogamy historically.[44] In both cohorts, fasting blood samples were collected and many health-related phenotypes, including IOP and CCT as well as environmental exposures were measured. Specifically, serum glucose, fasting insulin and HbA1c were measured. CCT was measured using an ultrasound pachymeter (Heidelberg Engineering; Heidelberg, Germany). IOP was measured with a tonopen (Reichert Technologies; Buffalo, NY).

Genetic correlations in the Orkney and Shetlandic Cohorts

We used SOLAR (Sequential Oligogenic Linkage Analysis Routines) to decompose phenotypic covariances for IOP, CCT and diabetes-related serum biomarkers from our island cohorts into environmental, phenotypic and genetic components using pedigree data. We used measures averaged between both eyes of a participant. We excluded measures from eyes with a history of surgery that might affect CCT or IOP measurements and from participants with keratoconus. HbA1c values from individuals with diabetes or FBS >7mmol/l were also excluded. IOPs were not adjusted for CCT or transformed but were adjusted for age and sex. CCT, adjusted for age and sex, underwent z-score transformation while FBS, HbA1c and FI underwent rank transformation, with FI undergoing natural log transformation first. All serum diabetes biomarkers were further adjusted for sex, age, age^2 and BMI. P-values < 0.0042 were considered significant to correct for the multiple comparisons made (2 glaucoma traits × 3 diabetes traits x 2 cohorts).

Results:

Genetic correlation between the various glaucoma-related quantitative traits and POAG revealed significant trends (**Table 2**). There was a positive genetic association between IOP measured in the IGGC and POAG as previously reported (r_g = 0.45; Standard Error (SE) = 0.12;

p = 3.0E-04).[24] Similarly there were strong positive genetic correlations between IOPcc and POAG (r_g = 0.50; SE = 0.09; p = 5.5E-08) and between IOPg and POAG (r_g = 0.60; SE = 0.15; p = 4.3E-05). None of the corneal features (CCT, CH or CRF) showed significant genetic correlation with CDR (p≥0.13) or POAG (p≥0.07). Interestingly, while CCT showed strong positive genetic correlations with IOPg (r_g=0.58; SE=0.07; p=1.8E-15) and IOPg (r_g = 0.48; SE = 0.07; p=3.7E-12), it did not show significant genetic correlation with IOPcc (r_g = 0.07; SE = 0.05; p=0.21). Furthermore, there was also a strong positive genetic correlation between CDR and POAG (r_g = 0.57; SE = 0.09; p = 2.8E-10). IOPcc showed a positive genetic correlation with CDR (r_g = 0.16; SE = 0.05; p = 9.3E-04) that was not significant after correcting for multiple comparisons. We found strong genetic correlations between IOP measured in various ways in the IGGC as well as between IOPg with the following corneal biophysical traits: CCT, CH and CRF (range of r_g = 0.31 - 0.81; p≤3.2E-07).

Next, we examined the genetic correlations between BMI, blood lipid traits and glaucoma-related traits (**Table 3**) as well as the genetic correlations between diabetes- and glaucoma-related traits (**Table 4)**. Overall, these results were null after correction for multiple comparisons. Notably, there were non-significant inverse genetic correlations between HbA1c and POAG (r_g = -0.31; SE = 0.14; p = 0.02) and between T2D and POAG (r_g = -0.14; SE = 0.10; p = 0.16).

The ORCADES and VIKING cohorts offered an opportunity to assess the phenotypic correlations between measured glaucoma-related traits and measured serum biomarkers related to diabetes as well as genotypic correlations based on pedigree information, as opposed to genetic biomarkers (Supplemental **Tables 2** and **3**). Consistent with classic twin studies,[39] the heritability of CCT was high (range: 0.78-0.85). Heritability for IOP was 0.13-0.14 in ORCADES and 0.25 in the VIKING study. Phenotypic correlations were very low (<6%) between CCT or IOP and measured diabetes-related serum biomarkers. We found no statistically significant genetic or environmental correlations between diabetes- and glaucoma-related traits after correction for multiple testing in both cohorts (**Supplemental Tables 2** and **3**). In the VIKING cohort, there was a strong genetic correlation between IOP and CCT (r_g = 0.45; p = 9.7E-06). In both cohorts, a modest phenotypic correlation (r_p) between IOP and CCT was observed (r_p = 0.16; p = 7.8E-08 in ORCADES; r_p = 0.26; p = 3.3E-25 in the VIKING study).

Discussion

Using a genome-wide genetic correlation approach, we found no significant relationship between diabetes- and glaucoma-related traits after adjustment for multiple comparisons. These null results must be assessed in context of the power of this study to find significant associations. A consensus estimate of "good" power is based on the square root of the product of the heritability and sample size for the traits having a value >4500.[41] The power was considered to be "good" or better for 47 out of 56 bivariate analyses between quantitative diabetes- and quantitative glaucoma-related traits (see **Supplemental Table 1).** There was one nominal positive association between IOP measured in the IGGC and FBS with subpar power (r_g =0.23; p=0.0075; power product=3917) but more adequately powered associations between IOPg and FBS and IOPcc and FBS were definitely null (p≥0.47; power product ≥ 6772; see **Table 4** and **Supplemental Table 1**). T2D did not show any significant genetic correlations with any of the seven quantitative glaucoma-related traits (p≥0.16) and for all of these bivariate analyses there was at least "good" power to observe such an association (power product ≥ 4800; Supplemental Table 3). POAG and T2D are categorical traits and the analysis for genetic correlation between

them was slightly underpowered (power product=3957); yet, the result was in the inverse direction (r_g=-0.14) and not significant (p=0.16). Our findings using GWAS statistics were consistent with individual level data from two population pedigrees and do not support a genetic relationship between diabetes and glaucoma.

Our result showing a non-significant inverse genetic correlation between T2D and POAG runs contrary to the significant positive correlation between these quantitative traits in a Japanese population.[25] The numbers of cases in the genome-wide datasets were comparable between the Asian and our European sample so power differences were unlikely but there could be differences in genetic structure between these groups that account for these differences. For example, *LOXL1* was found to be a genome-wide marker for POAG in Japanese subjects,[25] but to date *LOXL1* markers are not associated with POAG in European-derived Caucasians.[45] Using the same Japanese population, Kinai et al. did not find significant genetic correlations between diabetes quantitative traits (HDL, LDL, TG, blood sugar, and HbA1c) and glaucoma, a finding consistent with our results.[26] Furthermore, in a US-based multiethnic population, a panel of genome-wide genetic biomarkers for T2D were not associated with POAG.[27]

Several diabetes quantitative traits are positively related to IOP in epidemiological studies;[1-6] yet, we find no genetic correlations between these quantitative diabetes traits and IOP. Overall, while CCT is increased in patients with diabetes based on several studies,[46-48] this corneal feature only partially mediated IOP variation in a study from Singapore.[6] While CCT is a static biophysical parameter, CH and CRF are dynamic biomechanical properties that are also affected by diabetes control.[49, 50] Overall, accounting for CCT, CH and CRF may not completely explain how the diabetic process leads to increased IOP as measured by Goldman applanation tonometry. Nonetheless, the large UK BioBank study suggests there is no relationship between self-reported diabetes and cornea-compensated IOP.[9] Of course, both epidemiological[51] and genetic correlation analysis[24] strongly link IOP to POAG risk, and our study affirms the latter regardless of how IOP is measured. Yet the genetic correlations between any corneal phenotype (CCT, CRF and CH) and POAG are not significant. Furthermore, while genetic correlations between IOP measured in the IGGC and corneal phenotypes and between IOPg and corneal phenotypes are all high, there was no correlation between IOPcc and CCT. Overall these data suggest that from a genetic perspective CCT, CH and CRF quantify features unrelated to POAG, although they may be related to POAG phenotypically.

The epidemiological association between diabetes and glaucoma is somewhat more controversial but most studies indicate a positive association between the two conditions.[52] Our genetic correlation study, which is relatively free of bias related to reverse causation or disease detection, indicates a non-significant inverse genetic correlation between T2D and POAG. Furthermore, genetic correlations between IOP and T2D and between CDR and T2D are also null despite adequate power (power product ≥ 4800; **Supplemental Table 1**). Notably, we found strong genetic correlations between CDR and POAG despite only modest power (power product = 4400; **Supplemental Table 1**) and modest but non-significant correlations between CDR and IOP, suggesting that, from a genetic perspective, T2D genetic markers are largely not shared with POAG in European populations. These genetic findings may not be applicable to people of other ancestry but do seem adequately powered to address our study question and call for more prospective study of the relationship between diabetes and POAG using a population that is free of disease at baseline and is systematically monitored for both conditions.

Several longitudinal studies found a modest positive association between measured BMI and IOP,[53-55] while epidemiological studies of the relation between BMI and incident POAG had

mixed results.[56, 57] Furthermore some studies suggest that components of the metabolic syndrome are associated with open-angle glaucoma[58] but this association may vary by BMI status.[59] BMI is a readily obtainable phenotype with the largest summary GWAS data set available among the traits we studied.[33] There is strong genetic correlation between BMI and T2D (r_g=0.35; SE=0.04; p=4.0E-15; **Supplemental Table 4**) but no significant correlation between BMI and any of the glaucoma-related traits (p≥0.099; **Table 3**). These findings suggest that if BMI or metabolic syndrome plays a role in POAG pathogenesis, they may do so through intermediary effects on the glaucomatous process that are not measured in this study.

While these results do not support genetic correlations between diabetes and glaucoma, there are several non-genetic explanations that can be advanced in support of a positive relation between diabetes and glaucoma. For example, it is possible that hyperglycemia leads to the accumulation of advanced-glycation end products[60] and fibronectin production[61] in the trabecular meshwork leading to increased IOP in patients with T2D. Several reports indicate that experimental diabetes exacerbates IOP-induced optic damage;[62-64] however, there is contrary evidence that hyperglycemia was neuroprotective in a rodent model of glaucoma.[65] Finally there is an anecdotal report of a rhesus monkey with spontaneous diabetes, elevated IOP, diabetic retinopathy and glaucoma.[66]

This study has strengths and weaknesses. Strengths include the use of LD score regression, a novel unbiased approach, to assess correlations between many traits where strong positive associations are suspected such as IOP and POAG[24] and others where there is controversy such as T2D and POAG.[25,26] Furthermore, our genetic correlation analysis between diabetes and glaucoma was extensive as we considered nine diabetes- and eight glaucoma-related traits. We included some studies where the genetic architecture for continuous traits were ascertained in populations where the prevalence of the respective related diseases (T2D and POAG) was minimized. Such approaches allow for the unbiased detection of novel physiologic loci that might be disease-related as well cross-correlated with another disease. The absence of major genetic correlations between diabetes- and glaucoma-related traits is corroborated by pedigree data obtained in two cohorts. In addition, we leveraged the largest available samples of genetic data on diabetes- and glaucoma-related traits which were largely adequately powered. The cross-correlations within diabetes traits and within glaucoma traits produced expected results. For example, we estimated a strong inverse relation between HDL and T2D (r_g = -0.40; SE = 0.06; p = 4.2E-11; **Supplemental Table 4**), and a strong positive genetic correlation between IOP measured in various ways and POAG, as previously reported.[24] Weaknesses include the fact that the study was limited to European populations although some, but not all, data from Japan are consistent with our findings.[26] Second, the absence of a statistically significant genetic correlation does not rule out that a minority of genes are truly shared between diabetes- and glaucoma-related traits.

In summary, we found no genetic correlations between comprehensive sets of diabetes- and glaucoma-related traits. These findings were supported in analyses from two island-based cohorts designed to estimate genetic, environmental and phenotypic correlations in directly measured traits that is informed by pedigree data. T2D and related quantitative traits also do not share significant genome-wide SNP heritability with POAG or its related traits. It is therefore reasonable to consider non-genetic factors, including ones that affect the biomechanical properties of the cornea and perhaps even the optic nerve, as mediating the epidemiological associations between diabetes and elevated IOP or POAG. These findings have important implications for our understanding of POAG.

Acknowledgements/Disclosures:

a: Funding/Support: NIH EY015473 (LRP), P30 EY014104 (JLW) and EY022305 (JLW) supported this work. AK is supported by a Moorfields Eye Charity Career Development Fellowship. SM was supported by an Australian Research Council Fellowship. SM, DAM and AWH acknowledge grants 1150144 and 1116360 from the Australian National Health and Medical Research Council.

b: Financial disclosures: The authors have no conflicts of interest to declare with respect to the work described. Unrelated to this work, Dr. Pasquale is a consultant for Bausch & Lomb, Verily and Eyenovia. Anthony Khawaja is a consultant to Allergan and Novartis, has received travel expenses from Thea and has received lecturing fees from Grafton Optical.

c: Other acknowledgements:

ORCADES Acknowledgements:

The Orkney Complex Disease Study (ORCADES) was supported by the Chief Scientist Office of the Scottish Government (CZB/4/276, CZB/4/710), a Royal Society URF to J.F.W., the MRC Human Genetics Unit quinquennial programme "QTL in Health and Disease", Arthritis Research UK and the European Union framework program 6 EUROSPAN project (contract no. LSHG-CT-2006-018947). DNA extractions were performed at the Wellcome Trust Clinical Research Facility in Edinburgh. We would like to acknowledge the invaluable contributions of the research nurses in Orkney, the administrative team in Edinburgh and the people of Orkney.

VIKING Acknowledgements:
The Viking Health Study – Shetland (VIKING) was supported by the MRC Human Genetics Unit quinquennial programme grant "QTL in Health and Disease". DNA extractions and genotyping were performed at the Edinburgh Clinical Research Facility, University of Edinburgh. We would like to acknowledge the invaluable contributions of the research nurses in Shetland, the administrative team in Edinburgh and the people of Shetland.

References

1. Zhao D, Cho J, Kim MH, Friedman DS, Guallar E. Diabetes, fasting glucose, and the risk of glaucoma: a meta-analysis. *Ophthalmology* 2015;122(1):72-8.
2. Yokomichi H, Kashiwagi K, Kitamura K, et al. Evaluation of the associations between changes in intraocular pressure and metabolic syndrome parameters: a retrospective cohort study in Japan. *BMJ Open* 2016;6(3):e010360.
3. Hashemi H, Khabazkhoob M, Emamian MH, Shariati M, Yekta A, Fotouhi A. Distribution of intraocular pressure and its determinants in an Iranian adult population. *Int J Ophthalmol* 2016;9(8):1207-14.
4. Cohen E, Kramer M, Shochat T, Goldberg E, Krause I. Relationship between serum glucose Levels and intraocular pressure, a population-based cross-sectional study. *J Glaucoma* 2017;26(7):652-6.
5. Bekmez S, Kocaturk T. Higher intraocular pressure levels associated with lower hysteresis In type 2 diabetes. *Open Ophthalmol J* 2018;12:29-33.
6. Luo XY, Tan NYQ, Chee ML, et al. Direct and indirect associations between diabetes and intraocular pressure: The Singapore Epidemiology of Eye Diseases Study. *Invest Ophthalmol Vis Sci* 2018;59(5):2205-11.
7. Goldich Y, Barkana Y, Gerber Y, et al. Effect of diabetes mellitus on biomechanical parameters of the cornea. *J Cataract Refract Surg* 2009;35(4):715-9.
8. Scheler A, Spoerl E, Boehm AG. Effect of diabetes mellitus on corneal biomechanics and measurement of intraocular pressure. *Acta Ophthalmol* 2012;90(6):e447-51.
9. Chan MP, Grossi CM, Khawaja AP, et al. Associations with Intraocular Pressure in a Large Cohort: Results from the UK Biobank. *Ophthalmology* 2016;123(4):771-82.
10. Zhao YX, Chen XW. Diabetes and risk of glaucoma: systematic review and a meta-analysis of prospective cohort studies. *Int J Ophthalmol* 2017;10(9):1430-5.
11. Hou H, Shoji T, Zangwill LM, et al. Progression of primary open-angle glaucoma in diabetic and nondiabetic patients. *Am J Ophthalmol* 2018;189:1-9.
12. Pimentel LG, Gracitelli CP, da Silva LS, Souza AK, Prata TS. Association between glucose levels and intraocular pressure: Pre- and postprandial analysis in diabetic and nondiabetic patients. *J Ophthalmol* 2015;2015:832058.
13. Wu CJ, Fang WH, Kao TW, et al. Postprandial Glucose as a Risk Factor for Elevated Intraocular Pressure. *PloS One* 2016;11(12):e0168142.
14. Chun YH, Han K, Park SH, et al. Insulin resistance is associated with intraocular pressure elevation in a non-obese Korean population. *PLoS One* 2015;10(1):e112929.
15. Fujiwara K, Yasuda M, Ninomiya T, et al. Insulin Resistance Is a Risk Factor for Increased Intraocular Pressure: The Hisayama Study. *Invest Ophthalmol Vis Sci* 2015;56(13):7983-7.
16. Klein BE, Klein R, Linton KL. Intraocular pressure in an American community. The Beaver Dam Eye Study. *Invest Ophthalmol Vis Sci* 1992;33(7):2224-8.
17. Colditz GA, Willett WC, Stampfer MJ, et al. Weight as a risk factor for clinical diabetes in women. *Am J Epidemiol* 1990;132:501-13.
18. Colditz GA, Willett WC, Rotnitzky A, Manson JE. Weight gain as a risk factor for clinical diabetes mellitus in women. *Ann Intern Med* 1995;122(7):481-6.
19. Hu FB, Manson JE, Stampfer MJ, et al. Diet, lifestyle, and the risk of type 2 diabetes mellitus in women. *N Engl J Med 2001*;345(11):790-7.

20. Chua J, Chee ML, Chin CWL, et al. Inter-relationship between ageing, body mass index, diabetes, systemic blood pressure and intraocular pressure in Asians: 6-year longitudinal study. *Br J Ophthalmol* 2018;103(2):196-202.
21. Bulik-Sullivan B, Finucane HK, Anttila V, et al. An atlas of genetic correlations across human diseases and traits. *Nat Genet* 2015;47(11):1236-41.
22. Bulik-Sullivan BK, Loh PR, Finucane HK, et al. LD Score regression distinguishes confounding from polygenicity in genome-wide association studies. *Nat Genet* 2015;47(3):291-5.
23. Pickrell JK, Berisa T, Liu JZ, Segurel L, Tung JY, Hinds DA. Detection and interpretation of shared genetic influences on 42 human traits. *Nat Genet* 2016;48(7):709-17.
24. Aschard H, Kang JH, Iglesias AI, et al. Genetic correlations between intraocular pressure, blood pressure and primary open-angle glaucoma: a multi-cohort analysis. *Eur J Hum Genet* 2017;25(11):1261-7.
25. Shiga Y, Akiyama M, Nishiguchi KM, et al. Genome-wide association study identifies seven novel susceptibility loci for primary open-angle glaucoma. *Hum Mol Genet* 2018;27(8):1486-96.
26. Kanai M, Akiyama M, Takahashi A, et al. Genetic analysis of quantitative traits in the Japanese population links cell types to complex human diseases. *Nat Genet* 2018;50(3):390-400.
27. Shen L, Walter S, Melles RB, Glymour MM, Jorgenson E. Diabetes pathology and risk of primary open-angle glaucoma: Evaluating causal mechanisms by using genetic information. *Am J Epidemiol* 2016;183(2):147-55.
28. Zheng J, Erzurumluoglu AM, Elsworth BL, et al. LD Hub: a centralized database and web interface to perform LD score regression that maximizes the potential of summary level GWAS data for SNP heritability and genetic correlation analysis. *Bioinformatics* 2017;33(2):272-9.
29. Scott RA, Lagou V, Welch RP, et al. Large-scale association analyses identify new loci influencing glycemic traits and provide insight into the underlying biological pathways. *Nat Genet* 2012;44(9):991-1005.
30. Saxena R, Hivert MF, Langenberg C, et al. Genetic variation in GIPR influences the glucose and insulin responses to an oral glucose challenge. *Nat Genet* 2010;42(2):142-8.
31. Soranzo N, Sanna S, Wheeler E, et al. Common variants at 10 genomic loci influence hemoglobin A(1)(C) levels via glycemic and nonglycemic pathways. *Diabetes* 2010;59(12):3229-39.
32. Teslovich TM, Musunuru K, Smith AV, et al. Biological, clinical and population relevance of 95 loci for blood lipids. *Nature* 2010;466(7307):707-13.
33. Locke AE, Kahali B, Berndt SI, et al. Genetic studies of body mass index yield new insights for obesity biology. *Nature* 2015;518(7538):197-206.
34. Morris AP, Voight BF, Teslovich TM, et al. Large-scale association analysis provides insights into the genetic architecture and pathophysiology of type 2 diabetes. *Nat Genet* 2012;44(9):981-90.
35. Hysi PG, Cheng CY, Springelkamp H, et al. Genome-wide analysis of multi-ancestry cohorts identifies new loci influencing intraocular pressure and susceptibility to glaucoma. *Nat Genet* 2014;46(10):1126-30.
36. Springelkamp H, Hohn R, Mishra A, et al. Meta-analysis of genome-wide association studies identifies novel loci that influence cupping and the glaucomatous process. *Nat Commun* 2014;5:4883.

37. Bailey JN, Loomis SJ, Kang JH, et al. Genome-wide association analysis identifies TXNRD2, ATXN2 and FOXC1 as susceptibility loci for primary open-angle glaucoma. *Nat Genet* 2016;48(2):189-94.
38. Khawaja AP, Cooke Bailey JN, Wareham NJ, et al. Genome-wide analyses identify 68 new loci associated with intraocular pressure and improve risk prediction for primary open-angle glaucoma. *Nat Genet* 2018;50(6):778-82.
39. Toh T, Liew SH, MacKinnon JR, et al. Central corneal thickness is highly heritable: the twin eye studies. *Invest Ophthalmol Vis Sci* 2005;46(10):3718-22.
40. Blanco-Gomez A, Castillo-Lluva S, Del Mar Saez-Freire M, et al. Missing heritability of complex diseases: Enlightenment by genetic variants from intermediate phenotypes. *BioEssays* 2016;38(7):664-73.
41. Anttila V, Bulik-Sullivan B, Finucane HK, et al. Analysis of shared heritability in common disorders of the brain. *Science* 2018;360(6395). pii: eaap8757. doi: 10.1126/science.aap8757
42. Finucane HK, Bulik-Sullivan B, Gusev A, et al. Partitioning heritability by functional annotation using genome-wide association summary statistics. *Nat Genet* 2015;47(11):1228-35.
43. McQuillan R, Leutenegger AL, Abdel-Rahman R, et al. Runs of homozygosity in European populations. *Am J Hum Genet* 2008;83(5):359-72.
44. Clegg FJ. Some factors affecting marital isonymy in three Outer Hebridean parishes. *Ann Hum Biol* 2001;28(4):455-66.
45. Wiggs JL, Yaspan B, Hauser M, et al. Common variants at 9p21 and 8q22 are associated with increased susceptibility to optic nerve degenerationin glaucoma. *PLoS Genetics* 2012;8(4):e1002654.
46. Storr-Paulsen A, Singh A, Jeppesen H, Norregaard JC, Thulesen J. Corneal endothelial morphology and central thickness in patients with type II diabetes mellitus. *Acta Ophthalmol* 2014;92(2):158-60.
47. Briggs S, Osuagwu UL, AlHarthi EM. Manifestations of type 2 diabetes in corneal endothelial cell density, corneal thickness and intraocular pressure. *J Biomed Res* 2015;30 doi: 10.7555/JBR.29.20140075.
48. Sanchis-Gimeno JA, Alonso L, Rahhal M, Bastir M, Perez-Bermejo M, Belda-Salmeron L. Corneal thickness differences between type 2 diabetes and non-diabetes subjects during preoperative laser surgery examination. *Journal Diabetes Complications* 2017;31(1):209-12.
49. Perez-Rico C, Gutierrez-Ortiz C, Gonzalez-Mesa A, Zandueta AM, Moreno-Salgueiro A, Germain F. Effect of diabetes mellitus on Corvis ST measurement process. *Acta Ophthalmol* 2015;93(3):e193-8.
50. Yazgan S, Celik U, Kaldirim H, et al. Evaluation of the relationship between corneal biomechanic and HbA1C levels in type 2 diabetes patients. *Clin Ophthalmol* 2014;8:1549-53.
51. Sommer A, Tielsch JM, Katz J, et al. Relationship between intraocular pressure and primary open angle glaucoma among white and black Americans. The Baltimore Eye Survey. *Arch Ophthalmol* 1991;109(8):1090-5.
52. Tham YC, Cheng CY. Associations between chronic systemic diseases and primary open angle glaucoma: an epidemiological perspective. Clin Exp Ophthalmol 2017;45(1):24-32.
53. Wu SY, Nemesure B, Hennis A, Leske MC. Nine-year changes in intraocular pressure: the Barbados Eye Studies. *Arch Ophthalmol* 2006;124(11):1631-6.

54. Zhao D, Kim MH, Pastor-Barriuso R, et al. A Longitudinal Study of Association between adiposity markers and intraocular pressure: The Kangbuk Samsung Health Study. *PLoS One* 2016;11(1):e0146057.
55. Han X, Yang T, Zhang J, et al. Longitudinal changes in intraocular pressure and association with systemic factors and refractive error: Lingtou Eye Cohort Study. *BMJ Open* 2018;8(2):e019416.
56. Pasquale LR, Willett WC, Rosner BA, Kang JH. Anthropometric measures and their relation to incident primary open-angle glaucoma. *Ophthalmology* 2010;117(8):1521-9.
57. Springelkamp H, Wolfs RC, Ramdas WD, et al. Incidence of glaucomatous visual field loss after two decades of follow-up: the Rotterdam Study. *Eur J Epidemiol* 2017;32(8):691-9.
58. Newman-Casey PA, Talwar N, Nan B, Musch DC, Stein JD. The relationship between components of metabolic syndrome and open-angle glaucoma. *Ophthalmology* 2011;118(7):1318-26.
59. Kim HA, Han K, Lee YA, Choi JA, Park YM. Differential association of metabolic risk factors with open angle glaucoma according to obesity in a Korean population. *Sci Rep* 2016;6:38283.
60. Park CH, Kim JW. Effect of advanced glycation end products on oxidative stress and senescence of trabecular meshwork cells. *Kor J Ophthalmol* 2012; 26(2):123-31.
61. Sato T, Roy S. Effect of high glucose on fibronectin expression and cell proliferation in trabecular meshwork cells. *Invest Ophthalmol Vis Sci* 2002;43(1):170–5.
62. Kanamori A, Nakamura M, Mukuno H, et al. Diabetes has an additive effect on neural apoptosis in rat retina with chronically elevated intraocular pressure. *Curr Eye Res* 2004; 28(1):47–54.
63. Chysostomous V, vanWijngaarden P, Steinberg GR, Crowston JG. A short term high-fat high-sucrose diet in mice impairs optic nerve recovery after injury and this is not reversed by exercise. *Exp Eye Res* 2017; 162:104-9.
64. Aleman-Flores R, Mompeo-Corredera B. Glial Cells and Retinal Nerve Fibers Morphology in the Optic Nerves of Streptozotocin-induced Hyperglycemic Rats. J Ophthalmic Vis Res 2018;13(4):433-8.
65. Ebneter A, Chidlow G, Wood JP, Casson RJ. Protection of retinal ganglion cells and the optic nerve during short-term hyperglycemia in experimental glaucoma. *Arch Ophthalmol* 2011; 129(10);1337-44
66. Johnson MA, Lutty GA, McLeod DS, et al. Ocular structure and function in an aged monkey with spontaneous diabetes mellitus. *Exp Eye Res* 2005;80(1):37-42.