Received: 9 April 2024 | Accepted: 22 June 2024

DOI: 10.1111/aos.16737

Acta Ophthalmologica

ORIGINAL ARTICLE

Longitudinal associations of retinal vessel morphology with intraocular pressure and blood pressure at follow-up visit—Findings from a Danish eye and vision cohort, Project FOREVER

Josefine Freiberg[1] | **Roshan Welikala**[2] | **Jens Rovelt**[1] | **Sarah A. Barman**[2] | **Christopher G. Owen**[3] | **Alicja R. Rudnicka**[3] | **Miriam Kolko**[1,4] | **on behalf of the FOREVER consortium**

[1]Department of Drug Design and Pharmacology, University of Copenhagen, Copenhagen, Denmark

[2]School of Computer Science and Mathematics, Kingston University, Surrey, UK

[3]Population Health Research Institute, St. George's, University of London, London, UK

[4]Department of Ophthalmology, Copenhagen University Hospital, Rigshospitalet, Glostrup, Copenhagen, Denmark

Correspondence
Miriam Kolko, Department of Drug Design and Pharmacology, University of Copenhagen, Copenhagen, Denmark.
Email: miriamk@sund.ku.dk

Abstract

Purpose: To characterise the retinal vasculometry of a Danish eye and vision cohort and examine associations with systolic blood pressure (BP), diastolic BP, mean arterial BP, and intraocular pressure (IOP).

Design: Longitudinal study.

Methods: The retinal vasculature of fundus images from the FOREVER (Finding Ophthalmic Risks and Evaluating the Value of Eye exams and their predictive Reliability) cohort was analysed using a fully automated image analysis program. Longitudinal associations of retinal vessel morphology at follow-up visit with IOP (baseline and follow-up) and BP (follow-up) were examined using multilevel linear regression models adjusting for age, sex and retinal vasculometry at baseline as fixed effects and person as random effect. Width measurements were additionally adjusted for the spherical equivalent.

Results: A total of 2089 subjects (62% female) with a mean age of 61 (standard deviation 8) years and a mean follow-up period of 4.1 years (SD 0.6 years) were included. The mean arteriolar diameter was approximately 20% thinner than the mean venular diameter, and venules were about 21%–23% less tortuous than arterioles. BP at follow-up was associated with decreased arteriolar diameter from baseline to follow-up. After adjusting for baseline IOP, IOP at follow-up was associated with increased arteriolar tortuosity above baseline (0.59%, 95% CI 0.08–1.10, *p*-value 0.024).

Conclusion: In a Danish eye and vision cohort, variations in BP and alterations in IOP over time were associated with changes in the width and tortuosity of retinal vessels. Our findings contribute novel insights into retinal vascular alterations over time.

KEYWORDS
automated retinal image analysis, longitudinal cohort study, retinal vasculometry

1 | INTRODUCTION

Systemic as well as ocular conditions can manifest in the retina, making understanding retinal vascular health a valuable tool for assessing risk markers of disease. The retina is often referred to as a mirror of the brain and circulatory system (Kumar, 2018; London et al., 2013; Ptito et al., 2021), and pathophysiological changes in retinal vasculometry are widely recognised as being associated with various conditions, such as systemic hypertension (Modi & Arsiwalla, 2024; Wong & Mitchell, 2004).

Retinal fundus images provide a unique and non-invasive way to visualise the vascular system, and it is routinely included in eye examinations (Mishra & Tripathy, 2023). In recent years, the utilisation of retinal fundus images in optician settings, particularly in

the United Kingdom and Scandinavia, has created large data sources. These data sources provide detailed information and potential insights into the health of the microcirculatory system. As people often revisit their opticians when refractive alterations occur, their eye exams are a valuable resource for studying vascular health and, in particular, retinal vasculometry changes over time. In 2022, a new eye and vision cohort, Project FOREVER (Finding Ophthalmic Risk and Evaluating the Value of Eye exams and their predictive Reliability), was initiated, collecting comprehensive data from eye examinations from optician customers in Denmark (Freiberg, Rovelt, et al., 2023). The FOREVER cohort provides insight into the eye health of the Danish population as it contains longitudinal data from eye examinations, including retinal fundus images from optician customers across Denmark (Freiberg, Rovelt, et al., 2023).

Traditionally, subjective assessments of retinal fundus images, reliant on observer expertise, have been the norm, posing challenges in objective image comparison as well as scaling up for large population-based studies. However, objective and robust scientific methods are essential for research investigating retinal vasculometry as a potential biomarker for diseases. To address this need, software tools and algorithms have been developed for automated retinal vessel analysis and validated against human observers (Fraz et al., 2015; Zhou et al., 2022). QUARTZ (Quantitative Analysis of Retinal vessel Topology and siZe) is an automated image analysis software that creates retinal vessel maps and provides quantitative data on vasculometry measurements (Fraz et al., 2015). QUARTZ has previously been utilised in large-scale epidemiological studies exploring disease biomarkers and investigating the link between eye-, systemic-, and vascular health (Owen et al., 2019; Rudnicka et al., 2020, 2022; Tapp et al., 2019, 2020). Based on data from the UK Biobank and the European Prospective Investigation into Cancer (EPIC) Norfolk study, cross-sectional associations between retinal vasculometry and blood pressure (BP), as well as cardiovascular disease, diabetes, body mass index, and glaucoma have been examined previously, showing strong associations with a variety of diseases and risk factors (Owen et al., 2019; Rudnicka et al., 2020, 2022; Tapp et al., 2019, 2020). However, few longitudinal studies have examined changes in retinal vasculometry over time. In this study, we aimed to characterise the retinal vasculometry of the FOREVER cohort over time by employing the automated image analysis software QUARTZ. We investigated longitudinal changes in retinal vasculometry with BP at follow-up visit and intraocular pressure (IOP) from both baseline and follow-up visit, providing novel insights into possible vascular alterations over time.

2 | METHODS

Data from the Danish eye and vision cohort study, Project FOREVER, were analysed. A detailed description of the methodology and design of Project FOREVER have been described previously by Freiberg, Rovelt, et al. (2023). The study was approved by the National Committee on Health Research Ethics, Denmark (project ID H-21026000), and participants provided written informed consent before enrolment (Freiberg, Rovelt, et al., 2023).

2.1 | Physical examination

A subgroup of participants from the FOREVER cohort, aged above 50, underwent two consecutive eye examinations with a follow-up period of at least 3 years. BP was measured using a Microlife® BP B6 Connect BP monitor at the follow-up visit. Using the Microlife® Average Mode (MAM) function, the average of three consecutive measurements was calculated (Freiberg, Rovelt, et al., 2023). BP was measured in mmHg. Mean arterial BP was calculated based on systolic BP and diastolic BP, with mean arterial BP = diastolic BP + 1/3 × (systolic BP − diastolic BP) (DeMers & Wachs, 2024). The optician was instructed to measure BP 30 min after smoking, physical activity, food or coffee intake.

Of relevance to this study, eye examinations carried out in the optician stores included at both visits: Refraction measured with auto kerato-refractometer (TopCon KR-800), IOP and corneal thickness measured using non-contract tonometry and pachymeter (Canon TX-20-P), and 45° field-of-view, macular-centred, non-mydriatic retinal fundus images captured with the Canon CR-2 AF using Canon EOS 70D and Canon EOS 80D cameras (see Figure 1 for illustration of data collection) (Freiberg, Rovelt, et al., 2023; Freiberg, Welikala, et al., 2023).

The spherical equivalent refractive error was calculated as sphere power plus half the cylinder power (Enaholo et al., 2024). Best visual acuity was measured on the LogMAR scale. IOP was measured in mmHg, and corneal thickness was in μm (Freiberg, Rovelt, et al., 2023).

2.2 | Retinal image processing

The retinal vasculometry of retinal fundus images was analysed using the QUARTZ software. QUARTZ has previously been calibrated and validated to the FOREVER image dataset using an anonymised FOREVER Pilot dataset of 3682 images and demonstrated good accuracy and reliability across all quantified parameters (Freiberg, Welikala, et al., 2023). For a thorough description of the validation of QUARTZ on the FOREVER Pilot dataset, see Freiberg, Welikala, et al. (2023). The level of performance of the different retinal vasculometry parameters measured was aligned with or superior to the previous performance of QUARTZ when evaluated on the UK Biobank retinal image dataset (Freiberg, Welikala, et al., 2023; Welikala et al., 2017).

The width and tortuosity of all arterioles and venules were obtained from the entire retinal image by analysing the images using QUARTZ. Only images of adequate quality and segments with widths within 4 standard deviations (SDs) from the mean of all widths were included. Segments without information on tortuosity and

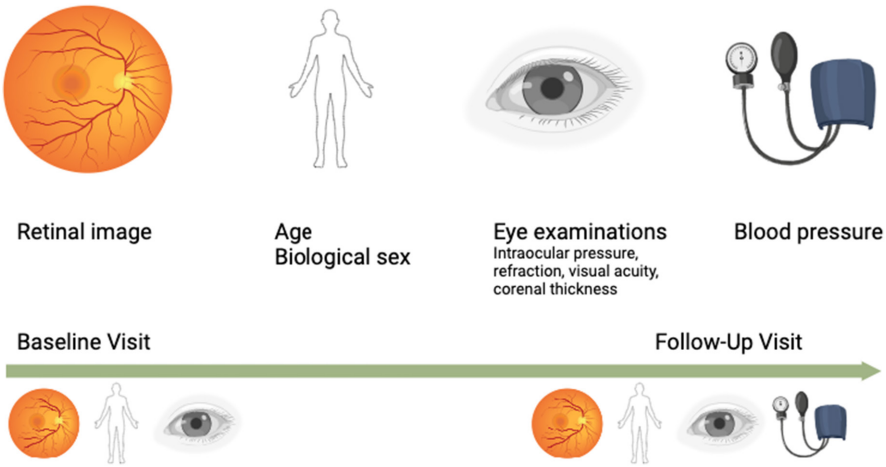

FIGURE 1 Illustration of data collection. At baseline and follow-up visit, retinal images, information about age and sex and eye examinations including intraocular pressure, refraction, visual acuity, and corneal thickness were collected. At follow-up visit, blood pressure was measured additionally. Figure created with Biorender.

segments with an arteriole or venule probability below 0.80 were excluded from the analysis. For every image, the mean width and mean tortuosity for arterioles and venules were calculated and weighted by the segment length of each vessel. Width measurements were provided in pixels, and tortuosity was quantified using arbitrary units.

2.3 | Statistical analysis

The software R and RStudio were used for statistical analysis (R Core Team, 2020). Retinal arteriolar and venular widths were normally distributed, whereas tortuosity measures were positively skewed and log-transformed.

Cross-sectional analyses were conducted using data from the most recent visit. Multilevel linear regression models were employed to investigate associations between retinal vasculometry and systolic BP, diastolic BP, mean arterial BP or IOP. Model 1 adjusted for age and sex as fixed effects and person as random effect, allowing for right and left eye data from the same person to contribute to the analyses. For width measurements (in pixels), model 2 was further adjusted for spherical equivalent at follow-up as fixed effect to investigate the stability of width associations with risk factors. Age, systolic BP, diastolic BP, mean arterial BP, and IOP were included as continuous variables. For both cross-sectional and longitudinal analyses, associations were estimated per decade rise in age and per 10 mmHg rise in systolic BP, diastolic BP and mean arterial BP. Tortuosity associations with covariates were converted to percentage differences due to the log transformation of tortuosity (the dependent variable) per specified rise in covariates or as the percentage difference between groups.

Data from baseline and follow-up visits at least 3 years later were used for longitudinal analyses. Pearson's correlation coefficient was computed to explore the linear relationship between baseline and follow-up retinal vasculometry measures and IOP. Retinal vasculometry at follow-up (adjusted for baseline values) with IOP at baseline and systolic BP, diastolic BP, mean arterial BP, and IOP at follow-up were examined and adjusted for the same confounders as in model 1 and 2 (model 3 and 4).

A similar approach was used to examine the retinal vasculometry at follow-up (adjusted for baseline values) and associations with change in IOP above baseline (models 5 and 6), adjusting for the same confounders as in models 1–4.

3 | RESULTS

3.1 | Descriptive statistics

A total of 2089 subjects were included in the analysis, with the majority being female (63%).

3.1.1 | Baseline

At the first visit, the mean age was 61 (SD 8) years. Between 1714 and 1978 subjects had ocular or retinal vascular measurements. The mean IOP, spherical equivalent, best corrected visual acuity, and corneal thickness were similar for the left and right eye (see Table 1). For both eyes, the width of the arterioles was approximately 20% narrower than the venules, while the venules were between 21% and 23% less tortuous than the arterioles (see Table 1 for absolute numbers).

3.1.2 | Follow-up visit

The mean difference between baseline and follow-up visits was 4.1 years (SD 0.6 years), with a minimum follow-up of 3.0 and a maximum of 6.3 years. 1755 participants had their BP measured with a mean systolic BP of 142 mmHg (SD 19), a mean diastolic BP of 82 mmHg (SD 10) and a mean arterial BP of 102 mmHg (SD 12). Between 1812 and 2060 subjects had ocular or vascular measurements. All ocular measurements were similar for the right and left eye (see Table 1). The arterioles were thinner (by 19%–20%) than venules, and venules were

TABLE 1 Characteristics of the population at baseline and follow-up visit with age, sex, blood pressure, ocular measurements and vasculometry.

Characteristics				
		Baseline		Follow-up
Participants, *n*		2089		2089
Age in years, mean (SD)		61 (8)		65 (8)
Sex, *n* (% female)		1309 (63)		1309 (63)
Blood pressure				
Participants, *n*				1755
Systolic blood pressure in mmHg, mean (SD)				142 (19)
Diastolic blood pressure in mmHg, mean (SD)				82 (10)
Mean arterial pressure in mmHg, mean (SD)				102 (12)
	Baseline		**Follow-up**	
	N	**Mean (SD)**	***N***	**Mean (SD)**
Ocular measurements				
IOP OD in mmHg	1861	16.1 (3.5)	2027	15.9 (3.4)
IOP OS in mmHg	1867	16.1 (3.5)	2028	16.0 (3.4)
Δ IOP OD in mmHg			1812	−0.12 (2.52)
Δ IOP OS in mmHg			1818	−0.11 (2.57)
Refraction OD, spheric equivalent	1714	−0.4 (2.5)	1840	−0.2 (2.5)
Refraction OS, spheric equivalent	1717	−0.3 (2.5)	1828	−0.2 (2.5)
Best visual acuity OD, LogMar scale	1963	1.0 (0.2)	2050	1.0 (0.2)
Best visual acuity OD, LogMar scale	1970	1.0 (0.2)	2060	1.0 (0.2)
Corneal thickness OD in µm	1893	549 (35)	2000	554 (34)
Corneal thickness OS in µm	1894	549 (33)	2002	553 (35)
Retinal vasculometry				
Arteriolar width OD in pixels	1975	16.04 (1.69)	1922	16.15 (1.78)
Arteriolar width OS in pixels	1948	16.06 (1.63)	1901	16.24 (1.85)
Venular width OD in pixels	1978	20.06 (2.32)	1934	20.17 (2.35)
Venular width OS in pixels	1952	20.07 (2.33)	1915	20.16 (2.45)
Arteriolar tortuosity OD[a]	1958	1.38 (0.51)	1889	1.38 (0.52)
Arteriolar tortuosity OS[a]	1931	1.39 (0.50)	1868	1.40 (0.52)
Venular tortuosity OD[a]	1961	1.06 (0.30)	1901	1.07 (0.30)
Venular tortuosity OS[a]	1935	1.10 (0.29)	1882	1.10 (0.30)
Δ Arteriolar width OD in pixels			1842	0.12 (1.75)
Δ Arteriolar width OS in pixels			1796	0.19 (1.73)
Δ Venular width OD in pixels			1856	0.14 (2.07)
Δ Venular width OS in pixels			1812	0.10 (2.16)
Δ Arteriolar tortuosity ($\times 10^3$) OD[a]			1811	0.00 (0.39)
Δ Arteriolar tortuosity ($\times 10^3$) OS[a]			1766	0.00 (0.37)
Δ Venular tortuosity ($\times 10^3$) OD[a]			1825	0.02 (0.22)
Δ Venular tortuosity ($\times 10^3$) OS[a]			1782	0.01 (0.23)

Note: Blood pressure measurements and changes in intraocular pressure and retinal vasculometry measures were only provided for follow-up visits. Ocular measurements and vasculometry were provided for both eyes.

Abbreviations: OD, right eye; OS, left eye; SD, standard deviation; Δ, change from baseline to follow-up.

[a]Tortuosity = log(tortuosity × 10^3).

less tortuous (by 21%–22%) than arterioles. Between 1766 and 1856 subjects had data for IOP or retinal vasculometry measures from both baseline and follow-up visits. Mean IOP for both eyes decreased at the follow-up visit compared to the baseline visit, whereas all vasculometry measures increased at the follow-up visit (see Table 1 for exact numbers).

3.2 | Cross-sectional

3.2.1 | Width

Cross-sectional associations remained stable with increasing levels of adjustment (i.e., comparing Model 1 with Model 2 in Table 2). The absolute difference in

TABLE 2 Cross-sectional analyses at follow-up: absolute differences in arteriolar and venular vessel width from multilevel modelling adjusted for age and sex as fixed effects and person as random effects (Model 1). Model 2 further adjusted for spherical equivalent at follow-up. Estimates in pixels with 95% CI and *p*-value.

	Absolute difference in arteriolar width in pixels (95% CI)				Absolute difference in venular width in pixels (95% CI)			
	Model 1		Model 2		Model 1		Model 2	
Risk marker	Estimate	*p*-value	Estimate	*p*-value	Estimate	*p*-value	Estimate	*p*-value
Age per decade	−0.07 (−0.15 to 0.02)	0.136	−0.14 (−0.23 to −0.05)	**0.003**	0.18 (0.06 to 0.30)	**0.003**	0.00 (−0.12 to 0.12)	0.978
Sex (male)	−0.19 (−0.34 to −0.05)	**0.009**	−0.19 (−0.35 to −0.04)	**0.013**	−0.16 (−0.36 to 0.04)	0.107	−0.16 (−0.36 to 0.05)	0.130
SBP per 10 mmHg	−0.14 (−0.18 to −0.10)	$\mathbf{1.0\times10^{-11}}$	−0.13 (−0.17 to −0.09)	$\mathbf{2.1\times10^{-9}}$	−0.01 (−0.07 to 0.04)	0.656	0.00 (−0.05 to 0.06)	0.944
DBP per 10 mmHg	−0.32 (−0.39 to −0.24)	$\mathbf{1.3\times10^{-16}}$	−0.29 (−0.37 to −0.21)	$\mathbf{5.2\times10^{-13}}$	−0.06 (−0.16 to 0.05)	0.270	−0.02 (−0.13 to 0.08)	0.695
MAP per 10 mmHg	−0.26 (−0.32 to −0.20)	$\mathbf{3.0\times10^{-16}}$	−0.24 (−0.31 to −0.18)	$\mathbf{7.9\times10^{-13}}$	−0.04 (−0.13 to 0.05)	0.395	−0.01 (−0.1 to 0.08)	0.854
IOP per mmHg	−0.03 (−0.05 to −0.01)	**0.007**	−0.02 (−0.04 to −0.00)	**0.031**	−0.01 (−0.04 to 0.01)	0.259	0.00 (−0.03 to 0.02)	0.885

Note: Statistically significant *p*-values are highlighted in bold.
Abbreviations: DBP, diastolic blood pressure; IOP, intraocular pressure; MAP, mean arterial blood pressure; SBP, systolic blood pressure.

TABLE 3 Cross-sectional analyses at follow-up: Percentage differences in arteriolar and venular vessel tortuosity from multilevel modelling adjusted for age and sex as fixed effects and person as random effects (Model 1).

	Percentage difference in arteriolar tortuosity (95% CI)		Percentage difference in venular tortuosity (95% CI)	
	Model 1		Model 1	
Risk marker	Estimate	*p*-value	Estimate	*p*-value
Age per decade	2.64 (0.10 to 5.19)	**0.042**	−1.08 (−2.46 to 0.31)	0.129
Sex (male)	0.13 (−4.01 to 4.45)	0.952	−2.59 (−4.82 to −0.31)	**0.026**
SBP per 10 mmHg	0.97 (−0.23 to 2.17)	0.112	0.65 (0.00 to 1.31)	0.051
DBP per 10 mmHg	1.00 (−1.21 to 3.22)	0.374	−0.32 (−1.53 to 0.90)	0.608
MAP per 10 mmHg	1.25 (−0.61 to 3.11)	0.187	0.38 (−0.65 to 1.40)	0.471
IOP per mm Hg	0.09 (−0.48 to 0.66)	0.761	0.07 (−0.25 to 0. 39)	0.672

Note: Statistically significant *p*-values are highlighted in bold.
Abbreviations: CI, confidence interval; DBP, diastolic blood pressure; IOP, intraocular pressure; MAP, mean arterial blood pressure; SBP, systolic blood pressure.

arteriolar width decreased with age, and males had narrower arterioles than females (Table 2). Per 10 mmHg rise in systolic BP, arteriolar width decreased by −0.13 pixels (95% CI −0.17 to −0.09 pixels, $p<0.001$, model 2) and by −0.02 pixels per mmHg increase in IOP (95% CI −0.04 to −0.00 pixels, $p<0.05$, model 2). Venular width increased with age (0.18 pixels per 10 years [95% CI 0.06–0.30, $p<0.01$, model 1]). However, the age-related association was not significant after adjusting for spherical equivalent (model 2). None of the other variables (systolic BP, diastolic BP, mean arterial BP and IOP) were associated with significant differences in venular width (Table 2).

3.2.2 | Tortuosity

Arteriolar tortuosity increased with age (2.64% increase per 10 years, 95% CI 0.10–5.19, *p*-value <0.05), whereas venules were less tortuous for men than women (−2.59%, 95% CI −4.82 to −0.31%, *p*-value <0.05). For the other parameters, we observed no significant differences in tortuosity (Table 3).

3.3 | Longitudinal

3.3.1 | Width

Results from longitudinal associations revealed stability in effect sizes with different levels of adjustment (i.e., comparing model 3 with model 4 in Table 4). The correlation coefficient between baseline and follow-up retinal diameter was 0.44 and 0.55 for arterioles and venules, respectively. Increasing age at baseline was significantly associated with a reduction in arteriolar width above baseline (model 4). There was no statistically significant association with sex. All BP measurements at follow-up were associated with a negative change in arteriolar width above baseline. IOP at follow-up was associated with arteriolar width above baseline; however, it was only significant in model 3.

TABLE 4 Longitudinal analyses: Associations between absolute change in width above baseline per specified rise in risk markers. Model 3 adjusted for age, sex, and vessel width at baseline as fixed effect and person as random effect. Model 4 further adjusted for spherical equivalent at follow-up as fixed effect.

	Absolute change in arteriolar width in pixels above baseline (95% CI)				Absolute change in venular width in pixels above baseline (95% CI)			
	Model 3		Model 4		Model 3		Model 4	
Risk marker	Estimate	*p*-value	Estimate	*p*-value	Estimate	*p*-value	Estimate	*p*-value
Age baseline per decade	−0.08 (−0.16–0.00)	0.053	−0.15 (−0.23 to −0.06)	**0.001**	0.15 (0.04 to 0.25)	**0.006**	0.00 (−0.11 to 0.10)	0.945
Sex baseline (male)	−0.13 (−0.26 to 0.00)	0.055	−0.13 (−0.27 to 0.01)	0.064	−0.10 (−0.28 to 0.08)	0.262	−0.11 (−0.29 to 0.07)	0.218
SBP follow-up per 10 mmHg	−0.13 (−0.16 to −0.09)	**8.7×10^{-11}**	−0.12 (−0.16 to −0.08)	**3.7×10^{-9}**	−0.02 (−0.07 to 0.03)	0.519	−0.01 (−0.06 to 0.04)	0.767
DBP follow-up per 10 mmHg	−0.28 (−0.35 to −0.21)	**5.5×10^{-15}**	−0.26 (−0.33 to −0.19)	**2.9×10^{-12}**	−0.05 (−0.14 to 0.04)	0.270	−0.02 (−0.12 to 0.07)	0.607
MAP follow-up per 10 mmHg	−0.23 (−0.29 to −0.17)	**9.0×10^{-15}**	−0.22 (−0.28 to −0.16)	**3.0×10^{-12}**	−0.04 (−0.11 to 0.04)	0.340	−0.02 (−0.10 to 0.06)	0.659
IOP baseline per mmHg	−0.02 (−0.03 to 0.00)	0.080	−0.01 (−0.03 to 0.01)	0.177	0.00 (−0.02 to 0.02)	0.824	0.00 (−0.02 to 0.02)	0.882
IOP follow-up per mmHg	−0.02 (−0.04 to −0.01)	**0.008**	−0.02 (−0.03 to 0.00)	0.072	−0.02 (−0.04 to 0.00)	0.089	−0.01 (−0.03 to 0.01)	0.526

Note: Statistically significant *p*-values are highlighted in bold.

Abbreviations: CI, confidence interval; DBP, diastolic blood pressure; IOP, intraocular pressure; MAP, mean arterial blood pressure; SBP, systolic blood pressure.

TABLE 5 Longitudinal analyses: Associations between absolute change in width above baseline and absolute change in intraocular pressure above baseline. Model 5 adjusted for age, biological sex, vessel width, and intraocular pressure at baseline as fixed effect and person as random effect. Model 6 further adjusted for spherical equivalent at follow-up as fixed effect. Estimates in pixels with 95% CI and *p*-value.

	Absolute change in arteriolar width in pixels at follow-up (95% CI)				Absolute change in venular width in pixels from at follow-up (95% CI)			
	Model 5		Model 6		Model 5		Model 6	
Risk marker	Estimate	*p*-value	Estimate	*p*-value	Estimate	*p*-value	Estimate	*p*-value
Age per decade	−0.10 (−0.18 to −0.01)	**0.024**	−0.16 (−0.25 to −0.06)	**0.001**	0.14 (0.03 to 0.25)	**0.013**	−0.01 (−0.12 to 0.10)	0.869
Sex (male)	−0.14 (−0.28 to 0.00)	0.056	−0.14 (−0.29 to 0.01)	0.059	−0.12 (−0.30 to 0.06)	0.203	−0.13 (−0.31 to 0.06)	0.184
IOP BL per mmHg	−0.01 (−0.02 to 0.01)	0.595	−0.01 (−0.03 to 0.01)	0.592	0.01 (−0.02 to 0.03)	0.492	0.01 (−0.02 to 0.03)	0.480
IOP FU per mmHg	−0.02 (−0.04 to 0.00)	0.059	−0.01 (−0.03 to 0.01)	0.253	−0.02 (−0.04 to 0.00)	0.084	−0.01 (−0.04 to 0.01)	0.358

Note: Statistically significant *p*-values are highlighted in bold.

Abbreviations: CI, confidence interval; IOP, intraocular pressure.

Concerning the change in venular width above baseline, we found no significant associations for any risk markers after adjusting for spherical equivalent (model 4).

The correlation coefficient between IOP at baseline and follow-up was 0.70. When adjusting for both IOP at baseline and follow-up in the same model (models 5 and 6), the estimate for IOP at follow-up reflected the change in diameter associated with the change in IOP above baseline. As for a change in width above baseline and association with a change in IOP above baseline, we found no significant associations for arterioles or venules (Table 5).

3.3.2 | Tortuosity

The correlation coefficient between baseline and follow-up tortuosity was 0.66 and 0.56 for arterioles and venules, respectively. The arteriolar tortuosity above baseline increased with age (2.26% per decade [95% 0.40–4.13, *p*-value <0.05]). Neither BP, IOP at follow-up, nor IOP at baseline were significantly associated with a change in arteriolar tortuosity above baseline (model 3, Table 6).

As for a change in venular tortuosity above baseline, it was significantly associated with sex. Compared with females, males had a negative change in venular tortuosity

TABLE 6 Longitudinal analyses: Associations between percentage change in tortuosity above baseline and various risk factors. Model 3 adjusted for age, sex, and tortuosity at baseline as fixed effects and person as random effect. Estimates in percentage with 95% CI and *p*-value.

	Percentage change in arteriolar tortuosity above baseline (95% CI)		Percentage change in venular tortuosity above baseline (95% CI)	
	Model 3		Model 3	
Risk factor	**Estimate, % (95% CI)**	***p*-value**	**Estimate, % (95% CI)**	***p*-value**
Age per decade	2.26 (0.40 to 4.13)	**0.017**	−1.00 (−2.10 to 0.10)	0.075
Sex (male)	1.03 (−2.04 to 4.19)	0.515	−1.91 (−3.69 to −0.09)	**0.040**
SBP per 10 mmHg	0.83 (−0.04 to 1.71)	0.062	0.36 (−0.16 to 0.88)	0.177
DBP per 10 mmHg	0.88 (−0.74 to 2.49)	0.288	−0.30 (−1.51 to 0.91)	0.627
MAP per 10 mmHg	1.08 (−0.28 to 2.44)	0.119	0.36 (−0.66 to 1.38)	0.491
IOP baseline per mmHg	−0.14 (−0.55 to 0.27)	0.509	0.14 (−0.11 to 0.39)	0.276
IOP follow-up per mmHg	0.31 (−0.09 to 0.72)	0.129	−0.08 (−0.32 to 0.17)	0.542

Note: Statistically significant *p*-values are highlighted in bold.
Abbreviations: CI, confidence interval; DBP, diastolic blood pressure; IOP, intraocular pressure; MAP, mean arterial blood pressure; SBP, systolic blood pressure.

TABLE 7 Longitudinal analyses: Associations between percentage change in tortuosity above baseline and percentage change in intraocular pressure above baseline. Model 5 adjusted for age, sex, tortuosity at baseline and intraocular pressure at baseline as fixed effect, and person as random effect. Estimates in percentage with 95% CI and *p*-value.

	Percentage change in arteriolar tortuosity above baseline (95% CI)		Percentage change in venular tortuosity above baseline (95% CI)	
	Model 5		Model 5	
Risk factor	**Estimate % (95% CI)**	***p*-value**	**Estimate % (95% CI)**	***p*-value**
Age per decade	**2.18 (0.19 to 4.19)**	**0.032**	−1.25 (−2.43 to −0.06)	**0.039**
Sex (male)	1.67 (−1.64 to 5.08)	0.327	−1.88 (−3.80 to 0.08)	0.059
IOP baseline per mmHg	−0.41 (−0.91 to 0.09)	0.106	0.32 (0.00 to 0.63)	**0.047**
IOP follow-up per mmHg	0.59 (0.08 to 1.10)	**0.024**	−0.32 (−0.63 to 0.00)	0.051

Note: Statistically significant *p*-values are highlighted in bold.
Abbreviations: CI, confidence interval; IOP, intraocular pressure.

above baseline by −1.91% (95% CI −3.69 to −0.09, *p*-value <0.05) (Table 6).

When adjusting for IOP at baseline and follow-up in the same model (models 5 and 6), the estimate for IOP at follow-up reflected the change in tortuosity associated with the change in IOP above baseline. We found significant associations for both arteriolar and venular tortuosity and longitudinal changes in IOP. For arterioles, we found that an increase in 1 mmHg IOP at follow-up adjusted for baseline IOP was associated with 0.59% (95% 0.08 to 1.10, *p*-value <0.05) increase in tortuosity above baseline. For venules, an increase of 1 mmHg baseline IOP was associated with a 0.32% (95% CI 0.00–0.63, *p*-value <0.05) change in tortuosity above baseline after adjustment for follow-up IOP (Table 7).

4 | DISCUSSION

This paper provides insights into the interplay between retinal vasculometry and both IOP and BP, particularly by providing novel insights into longitudinal changes in retinal vasculometry. To establish the validity of the cohort, we examined cross-sectional associations to ensure that the associations were in the same direction as previously documented in other cohorts.

In our cross-sectional analysis, we found that mean arteriolar width decreased with increasing systolic BP (−0.80% per 10 mmHg), diastolic BP (−1.79% per 10 mmHg) and mean arterial BP (−1.48 per 10 mmHg). These findings are consistent with previous studies in retinal vasculometry using the QUARTZ software. Based on a subset of 54 714 subjects from the UK Biobank, decreased arteriolar width was associated with increased systolic BP (−1.02% per 10 mmHg), diastolic BP (−1.89% per mmHg) and mean arterial BP (−1.72% per mmHg) (Tapp et al., 2019). Another study based on cross-sectional data from 5947 subjects from the EPIC-Norfolk study, found similar results, showing that arteriolar widths decreased with systolic BP (−0.67% per 10 mmHg) and diastolic BP (−1.39% per 10 mmHg) (Owen et al., 2019). Other studies have demonstrated similar associations using different software for automated image analysis; arterioles being 7%–9% narrower in hypertensive compared to normotensive subjects (Robertson et al., 2020), arteriolar diameter decreased in patients with controlled and uncontrolled hypertension compared with healthy controls (2.7% and 4.7%) (Wong et al., 2003), and narrower arterioles associated with increased mean arterial BP associated (Liew et al., 2008).

In line with previous findings from the UK Biobank cohort (Tapp et al., 2019), we found an increase in arteriolar tortuosity with advancing age. However, diverging

from these trends, a study conducted in an Asian population presented an inverse relationship, indicating a decline in arteriolar tortuosity with increasing age (Cheung et al., 2011).

Given that increased tortuosity is one of the manifestations of hypertensive retinopathy (Badawi et al., 2022), we expected to find supporting evidence in our dataset. However, we could not confirm significant associations between tortuosity and BP, which might be related to the limited sample size in our cross-sectional study. The association between tortuosity and BP remains inconclusive, as larger-scale studies (Owen et al., 2019; Tapp et al., 2019) propose a positive correlation between increased tortuosity and BP, while smaller studies (Cheung et al., 2011; Taarnhøj et al., 2008) suggest an inverse association. Longitudinal studies investigating tortuosity are sparse and have not provided strong evidence of an association with BP (Xue et al., 2023). The ambiguity in findings regarding tortuosity may arise from diverse software and different measurement approaches when determining the tortuosity of vessels, contributing to the unclear relationship between tortuosity and BP.

However overall, the ability to replicate the cross-sectional findings indicating associations between retinal vessel width and BP, particularly as the most extensively studied parameter concerning vasculometry outcomes, provided validity to the vasculometry findings in the FOREVER cohort.

Results from our longitudinal association studies yielded several insights into the change in vascular measurements above baseline. Previous longitudinal studies have explored the relationship between retinal vasculometry and the risk of later development of hypertension. A longitudinal study showed that narrower arterioles were associated with an increased risk of developing severe hypertension 10 years later (Wang et al., 2008). Similarly, studies using data from the Beaver Dam Eye Study, the Atherosclerosis Risk in Communities Study, and the Rotterdam study have shown that narrower arterioles are associated with increased risk of hypertension at 10, 6 and 6.6 years of follow-up, respectively (Ikram et al., 2006; Wong, Klein, et al., 2004; Wong, Shankar, et al., 2004).

We found that longitudinal associations between IOP and tortuosity depended on IOP both at baseline and follow-up. Accounting for the influence of IOP at baseline, our analysis showed that an increase in IOP at follow-up was associated with a subsequent increase in arteriolar tortuosity compared with baseline. Similarly, when adjusting for IOP at follow-up, higher IOP at baseline was associated with more tortuous venules at follow-up compared with baseline. These findings indicate that changes in IOP over time may alter vessel tortuosity. To our knowledge, the longitudinal changes in retinal vasculometry with IOP have not been studied previously, but a cross-sectional study based on the Singapore Malay Eye Study has shown that arteriolar tortuosity decreased with increasing IOP (Wu et al., 2013). A finding that we could not confirm in our cross-sectional analysis.

Our cross-sectional findings of narrower arterioles associated with increased IOP align with other studies investigating the associations between glaucoma subtypes and vasculometry. Based on data from the EPIC—Norfolk study, high-tension open-angle glaucoma has been associated with narrower arterioles (Rudnicka et al., 2020). Moreover, outcomes from the Blue Mountains Eye Study indicated significantly narrower vessels in glaucomatous eyes compared with non-glaucomatous eyes and those with ocular hypertension only (Mitchell et al., 2005). The observed combination of increased IOP and diminished arteriolar width may result in reduced retinal perfusion in ocular hypertensive eyes, leading to compromised blood flow to ocular tissues. Combined with longitudinal associations between increased arteriolar tortuosity and IOP at follow-up adjusted for IOP at baseline (model 5), our findings support the vascular theory of glaucoma, suggesting that inadequate blood supply to retinal ganglion cells may contribute to the development and progression of glaucoma (Chan et al., 2017; Moore et al., 2008) or that vascular autoregulation caused by the loss of retinal ganglion cells may lead to decreased blood supply to the retina (Mitchell et al., 2005). However, as ocular hypertension without glaucoma exists, this may only constitute a partial explanation of the pathophysiology of glaucoma, necessitating further investigation through alternative study designs.

Interestingly, our study identified a decrease in IOP from baseline to follow-up (Table 1). However, the decrease is minimal and is not considered clinically relevant. Also, the large standard deviations of the estimates suggest substantial variability in individual IOP changes among the participants. Additionally, IOP exhibits diurnal fluctuations which could affect the measurements. We hypothesise that the negative mean IOP change could be attributed to the nature of our cohort, which is compromised by individuals visiting optician shops. When elevated IOP is detected in an optician setting, individuals are likely referred for further evaluation by an ophthalmologist and prescribed IOP-lowering treatment. We were not able to account for this in our analysis, as we did not have information on prescribed medicine or potential glaucoma diagnosis. Furthermore, the associations between changes in IOP and retinal vessel width were inconclusive. This may be attributed to the relatively short average follow-up period.

We adjusted the longitudinal models for spherical equivalent, as the vessel width parameters were measured in pixels rather than microns. The aim was to ensure that the associations observed with other risk factors remained stable and independent of refractive changes. A longer axial length and increased myopic refractions have been shown to be associated with a narrower width of arterioles (Lim et al., 2011). Hypermetropic eyes, with a shorter axial length, position the retina closer to the camera, potentially causing artefactually increased vessel widths. Conversely, myopic eyes may show artefactually decreased widths due to the retina being positioned further away. Note that we adjusted for spherical equivalent because we did not have information on axial length. However, we did not adjust for refraction when examining tortuosity, as the distance of the retina to the camera should not materially affect the estimation of tortuosity. We are aware that in subjects who have undergone cataract surgery with the replacement of

the lens, the spherical equivalent may not correctly reflect the anatomy of the eye. We were not able to account for this in our analysis, as we did not have information on cataract surgery.

We did not have information on prescribed antihypertensive treatment and hence were not able to account for this in our analysis. We adjusted follow-up measurements of retinal vasculometry for baseline values in the longitudinal analyses but did not adjust change between baseline and follow-up for baseline measurements in order to limit the role of bias. The longitudinal analysis could be approached differently by examining change in retinal vessel outcomes as dependent variable, but the approach depends on the research question. We were interested in investigating exposure relationship rather than causal pathways to an outcome, and hence the follow-up of retinal vasculometry was regressed against baseline values. The limited sample size of our cross-sectional analyses may have limited replication of previous findings concerning tortuosity and venular width from the UK Biobank cohort. Despite the limitations, our study design benefits from its strength in incorporating data from multiple visits.

5 | CONCLUSION

In conclusion, we found narrower arteriolar width associated with increased BP and IOP in agreement with previous cross-sectional findings. Increased BP at follow-up was associated with decreased arteriolar width above baseline. Increased IOP at follow up adjusted for baseline IOP was associated with increased arteriolar tortuosity above baseline. Our findings indicate that variations in BP and alterations in IOP over time are associated with changes in the width and tortuosity of retinal However, the findings cannot be generalised to other non-white ethnicities or age groups beyond those included in this study. Further studies examining longitudinal associations between retinal vasculometry and hemodynamic risk factors are needed to understand the ageing process and vascular aetiology of chronic diseases such as glaucoma.

ACKNOWLEDGEMENTS

We would like to thank all the participants who contributed to Project FOREVER. Special thanks are extended to the management and staff of Synoptik A/S for their dedicated efforts in data collection and their valuable collaboration. We appreciate the insights provided by Fight for Sight Denmark into patients' perspectives. We would like to thank Charlotte Taul and Conny Graae for their ongoing help and project support. We would like to thank the Department of Ophthalmology, Rigshospitalet-Glostrup for supporting the project. Lastly, our sincere thanks go to the University of Copenhagen and the Department of Drug Design and Pharmacology for their support, infrastructure, and hosting of Project FOREVER.

FOREVER consortium: The FOREVER consortium is led by Miriam Kolko (miriamk@sund.ku.dk). The steering group members are Miriam Kolko, Christian Torp-Pedersen, Marianne Benn, Søren Brunak, Morten la Cour, Janne Tolstrup, Annette Slyngborg, and Jens Rovelt. Associated partners are Kristoffer Sølvsten Burgdorf, Anders Bjørholm Dahl, Josefine Vilsbøll Sudgaard, and Piotr Jaroslaw Chmura. Ph.D. fellows are Josefine Freiberg, Marcel Reimann and Jens Rovelt. Ophthalmologists who are employed on Project FOREVER are Dorte Nellemann Thornit and Kim Holmgaard. International partners involved in Project FOREVER are Paul Foster, Gus Gazzard, David P. Crabb, Ananth Viswanathan, Sarah Barman, Christopher G. Owen, and Alicja R. Rudnicka.

FUNDING INFORMATION

The Synoptik Foundation has funded Project FOREVER. An additional grant has been obtained from the A. P. Møller foundation, the Velux foundation and the Danish Life Science Cluster. The funders had no role in data analysis, decision to publish, or preparation of the manuscript.

ORCID

Josefine Freiberg https://orcid.org/0000-0003-1520-7226
Jens Rovelt https://orcid.org/0000-0002-4114-0267
Miriam Kolko https://orcid.org/0000-0001-8697-0734

REFERENCES

Badawi, S.A., Fraz, M.M., Shehzad, M., Mahmood, I., Javed, S., Mosalam, E. et al. (2022) Detection and grading of hypertensive retinopathy using vessels tortuosity and arteriovenous ratio. *Journal of Digital Imaging*, 35(2), 281–301. Available from: https://doi.org/10.1007/s10278-021-00545-z

Chan, K.K.W., Tang, F., Tham, C.C.Y., Young, A.L. & Cheung, C.Y. (2017) Retinal vasculature in glaucoma: a review. *BMJ Open Ophthalmology*, 1(1), e000032. Available from: https://doi.org/10.1136/bmjophth-2016-000032

Cheung, C.Y.-L., Zheng, Y., Hsu, W., Lee, M.L., Lau, Q.P., Mitchell, P. et al. (2011) Retinal vascular tortuosity, blood pressure, and cardiovascular risk factors. *Ophthalmology*, 118(5), 812–818. Available from: https://doi.org/10.1016/j.ophtha.2010.08.045

DeMers, D. & Wachs, D. (2024) Physiology, mean arterial pressure. In: *StatPearls*. Treasure Island: StatPearls Publishing. Copyright © 2024, StatPearls Publishing LLC.

Enaholo, E.S., Musa, M.J. & Zeppieri, M. (2024) The spherical equivalent. In: *StatPearls*. Treasure Island: StatPearls Publishing. Copyright © 2024, StatPearls Publishing LLC.

Fraz, M.M., Welikala, R.A., Rudnicka, A.R., Owen, C.G., Strachan, D.P. & Barman, S.A. (2015) QUARTZ: quantitative analysis of retinal vessel topology and size—an automated system for quantification of retinal vessels morphology. *Expert Systems with Applications*, 42(20), 7221–7234. Available from: https://doi.org/10.1016/j.eswa.2015.05.022

Freiberg, J., Rovelt, J., Gazzard, G., la Cour, M. & Kolko, M. (2023) Finding ophthalmic risk and evaluating the value of eye exams and their predictive reliability (FOREVER)—a cohort study in a Danish high street optician setting: design and methodology. *Acta Ophthalmologica*, 102, 80–90. Available from: https://doi.org/10.1111/aos.15693

Freiberg, J., Welikala, R.A., Rovelt, J., Owen, C.G., Rudnicka, A.R., Kolko, M. et al. (2023) Automated analysis of vessel morphometry in retinal images from a Danish high street optician setting. *PLoS One*, 18(8), e0290278. Available from: https://doi.org/10.1371/journal.pone.0290278

Ikram, M.K., Witteman, J.C.M., Vingerling, J.R., Breteler, M.M.B., Hofman, A. & Jong, P.T.V.M. (2006) Retinal vessel diameters and risk of hypertension. *Hypertension*, 47(2), 189–194. Available from: https://doi.org/10.1161/01.HYP.0000199104.61945.33

Kumar, V. (2018) Eye is the window to the brain pathology. *Current Advances in Ophthalmology*, 1(1), 3–4. Available from: https://doi.org/10.29199/2638-9940/caop-101013

Liew, G., Sharrett, A.R., Wang, J.J., Klein, R., Klein, B.E.K., Mitchell, P. et al. (2008) Relative importance of systemic determinants of retinal arteriolar and venular caliber: the atherosclerosis risk in communities study. *Archives of Ophthalmology*, 126(10), 1404–1410. Available from: https://doi.org/10.1001/archopht.126.10.1404

Lim, L.S., Cheung, C.Y.-L., Lin, X., Mitchell, P., Wong, T.Y. & Mei-Saw, S. (2011) Influence of refractive error and axial length on retinal vessel geometric characteristics. *Investigative Ophthalmology & Visual Science*, 52(2), 669–678. Available from: https://doi.org/10.1167/iovs.10-6184

London, A., Benhar, I. & Schwartz, M. (2013) The retina as a window to the brain—from eye research to CNS disorders. *Nature Reviews Neurology*, 9(1), 44–53. Available from: https://doi.org/10.1038/nrneurol.2012.227

Mishra, C. & Tripathy, K. (2023) Fundus camera. In: *StatPearls*. Treasure Island: StatPearls Publishing. Copyright © 2023, StatPearls Publishing LLC.

Mitchell, P., Leung, H., Wang, J.J., Rochtchina, E., Lee, A.J., Wong, T.Y. et al. (2005) Retinal vessel diameter and open-angle glaucoma: the Blue Mountains Eye Study. *Ophthalmology*, 112(2), 245–250. Available from: https://doi.org/10.1016/j.ophtha.2004.08.015

Modi, P. & Arsiwalla, T. (2024) Hypertensive retinopathy. In: *StatPearls*. Treasure Island: StatPearls Publishing. Copyright © 2024, StatPearls Publishing LLC.

Moore, D., Harris, A., Wudunn, D., Kheradiya, N. & Siesky, B. (2008) Dysfunctional regulation of ocular blood flow: a risk factor for glaucoma? *Clinical Ophthalmology*, 2(4), 849–861. Available from: https://doi.org/10.2147/opth.s2774

Owen, C.G., Rudnicka, A.R., Welikala, R.A., Fraz, M.M., Barman, S.A., Luben, R. et al. (2019) Retinal vasculometry associations with cardiometabolic risk factors in the European prospective investigation of cancer-Norfolk study. *Ophthalmology*, 126(1), 96–106. Available from: https://doi.org/10.1016/j.ophtha.2018.07.022

Ptito, M., Bleau, M. & Bouskila, J. (2021) The retina: a window into the brain. *Cells*, 10(12), 3269. Available from: https://doi.org/10.3390/cells10123269

R Core Team. (2020) *R: a language and environment for statistical computing*. Vienna, Austria: R Foundation for Statistical Computing. Available from: https://www.R-project.org/

Robertson, G., Fleming, A., Williams, M.C., Trucco, E., Quinn, N., Hogg, R. et al. (2020) Association between hypertension and retinal vascular features in ultra-widefield fundus imaging. *Open Heart*, 7(1), e001124. Available from: https://doi.org/10.1136/openhrt-2019-001124

Rudnicka, A.R., Owen, C.G., Welikala, R.A., Barman, S.A., Whincup, P.H., Strachan, D.P. et al. (2020) Retinal vasculometry associations with glaucoma: findings from the European prospective investigation of cancer–Norfolk eye study. *American Journal of Ophthalmology*, 220, 140–151. Available from: https://doi.org/10.1016/j.ajo.2020.07.027

Rudnicka, A.R., Welikala, R., Barman, S., Foster, P.J., Luben, R., Hayat, S. et al. (2022) Artificial intelligence-enabled retinal vasculometry for prediction of circulatory mortality, myocardial infarction and stroke. *The British Journal of Ophthalmology*, 106(12), 1722–1729. Available from: https://doi.org/10.1136/bjo-2022-321842

Taarnhøj, N.C.B.B., Munch, I.C., Sander, B., Kessel, L., Hougaard, J.L., Kyvik, K. et al. (2008) Straight versus tortuous retinal arteries in relation to blood pressure and genetics. *British Journal of Ophthalmology*, 92(8), 1055–1060. Available from: https://doi.org/10.1136/bjo.2007.134593

Tapp, R.J., Owen, C.G., Barman, S.A., Welikala, R.A., Foster, P.J., Whincup, P.H. et al. (2019) Associations of retinal microvascular diameters and tortuosity with blood pressure and arterial stiffness: United Kingdom biobank. *Hypertension*, 74(6), 1383–1390. Available from: https://doi.org/10.1161/hypertensionaha.119.13752

Tapp, R.J., Owen, C.G., Barman, S.A., Welikala, R.A., Foster, P.J., Whincup, P.H. et al. (2020) Retinal vascular tortuosity and diameter associations with adiposity and components of body composition. *Obesity (Silver Spring)*, 28(9), 1750–1760. Available from: https://doi.org/10.1002/oby.22885

Wang, J.J., Rochtchina, E., Liew, G., Tan, A.G., Wong, T.Y., Leeder, S.R. et al. (2008) The long-term relation among retinal arteriolar narrowing, blood pressure, and incident severe hypertension. *American Journal of Epidemiology*, 168(1), 80–88. Available from: https://doi.org/10.1093/aje/kwn100

Welikala, R.A., Fraz, M.M., Habib, M.M., Daniel-Tong, S., Yates, M., Foster, P.J. et al. (2017) Automated quantification of retinal vessel morphometry in the UK biobank cohort. In: *2017 Seventh International Conference on Image Processing Theory, Tools and Applications (IPTA)*.

Wong, T.Y., Klein, R., Klein, B.E.K., Meuer, S.M. & Hubbard, L.D. (2003) Retinal vessel diameters and their associations with age and blood pressure. *Investigative Ophthalmology & Visual Science*, 44(11), 4644–4650. Available from: https://doi.org/10.1167/iovs.03-0079

Wong, T.Y., Klein, R., Sharrett, A.R., Duncan, B.B., Couper, D.J., Klein, B.E.K. et al. (2004) Retinal arteriolar diameter and risk for hypertension. *Annals of Internal Medicine*, 140(4), 248–255. Available from: https://doi.org/10.7326/0003-4819-140-4-200402170-00006

Wong, T.Y. & Mitchell, P. (2004) Hypertensive retinopathy. *The New England Journal of Medicine*, 351(22), 2310–2317. Available from: https://doi.org/10.1056/NEJMra032865

Wong, T.Y., Shankar, A., Klein, R., Klein, B.E.K. & Hubbard, L.D. (2004) Prospective cohort study of retinal vessel diameters and risk of hypertension. *BMJ*, 329(7457), 79. Available from: https://doi.org/10.1136/bmj.38124.682523.55

Wu, R., Cheung, C.Y.-L., Saw, S.M., Mitchell, P., Aung, T. & Wong, T.Y. (2013) Retinal vascular geometry and glaucoma: the Singapore Malay eye study. *Ophthalmology*, 120(1), 77–83. Available from: https://doi.org/10.1016/j.ophtha.2012.07.063

Xue, C.C., Li, C., Hu, J.F., Wei, C.C., Wang, H., Ahemaitijiang, K. et al. (2023) Retinal vessel caliber and tortuosity and prediction of 5-year incidence of hypertension. *Journal of Hypertension*, 41(5), 830–837. Available from: https://doi.org/10.1097/hjh.0000000000003406

Zhou, Y., Wagner, S.K., Chia, M.A., Zhao, A., Woodward-Court, P., Xu, M. et al. (2022) AutoMorph: automated retinal vascular morphology quantification via a deep learning pipeline. *Translational Vision Science & Technology*, 11(7), 12. Available from: https://doi.org/10.1167/tvst.11.7.12

How to cite this article: Freiberg, J., Welikala, R., Rovelt, J., Barman, S.A., Owen, C.G., Rudnicka, A.R. et al. (2024) Longitudinal associations of retinal vessel morphology with intraocular pressure and blood pressure at follow-up visit—Findings from a Danish eye and vision cohort, Project FOREVER. *Acta Ophthalmologica*, 00, 1–10. Available from: https://doi.org/10.1111/aos.16737