RESEARCH ARTICLE

Retinal Biomarkers of Cerebral Small Vessel Disease: A Systematic Review

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Abstract

Introduction

Cerebral Small Vessel Disease (CSVD), a progressive degenerative disorder of small caliber cerebral vessels, represents a major contributor to stroke and vascular dementia incidence worldwide. We sought to conduct a systematic review of the role of retinal biomarkers in diagnosis and characterization of CSVD.

Methods

We conducted a systematic review of MEDLINE, PubMed, Scopus, the Cochrane Library Database, and Web of Science. We identified studies of sporadic CSVD (including CSVD not otherwise specified, Cerebral Amyloid Angiopathy, and Hypertensive Arteriopathy) and the most common familial CSVD disorders (including CADASIL, Fabry disease, and MELAS). Included studies used one or more of the following tools: visual fields assessment, fundus photography, Optical Coherence Tomography and OCT Angiography, Fluorescein Angiography, Electroretinography, and Visual Evoked Potentials.

Results

We identified 48 studies of retinal biomarkers in CSVD, including 9147 cases and 12276 controls. Abnormalities in retinal vessel diameter (11 reports, n = 11391 participants), increased retinal vessel tortuosity (11 reports, n = 617 participants), decreased vessel fractal dimension (5 reports, n = 1597 participants) and decreased retinal nerve fiber layer thickness (5 reports, n = 4509 participants) were the biomarkers most frequently associated with CSVD. We identified no reports conducting longitudinal retinal evaluations of CSVD, or systematically evaluating diagnostic performance.

Conclusion

Multiple retinal biomarkers were associated with CSVD or its validated neuroimaging biomarkers. However, existing evidence is limited by several shortcomings, chiefly small sample size and unstandardized approaches to both biomarkers' capture and CSVD

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characterization. Additional larger studies will be required to definitively determine whether retinal biomarkers could be successfully incorporated in future research efforts and clinical practice.

Introduction

Cerebral Small Vessel Disease (CSVD) is a progressive, age-related degenerative disorder of the small caliber vessels of the Central Nervous System (CNS) [1–3]. Due to progressive accumulation of microvascular lesions over time, it is responsible for almost 20% of ischemic stroke and over 80% of all hemorrhagic stroke [4]. CSVD also represents the second most common form of dementia, following Alzheimer's disease [1,5]. The vast majority of CSVD cases are sporadic in nature, presenting without a clear familial inheritance pattern (Table 1).

Most sporadic CSVD cases (over 90% of all CSVD diagnoses) are accounted for by two progressive, aging-related disorders: Cerebral Amyloid Angiopathy (CAA) and Hypertensive Arteriopathy (HTNA) [1]. Rare familial forms occurring on a hereditary basis (usually monogenic autosomal dominant) have also been identified, with the most frequently reported being Cerebral Autosomal Dominant Arteriopathy with Subcortical Infarcts and Leukoencephalopathy (CADASIL), Fabry disease and Mitochondrial Encephalomyopathy, Lactic Acidosis and Stroke-like episodes (MELAS) syndrome [1,6]. Other rarer subtypes of CSVD include infectious and immune-mediate forms (Table 1).

Table 1. Cerebral small vessel disease subtypes.

Sporadic CSVD	Familial (Hereditary) CSVD
Age-related	CADASIL
• CAA	CARASIL
• HTNA	MELAS
Immune-mediated	Fabry Disease
• Primary CNS Vasculitis	CSVD due to Type IV Collagen Disease
• Secondary CNS Vasculitis	Retinal Vasculopathy with Cerebral Leukoencephalopathy
• ANCA-associated vasculitis	Hereditary CAA
• Hypersensitivity vasculitis	• Dutch Variant Hereditary CAA
• CNS Vasculitis due to SLE	• Flemish Variant Hereditary CAA
• CNS Vasculitis due to Sjogren	• Italian Variant Hereditary CAA
• Rheumatoid Vasculitis	• Piedmont Variant Hereditary CAA
• CNS Vasculitis due to MCTD	• Arctic Variant Hereditary CAA
• CNS Vasculitis due to Behçet	• Icelandic Variant Hereditary CAA
Infectious	• Iowa Variant Hereditary CAA
• HIV CNS Vasculitis	• Meningovascular Amyloidosis
• Meningovascular Syphilis	
• CMV Vasculitis	
• VZV Vasculitis	
• HBV and HCV Vasculitis	
• Cerebral malaria	

Abbreviations: CAA = Cerebral Amyloid Angiopathy, CADASIL = Cerebral Autosomal Dominant Arteriopathy with Subcortical Infarcts and Leukoencephalopathy, CARASIL = Cerebral Autosomal Recessive Arteriopathy with Subcortical Infarcts and Leukoencephalopathy, CNS = Central Nervous System, CSVD = Cerebral Small Vessel Disease, HTNA = Hypertensive Arteriopathy, MCTD = Mixed Connective Tissues Disease, MELAS = Mitochondrial Encephalomyopathy, Lactic Acidosis and Stroke-like episodes, SLE = Systemic Lupus Erythematosus.

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While definitive diagnosis of sporadic and familial forms of CSVD requires histopathological examination of brain tissue, in clinical practice the diagnostic gold standard is identification of typical CSVD-related ischemic and hemorrhagic lesions on MRI brain imaging. The neuroimaging biomarkers most commonly associated with CSVD include white matter hyperintensities (also referred to as leukoaraiosis), lacunar infarcts, dilated perivascular spaces, cortical superficial siderosis, and cerebral microbleeds [7]. However, these findings are consistent with the presence of irreversible ischemic or hemorrhagic CNS damage, and are, therefore, of limited use in the diagnosis and monitoring of the preclinical and minimally symptomatic stages of CSVD [2]. In addition, financial (scan and personnel costs) and logistical (availability of equipment and expertise) limitations prevent the widespread use of MRI neuroimaging in early screening for CSVD and monitoring of disease progression and response to treatment over time [1].

The retina contains CNS neurons and a small vessel network displaying close anatomical and physiological parallels with the corresponding cerebral neurovascular unit [8]. It is, therefore, conceivable that non-invasive evaluation of retinal neurons and vessels may provide novel biomarkers for CSVD diagnosis and staging [9,10]. Because retinal biomarkers provide information on tissue structure and function at the microscopic level, they may allow for diagnosis of CSVD in earlier, less symptomatic or asymptomatic stages. Finally, retinal biomarkers compare very favorably with MRI-based neuroimaging in terms of equipment availability, operating costs, and expertise required to gather data [8]. Therefore, they may allow for large-scale screening for CSVD in at-risk population (e.g. elderly individuals), in a way MRI neuroimaging cannot due to prohibitive costs and insufficient number of scanners and trained personnel available.

To date, several studies have tested this overall hypothesis using a variety of different technologies, including visual fields (VF) assessment, fundus photography, Optical Coherence Tomography (OCT) and OCT Angiography (OCTA), Fluorescein Angiography (FA), Electroretinography (ERG), and Visual Evoked Potentials (VEP) [9,10]. All these biomarker acquisition modalities offer a variety of potential advantages over MRI neuroimaging, including widespread availability as part of routine medical care and ability to evaluate neurons and blood vessels at the microscopic level (which is currently possible only in a very limited fashion with MRI neuroimaging) [8].

To date, no retinal biomarkers have emerged as candidates for adoption into routine diagnostic or clinical care practice for CSVD. The present systematic review aims to evaluate existing evidence on the performance of retinal biomarkers in the diagnosis and staging of different forms of CSVD. Our primary goal is to identify retinal biomarkers demonstrating associations with: 1) CSVD diagnoses (in affected individuals vs. healthy controls); 2) established neuroimaging markers of CSVD; 3) acute stroke risk or cognitive decline secondary to CSVD. We also sought to identify studies reporting diagnostic performance for different CSVD disorders, whether in initial screening or longitudinal monitoring. Finally, we evaluated the strengths and gaps in currently available evidence on a disease and technology-specific basis in order to better inform future research efforts.

Material and methods

Review rationale and overall design

This systematic review was conducted on the basis of a pre-specified protocol and designed in agreement with the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) 2020 guidelines [11]. We chose to focus on studies of CSVD, in both its sporadic (CSVD not otherwise specified, CAA, or HTNA) and most common familial (CADASIL,

Fabry disease, and MELAS) forms [1,6]. The inclusion of MELAS disease (primarily a mitochondrial disease) in the present analyses is motivated by findings indicating small vessel vasculopathy secondary to energy failure as central to the characteristic ischemic events in this condition [12]. For sporadic CSVD forms, we focused on studies utilizing established neuropathological or neuroimaging criteria for diagnosis [1,5]. For familial CSVD forms, we focused on studies with confirmed genetic diagnoses and consistent clinical and neuroimaging phenotypes [6].

We pre-specified inclusion of the following retinal evaluation modalities: visual fields (VF) assessment, fundus photography, Optical Coherence Tomography (OCT) and OCT Angiography (OCTA), Fluorescein Angiography (FA), Electroretinography (ERG), and Visual Evoked Potentials (VEP) [13]. Our primary pre-specified objective was to identify retinal biomarkers distinguishing CSVD cases from controls. As a secondary objective, we sought to determine whether retinal biomarkers (individually or in combination) were found to be associated with either: 1) neuroimaging markers of CSVD severity; or 2) clinical metrics of acute stroke risk or cognitive decline secondary to CSVD. We defined CSVD-related MRI markers according to the Standards for Reporting Vascular Changes on Neuroimaging (STRIVE) guidelines [7]. All analyses were conducted using publicly available summary data, without any access to individual level data. As such, no institutional review board approval or informed patient consent was required.

Search strategy

We conducted an online literature search using the following publicly accessible databases: Medical Literature Analysis and Retrieval System Online (MEDLINE), PubMed, Scopus, the Cochrane Library Database, and Web of Science. Please refer to Supporting Information (S1 File) for details on the Search Strategy. We restricted our search to studies published in English. Following initial database queries, results were harmonized in a single list of publications. We then manually reviewed references of relevant articles to identify additional potentially relevant publications via forward citation search. After completing this step, the initial publication list was pruned from duplicate entries (Fig 1). We then reviewed study abstracts to identify studies that met the following criteria: 1) included original data from human participants; 2) investigated one or more retinal biomarkers generated using the pre-specified methodologies; 3) compare the distribution of retinal biomarkers across CSVD patients, between CSVD patients and healthy controls, across patient groups identified by established CSVD neuroimaging markers, or across patient groups identified by stroke risk and/or cognitive performance measures. In order to qualify for a diagnosis of CSVD, participants in a study had to be present with either: 1) CSVD-related lacunar ischemic stroke; 2) CSVD-related spontaneous intracerebral hemorrhage; 3) CSVD-related cognitive decline fulfilling criteria for Vascular contributions to Cognitive Impairment and Dementia (VCID). We specifically excluded studies that did not confirm that stroke or cognitive decline were attributable to CSVD based on current diagnostic criteria [14,15]. We specifically excluded the following article types: 1) review studies; 2) individual case studies; 3) study protocols; 4) conference presentations, abstracts, or summaries; 5) comments on original research that did not present novel peer-review findings; 6) editorial commentaries, viewpoints, and other opinion pieces. For previously published systematic meta-analyses, we separately evaluated each included study (if not already identified as part of our search strategy) for inclusion in our systematic review. When a determination about meeting inclusion and exclusion criteria could not be reached via abstract review, studies were marked for full-text review.
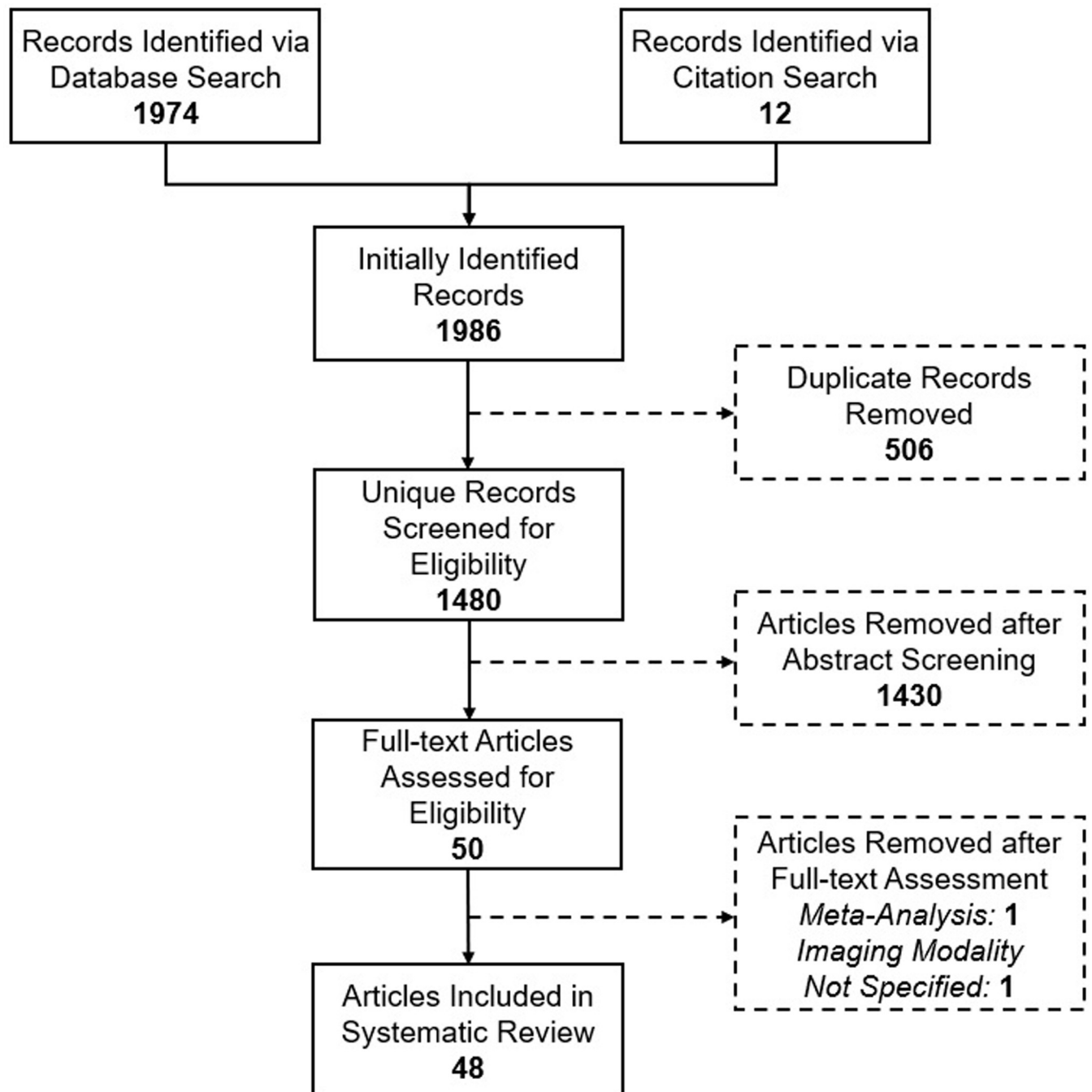
Fig 1. Search strategy flow chart.

Data extraction

Following initial screening of potentially relevant publications, eligibility for inclusion in the present review was confirmed via full-text review. We pre-specified for extraction, from each individual article, the following data points: authors, publication year, pre-specified study aim / hypotheses, study type, number of patients and controls, participants' demographics (number of male vs. female, mean age), participant selection criteria, CSVD diagnostic criteria

employed, MRI neuroimaging (if applicable), cognitive performance evaluation (if applicable), genetic diagnostic testing (if applicable), retinal evaluation modality, device and imaging settings, image quality control procedures, retinal biomarkers extracted and extraction methodology, outcomes of interest, and statistical modeling methods. Data extraction was conducted by two separate reviewers (ZT and JC) independently and blinded to each other. All extracted data points were cross-checked, and disagreements reconciled via joint evaluation by a board-certified optometrist with expertise in ocular imaging (EZB) and a board-certified neurologist with expertise in CSVD (AB).

Study quality assessment

We performed systematic assessment of study quality for eligible publications in agreement with the STrengthening the Reporting of OBservational studies in Epidemiology (STROBE) recommendations as qualifying items [16]. We used the STROBE checklist to asses study quality based on whether or not individual recommendation items were successfully addressed, with a final score ranging from 0 (none of the recommendations addressed) to 22 (all recommendations addressed). We separately scored studies of OCT and OCTA markers in CSVD using the Advised Protocol for OCT Study Terminology and Elements (APOSTEL) v2.0 recommendations, which provide an optimal framework for design, execution, and reporting of results in quantitative OCT/OCTA studies [17]. Using an identical procedure as for the STROBE study quality score, we assigned individual publications values ranging from 0 (none of the recommendations addressed) to 9 (all recommendations addressed). We did not identify specific recommendations for other retinal evaluation methodologies that could be applied to evaluate study quality. Of note, study quality was evaluated after the final list of included publications was generated and, therefore, had no impact on whether individual articles were included or excluded from the present analyses.

Data analysis

Based on prior reviews on similar topics, we expected to identify a small number of studies investigating each individual form of CSVD with a specific retinal evaluation modality [8,9,18–20]. In addition, we expected included studies to report on a variety of retinal biomarkers with widely differing definitions. We, therefore, did not pre-specify methods for meta-analysis of published evidence, but rather opted to focus on a systematic presentation of results. We chose to collate all associations between retinal biomarkers and CSVD disorders, subdivided by disease of interest and data acquisition modality.

Results

Search results

Our initial automated searches of online repositories identified a total of 1974 reports fitting the search criteria. We identified an additional 12 reports via manual examination and automated cross-reference of citations from publications identified via our search strategy. After elimination of 506 duplicated records, we screened for eligibility 1480 publications (Fig 1). A total of 1430 reports were excluded after manual review of abstracts for failing to satisfy all inclusion and exclusion criteria. We therefore conducted full-text manual review of 50 papers. Among these, one was excluded as it presented a meta-analysis of previously published primary data. As per our pre-specified methodology, we included all meta-analyzed studies (if they individually met our eligibility criteria) in our review. Another publication was excluded
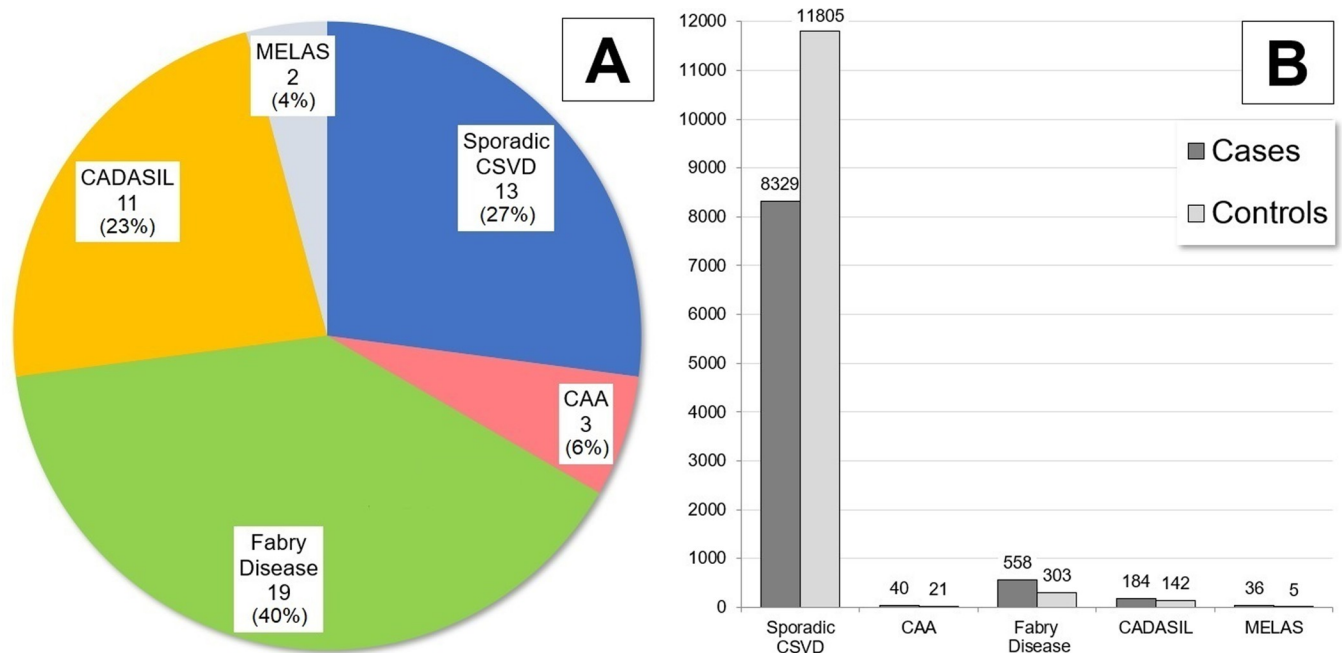
Fig 2. Number and sample size of studies included in systematic review. Panel A: Number and percentages of studies included in the present systematic review, based on CSVD disorder of interest. Panel B: Number of affected (cases) and healthy (controls) individuals participating in studies included in the present systematic review, based on CSVD disorder of interest. Abbreviations: CAA = Cerebral Amyloid Angiopathy, CADASIL = Cerebral Autosomal Dominant Arteriopathy with Subcortical Infarcts and Leukoencephalopathy, CSVD = Cerebral Small Vessel Disease, MELAS = Mitochondrial Encephalomyopathy, Lactic Acidosis, and Stroke-like episodes syndrome.

as the retinal evaluation modality employed could not be definitively ascertained. We therefore included 48 separate published reports of studies of retinal biomarkers in CSVD (Fig 1).

Studies included in systematic review

We present information on the number and percentage of studies focusing on CSVD in general (henceforth referred to as "sporadic", to distinguish from familial monogenic forms), CAA, CADASIL, Fabry disease and MELAS in Fig 2 (Panel A).

It is worth nothing that while three papers specifically applied diagnostic criteria to enroll patients with CAA, none of the remaining papers focused on sporadic CSVD applied criteria specific to either CAA or HTNA. Rather, these studies defined participants on the basis of clinical presentation and neuroimaging as being diagnosed with CVSD (or similar terminology), without further specification. As visually illustrated in Fig 2 (Panel B), individuals enrolled in these studies of sporadic CSVD represented the overwhelming majority among participants included in the present systematic review, since they accounted for 8329 of 9147 cases (91%), and 11805 of 12276 control (96%).

We present in Table 2 information on retinal evaluation modalities employed by studies included in our systematic review, based on the CSVD disorder of interest.

The vast majority of studies employed a single imaging modality, with only a handful incorporating multiple techniques, and none including all those considered for inclusion in our systematic review. Fig 3 provides a summary of the findings from our systematic review in terms of retinal biomarkers identified.

Vascular retinal biomarkers represented the largest number of studies reporting positive associations, especially for: 1) changes in diameter of retinal vessels (11 reported associations

Table 2. Summary of imaging modalities and CSVD disorders for studies included in systematic review.

		CSVD Disorders				
		Sporadic CSVD	**CAA**	**Fabry Disease**	**CADASIL**	**MELAS**
Imaging Modalities	**Fundus Photography**	**10** Ref: [21–30]	**3** Ref: [31–33]	**11** Ref: [34–44]	**5** Ref: [45–49]	**1** Ref: [50]
	OCT	**2** Ref: [51,52]	**2** Ref: [31,33]	**5** Ref: [34,37–39,53]	**6** Ref: [45,48,54–57]	**1** Ref: [58]
	OCTA	**2** Ref: [51,59]	**1** Ref: [31]	**8** Ref: [37,44,53,60–64]	**1** Ref: [55]	-
	FA	-	**1** Ref: [32]	-	**3** Ref: [45,47,48]	-
	ERG	-	-	**1** Ref: [37]	**2** Ref: [65,66]	**1** Ref: [50]
	VEP	-	-	-	**2** Ref: [48,57]	-
	VF Assessment	-	-	**3** Ref: [36,67,68]	**2** Ref: [47,48]	-

Table presents the number of studies employing specific imaging modalities in each CSVD disorder of interest for the present systematic review. Several studies employed multiple imaging modalities, please refer to the Results section for details. Abbreviations: CAA = Cerebral Amyloid Angiopathy, CADASIL = Cerebral Autosomal Dominant Arteriopathy with Subcortical Infarcts and Leukoencephalopathy, CSVD = Cerebral Small Vessel Disease, ERG = Electroretinography, FA = Fluorescein Angiography, MELAS = Mitochondrial Encephalomyopathy, Lactic Acidosis, and Stroke-like episodes syndrome, OCT = Optical Coherence Tomography, OCTA = Optical Coherence Tomography Angiography, VEP = Visual Evoked Potentials, VF = Visual Field.

among 11391 participants); 2) increased retinal vessel tortuosity (11 reported associations among 617 participants) and decreased vessel fractal dimension (5 reported associations among 1597 participants), both established markers of progressive chronic retinal angiopathy [20]. Among neuronal retinal biomarkers, decreased retinal nerve fiber layer (RNFL) thickness was the only one to be associated with multiple CSVD disorders (5 reported associations among 4509 participants).

We initially planned to compare results on retinal biomarkers across different forms of CSVD to determine whether consistent association patterns emerged, potentially indicating shared pathophysiological mechanisms. However, we found no biomarkers displaying consistent associations across all or even most CSVD disorders of interest. Based on findings from Table 2 and Fig 3, this observation most likely reflects limited overlap in choice of retinal evaluation technologies and specific biomarkers across different studies, rather than underlying biological heterogeneity.

Study quality assessment

The median quality score based on STROBE recommendations [16] for included studies was 15/22, with inter-quartile range of 11/22 to 19/22. Most studies lost points for failing to appropriately describe study design in title or abstract; failing to explain rationale for study sample size; inadequate explanations provided regarding controlling for potential sources of bias; and inadequate discussion of the generalizability of results. Among 16 studies presenting results of retinal OCT-based imaging in CSVD patients, the median quality score based on APOSTEL v2.0 recommendations [17] was 4/9, with inter-quartile range of 2/9 to 6/9. Most studies lost points for failing to clearly document scanning protocol; acquisition devices (either hardware and/or software); and acquisition settings. Overall, our study quality assessment did raise concerns about a substantial proportion of studies failing to provide detailed information on key aspects of study design (especially sample size and projected power) and study execution, primarily in terms of details pertaining to hardware, software, and parameters used for data acquisition.

Retinal biomarkers in sporadic CSVD

We identified a total of 13 studies investigating the association between retinal biomarkers and sporadic CSVD (Table 3). These studies included a total of 8329 sporadic CSVD patients and
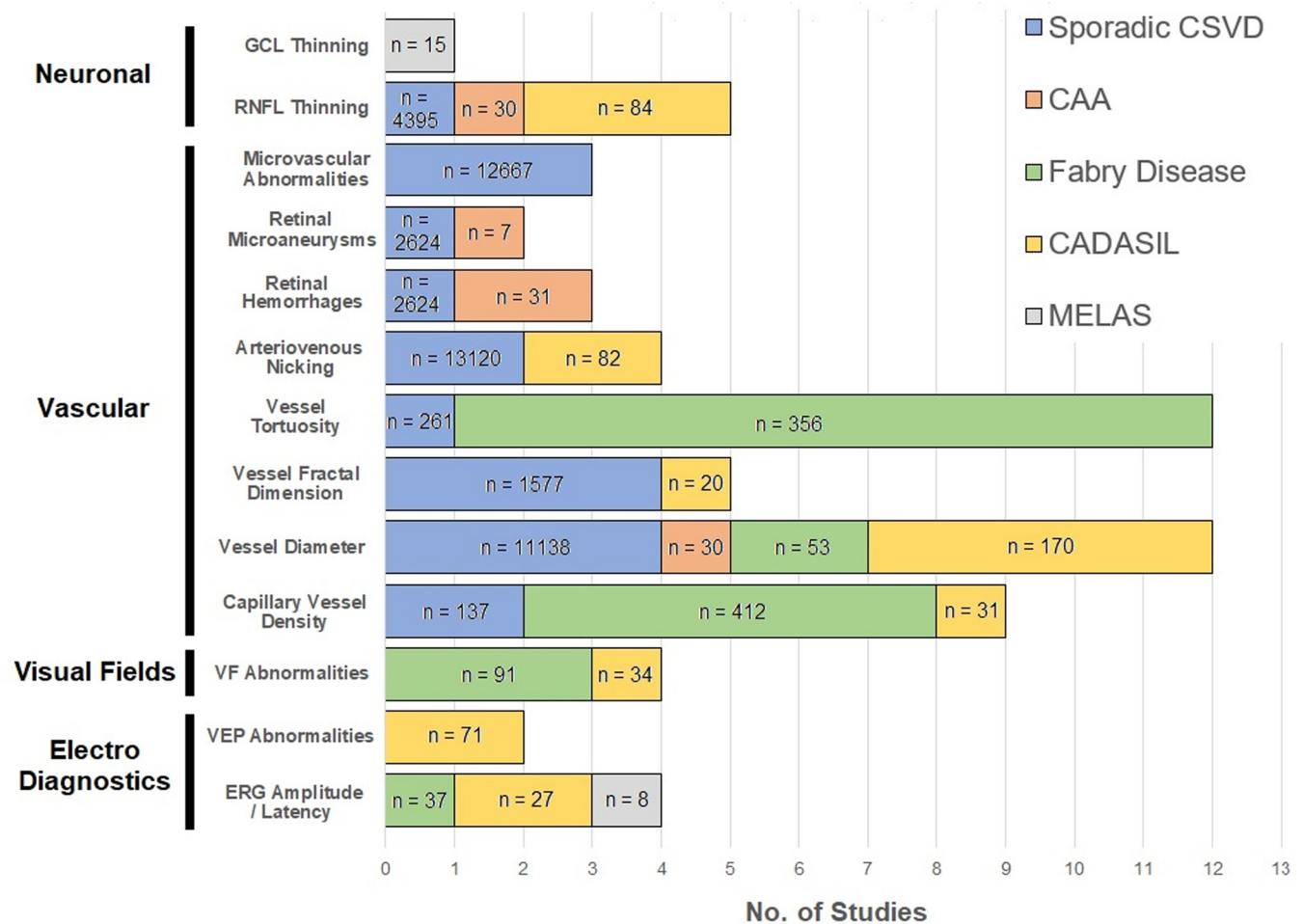
Fig 3. Retinal biomarkers identified in systematic review. Figure presents the number of studies identifying specific retinal biomarkers as associated with each CSVD disorder of interest for the present systematic review. For each CSVD disorder, the number of individuals included in studies reporting association of a specific biomarker is reported (denoted as n). Abbreviations: CAA = Cerebral Amyloid Angiopathy, CADASIL = Cerebral Autosomal Dominant Arteriopathy with Subcortical Infarcts and Leukoencephalopathy, CSVD = Cerebral Small Vessel Disease, ERG = Electroretinography, GCL = Ganglion Cell Layer, MELAS = Mitochondrial Encephalomyopathy, Lactic Acidosis, and Stroke-like episodes syndrome, RNFL = Retinal Nerve Fiber Layer, VEP = Visual Evoked Potentials, VF = Visual Field.

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11805 controls. The median number of CSVD patients per study was 262 (range 24–4395) and the median number of controls per study was 814 (range 20–10158). We identified 10 reports using fundus photography [21–30], one study using OCT [52], one using OCTA [59], and one combining OCT and OCTA [51]. We found no report leveraging VF assessment, FA or ERG to investigate sporadic CSVD. A total of 4 of 13 studies (31%) utilized ischemic stroke as diagnostic criterion for sporadic CSVD. Evaluation of one or more CSVD neuroimaging biomarkers was included in 7 of 13 studies (54%). Only three studies (21%) utilized vascular cognitive impairment as eligibility criterion. The majority of publications (10 of 13, 77%) provided full details of imaging device and methodology. Only 5 of 13 studies (38%) reported systematically performing eye dilation as part of their methodology, although an additional 5 of 13 (38%) utilized exclusively non-mydriatic fundus cameras designed for image acquisition without requirement for pupil dilation. Sporadic CSVD studies utilizing fundus photography identified arterial and venular fractal dimensions [22–24] or arteriolar and venular caliber [25,27,30] as associated with WMH, lacunar infarcts, or cerebral microbleeds. Four studies [27–30] reported

Table 3. Summary of design, patient characteristics, methodology and results for studies of sporadic CSVD.

Study	Year	Design	CSVD Phenotype(s)	No. Participants	Imaging Modality	Device	Software	Pupil Dilation	Biomarkers Identified
Abdelhak et al.	2020	Case-Control	Lacunar Stroke VCID	24 subjects 20 controls	OCT	Heidelberg Spectralis	Heidelberg Spectralis Version 6.2	No	• Retinal Artery Mean Wall Thickness • Retinal Artery Wall to Lumen Ratio
Cheung et al.	2010	Cross-sectional	Lacunar Ischemic Stroke	392 subjects	Fundus Photography	Fundus Camera (Not specified)	Semi-automated program (International Retinal Imaging Software [IRIS-Fractal])	Yes	• Retinal Vessel fractal dimension
Doubal et al.	2010	Case-Control	Lacunar Ischemic Stroke	86 subjects 80 controls	Fundus Photography	CRDGi Canon	Matlab	Yes	• Retinal Vessel fractal dimension
McGrory et al.	2019	Cross-sectional	Ischemic Stroke Lacunar Infarct WMH	758 subjects	Fundus Photography	CRDGi Canon	VAMPIRE	No (non-mydriatic camera)	• Arteriolar fractal dimension • Venular fractal dimension
Hilal et al.	2014	Cross-sectional	Lacunar Infarcts WMH Cerebral Microbleeds	261 subjects	Fundus Photography	Fundus Camera (Not specified)	Semi-automated program (Singapore I Vessel Assessment (v3.0))	Yes	• Retinal Vessel fractal dimension • Retinal Vessel tortuosity
Ikram et al.	2006	Cohort	Lacunar Infarcts WMH	490 subjects	Fundus Photography	Topcon Camera	Semi-automated program (Retinal Analysis, Optimate)	Yes	• Retinal Venular Dilation
Kim et al.	2011	Cohort	Lacunar Infarcts WMH	4395 subjects	Fundus Photography	EOS D60 Canon Camera	N/A	No (non-mydriatic camera)	• RNFL Wedge-like Defect
Kwa et al.	2002	Cohort	Lacunar Infarct WMH	108 subjects	Fundus Photography	Optimed	N/A	Yes	• Retinal microvascular abnormalities • Retinal arterial narrowing • Retinal arterial sclerosis • Retinal exudates
Lee et al.	2019	Cross-sectional	VCID	1077 subjects 1547 controls	Fundus Photography	Fundus Camera (Not specified)	Only retinal arteriolar diameter calculated via semi-automated system	No (non-mydriatic camera)	• Retinopathy • Retinal microaneurysms • Retinal hemorrhages • AV nicking
Lee et al.	2020	Cross-sectional	AD Cognitive Impairment VCID	60 subjects	OCT OCTA	Topcon DRI Triton	Native OCT software (IMAGEnet 6 V.1.14.8538)	No	• Retinal Capillary Density
Shu et al.	2020	Cross-sectional	Ischemic stroke or TIA	263 subjects	Fundus Photography	KOWA nonmyd7	N/A	No (non-mydriatic camera)	• Retinopathy score • Retinal microvascular abnormalities
Yatsuya et al.	2010	Cohort	Ischemic stroke	10,496 subjects with 338 incident strokes	Fundus Photography	Canon CR-45UAF	"Computer-assisted approach"	No (non-mydriatic camera)	• Retinal Arteriolar Diameter • Retinal Venular Diameter • Retinal Focal Arteriolar Narrowing • AV nicking • Retinal microvascular abnormalities

(*Continued*)

Table 3. (Continued)

Study	Year	Design	CSVD Phenotype(s)	No. Participants	Imaging Modality	Device	Software	Pupil Dilation	Biomarkers Identified
Wang et al.	2021	Case-Control	Lacunar Infarcts WMH	47 subjects 30 controls	OCTA	RTVue-XR OptoVue AVANTI	Native OCT software (AVANTI)	No	• Macular Superficial Capillary Plexus Vessel Density • Radial Peripapillary Capillary Vessel Density

Abbreviations: CSVD = Cerebral Small Vessel Disease, OCT = Optical Coherence Tomography, OCTA = Optical Coherence Tomography Angiography,

TIA = Transient Ischemic Attack, VCID = Vascular Contributions to Cognitive Impairment and Dementia, WMH = White Matter Hyperintensities.

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associations between retinal markers of retinopathy (retinal hemorrhages, AV nicking, microvascular abnormalities) as associated with WMH and/or lacunar infarcts. A single large study [26] reported higher prevalence among CSVD patients (compared to healthy controls) of RNFL wedge-shaped defects on fundus photography, a semi-quantitative marker of focal nerve fiber damage [69]. Regarding OCT imaging, one study [51] reported no associations with retinal measurements, while another [52] reported increased arteriolar thickness, quantified as Mean Wall Thickness (MWT) or Wall-to-Lumen Ratio (WLR), among CSVD patients compared to controls. The latter report also identified an association between arteriolar WLR and WMH severity on MRI, as well as with select cerebrospinal fluid biomarkers. Sporadic CSVD studies collecting OCTA images [51,59] found lower retinal capillary density in the peripapillary network in patients with CSVD, which was also associated with WMH on MRI.

Retinal biomarkers in CAA

We identified a total of three studies investigating the association between retinal biomarkers and CAA (Table 4). One study employed fundus photography, OCT and OCTA

Table 4. Summary of design, patient characteristics, methodology and results for studies of CAA.

Study	Year	Design	CSVD Phenotype(s)	No. Participants	Imaging Modality	Device	Software	Pupil Dilation	Biomarkers Identified
Alber et al.	2021	Case-Control	Sporadic CAA Cerebral Microbleeds WMH Memory Performance	12 subjects 12 controls	Fundus Photography	RTVue-XR OptoVue AVANTI	Native OCT software (AVANTI)	Yes	• Retinal hemorrhages
				12 subjects 12 controls	OCT	RTVue-XR OptoVue AVANTI	Native OCT software (AVANTI)	Yes	• None
				12 subjects 12 controls	OCTA	RTVue-XR OptoVue AVANTI	Native OCT software (AVANTI)	Yes	• None
Lee et al.	2009	Cross-Sectional	Sporadic CAA	7 patients	Fundus Photography	N/A	N/A	Yes	• Retinal hemorrhages • Retinal microaneurysms
				7 patients	Fluorescein Angiography	N/A	N/A	Yes	• Retinal hemorrhages • Retinal microaneurysms.
van Etten et al.	2020	Case-Control	Hereditary (Dutch Mutation) CAA	21 subjects 9 controls	Fundus Photography	Topcon TRC-50DX	N/A	Yes	• Retinal Arteriolar Narrowing
				21 subjects 9 controls	OCT	Heidelberg Spectralis	Heidelberg Eye Explorer v1.9.10.0	Yes	• RNFL thickness

Abbreviations: CAA = Cerebral Amyloid Angiopathy, CSVD = Cerebral Small Vessel Disease, OCT = Optical Coherence Tomography, OCTA = Optical Coherence Tomography Angiography, WMH = White Matter Hyperintensities.

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concomitantly in a case-control design of 12 patients with possible or probable CAA (based on the validated Boston criteria) and 12 healthy controls [31]. Although this study identified no differences in retinal biomarkers between CAA cases and controls, retinal microbleeds were associated with episodic memory performance among CAA patients. Another study combined fundus photography with FA to examine a consecutive series of seven patients admitted with CAA-related intracerebral hemorrhage (as defined using the Boston criteria) [32]. Investigators found multiple dot and blot retinal hemorrhages on fundus photography and retinal microaneurysm in at least one eye for each CAA patient. The third study jointly employed fundus photography and OCT to conduct a case-control analysis of 21 carriers of the Dutch-mutation variant of Hereditary CAA (8 pre-symptomatic individuals without history of stroke or cognitive decline, and 13 symptomatic patients) and 9 healthy controls [33]. Retinal arteriolar narrowing was more common among mutation carriers (both symptomatic and asymptomatic) than controls. Peripapillary RNFL thickness was lower in symptomatic patients compared to controls, but not among pre-symptomatic individuals.

Retinal biomarkers in fabry disease

We identified a total of 19 studies of retinal biomarkers in Fabry disease (Table 5). These studies included in total 558 affected individuals and 303 healthy controls. The median number of Fabry disease patients per study was 28 (range 8–57) and the median number of healthy

Table 5. Summary of design, patient characteristics, methodology and results for studies of Fabry disease.

Study	Year	Design	CSVD Phenotype(s)	No. Participants	Imaging Modality	Device	Software	Pupil Dilation	Biomarkers Identified
Atiskova et al.	2019	Case-control	Fabry Disease	27 subjects 27 controls	Fundus Photography	Heidelberg Spectralis	ImageJ	No	• Macular hyper-reflective foci • Retinal vessel tortuosity
				27 subjects 27 controls	OCT	Heidelberg Spectralis	ImageJ	Yes	• None
Bacherini et al.	2021	Case-control	Fabry Disease	13 subjects 13 controls	OCTA	Nidek RS-3000 Advance 2	Native OCT software (AngioScan)	Yes	• Superficial capillary plexus vessel density • Deep capillary plexus vessel density
Cakmak et al.	2020	Case-control	Fabry Disease	25 subjects 37 controls	OCTA	RTVue-XR OptoVue Avanti	Native OCT software (AVANTI v 2018.0.0.18)	Yes	• Superficial capillary plexus vessel density • Deep capillary plexus vessel density • Foveal avascular zone area
Cennamo et al.	2019	Case-control	Fabry Disease	54 subjects 70 controls	OCTA	RTVue-XR OptoVue Avanti	Native OCT software (AngioAnalytic)	No	• Superficial capillary plexus vessel density • Deep capillary plexus vessel density
Cennamo et al.	2020	Cross-sectional	Fabry Disease	50 subjects	OCTA	RTVue-XR OptoVue Avanti	Native OCT software (ReVue XR v2017.1.0.151 & AngioAnalytic)	No	• None
Dogan et al.	2020	Case-control	Fabry Disease	38 subjects 40 controls	OCTA	RTVue-XR OptoVue Avanti	Native OCT software (AVANTI v 2016.2.0)	Yes	• Central macular thickness • Deep capillary plexus vessel density • Choriocapillaris flow area.

(*Continued*)

Table 5. (Continued)

Study	Year	Design	CSVD Phenotype(s)	No. Participants	Imaging Modality	Device	Software	Pupil Dilation	Biomarkers Identified
Fledelius et al.	2015	Cross-sectional	Fabry Disease	37 subjects	Fundus Photography	N/A	N/A	Yes	• CRAO • Retinal arterial narrowing • Retinal arterial tortuosity • Retinal venous tortuosity
Lin et al.	2021	Case-control	Fabry Disease	26 subjects 28 controls	OCT	Zeiss Cirrus HD 5000	Native OCT software (Angioplex)	Yes	• Choroidal thickness
				26 subjects 28 controls	OCTA	Zeiss Cirrus HD 5000	Native OCT software (Angioplex)	Yes	• Macular vessel length • Superficial capillary plexus vessel density • Superficial capillary plexus vessel length
Michaud	2019	Cross-sectional	Fabry Disease	28 subjects	Fundus Photography	Canon Camera	N/A	Yes	• Retinal vessel tortuosity
				28 subjects	Visual Field	FDT Welch Allyn	N/A	Yes	• Unspecified VF defects
Minnella et al.	2019	Case-control	Fabry Disease	20 subjects 17 controls	Fundus Photography	Topcon DRI Triton	Matlab	Yes	• Retinal vessel tortuosity
				20 subjects 17 controls	OCT	Topcon DRI Triton	Native OCT software	Yes	• None
				20 subjects 17 controls	OCTA	Topcon DRI Triton	Native OCT software	Yes	• Superficial capillary plexus vessel density • Deep capillary plexus vessel density • Perifoveal blood flow • Foveal avascular zone area
				20 subjects 17 controls	ERG	N/A	N/A	Yes	• Decreased ERG response amplitude
Morier et al.	2010	Cross-sectional	Fabry Disease	23 subjects	Fundus Photography	Kowa AD 5mp camera	N/A	Yes	• Retinal vessel tortuosity
				23 subjects	OCT	Zeiss Stratus	N/A	Yes	• None
San Román et al.	2017	Cross-sectional	Fabry Disease	10 subjects	Fundus Photography	Zeiss Visucam Pro	Custom engineered software [40]	Yes	• Retinal vessel tortuosity • Retinal venous tortuosity • Retinal arterial tortuosity
				10 subjects	OCT	Zeiss Cirrus HD 5000	N/A	Yes	• None
Sodi et al.	2013	Case-control	Fabry Disease	35 subjects 35 controls	Fundus Photography	Zeiss TF 450 Plus Canon CF 60 UVI Topcon TRC-50VT	Custom engineered software	Yes	• Retinal vessel tortuosity
Sodi et al.	2019	Case-control	Fabry Disease	11 subjects 11 controls	Fundus Photography	Zeiss TF 450 Plus	Custom engineered software [40]	Yes	• Retinal vessel tortuosity • Retinal venous tortuosity • Retinal arterial tortuosity
Sodi et al.	2020	Cross-sectional	Fabry Disease	18 subjects	Fundus Photography	Imagine Eyes rtx1, i2k Align Retina software	Native rtx1 camera software	No	• Retinal Venous Tortuosity

(*Continued*)

Table 5. (Continued)

Study	Year	Design	CSVD Phenotype(s)	No. Participants	Imaging Modality	Device	Software	Pupil Dilation	Biomarkers Identified
Sodi et al.	2021	Case-control	Fabry Disease	8 subjects 8 controls	Fundus Photography	N/A	Custom engineered software [40]	Yes	• Retinal arterial diameter
Wiest et al.	2021	Cross-sectional	Fabry Disease	57 subjects	Fundus Photography	Optos	N/A	No	• Retinal vessel tortuosity
				57 subjects	OCTA	Zeiss PLEX Elite 9000	Native OCT software (v2.0.1.47652; Macular Density v0.7.1)	No	• Retinal vessel tortuosity • Superficial capillary plexus vessel density
Orssaud et al.	2003	Cross-sectional	Fabry Disease	32 subjects	Visual Field	Goldmann	N/A	Yes	• Enlarged blind spot
Pitz et al.	2009	Cross-sectional	Fabry Disease	31 subjects	Visual Field	HVF 30–2	N/A	No	• Heterogenous VF defects

Abbreviations: CSVD = Cerebral Small Vessel Disease, ERG = Electroretinography, OCT = Optical Coherence Tomography, OCTA = Optical Coherence Tomography Angiography, WMH = White Matter Hyperintensities.

controls per study was 27 (range 8–70). Among included studies, 11 employed fundus photography [34–44], five used OCT [34,37–39,53], eight used OCTA [37,44,53,60–64], three leveraged VF assessment [36,67,68] and one presented results of ERG testing [37]. FA and VEP were the only imaging methodology not employed in published reports. Adequately detailed information on device and methodology used was provided by 17 of 19 studies (89%), and eye dilation was performed in 15 of 19 studies (79%). In studies using fundus photography, investigators repeatedly found associations between Fabry disease and several retinal vascular abnormalities, including retinal vessel tortuosity [34,39,40], retinal arteriolar narrowing [35], and decreased retinal arteriolar diameter [43]. There were no retinal biomarkers emerging as associated with Fabry disease diagnosis or severity in the five identified studies incorporating OCT imaging. Among eight studies conducting OCTA imaging, vessel density and foveal avascular zone area were most frequently reported as associated with Fabry disease diagnosis or severity. Six studies found decreased vessel density in the deep and/or superficial capillary plexus in Fabry patients [37,44,53,60,61,64]. One study reported vessel density as increased in the deep capillary plexus and decreased in the superficial capillary plexus [63]. Another study found no association between any retinal vessel density metrics and Fabry disease [62]. Regarding foveal avascular zone area, three papers [53,60,64] reported no difference between Fabry cases and controls and two papers [37,61] reported enlargement in affected individuals. Less frequently reported OCTA biomarkers found to be associated with Fabry disease were choriocapillaris flow area [64], perifoveal flow area [37], and macular vessel average length [53]. Studies incorporating VF assessment reported heterogenous abnormalities in Fabry disease patients, including multiple unspecified defects [36], blind spot enlargement [67,68], and scattered central scotomas [68]. One study of Fabry disease using ERG reported decreased in ERG mean amplitude among affected individuals [37].

Retinal biomarkers in CADASIL

We identified 11 studies of retinal biomarkers in CADASIL, including 184 affected individuals and 142 healthy controls (Table 6). The median number of CADASIL patients per study was 30 (range 3–38) and the median number of controls per study was 16 (range 4–27). We identified five studies employing fundus photography [45–49], six employing OCT [45,48,54–57]

Table 6. Summary of design, patient characteristics, methodology and results for studies of CADASIL.

Study	Year	Design	CSVD Phenotype(s)	No. Participants	Imaging Modality	Device	Software	Pupil Dilation	Biomarkers Identified
Alten et al.	2014	Case-control	CADASIL	14 subjects 14 controls	Fundus Photography	Zeiss Visucam	ImageJ (semi-automated plugin)	No	• Arterio-venous nicking • Retinal venous dilation
				14 subjects 14 controls	OCT	Heidelberg Spectralis	Heidelberg Eye Explorer software	No	• RNFL thickness • Retinal vessel diameter
				14 subjects 14 controls	FA	Heidelberg Spectralis	N/A	No	• None
Cavallari et al.	2011	Cross-sectional	CADASIL WMH	10 subjects 10 controls	Fundus Photography	N/A	ImageJ (with FracLac plugin)	No	• Retinal vessel fractal dimension
Cumurciuc et al.	2004	Cross-sectional	CADASIL	18 subjects	Fundus Photography	N/A	N/A	No	• Heterogenous retinal abnormalities
				18 subjects	Visual Field	N/A	N/A	No	• No VF defects
				18 subjects	FA	N/A	N/A	No	• Heterogenous retinal findings
Fang et al.	2017	Case-control	CADASIL	27 subjects 27 controls	OCT	Heidelberg Spectralis	Heidelberg Eye Explorer software	No	• Choroidal thickness • Retinal arterial diameter • Retinal venous diameter • Arterio-venous Wall thickness
Nelis et al.	2018	Case-control	CADASIL	11 subjects 21 controls	OCT	Heidelberg Spectralis	Heidelberg Eye Explorer software	Yes	• None
				11 subjects 21 controls	OCTA	RTVue-XR OptoVue Avanti	ImageJ (v 1.51n)	Yes	• Deep capillary plexus vessel density
Parisi et al.	2000	Case-Control	CADASIL	3 subjects 4 controls	ERG	BM 6000 Ganzfeld	Native BM 6000 software	Yes	• Delayed PERG responses
Parisi et al.	2003	Case-Control	CADASIL	6 subjects 14 controls	ERG	BM 6000 Ganzfeld [66]	Native BM 6000 software [66]	Yes	• Delayed ERG, OP and PERG responses
Parisi et al.	2007	Case-control	CADASIL	6 subjects 16 controls	OCT	Humphrey OCT3	Native OCT software	Yes	• RNFL thickness
Pretegiani et al.	2013	Cross-sectional	CADASIL	34 subjects	Fundus Photography	N/A	N/A	No	• Retinal arteriolar narrowing • Retinal venous dilation
				34 subjects	OCT	Zeiss Stratus 3000 OCT	N/A	No	• RNFL thickness
				34 subjects	Visual Field	Automated Perimetry or Goldmann Perimetry	N/A	No	• Heterogenous visual field defects
				34 subjects	FA	N/A	N/A	No	• Heterogenous retinal findings
				34 subjects	VEP	N/A	N/A	No	• Heterogeneous VEP abnormalities
Roine et al.	2006	Cross-sectional	CADASIL	38 subjects 16 controls	Fundus Photography	N/A	Olympus DP-SOFT v 3.2	Yes	• Retinal arteriolar narrowing • Arterio-venous ratios • Arterio-venous nicking

(Continued)

Table 6. (Continued)

Study	Year	Design	CSVD Phenotype(s)	No. Participants	Imaging Modality	Device	Software	Pupil Dilation	Biomarkers Identified
Rufa et al.	2011	Case-control	CADASIL	17 subjects 20 controls	OCT	Zeiss Stratus 3000 OCT	Peripapillary fast RNFL program (v 3.0)	Yes	• RNFL thickness
				17 subjects 20 controls	VEP	N/A	N/A	Yes	• Heterogeneous VEP abnormalities

Abbreviations: CADASIL = Cerebral Autosomal Dominant Arteriopathy with Subcortical Infarcts and Leukoencephalopathy, CSVD = Cerebral Small Vessel Disease, ERG = Electroretinography, FA = Fluorescein Angiography, OCT = Optical Coherence Tomography, OCTA = Optical Coherence Tomography Angiography, OP = Oscillatory Potentials, PERG = Pattern Electroretinography, RNFL = Retinal Nerve Fiber Layer, VEP = Visual Evoked Potentials, WMH = White Matter Hyperintensities.

and one OCTA [55], two studies presenting VF assessment results [47,48], three including FA results [45,47,48], two conducting ERG [65,66], and two including VEP data [48,57]. We determined that 6 of 11 studies (55%) provided detailed information on both the device utilized and methodology. Pupil dilation was performed and adequately reported from a methodological standpoint in 6 of 11 studies (55%). Among fundus photography studies, microvascular abnormalities were found to be associated with CADASIL diagnosis, including, specifically, arteriolar narrowing [48,49] and AV nicking [45,49]. One study reported lower retinal vessel fractal dimensions in CADASIL cases compared to controls [46]. OCT imaging identified decreased RNFL thickness as associated with CADASIL in 3 of 6 studies, either in all quadrants [48,56] or specifically in the temporal quadrant [57]. A single OCTA study identified decreased vessel density in the deep retinal plexus in CADASIL patients compared to healthy controls [55]. In two cross-sectional studies performing VF assessment in CADASIL patients there were no specific abnormalities found to be consistently present in affected individuals, though a number of isolated heterogenous abnormalities were reported [47,48]. Similarly, the results of three studies employing FA were notable for isolated findings (RPE changes, scattered drusen) in a handful of affected individuals [45,47,48]. Two case-control studies employing ERG identified delayed ERG, oscillatory potential (OP) and pattern ERG (PERG) responses [65,66]. Both studies employing VEP reported asymmetric P100 latency and bilateral increase in P100 delay, but these findings were present in less than half of affected individuals [48,57].

Retinal biomarkers in MELAS

We found two studies investigating retinal biomarkers in MELAS (Table 7). An older study presents fundus photography and ERG data from 26 individuals with genetically confirmed MELAS diagnosis [50]. The investigators identified paramacular RPE atrophy in 10 of 26 patients (38%), and found decreased ERG response amplitude, increased latency, or both in 7 of 8 patients who underwent electrodiagnostic evaluation (88%) A more recent study performed OCT imaging on 10 affected individuals and 5 healthy controls [58]. The investigators found lower GCL thickness among MELAS patients compared to controls (after adjusting for prior episodes of transient homonymous hemianopia potentially accounting for direct retinal disease involvement). Lower GCL thickness was also associated with longer disease duration among affected individuals.

Discussion

In this systematic review we identified 48 studies investigating associations between retinal biomarkers and different forms of CSVD, including a total of 21423 participants (9147 CSVD

Table 7. Summary of design, patient characteristics, methodology and results for studies of MELAS.

Study	Year	Design	CSVD Phenotype(s)	No. Participants	Imaging Modality	Device	Software	Pupil Dilation	Biomarkers Identified
Latvala et al.	2002	Cross-sectional	MELAS	26 subjects	Fundus Photography	Canon FC-60Z connected to Kodak digital camera system	N/A	Yes	• RPE atrophy
				8 subjects	ERG	Nicolet Viking II	N/A	Yes	• Decreased ERG amplitude • Increased ERG latency
Shinkai et al.	2021	Cross-sectional	MELAS	10 subjects 5 controls	OCT	Nidek RS-3000 Advance	Native OCT software (v 1.5.5.0)	No	• GCL thickness

Abbreviations: CSVD = Cerebral Small Vessel Disease, ERG = Electroretinography, MELAS = Mitochondrial Encephalomyopathy, Lactic Acidosis, and Stroke-like episodes syndrome, OCT = Optical Coherence Tomography.

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cases and 12276 healthy controls). Overall, our review identified multiple reported associations between retinal biomarkers and CSVD-related clinical outcomes and neuroimaging metrics. From a purely theoretical standpoint, retinal biomarkers could therefore replace (or at least complement) neuroimaging in initial diagnosis and longitudinal follow-up of CSVD, owing to: 1) lower costs associated with acquisition and operation of retinal evaluation scanners vs. MRI scanners; 2) availability of multiple options (fundus photography, OCT, OCTA) for retinal evaluation using portable devices; 3) relative availability of personnel trained to perform retinal vs. neuroimaging evaluations, and interpret study results [8]. These advantages would also make screening of asymptomatic at risk individuals potentially feasible, in a way that MRI-based neuroimaging is currently not capable of. However, published evidence falls short of clearly quantifying the diagnostic performance sensitivity of these biomarkers; thus, we could not definitively assess their relevance and yield regarding future research studies and clinical practice. Therefore, our systematic review highlights the need for larger, more adequately powered and specifically designed studies in order to address these open questions.

We found substantial heterogeneity in sample size across included studies, ranging from small cross-sectional surveys including a handful of cases to large cohort studies with thousands of participants. Larger studies utilizing a cohort format would generally be expected to provide more robust information on the association between retinal biomarkers and CSVD. However, the large cohort studies included in our review exclusively utilized fundus photography for retinal evaluation, thus being unable to access insights provided by more recent studies employing OCT/OCTA to study neurodegenerative and neurovascular disorders [8]. Indeed, most participating studies employed a single retinal evaluation modality, with only a handful combining two or more modalities. Therefore, our findings point to the need for large, adequately powered studies of retinal biomarkers that utilize validated standardized methodologies for capture of CSVD-related clinical outcomes and neuroimaging markers [1]. Additional desirable features for planned future studies of retinal biomarkers in CSVD include the use of multiple retinal evaluation technologies and longitudinal evaluation of biomarkers (both retinal and neuroimaging) over time, to be correlated with CSVD clinical progression in the form of subsequent stroke risk and cognitive decline. Eventually, these longitudinal studies incorporating parallel, repeat retinal and brain imaging over time would also be instrumental in clarify the biological relationships existing between neuronal and microvascular changes occurring in different anatomical locations [8].

Our findings also emphasize the importance of adopting more standardized approaches to study design (with specific emphasis on longitudinal retinal evaluation and estimation of

adequate sample sizes on the basis of robust power calculations) and execution (emphasizing careful and detailed reporting of hardware, software, protocols, and data acquisition parameters employed). Of note, detailed recommendations for design, execution, and results' reporting are currently available only for OCT/OCTA studied (in the form of the APOSTEL 2.0 recommendations) [17]. Future CSVD studies employing other technologies would benefit from consensus-driven formulation of similar guidelines, which would in turn enhance scientific rigor and reproducibility of reported findings.

The vast majority of participants in this systematic review were enrolled in studies investigating sporadic CSVD. In all included reports, a diagnosis was made via a combination of clinical history (usually CSVD-related lacunar stroke) and neuroimaging (usually lacunes or white matter disease). However, none of the included studies conducted subtyping of sporadic CSVD to determine the relative prevalence of its two most common subtypes, CAA and HTNA. While we did identify dedicated studies of retinal biomarkers in CAA, HTNA has so far not yet been evaluated in-depth. In addition, variations in CSVD diagnostic criteria resulted in heterogeneity in clinical severity, ranging from asymptomatic, to minor stroke, to advanced cognitive decline or severe stroke. Finally, the overwhelming majority of studies focused only on WMH and lacunar infarcts as neuroimaging CSVD markers, while neglecting all others [7]. Taken together, published evidence supports an association between retinal microvasculature abnormalities and sporadic CSVD, whether quantified as discrete findings (microvascular abnormalities, defined as presence of hemorrhages, arteriolar narrowing, venular dilation, or AV nicking), vessel diameter, or fractal dimension. As previously mentioned, published studies have yet to provide reliable estimates for the diagnostic performance of retinal biomarkers in CSVD diagnosis or staging, a key prerequisite for more widespread application to research endeavors and introduction in clinical practice.

Despite the relevance of CAA as a major contributor to stroke incidence and cognitive decline [70,71], we found only three studies investigating retinal biomarkers in CAA that included 61 participants in total. While accounting for limitations due to very small sample size, these studies identified retinal hemorrhages as potentially sensitive markers of CAA and correlated them with hemorrhagic CNS disease burden. However, similar findings were identified upon reviewing studies focusing on sporadic CSVD at large, as mentioned above. It remains to be determined whether retinal hemorrhages represent specific retinal biomarkers for CAA (as opposed to HTNA) that were included in studies of CSVD at large due to incomplete clinical characterization. It is alternatively possible that retinal hemorrhages represent shared retinal biomarkers in all forms of CSVD, regardless of subtype. No OCT or OCTA derived retinal biomarker associated with CAA has emerged to date, although limited sample size and substantial differences in methodology and approaches are likely responsible. Indeed, larger, more adequately powered studies of CAA including at minimum fundus photography, OCT and OCTA are warranted based on findings from this systematic review.

Despite being an uncommon diagnosis in clinical practice, reports investigating retinal biomarkers in Fabry disease accounted for 40% of studies included in this systematic review, and included 861 participants in total. Investigators also reported on a wide array of retinal evaluation modalities for this CSVD subtype, with only fluorescein angiography and VEP lacking dedicated studies. Overall, our review findings indicate that retinal microvascular abnormalities are frequently identified in Fabry disease as either discrete abnormalities or decreased vessel density and may therefore represent sensitive biomarkers. However, similar findings were reported in studies of sporadic CSVD (as reported above) and may therefore not be specific to Fabry disease. Additional studies are warranted to expand upon these observations and categorize retinal vascular biomarkers in a systematic fashion, ideally combining different methodologies in each study to increase likelihood of identifying patterns specific to this condition.

We found a substantial number of studies (23% of total) investigating retinal biomarkers in CADASIL. This CSVD disorder was also the only condition with published reports for all retinal evaluation modalities considered in this systematic review. Taken together, available evidence indicates that retinal microvascular changes and decreased RNFL thickness may represent sensitive markers for CADASIL. As previously discussed for Fabry disease, these or very similar findings have also been reported in sporadic CSVD, raising concerns about their diagnostic specificity. Electrodiagnostic studies (ERG and VEP) also uncovered a variety of abnormal findings in CADASIL patients, though not consistently and only in a subset of affected individuals. Larger studies are warranted to clarify the diagnostic performance of retinal vascular measures in CADASIL and to systematically assess the relevance of previously identified electrodiagnostic abnormalities.

We included MELAS as a CSVD disorder in our systematic review on the basis of prior reports implicating small vessel vasculopathy in the pathogenesis of stroke associated with this condition. We identified only two studies of retinal biomarkers in MELAS that did not find definitive associations. Therefore, there is currently no evidence as to whether MELAS-related small vessel vasculopathy can be identified in the retina. Currently published findings (albeit limited in terms of small sample size) support the hypothesis of retinal involvement in MELAS, thus warranting additional, larger studies of its impact on neuronal and vascular biomarkers.

Conclusions

In this systematic review we identified associations between several retinal biomarkers and CSVD-related clinical outcomes (stroke and cognitive impairment) and neuroimaging findings (chiefly white matter disease, lacunes, and cerebral microbleeds). Retinal microvascular abnormalities identified via either fundus photography, OCT or OCTA have so far generated the largest amount of published evidence for association with CSVD. However, definitive evidence on the performance of retinal biomarkers in diagnosing CSVD and following its progression over time is currently lacking. The majority of published studies also suffered from several methodological limitations, chiefly small sample sizes and inadequate reporting of key factors in study design, protocols for data capture, and analytical methods. Larger, adequately powered studies employing standardized methodologies for both retinal evaluation and CSVD characterization (ideally incorporating repeated measurements over time) are therefore required to definitively establish the potential impact of these technologies in future research efforts and clinical practice.

Supporting information

S1 Checklist. Retinal imaging in CSVD review—PRISMA checklist.
(PDF)

S1 File. Search strategy. Terms, syntax, and databases used to perform systematic review of scientific literature.
(DOCX)

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References

1. Cannistraro RJ, Badi M, Eidelman BH, Dickson DW, Middlebrooks EH, Meschia JF. CNS small vessel disease: A clinical review. Neurology. 2019; 92(24):1146–56. Epub 2019/05/31. https://doi.org/10.1212/WNL.0000000000007654 PMID: 31142635; PubMed Central PMCID: PMC6598791.

2. Li Q, Yang Y, Reis C, Tao T, Li W, Li X, et al. Cerebral Small Vessel Disease. Cell transplantation. 2018; 27(12):1711–22. Epub 2018/09/27. https://doi.org/10.1177/0963689718795148 PMID: 30251566; PubMed Central PMCID: PMC6300773.

3. Ostergaard L, Engedal TS, Moreton F, Hansen MB, Wardlaw JM, Dalkara T, et al. Cerebral small vessel disease: Capillary pathways to stroke and cognitive decline. Journal of cerebral blood flow and metabolism: official journal of the International Society of Cerebral Blood Flow and Metabolism. 2016; 36 (2):302–25. Epub 2015/12/15. https://doi.org/10.1177/0271678X15606723 PMID: 26661176; PubMed Central PMCID: PMC4759673.

4. Pantoni L. Cerebral small vessel disease: from pathogenesis and clinical characteristics to therapeutic challenges. Lancet neurology. 2010; 9(7):689–701. https://doi.org/10.1016/S1474-4422(10)70104-6 PMID: 20610345.

5. Shi Y, Wardlaw JM. Update on cerebral small vessel disease: a dynamic whole-brain disease. Stroke and vascular neurology. 2016; 1(3):83–92. Epub 2017/09/30. https://doi.org/10.1136/svn-2016-000035 PMID: 28959468; PubMed Central PMCID: PMC5435198.

6. Federico A, Di Donato I, Bianchi S, Di Palma C, Taglia I, Dotti MT. Hereditary cerebral small vessel diseases: a review. Journal of the neurological sciences. 2012; 322(1–2):25–30. Epub 2012/08/08. https://doi.org/10.1016/j.jns.2012.07.041 PMID: 22868088.

7. Wardlaw JM, Smith EE, Biessels GJ, Cordonnier C, Fazekas F, Frayne R, et al. Neuroimaging standards for research into small vessel disease and its contribution to ageing and neurodegeneration. Lancet neurology. 2013; 12(8):822–38. https://doi.org/10.1016/S1474-4422(13)70124-8 PMID: 23867200; PubMed Central PMCID: PMC3714437.

8. Kashani AH, Asanad S, Chan JW, Singer MB, Zhang J, Sharifi M, et al. Past, present and future role of retinal imaging in neurodegenerative disease. Progress in retinal and eye research. 2021; 83:100938. Epub 2021/01/19. https://doi.org/10.1016/j.preteyeres.2020.100938 PMID: 33460813; PubMed Central PMCID: PMC8280255.

9. Liao H, Zhu Z, Peng Y. Potential Utility of Retinal Imaging for Alzheimer's Disease: A Review. Frontiers in aging neuroscience. 2018; 10:188. Epub 2018/07/11. https://doi.org/10.3389/fnagi.2018.00188 PMID: 29988470; PubMed Central PMCID: PMC6024140.

10. London A, Benhar I, Schwartz M. The retina as a window to the brain-from eye research to CNS disorders. Nature reviews Neurology. 2013; 9(1):44–53. Epub 2012/11/21. https://doi.org/10.1038/nrneurol.2012.227 PMID: 23165340.

11. Page MJ, McKenzie JE, Bossuyt PM, Boutron I, Hoffmann TC, Mulrow CD, et al. The PRISMA 2020 statement: An updated guideline for reporting systematic reviews. PLoS medicine. 2021; 18(3): e1003583. Epub 2021/03/30. https://doi.org/10.1371/journal.pmed.1003583 PMID: 33780438; PubMed Central PMCID: PMC8007028.

12. Mancuso M, Arnold M, Bersano A, Burlina A, Chabriat H, Debette S, et al. Monogenic cerebral small-vessel diseases: diagnosis and therapy. Consensus recommendations of the European Academy of Neurology. European journal of neurology: the official journal of the European Federation of Neurological Societies. 2020; 27(6):909–27. Epub 2020/03/21. https://doi.org/10.1111/ene.14183 PMID: 32196841.

13. Ghannam ASB, Subramanian PS. Neuro-ophthalmic manifestations of cerebrovascular accidents. Current opinion in ophthalmology. 2017; 28(6):564–72. Epub 2017/10/07. https://doi.org/10.1097/ICU.0000000000000414 PMID: 28984724.

14. Sachdev P, Kalaria R, O'Brien J, Skoog I, Alladi S, Black SE, et al. Diagnostic criteria for vascular cognitive disorders: a VASCOG statement. Alzheimer Dis Assoc Disord. 2014; 28(3):206–18. https://doi.org/10.1097/WAD.0000000000000034 PMID: 24632990; PubMed Central PMCID: PMC4139434.

15. Arsava EM, Helenius J, Avery R, Sorgun MH, Kim GM, Pontes-Neto OM, et al. Assessment of the Predictive Validity of Etiologic Stroke Classification. JAMA neurology. 2017; 74(4):419–26. https://doi.org/10.1001/jamaneurol.2016.5815 PMID: 28241214; PubMed Central PMCID: PMC5470360.

16. von Elm E, Altman DG, Egger M, Pocock SJ, Gøtzsche PC, Vandenbroucke JP. The Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) statement: guidelines for reporting observational studies. PLoS medicine. 2007; 4(10):e296. Epub 2007/10/19. https://doi.org/10.1371/journal.pmed.0040296 PMID: 17941714; PubMed Central PMCID: PMC2020495.

17. Aytulun A, Cruz-Herranz A, Aktas O, Balcer LJ, Balk L, Barboni P, et al. APOSTEL 2.0 Recommendations for Reporting Quantitative Optical Coherence Tomography Studies. Neurology. 2021; 97(2):68–79. Epub 2021/04/30. https://doi.org/10.1212/WNL.0000000000012125 PMID: 33910937; PubMed Central PMCID: PMC8279566.

18. Britze J, Frederiksen JL. Optical coherence tomography in multiple sclerosis. Eye (London, England). 2018; 32(5):884–8. Epub 2018/02/03. https://doi.org/10.1038/s41433-017-0010-2 PMID: 29391574; PubMed Central PMCID: PMC5944645.

19. Petzold A, Balcer LJ, Calabresi PA, Costello F, Frohman TC, Frohman EM, et al. Retinal layer segmentation in multiple sclerosis: a systematic review and meta-analysis. Lancet neurology. 2017; 16 (10):797–812. Epub 2017/09/19. https://doi.org/10.1016/S1474-4422(17)30278-8 PMID: 28920886.

20. Wu HQ, Wu H, Shi LL, Yu LY, Wang LY, Chen YL, et al. The association between retinal vasculature changes and stroke: a literature review and Meta-analysis. International journal of ophthalmology. 2017; 10(1):109–14. Epub 2017/02/06. https://doi.org/10.18240/ijo.2017.01.18 PMID: 28149786; PubMed Central PMCID: PMC5225358.

21. Cheung N, Liew G, Lindley RI, Liu EY, Wang JJ, Hand P, et al. Retinal fractals and acute lacunar stroke. Annals of neurology. 2010; 68(1):107–11. Epub 2010/06/29. https://doi.org/10.1002/ana.22011 PMID: 20582985.

22. Doubal FN, MacGillivray TJ, Patton N, Dhillon B, Dennis MS, Wardlaw JM. Fractal analysis of retinal vessels suggests that a distinct vasculopathy causes lacunar stroke. Neurology. 2010; 74(14):1102–7. Epub 2010/04/07. https://doi.org/10.1212/WNL.0b013e3181d7d8b4 PMID: 20368631; PubMed Central PMCID: PMC2865776.

23. McGrory S, Ballerini L, Doubal FN, Staals J, Allerhand M, Valdes-Hernandez MDC, et al. Retinal microvasculature and cerebral small vessel disease in the Lothian Birth Cohort 1936 and Mild Stroke Study. Scientific reports. 2019; 9(1):6320. Epub 2019/04/21. https://doi.org/10.1038/s41598-019-42534-x PMID: 31004095; PubMed Central PMCID: PMC6474900.

24. Hilal S, Ong YT, Cheung CY, Tan CS, Venketasubramanian N, Niessen WJ, et al. Microvascular network alterations in retina of subjects with cerebral small vessel disease. Neuroscience letters. 2014; 577:95–100. Epub 2014/06/18. https://doi.org/10.1016/j.neulet.2014.06.024 PMID: 24937268.

25. Ikram MK, De Jong FJ, Van Dijk EJ, Prins ND, Hofman A, Breteler MM, et al. Retinal vessel diameters and cerebral small vessel disease: the Rotterdam Scan Study. Brain. 2006; 129(Pt 1):182–8. Epub 2005/12/01. https://doi.org/10.1093/brain/awh688 PMID: 16317022.

26. Kim M, Park KH, Kwon JW, Jeoung JW, Kim TW, Kim DM. Retinal nerve fiber layer defect and cerebral small vessel disease. Invest Ophthalmol Vis Sci. 2011; 52(9):6882–6. Epub 2011/07/28. https://doi.org/10.1167/iovs.11-7276 PMID: 21791593.

27. Kwa VI, van der Sande JJ, Stam J, Tijmes N, Vrooland JL. Retinal arterial changes correlate with cerebral small-vessel disease. Neurology. 2002; 59(10):1536–40. Epub 2002/11/27. https://doi.org/10.1212/01.wnl.0000033093.16450.5c PMID: 12451193.

28. Lee MJ, Deal JA, Ramulu PY, Sharrett AR, Abraham AG. Prevalence of Retinal Signs and Association With Cognitive Status: The ARIC Neurocognitive Study. J Am Geriatr Soc. 2019; 67(6):1197–203. Epub 2019/02/02. https://doi.org/10.1111/jgs.15795 PMID: 30706941; PubMed Central PMCID: PMC6698148.

29. Shu L, Liang J, Xun W, Yang H, Lu T. Prediction for the Total MRI Burden of Cerebral Small Vessel Disease With Retinal Microvascular Abnormalities in Ischemic Stroke/TIA Patients. Front Neurol. 2020; 11:268. Epub 2020/05/07. https://doi.org/10.3389/fneur.2020.00268 PMID: 32373049; PubMed Central PMCID: PMC7177024.

30. Yatsuya H, Folsom AR, Wong TY, Klein R, Klein BE, Sharrett AR. Retinal microvascular abnormalities and risk of lacunar stroke: Atherosclerosis Risk in Communities Study. Stroke; a journal of cerebral circulation. 2010; 41(7):1349–55. Epub 2010/06/05. https://doi.org/10.1161/STROKEAHA.110.580837 PMID: 20522816; PubMed Central PMCID: PMC2894269.

31. Alber J, Arthur E, Goldfarb D, Drake J, Boxerman JL, Silver B, et al. The relationship between cerebral and retinal microbleeds in cerebral amyloid angiopathy (CAA): A pilot study. Journal of the neurological sciences. 2021; 423:117383. Epub 2021/03/09. https://doi.org/10.1016/j.jns.2021.117383 PMID: 33684655.

32. Lee A, Rudkin A, Agzarian M, Patel S, Lake S, Chen C. Retinal vascular abnormalities in patients with cerebral amyloid angiopathy. Cerebrovasc Dis. 2009; 28(6):618–22. Epub 2009/10/24. https://doi.org/10.1159/000251173 PMID: 19851067.

33. van Etten ES, de Boer I, Steenmeijer SR, Al-Nofal M, Wermer MJH, Notting IC, et al. Optical coherence tomography detects retinal changes in hereditary cerebral amyloid angiopathy. European journal of neurology: the official journal of the European Federation of Neurological Societies. 2020; 27(12):2635–40. Epub 2020/09/08. https://doi.org/10.1111/ene.14507 PMID: 32894579; PubMed Central PMCID: PMC7702135.

34. Atiskova Y, Rassuli R, Koehn AF, Golsari A, Wagenfeld L, du Moulin M, et al. Retinal hyperreflective foci in Fabry disease. Orphanet journal of rare diseases. 2019; 14(1):296. Epub 2019/12/28. https://doi.org/10.1186/s13023-019-1267-2 PMID: 31878969; PubMed Central PMCID: PMC6933914.

35. Fledelius HC, Sandfeld L, Rasmussen Å K, Madsen CV, Feldt-Rasmussen U. Ophthalmic experience over 10 years in an observational nationwide Danish cohort of Fabry patients with access to enzyme replacement. Acta ophthalmologica. 2015; 93(3):258–64. Epub 2014/12/10. https://doi.org/10.1111/aos.12588 PMID: 25487570.

36. Michaud L. Longitudinal study on ocular manifestations in a cohort of patients with Fabry disease. PloS one. 2019; 14(6):e0213329. Epub 2019/06/28. https://doi.org/10.1371/journal.pone.0213329 PMID: 31246960; PubMed Central PMCID: PMC6597042 Genzyme Canada. Author received speaker fees from Shire on other topics (dry eye management). There are no patents, products in development or marketed products to declare. This does not alter our adherence to all the PLOS ONE policies on sharing data and materials.

37. Minnella AM, Barbano L, Verrecchia E, Martelli F, Pagliei V, Gambini G, et al. Macular Impairment in Fabry Disease: A Morpho-functional Assessment by Swept-Source OCT Angiography and Focal Electroretinography. Invest Ophthalmol Vis Sci. 2019; 60(7):2667–75. Epub 2019/06/27. https://doi.org/10.1167/iovs.18-26052 PMID: 31242288.

38. Morier AM, Minteer J, Tyszko R, McCann R, Clarke MV, Browning MF. Ocular manifestations of Fabry disease within in a single kindred. Optometry (St Louis, Mo). 2010; 81(9):437–49. Epub 2010/07/10. https://doi.org/10.1016/j.optm.2010.02.011 PMID: 20615758.

39. San Román I, Rodríguez ME, Caporossi O, Zoppetti C, Sodi A, Mecocci A, et al. COMPUTER ASSISTED RETINAL VESSEL TORTUOSITY EVALUATION IN NOVEL MUTATION FABRY DISEASE: Towards New Prognostic Markers. Retina. 2017; 37(3):592–603. Epub 2017/02/23. https://doi.org/10.1097/IAE.0000000000001177 PMID: 28225726.

40. Sodi A, Guarducci M, Vauthier L, Ioannidis AS, Pitz S, Abbruzzese G, et al. Computer assisted evaluation of retinal vessels tortuosity in Fabry disease. Acta ophthalmologica. 2013; 91(2):e113–9. Epub 2012/11/21. https://doi.org/10.1111/j.1755-3768.2012.02576.x PMID: 23164241.

41. Sodi A, Lenzetti C, Bacherini D, Finocchio L, Verdina T, Borg I, et al. Quantitative Analysis of Conjunctival and Retinal Vessels in Fabry Disease. Journal of ophthalmology. 2019; 2019:4696429. Epub 2019/05/17. https://doi.org/10.1155/2019/4696429 PMID: 31093369; PubMed Central PMCID: PMC6481025.

42. Sodi A, Germain DP, Bacherini D, Finocchio L, Pacini B, Marziali E, et al. IN VIVO OBSERVATION OF RETINAL VASCULAR DEPOSITS USING ADAPTIVE OPTICS IMAGING IN FABRY DISEASE.

Retina. 2020; 40(8):1623–9. Epub 2019/10/01. https://doi.org/10.1097/IAE.0000000000002648 PMID: 31568064.

43. Sodi A, Nicolosi C, Vicini G, Lenzetti C, Virgili G, Rizzo S. Computer-assisted retinal vessel diameter evaluation in Fabry disease. European journal of ophthalmology. 2021; 31(1):173–8. Epub 2019/11/14. https://doi.org/10.1177/1120672119886985 PMID: 31718270.

44. Wiest MRJ, Toro MD, Nowak A, Baur J, Fasler K, Hamann T, et al. Globotrioasylsphingosine Levels and Optical Coherence Tomography Angiography in Fabry Disease Patients. Journal of clinical medicine. 2021; 10(5). Epub 2021/04/04. https://doi.org/10.3390/jcm10051093 PMID: 33807900; PubMed Central PMCID: PMC7961664.

45. Alten F, Motte J, Ewering C, Osada N, Clemens CR, Kadas EM, et al. Multimodal retinal vessel analysis in CADASIL patients. PloS one. 2014; 9(11):e112311. Epub 2014/11/06. https://doi.org/10.1371/journal.pone.0112311 PMID: 25372785; PubMed Central PMCID: PMC4221286.

46. Cavallari M, Falco T, Frontali M, Romano S, Bagnato F, Orzi F. Fractal analysis reveals reduced complexity of retinal vessels in CADASIL. PloS one. 2011; 6(4):e19150. Epub 2011/05/11. https://doi.org/10.1371/journal.pone.0019150 PMID: 21556373; PubMed Central PMCID: PMC3083432.

47. Cumurciuc R, Massin P, Pâques M, Krisovic V, Gaudric A, Bousser MG, et al. Retinal abnormalities in CADASIL: a retrospective study of 18 patients. Journal of neurology, neurosurgery, and psychiatry. 2004; 75(7):1058–60. Epub 2004/06/18. https://doi.org/10.1136/jnnp.2003.024307 PMID: 15201374; PubMed Central PMCID: PMC1739116.

48. Pretegiani E, Rosini F, Dotti MT, Bianchi S, Federico A, Rufa A. Visual system involvement in CADASIL. Journal of stroke and cerebrovascular diseases: the official journal of National Stroke Association. 2013; 22(8):1377–84. Epub 2013/05/03. https://doi.org/10.1016/j.jstrokecerebrovasdis.2013.03.032 PMID: 23635925.

49. Roine S, Harju M, Kivelä TT, Pöyhönen M, Nikoskelainen E, Tuisku S, et al. Ophthalmologic findings in cerebral autosomal dominant arteriopathy with subcortical infarcts and leukoencephalopathy: a cross-sectional study. Ophthalmology. 2006; 113(8):1411–7. Epub 2006/08/01. https://doi.org/10.1016/j.ophtha.2006.03.030 PMID: 16877080.

50. Latvala T, Mustonen E, Uusitalo R, Majamaa K. Pigmentary retinopathy in patients with the MELAS mutation 3243A—>G in mitochondrial DNA. Graefe's archive for clinical and experimental ophthalmology = Albrecht von Graefes Archiv fur klinische und experimentelle Ophthalmologie. 2002; 240 (10):795–801. Epub 2002/10/25. https://doi.org/10.1007/s00417-002-0555-y PMID: 12397426.

51. Lee JY, Kim JP, Jang H, Kim J, Kang SH, Kim JS, et al. Optical coherence tomography angiography as a potential screening tool for cerebral small vessel diseases. Alzheimer's research & therapy. 2020; 12 (1):73. Epub 2020/06/13. https://doi.org/10.1186/s13195-020-00638-x PMID: 32527301; PubMed Central PMCID: PMC7291486.

52. Abdelhak A, Huss A, Brück A, Sebert U, Mayer B, Müller HP, et al. Optical coherence tomography-based assessment of retinal vascular pathology in cerebral small vessel disease. Neurological research and practice. 2020; 2:13. Epub 2020/12/17. https://doi.org/10.1186/s42466-020-00062-4 PMID: 33324919; PubMed Central PMCID: PMC7650138.

53. Lin Z, Pan X, Mao K, Jiao Q, Chen Y, Zhong Y, et al. Quantitative evaluation of retinal and choroidal changes in Fabry disease using optical coherence tomography angiography. Lasers in medical science. 2021. Epub 2021/01/08. https://doi.org/10.1007/s10103-020-03242-5 PMID: 33409749

54. Fang XJ, Yu M, Wu Y, Zhang ZH, Wang WW, Wang ZX, et al. Study of Enhanced Depth Imaging Optical Coherence Tomography in Cerebral Autosomal Dominant Arteriopathy with Subcortical Infarcts and Leukoencephalopathy. Chinese medical journal. 2017; 130(9):1042–8. Epub 2017/05/05. https://doi.org/10.4103/0366-6999.204935 PMID: 28469098; PubMed Central PMCID: PMC5421173.

55. Nelis P, Kleffner I, Burg MC, Clemens CR, Alnawaiseh M, Motte J, et al. OCT-Angiography reveals reduced vessel density in the deep retinal plexus of CADASIL patients. Scientific reports. 2018; 8 (1):8148. Epub 2018/05/29. https://doi.org/10.1038/s41598-018-26475-5 PMID: 29802397; PubMed Central PMCID: PMC5970147.

56. Parisi V, Pierelli F, Coppola G, Restuccia R, Ferrazzoli D, Scassa C, et al. Reduction of optic nerve fiber layer thickness in CADASIL. European journal of neurology: the official journal of the European Federation of Neurological Societies. 2007; 14(6):627–31. Epub 2007/06/02. https://doi.org/10.1111/j.1468-1331.2007.01795.x PMID: 17539939.

57. Rufa A, Pretegiani E, Frezzotti P, De Stefano N, Cevenini G, Dotti MT, et al. Retinal nerve fiber layer thinning in CADASIL: an optical coherence tomography and MRI study. Cerebrovasc Dis. 2011; 31 (1):77–82. Epub 2010/11/06. https://doi.org/10.1159/000321339 PMID: 21051887.

58. Shinkai A, Shinmei Y, Hirooka K, Tagawa Y, Nakamura K, Chin S, et al. Optical coherence tomography as a possible tool to monitor and predict disease progression in mitochondrial myopathy,

encephalopathy, lactic acidosis and stroke-like episodes. Mitochondrion. 2021; 56:47–51. Epub 2020/11/22. https://doi.org/10.1016/j.mito.2020.11.003 PMID: 33220496.

59. Wang X, Wei Q, Wu X, Cao S, Chen C, Zhang J, et al. The vessel density of the superficial retinal capillary plexus as a new biomarker in cerebral small vessel disease: an optical coherence tomography angiography study. Neurological sciences: official journal of the Italian Neurological Society and of the Italian Society of Clinical Neurophysiology. 2021; 42(9):3615–24. Epub 2021/01/13. https://doi.org/10.1007/s10072-021-05038-z PMID: 33432462.

60. Bacherini D, Vicini G, Nicolosi C, Tanini I, Lenzetti C, Finocchio L, et al. Optical Coherence Tomography Angiography for the Evaluation of Retinal Vasculature in Fabry Disease: Our Experience and Review of Current Knowledge. Front Neurol. 2021; 12:640719. Epub 2021/03/27. https://doi.org/10.3389/fneur.2021.640719 PMID: 33767663; PubMed Central PMCID: PMC7985262.

61. Cakmak AI, Atalay E, Cankurtaran V, Yaşar E, Turgut FH. Optical coherence tomography angiography analysis of fabry disease. International ophthalmology. 2020; 40(11):3023–32. Epub 2020/07/02. https://doi.org/10.1007/s10792-020-01486-2 PMID: 32607948.

62. Cennamo G, Montorio D, Santoro C, Cocozza S, Spinelli L, Di Risi T, et al. The Retinal Vessel Density as a New Vascular Biomarker in Multisystem Involvement in Fabry Disease: An Optical Coherence Tomography Angiography Study. Journal of clinical medicine. 2020; 9(12). Epub 2020/12/24. https://doi.org/10.3390/jcm9124087 PMID: 33352849; PubMed Central PMCID: PMC7766384.

63. Cennamo G, Di Maio LG, Montorio D, Tranfa F, Russo C, Pontillo G, et al. Optical Coherence Tomography Angiography Findings in Fabry Disease. Journal of clinical medicine. 2019; 8(4). Epub 2019/04/20. https://doi.org/10.3390/jcm8040528 PMID: 30999633; PubMed Central PMCID: PMC6517973.

64. Dogan C, Gonen B, Dincer MT, Mergen B, Kiykim E, Bakir A, et al. Evaluation of the reasons for the microvascular changes in patients with Fabry disease using optic coherence tomography angiography. European journal of ophthalmology. 2021; 31(6):3231–7. Epub 2020/11/24. https://doi.org/10.1177/1120672120974288 PMID: 33225739.

65. Parisi V, Pierelli F, Fattapposta F, Bianco F, Parisi L, Restuccia R, et al. Early visual function impairment in CADASIL. Neurology. 2003; 60(12):2008–10. Epub 2003/06/25. https://doi.org/10.1212/01.wnl.0000070411.13217.7e PMID: 12821756.

66. Parisi V, Pierelli F, Malandrini A, Carrera P, Olzi D, Gregori D, et al. Visual electrophysiological responses in subjects with cerebral autosomal arteriopathy with subcortical infarcts and leukoencephalopathy (CADASIL). Clinical neurophysiology: official journal of the International Federation of Clinical Neurophysiology. 2000; 111(9):1582–8. Epub 2000/08/30. https://doi.org/10.1016/s1388-2457(00)00366-7 PMID: 10964068.

67. Orssaud C, Dufier J, Germain D. Ocular manifestations in Fabry disease: a survey of 32 hemizygous male patients. Ophthalmic genetics. 2003; 24(3):129–39. Epub 2003/07/18. https://doi.org/10.1076/opge.24.3.129.15609 PMID: 12868031.

68. Pitz S, Grube-Einwald K, Renieri G, Reinke J. Subclinical optic neuropathy in Fabry disease. Ophthalmic genetics. 2009; 30(4):165–71. Epub 2009/10/27. https://doi.org/10.3109/13816810903148004 PMID: 19852573.

69. Jeoung JW, Park KH, Kim TW, Khwarg SI, Kim DM. Diagnostic ability of optical coherence tomography with a normative database to detect localized retinal nerve fiber layer defects. Ophthalmology. 2005; 112(12):2157–63. Epub 2005/11/18. https://doi.org/10.1016/j.ophtha.2005.07.012 PMID: 16290196.

70. Greenberg SM, Bacskai BJ, Hernandez-Guillamon M, Pruzin J, Sperling R, van Veluw SJ. Cerebral amyloid angiopathy and Alzheimer disease—one peptide, two pathways. Nature reviews Neurology. 2020; 16(1):30–42. Epub 2019/12/13. https://doi.org/10.1038/s41582-019-0281-2 PMID: 31827267.

71. Biffi A, Greenberg SM. Cerebral amyloid angiopathy: a systematic review. J Clin Neurol. 2011; 7(1):1–9. https://doi.org/10.3988/jcn.2011.7.1.1 PMID: 21519520; PubMed Central PMCID: PMC3079153.