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Original Article

Retinal vasculature – An imaging based morphological study

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ABSTRACT

Aim: Significant clinical associations during subtle and overt illness as well as proven effects of demographics and heredity on retinal vascular appearance call for a comprehensive analysis of retinal blood vessels that is more structured and objective than currant observer-driven techniques. Attending to this gap in information, the present study aims to describe the normal morphology of retinal arteries among healthy north Indian subjects.
Materials and methods: 800 digitalized fundus images from 400 north Indian subjects (180 M & 220 F; mean age 35 years) were quantitatively analyzed based on selective geometric and topographical parameters (distance, direction, tortuosity, branching) of the four retinal quadrant arteries through sophisticated semi automated digital asset management software techniques.
Results: Tortuosity and branching were more extensive in temporal than nasal quadrants, with a tendency for delayed on-start of arteriolar subdivisions, farther away from the optic disc and superior papillary trunks were positioned at keener angles than inferior trunks. A certain degree of dimorphism existed among eye sides and genders.
Conclusion: This study has elucidated retinal vascular geometrics of healthy Indians, a knowledge that can improve our understanding of regular features and natural variants in retinal vasculature. Morphological findings of this research will hopefully facilitate anatomists and clinicians in describing 'what's normal' in health and aid in differentiating it from the abnormal in case of disease. The potential utility of diagnostic and predictive fundus vascular screening in populations for appraisal of health risks mirrored in early retinal changes is a worthwhile direction to explore.

1. Introduction

The human retina is a light sensitive tissue lining the back of the eye which is nourished by two discrete circulatory systems—the retinal arteries nourished by the central retinal vessel and the choroidal blood vessels nourished by the posterior ciliary artery, both of which are derived from ophthalmic artery, which is the first branch of internal carotid

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artery.[1] At it emerges from the lamina cribrosa at the optic nerve head, the central retinal artery divides into superior and inferior papillary trunks, which in turn divide into four branched retinal arteries for the four retinal quadrants: namely the superior temporal, superior nasal, inferior temporal and inferior nasal arteries. These vessels carry important oxygen and nutrients and are vital to the healthy functioning of retina. Variations in the architectural framework of retinal vascular tree have often been observed and may occur normally or are indicative of underlying pathophysiological mechanisms. The retina is the only part of the central nervous system in which the micro vascular bed is visible to direct inspection. This makes it an ideal tissue to study distinctive pathological process of sequestration noninvasively in vivo and in a state of real life action.

Signs of systemic diseases are often reflected in the eye, which provides a window to microcirculation. Researchers have demonstrated associations between retinal vessel abnormalities and glaucoma, cataract, age-related macular degeneration and systemic diseases such as cardiovascular disease, hypertension, diabetes mellitus, renal disorders and stroke.[2,3] Ocular fundus images can provide valuable information on pathological changes of local ocular diseases and the early/first signs of systemic diseases. Therefore, analysis and interpretation of fundus images has become a necessary and important diagnostic and therapeutic tool in ophthalmology. These features are useful in revealing disease state in the form of measurable variables such as vessel length, width, branching pattern, color and tortuosity. Retinal vessels also serve as landmarks for image-guided laser treatments in choroidal neovascularization. These significant clinical associations call for a better understanding of retinal vascular morphology during health and in disease. As stated by Siatkowski, "*Increased knowledge is needed to guide ophthalmologists to a more appropriate and cost-effective treatment of people*". Published literature highlights the effects of demographics[4], heredity[5] and genes[6] on retinal blood vessels. Reliable methods that elucidate various vessel morphological characteristics are therefore needed which justifies the purpose of our research. The present study aims to describe the normal

Fig. 1 – Direction of sup &inf papillary trunk in a 360° fundus plane.

morphology of retinal arteries among North Indian subjects based on selective geometric and topographical analysis of vessels of the four retinal quadrants. To the best of the author's knowledge, such an in depth analysis of retinal vasculature through sophisticated semi automated techniques has not been done among Indian subjects till date. The retinal vascular morphology as revealed by this research will facilitate anatomists and clinicians in describing 'what's normal' in health and aid in differentiating it from the abnormal in case of disease.

2. Materials and methods

Color fundus images of 800 eyes from 400 North Indian subjects (180 M & 220 F; Mean age 34.80 ± 11.45; range 18–45 years) attending ophthalmology OPD and retina clinics of select tertiary hospitals and health care facilities including Subharti hospital Meerut, Sahara hospital, Lucknow, Allahabad eye clinic, Retina clinic Banaras Hindu University hospital, Varanasi, Uttar Pradesh etc. during 2011 through 2013 as part of the first author's PhD research program in anatomy.

2.1. *Inclusion criteria*

Adult male or female, generally 'healthy' and without any overt systemic or metabolic disorder, presenting with minor conditions of lid, conjunctiva, cornea or lens but with an essentially 'unaffected' retina. Criteria for 'healthy' or 'normalcy' were normal ophthalmic findings from fundoscopic grading done independently by trained ophthalmologists, especially normal appearance of the optic disc, normal visual fields, intraocular pressure and lack of significant retinal disorder.

2.2. *Exclusion criteria*

Individuals with evidence of any severe eye disease except refractive error, history of ocular trauma or intraocular surgery within the last 6 months, fulminate ocular inflammation or infection within the last 3 months, astigmatism more than +2.0 dioptres and ametropia of more than 65.0 dioptres. Also excluded were children, pregnant females, chronic diabetics or hypertensives and critically ill patients.

Fundus was photographed after duly taking informed consent (as per declaration of Helsinky).

2.3. *Inclusion criteria for fundus images*

Stereoscopic 35° color retinal photographs with the optic disc well focused within half a disc size off the center taken by mydriatic fundus camera (table-mounted Topcon 50-EX), image 1024 pixels, JPEG format, photographed by trained ophthalmologist and camera magnification quantified with a model to adjust for measurement error. This technique offers standardized assessment of fundus photographs, with sufficient reliability and high reproducibility. All selected images were of high quality, well focused, illuminated and without motion artifacts or noise. Clinical data on patients was entered on a database in context of demographic variables like age (in years), sex (male or female), eye side (right or left). The images were cross-examined by experienced clinical ophthalmologists to re-confirm their 'healthy retina' status in order to avoid confounding of results. Fundus images were

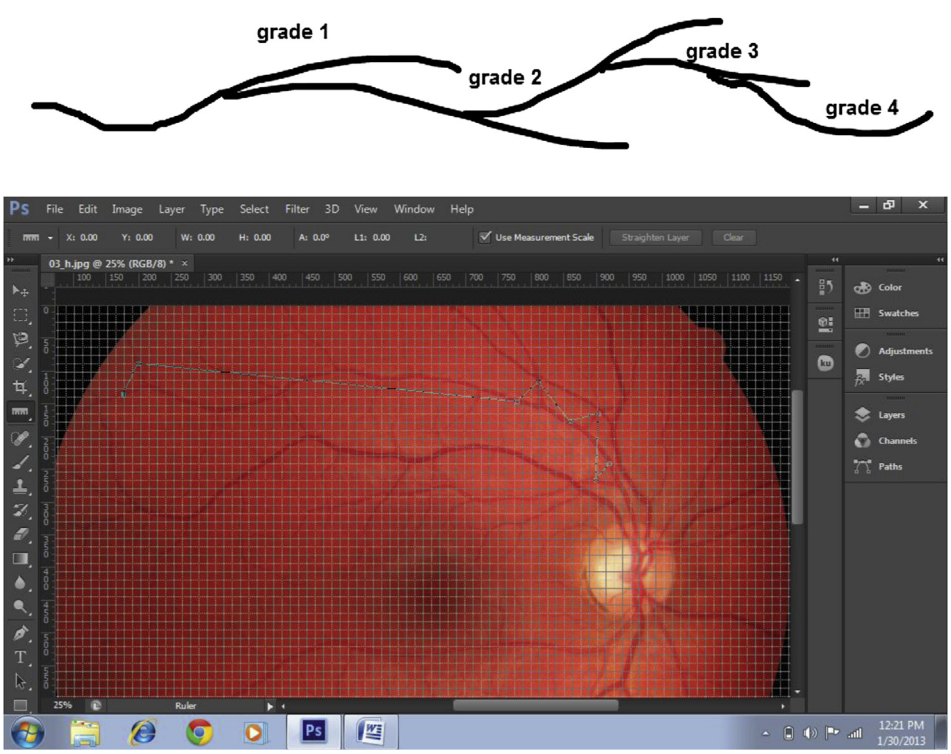

Fig. 2 – Branching pattern of vascular tree from main stem: grade 1 (first order branch); grade 2 (2nd order branch); grade 3 (3rd order branch); grade 4 (4th order branch).

digitalized and analyzed by semi automated software (Bridge CS6 Extended version, digital asset management software) similar to the technique used by others.[7] Arteries were distinguished from veins as per the standard guidelines for reading retinal images.[1] Various geometric features (1–4) of retinal arteries of all four quadrants of fundus viz. superior & inferior temporal & nasal were measured (Figs. 1–5).

1 *Direction*:

Direction of superior and inferior papillary trunks at the central retinal vessel bifurcation point on optic nerve head expressed as vessel angulation in a 360° fundus plane.

2 *Branching arcades*:

Segmental skeletonization of the arterial tree with respect to number of subdivisions. Each pair of branching points was regarded as one level up the branching arcade and sequential grades of branching were expressed as 1st order, 2nd order, 3rd order etc.

3 *Distance*:

Linear distance between the optic disc center and primary bifurcation point of artery expressed as a function of disc diameters (quantified to scale the number of optic disc diameters covered by the length).

4 *Tortuosity*:

Waviness of the artery assessed by qualitative grading based on a three level visual grading scale.

Grade 1 = straight (predominately straight vessels)
Grade 2 = wavy (moderate tortuosity, with one to two inflections of artery along the vascular start to end point)
Grade3 = tortuous (prominent tortuosity with three or more inflections)

Basic descriptive and interferential statistics was used to establish correlations between measurable parameters, using Statistical Package for Social Sciences version 17. The analysis comprised of frequency distributions, cross tabulations, Chi square test, Odds ratio and student's *t*-test. A P value of less than or equal to 0.05 was considered significant. The median values of the number of branching points was compared in subjects by Mann–Whitney test, tortuosity grades, papillary trunk direction, degree etc were analyzed. Adjustment for the effect of multiple variables was performed using multiple regression analysis. Means and standard deviations were compared using the *t*-test and frequencies were charted using the chi-squared test. Odds

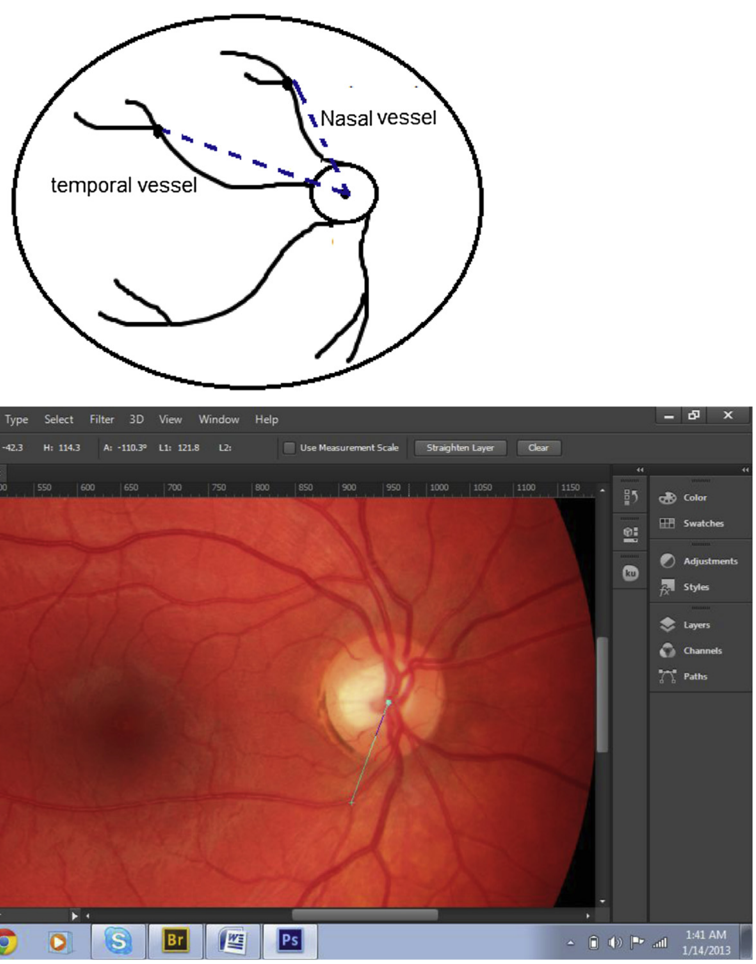

Fig. 3 – Linear distance between optic disc center to primary branching point.

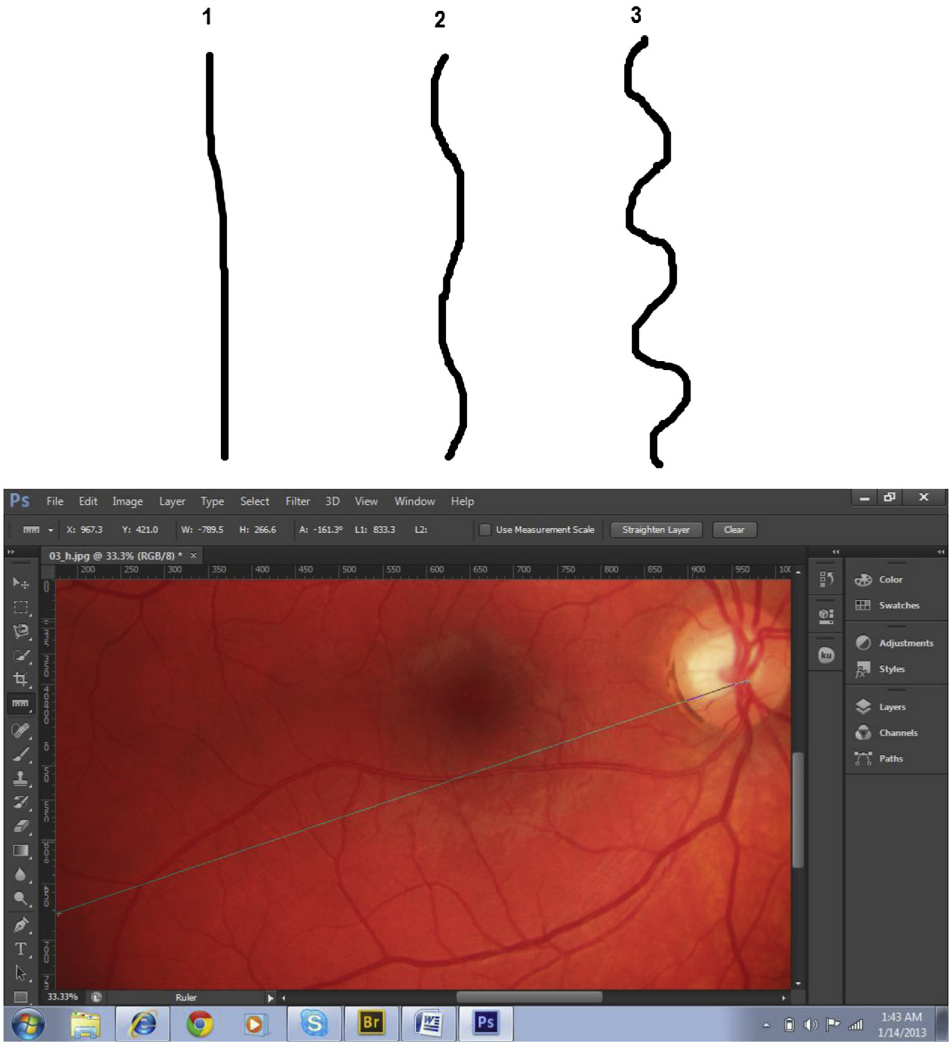

Fig. 4 – Grades of tortuosity of retinal blood vessel: 1 = straight/2 = wavy/3 = tortuous.

ratio was estimated at 95% confidence intervals from multiple logistic regressions, adjusting for potential confounding factors. Tests used for correlations were; Spearman coefficient of correlation for age, Mann–Whitney for gender, Wilcoxon sign rank for right and left eyes, Pearson correlation for correspondence between right and left eyes.

3. Results

The eyes of healthy Indian adults without overt illness showed the following characteristics of retinal vasculature:

- The superior papillary trunk was acute angled (an average of 50°) while the inferior papillary trunk was obtuse angled (an average of 122°) for both eyes (Table 1).
- Tortuosity was more in temporal than nasal vessels. Temporal arteries were equally divided between wavy and tortuous while nasal arteries were mostly straight.
- Branching was more extensive in temporal arteries (mostly second and third order branches) than nasal arteries (mostly first order branches).
- The mean tortuosity grade and branching points were more in left than right eyes (Tortuosity –1.86 for right eyes & 2.03 for left eyes and Branching –1.84 for right eyes & 2.29 for left eyes) (Table 1).
- The distance of primary bifurcation point from optic disc center was double for temporal arteries (average 2.01DD) as compared to nasal arteries (average 1.2DD) for both eyes (Table 1).
- Significant gender dimorphism was observed in superior nasal artery 'distance' (1.25 F vs 0.86 M) and inferior nasal artery 'tortuosity' (2.27 F vs 1.61 M), particularly for right eyes (Table 1).
- Significant lateral dimorphism between right and left eyes was observed in superior temporal artery 'tortuosity' & 'branching' and inferior nasal artery 'distance' (Table 2).
- Significant correlations with age were observed for superior papillary trunk 'direction'; superior temporal artery 'branching' in left eyes and superior nasal artery 'branching' in right eyes (Table 3).

4. Discussion

Several population based researches on retinal vascular morphology like The Rotterdam Study, The Wisconsin

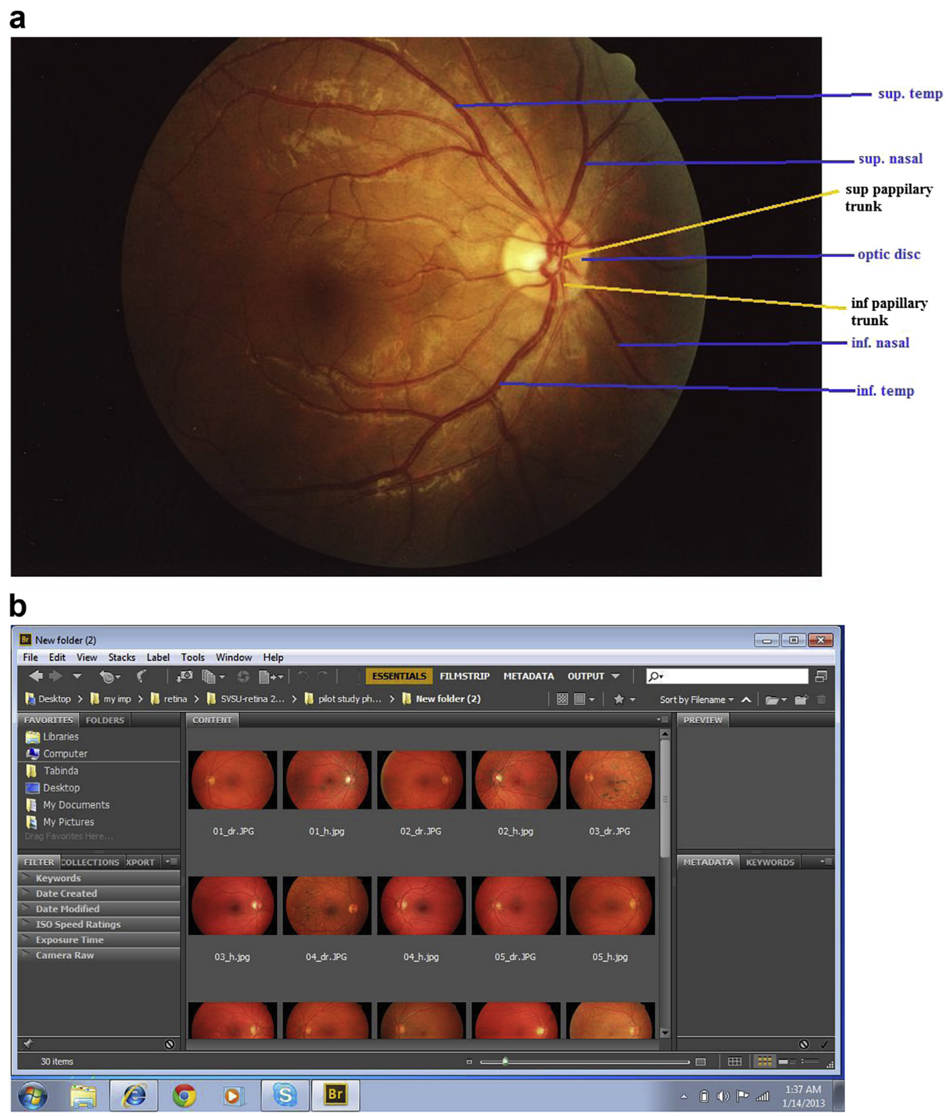

Fig. 5 – (a) Fundus image showing the four retinal blood vesels (superior temporal and superior nasal vessels emerging from the superior papillary trunk and inferior temporal and nasal vessels emerging from the inferior papillary trunk) at the optic disc. (b) Fundus image database uploaded in bridge CS6 digital asset management software.

Epidemiologic Study of Diabetic Retinopathy, The Cardiovascular Health Study The Beaver Dam Eye Study, The Blue Mountains Eye Study, The Atherosclerosis Risk in Communities Study evidence that retinal arterial changes are associated with increasing age, elevated blood pressure (past, current, and future), increasing degrees of macular diabetic edema, increasing severity of diabetic retinopathy, increased risk of coronary heart disease and increased mortality from Coronary Heart Disease and stroke.[2,5,8] Generalized arterial narrowing has been related to chronically elevated blood pressure levels. Researchers of the Atherosclerosis Risk in Communities Study elucidated the unique association between physical activity level, cardiovascular risk and retinal micro vascular signs.[8] Dynamic retinal vessel analysis is a well-established method for assessing vascular reactivity during normal conditions and after various physiological provocations like exercise, fasting, dieting and dehydration.[9] Lindley[10] concluded that retinal vessel appearances may provide a 'window' to the brain and help determine important underlying patho-physiological mechanisms of small vessel disease like stroke. The strongest recent evidence of role of genes on retinal vascular hereditability has been provided by Taarnhøj and co researchers during their research on the Danish twin population.[5,11] All these aforementioned studies have explored subtle or overt 'physical' changes in retinal microvasculature during frank disease or the preclinical stage of sickness. This study differs from the rest because it exclusively focuses on normal morphological features of retinal arteries in healthy Indian subjects, a facet that has yet not been thoroughly exposed in medical literature. Also, there are only a few studies similar to ours even in the west that probe into the vascular geometrics of healthy adults without overt disease like the one by Taarnhoj et al.[5,11] that was conducted on the Danish population. Similar to us, Hughes et al[12] used

Table 1 – Retinal microvasculature – gender wise comparisons, North Indians (*N* = 400, 180 M & 220 F).

Geometric parameters retinal arteries		N	Mean	Std. deviation	Minimum	Maximum	*p* value (Mann–Whitney test)
Superior pappilary trunk - direction (left eye)	Female	220	51.09	11.55	39.00	78.00	0.469
	Male	180	49.94	11.74	40.00	81.00	
	Total	400	50.58	11.50	39.00	81.00	
Inferior pappilary trunk – direction (left eye)	Female	220	119.55	15.17	100.00	156.00	0.978
	Male	180	121.00	18.10	100.00	160.00	
	Total	400	120.20	16.35	100.00	160.00	
Superior temporal vessel – tortuosity (teft eye)	Female	220	2.09	1.06	1.00	5.00	0.287
	Male	180	2.44	1.15	1.00	5.00	
	Total	400	2.25	1.10	1.00	5.00	
Superior nasal vessel – tortuosity (left eye)	Female	220	1.68	0.78	1.00	4.00	0.134
	Male	180	2.44	1.50	1.00	5.00	
	Total	400	2.03	1.21	1.00	5.00	
Inferior temporal vessel – tortuosity (left eye)	Female	220	1.86	1.28	1.00	5.00	0.168
	Male	180	2.17	1.04	1.00	4.00	
	Total	400	2.00	1.18	1.00	5.00	
Inferior nasal vessel – tortuosity (left eye)	Female	220	1.77	1.23	1.00	5.00	0.622
	Male	180	1.94	1.30	1.00	5.00	
	Total	400	1.85	1.25	1.00	5.00	
Superior temporal vessel – branching arcade (left eye)	Female	220	2.55	1.41	1.00	5.00	0.313
	Male	180	2.06	1.16	1.00	5.00	
	Total	400	2.33	1.31	1.00	5.00	
Superior nasal vessel – branching arcade (left eye)	Female	220	2.18	1.44	1.00	5.00	0.440
	Male	180	2.50	1.54	1.00	5.00	
	Total	400	2.33	1.47	1.00	5.00	
Inferior temporal vessel – branching arcade (left eye)	Female	220	2.27	1.16	1.00	5.00	0.284
	Male	180	2.78	1.44	1.00	5.00	
	Total	400	2.50	1.30	1.00	5.00	
Inferior nasal vessel – branching arcade (left eye)	Female	220	1.77	1.19	1.00	5.00	0.187
	Male	180	2.28	1.41	1.00	5.00	
	Total	400	2.00	1.30	1.00	5.00	
Superior temporal vessel – distance (left eye)	Female	220	2.10	1.09	.40	5.20	0.422
	Male	180	1.90	1.15	.70	5.20	
	Total	400	2.01	1.11	.40	5.20	
Superior nasal vessel – distance (left eye)	Female	220	0.91	0.53	.30	1.80	0.155
	Male	180	1.12	0.57	.30	1.80	
	Total	400	1.00	0.55	.30	1.80	
Inferior temporal vessel – distance (left eye)	Female	220	2.29	1.11	.60	3.70	0.254
	Male	180	2.13	0.55	1.50	3.30	
	Total	400	2.22	0.90	.60	3.70	
Inferior nasal vessel – distance (left eye)	Female	220	0.85	0.51	.30	2.40	0.583
	Male	180	1.07	0.74	.30	2.40	
	Total	400	0.95	0.62	.30	2.40	
Superior pappilary trunk – direction (right eye)	Female	220	53.59	18.84	23.00	95.00	0.172
	Male	180	47.72	19.35	23.00	114.00	
	Total	400	50.95	19.06	23.00	114.00	
Inferior pappilary trunk – direction (right eye)	Female	220	125.86	21.37	95.00	162.00	0.595
	Male	180	122.00	21.21	95.00	175.00	
	Total	400	124.13	21.11	95.00	175.00	
Superior temporal vessel – tortuosity (right eye)	Female	220	1.68	0.65	1.00	3.00	0.500
	Male	180	1.83	0.71	1.00	3.00	
	Total	400	1.75	0.67	1.00	3.00	
Superior nasal vessel – tortuosity (right eye)	Female	220	1.82	0.85	1.00	3.00	1.000
	Male	180	1.83	0.92	1.00	3.00	
	Total	400	1.83	0.87	1.00	3.00	
Inferior temporal vessel – tortuosity (right eye)	Female	220	1.82	0.66	1.00	3.00	0.397
	Male	180	2.00	0.69	1.00	3.00	
	Total	400	1.90	0.67	1.00	3.00	
Inferior nasal vessel – tortuosity (right eye)	Female	220	2.27	0.98	1.00	4.00	**0.027***
	Male	180	1.61	0.61	1.00	3.00	
	Total	400	1.98	0.89	1.00	4.00	

Table 1 – (continued)

Geometric parameters retinal arteries		N	Mean	Std. deviation	Minimum	Maximum	p value (Mann–Whitney test)
Superior temporal vessel – branching arcade (right eye)	Female	220	3.05	1.25	1.00	5.00	0.704
	Male	180	2.94	1.11	1.00	5.00	
	Total	400	3.00	1.18	1.00	5.00	
Superior nasal vessel – branching arcade (right eye)	Female	220	2.32	1.09	1.00	4.00	0.748
	Male	180	2.50	1.10	1.00	5.00	
	Total	400	2.40	1.08	1.00	5.00	
Inferior temporal vessel – branching arcade (right eye)	Female	220	2.55	1.30	1.00	5.00	0.768
	Male	180	2.39	1.04	1.00	4.00	
	Total	400	2.48	1.18	1.00	5.00	
Inferior nasal vessel – branching arcade (right eye)	Female	220	2.59	1.33	1.00	5.00	0.352
	Male	180	2.39	1.50	1.00	5.00	
	Total	400	2.50	1.40	1.00	5.00	
Superior temporal vessel – distance (right eye)	Female	220	2.41	1.42	.40	5.20	0.913
	Male	180	2.32	1.29	.70	5.20	
	Total	400	2.37	1.35	.40	5.20	
Superior nasal vessel – distance (right eye)	Female	220	1.25	0.61	.50	2.20	**0.033***
	Male	180	0.86	0.43	.30	1.70	
	Total	400	1.08	0.57	.30	2.20	
Inferior temporal vessel – distance (right eye)	Female	220	2.37	0.95	.30	3.70	0.495
	Male	180	2.58	0.94	.30	3.70	
	Total	400	2.46	0.94	.30	3.70	
Inferior nasal vessel – distance (right eye)	Female	220	1.56	1.13	.30	3.70	0.848
	Male	180	1.28	0.66	.50	3.00	
	Total	400	1.44	0.95	.30	3.70	

*p Value significant.

semi automated techniques to analyze digitized retinal photographs from healthy middle aged people, without clinical cardio vascular disease, diabetes, or hypertension. Arterioles were narrower and longer, had wider branching angles, and were more tortuous than venules and arteriolar narrowing was positively and independently associated with older age and elevated systolic blood pressure.

Quite interestingly, retinal vasculature is not only reflective of underlying health status and medical conditions; it even provides clues to an individual's birth status, maturity and surrounding environmental conditions. A population based study in preterm children found abnormal tortuosity and branching patterns in retina.[13] The fundus of childhood has a different morphology than adulthood in terms of length, branching, tortuosity and distribution of retinal vessels on the fundus surface.[14] In the Multi-Ethnic Study of Atherosclerosis using digital retinal images, chronic and acute associations between residential air pollution concentrations and retinal vessel diameters were found.[15] The Singapore Malay Eye Study concluded that myopic refractive errors and longer axial length of eyeball were associated with less tortuous arterioles and increased branching coefficients in both arterioles and venules.[16] In a retinal study by Michael et al,[17] an average arteriole branching coefficient of 1.31 was observed; with an average of 1.31 for men and 1.29 for women. No significant difference between eye sides or genders was found. The present study quantified branching arcades of retinal arteries based on progressive sequential subdivisions of the arterial tree and observed the branching mean of superior temporal, superior nasal, inferior temporal and inferior nasal arteries for right eyes of healthy subjects as 3, 2.40, 2.48 and 2.50 respectively and corresponding values for left eyes as 2.33, 2.33, 2.50 and 2.00. Our findings agree with Michael et al[17] to a certain extent; we observed no 'gender' differences for branching but 'eye side' differences were observed for superior temporal artery. Overall, the mean branching points were more in left than right eyes. The application of fractals and fractal growth processes to the branching blood vessels of normal human retinal circulation was introduced by Masters & Platt[18] and prominent studies have shown that the fractal nature of retinal vasculature is independent of age and sex.[19] This is in accordance with the findings of our research which observed that branching arcades of retinal arteries were unaffected by gender. However, in context of age, our findings stand for slight disagreement because we found that superior temporal and nasal artery 'branching' showed significant correlations with age.

Branching angle is the angle between two daughter vessels[20] and it is related to blood flow efficiency, energy cost of bulk flow, diffusion distance and greater competence in blood flow with lower amount of energy spent by the body. Zamir[20] proposed the optimal value for branching angle as 75° and increased angles have been related to decreased blood flow.[21] We found that the superior papillary trunk was acute angled (an average of 50°) while the inferior papillary trunk was obtuse angled (an average of 122°) for both eyes. Therefore, the branching angle of central retinal artery at modal bifurcation point into papillary trunks came to be 188° arithmetically. This discrepancy in values might be because the central retinal trunk or the papillary trunks are relatively 'huge' vessel bodies

Table 2 – Retinal microvasculature – side wise comparisons, North Indians (N = 400, 180 M & 220 F).

Geometric parameters retinal arteries		N	Mean	Std. deviation	Minimum	Maximum	Wilcoxon *p* value
Superior pappilary trunk – direction	Left side	400	50.58	11.50	39.00	81.00	0.968
	Right side	400	50.95	19.06	23.00	114.00	
	Total	800	50.76	15.64	23.00	114.00	
Inferior pappilary trunk – direction	Left side	400	120.20	16.35	100.00	160.00	0.212
	Right side	400	124.13	21.11	95.00	175.00	
	Total	800	122.16	18.87	95.00	175.00	
Superior temporal vessel – tortuosity	Left side	400	2.25	1.10	1.00	5.00	0.032*
	Right side	400	1.75	.67	1.00	3.00	
	Total	800	2.00	.94	1.00	5.00	
Superior nasal vessel – tortuosity	Left side	400	2.03	1.21	1.00	5.00	0.564
	Right side	400	1.83	.87	1.00	3.00	
	Total	800	1.93	1.05	1.00	5.00	
Inferior temporal vessel – tortuosity	Left side	400	2.00	1.18	1.00	5.00	0.739
	Right side	400	1.90	0.67	1.00	3.00	
	Total	800	1.95	0.95	1.00	5.00	
Inferior nasal vessel – tortuosity	Left side	400	1.85	1.25	1.00	5.00	0.563
	Right side	400	1.98	0.89	1.00	4.00	
	Total	800	1.91	1.08	1.00	5.00	
Superior temporal vessel – branching arcade	Left side	400	2.33	1.31	1.00	5.00	0.021*
	Right side	400	3.00	1.18	1.00	5.00	
	Total	800	2.66	1.28	1.00	5.00	
Superior nasal vessel – branching arcade	Left side	400	2.33	1.47	1.00	5.00	0.617
	Right side	400	2.40	1.08	1.00	5.00	
	Total	800	2.36	1.29	1.00	5.00	
Inferior temporal vessel – branching arcade	Left side	400	2.50	1.30	1.00	5.00	0.857
	Right side	400	2.48	1.18	1.00	5.00	
	Total	800	2.49	1.23	1.00	5.00	
Inferior nasal vessel – branching arcade	Left side	400	2.00	1.30	1.00	5.00	0.091
	Right side	400	2.50	1.40	1.00	5.00	
	Total	800	2.25	1.36	1.00	5.00	
Superior temporal vessel – distance	Left side	400	2.01	1.11	.40	5.20	0.205
	Right side	400	2.37	1.35	.40	5.20	
	Total	800	2.19	1.24	.40	5.20	
Superior nasal vessel – distance	Left side	400	1.00	0.55	.30	1.80	0.620
	Right side	400	1.08	0.57	.30	2.20	
	Total	800	1.04	0.56	.30	2.20	
Inferior temporal vessel – distance	Left side	400	2.22	0.90	.60	3.70	0.226
	Right side	400	2.46	0.94	.30	3.70	
	Total	800	2.34	0.92	.30	3.70	
Inferior nasal vessel – distance	Left side	400	0.95	0.62	.30	2.40	0.024*
	Right side	400	1.44	0.95	.30	3.70	
	Total	800	1.19	0.83	.30	3.70	

*p Value significant.

while the values proposed by Zamir stand in context of the much smaller arterioles further down the subdivision arcades. Sasongko[3] found increasing diabetes duration associated with increased arteriolar branching angle whereas angles were reduced in hypertension and low birth weight. In this study, we found superior papillary trunk angles significantly correlated with advancing age, with a tendency for wider angles among seniors. Decreased arterial branching angle has previously been associated with ageing,[17] which stands in accordance with our findings where widening of superior papillary trunk angles would automatically translate into the narrowing of central retinal arterial bifurcation angle.

Wide variations in retinal arterial tortuosity may mostly be explained by the effect of demographic and health indices.[5] Our findings established that age, gender and even ‘side’ of the eye may affect tortuosity patterns. In the present study, we found that the mean tortuosity grade was more in left than right eyes. Particularly, tortuosity for superior temporal artery exhibited lateral dimorphism. Also, temporal arteries were equally divided between wavy and tortuous while nasal arteries were mostly straight, similar to the observation of Bracher[22] who found generally more tortuous temporal than nasal vessels. Significant gender dimorphism was observed in inferior nasal artery tortuosity, with females having more tortuous arteries than males. Population based studies have established that less arteriolar tortuosity is independently associated with older age, higher blood pressure and higher body mass index.[23] Our findings disagree with earlier studies in context of having found no effect of age on retinal arterial tortuosity. Perhaps age does matter; but only after a certain period of time and maturity. Our subjects were robustly young (mean age 35 years) and healthy. Advancing age most likely takes its toll on retinal vasculature in old or diseased individuals, particularly chronic diabetics and hypertensives.

Table 3 – Retinal microvasculature – age correlations, North Indians (N = 400, 180 M & 220 F).

Spearman coefficient of correlation (with age)	Left eyes correlation coefficient (*r* value)	Left eyes *p* value	Right eyes correlation coefficient (*r* value)	Right eyes *p* value
Superior pappilary trunk – direction	−0.318*	0.048*	−0.027	0.871
Inferior pappilary trunk – direction	−0.028	0.865	−0.206	0.207
Superior temporal vessel – tortuosity	−0.074	0.655	0.150	0.363
Superior nasal vessel – tortuosity	−0.022	0.896	−0.024	0.885
Inferior temporal vessel – tortuosity	0.247	0.130	0.161	0.328
Inferior nasal vessel – tortuosity	−0.032	0.845	0.040*	0.809
Superior temporal vessel – branching arcade	0.435**	0.006*	−0.176	0.283
Superior nasal vessel – branching arcade	−0.192	0.240	−0.359*	0.025*
Inferior temporal vessel – branching arcade	−0.116	0.482	−0.064	0.700
Inferior nasal vessel – branching arcade	0.115	0.486	−0.245	0.132
Superior temporal vessel – distance	−0.209	0.202	0.078	0.638
Superior nasal vessel – distance	0.203	0.215	−0.224	0.170
Inferior temporal vessel – distance	−0.014	0.933	−0.107	0.518
Inferior nasal vessel – distance	−0.270	0.097	0.092	0.576

*p Value significant.

Another research had findings similar to ours and recorded no change in tortuosity with respect to age in healthy subjects without hypertension.[24] Taarnhøj[11] has quoted that retinal arterial tortuosity demonstrates strong heritability among first order blood relations; especially twins.

Distance of certain intuitive morphologic parameters from optic disc center has been found to be clinically relevant in conditions like glaucoma and retinopathy of prematurity.[25] Researchers examined retinal vascular tortuosity as a function of distance from the optic disk in infants with retinopathy of prematurity and found that mean cumulative tortuosity increased with distance from the disk margin. The pattern of perimetric loss in glaucoma and retinal nerve fiber layer thickness is associated with central retinal vessel trunk position expressed as a function of distance from optic disc border in disc diameters.[25] In the present study, we found the distance between disc center and prime arterial bifurcation point to be positively correlated to tortuosity and branching in retinal arteries of all four quadrants. Temporal arteries, owing to greater distances, were more tortuous and presented with higher grades of branching than nasal arteries.

In order to better understand the 'pathological', clinicians and anatomists need to know a little more about the 'normal' in context of retinal blood vessels among Indians, considering proven genetic and demographic influences on retinal microvasculature. This study is very appropriate in context of clinical research since it offers a foundation to quantitatively characterize retinal images with more objective measurements than current observer-driven techniques. Our technique, owing to its simple and direct application, has good reproducibility and can be further exploited for generation of automated algorithms that can detect abnormal features on digitalized fundus images. Also, we measured distances in disc diameters, a standardized measurement unit for retina-based studies. Additional strengths of this study include its cross sectional design with multi centric stratified sampling, sufficient cohort with multiple demographic and structural parameters, quantitative evaluation of vessels for all four quadrants and for both eyes (most contemporary studies have tackled retinal arteries or veins as an 'average' and right eyes have generally been considered representative).

Possible drawbacks include absence of simultaneous caliber assessments, measurement of 'only arteries' while excluding veins, considering their significant role in pathophysiology of disease states. Also, in this multivariate analysis, some other health indices could possibly have been included like body mass index, cholesterol level, smoking status, blood pressure and blood sugar levels, refraction error, axial length of eyeball, etc. to reveal correlations with local ocular and systemic risk factors.

5. Conclusion

The morphology of retinal blood vessels in North Indian subjects is not so different from other populations except for a few intuitive parameters, particularly tortuosity and branching patterns which seems natural owing to proven demographic and genetic influences on retinal vascular morphology. Broadly stating, tortuosity and branching are more extensive in temporal than nasal quadrants, with a tendency for delayed on-start of arteriolar subdivisions, farther away from the optic disc and superior papillary trunks are positioned at keener angles than inferior trunks. A certain degree of dimorphism exists among eye sides and genders; a finding which appears quite consistent morphologically. Considering arteries to be tubular channels directing the flow of fluid blood, we hypothesize that the natural dimorphism of retinal vascular architecture as observed in this study might determine microcirculatory efficacy and underlie unequal rates of progression of retinal lesions within two eyes of an individual or among 'seemingly comparable' individuals even though the exact mechanism for this remains unclear. The orientation of retinal blood vessels on the fundus plane is not merely 'a matter of chance' in the anatomic chronicle of the human body; rather, it has biological heralds and functional consequences. Direction, branching, distance and tortuosity; all determine the effectiveness and abundance of blood flow in the human retina that hold indispensable relevance for optimal vision and might be subtle indicators of micro vascular damage in disease states. This study has elucidated retinal vascular geometrics of healthy Indians, a knowledge that can improve our understanding of 'normal features' and 'natural variants' in retinal vasculature. Such an in depth morphological knowledge will facilitate an understanding of the pathogenesis of various disease phenomenon mirrored in early retinal changes and aid in planning timely and effective interventions. The potential utility of diagnostic and predictive fundus vascular screening for appraisal of health risks in populations is a worthwhile direction to explore. It needs to be comprehensively tackled in prospective studies in order to be used as a time and cost effective clinical tool for Indian ophthalmologists during upcoming years.

Conflicts of interest

All authors have none to declare.

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