CHAPTER 5

Retinal Nerve Fiber Layer thickness in human strabismic amblyopia

Colen TP, de Faber JT and Lemij HG
Binocular Vision & Strabismus Quarterly 2000;15(2):141-146

Abstract

Purpose Amblyopia is characterized by histopathological changes in the visual cortex and lateral geniculate nucleus. In the retina, however, no abnormalities have yet been reported. The purpose of this study was to compare the nerve fiber layer (NFL) thickness in the amblyopic eye with that in the sound eye of patients with strabismic amblyopia. As a practical implication, we investigated the validity of comparing Nerve Fiber Analyzer (NFA) measurements obtained in amblyopic eyes to the normative database built into the NFA.

Methods NFL thickness was measured with a third generation NFA, the GDx (Laser Diagnostic Technologies, San Diego, CA). This is a scanning laser polarimeter that has been designed for monitoring glaucoma. Twenty patients with strabismic amblyopia were imaged with the NFA. Patients had no nystagmus, neurological disease or glaucoma.

Results Nine patients had amblyopia in the right eye, and 11 patients in the left eye. The following NFL thickness parameters (all in microns) were compared: average thickness, superior maximum, inferior maximum, superior average, inferior average, nasal median and temporal median. In general, the sound eyes yielded higher thickness measures than the amblyopic eyes. These differences, however, were small and not statistically significant at the $\alpha = 0.05$ level.

Conclusions Measured with the NFA, there were no statistically significant differences in NFL thickness between the amblyopic and the sound eye in patients with strabismic amblyopia. Thus, when amblyopic eyes are measured with the NFA, the built-in normative database may serve as a reference to amblyopic data.

Introduction

Amblyopia is characterized by histopathological changes in the visual cortex and lateral geniculate nucleus (LGN).[1] Marked shrinkage of LGN-cells that receive input of the amblyopic eye are well described in human and non-human primates. The decrease in LGN cell size may be caused by retrograde inhibition by the striate cortex in strabismic amblyopia. In the retina of amblyopic eyes, however, no abnormalities have yet been reported.[2,3]

Since the introduction of the Nerve Fiber Analyzer (NFA), it is now possible to assess the thickness of the retinal nerve fiber layer (NFL) in vivo. The aim of this study was to investigate whether the retinal NFL in the amblyopic eye in strabismic amblyopia would differ in thickness as compared to the fellow eye.

Today, the NFA has become a widely used instrument for monitoring glaucoma. The thinner NFL in glaucoma can be quantitatively assessed which discriminates well between normals and glaucoma patients.[4-8] The latest version of the instrument, the NFA/GDx, has a built-in normative database that can be used to compare individual data with. This database consists of data of several hundred normal subjects subdivided into their various ethnic origins. Patients with any ocular disease includ-

ing amblyopia were not included in the database. It is unknown whether the normative database may serve as a valid reference in amblyopic eyes. Our study shows that this comparison is justified.

Methods

The Nerve Fiber Analyzer (NFA) NFL thickness was assessed with a third generation Nerve Fiber Analyzer, the so-called GDx (Laser Diagnostic Technologies, San Diego, CA). The NFA has been primarily designed for monitoring glaucoma. Changes of the retinal NFL that are characteristic for glaucoma, can be objectively quantified with the NFA.[5] A detailed description of the NFA has been published elsewhere.[9-11] In short, the NFA sends a polarized beam of laserlight into the eye, and analyses the amount of phase shift in the backscattered light. This phase shift, called retardation, is thought to arise from the specific lamellar orientation of the microtubules inside the retinal axons. The retinal NFL thickness value is then obtained by multiplying the degrees of retardation with 7.4, analogous to a monkey model where 7.4 μ retinal NFL thickness corresponded to 1 deg of retardation.[12]

In approximately 0.7 seconds, the peripapillary fundus is scanned with the standard 15 x 15 deg scanning angle and a resolution of 256 x 256 pixels. The data is used to compose a NFL thickness map of the same resolution, that is then color coded, and presented to the operator along with a reflectance fundus image for orientation. Next, the thickness map is automatically divided into 4 segments centered on the optic disc (superior 120 deg, inferior 120 deg, nasal 50 deg and temporal 70 deg). The operator positions a circle on the margin of the optic nerve head. Concentrically, a second circle with 1.75 times the diameter of the first is displayed automatically, excluding all pixels inside this second circle from analysis. Areas of blood vessels are a source of noise[13] and are therefore automatically excluded for analysis by the software.

For a quantitative approach, 14 standard parameters are available to the user. From these, the five absolute thickness parameters were selected, and in addition 2 non-standard parameters were derived by hand from existing standard parameters. The unit of measure of these 7 parameters is microns (μ). **Average thickness** is the average thickness of all pixels in all 4 segments together. **Superior average** and **Inferior average** is the average thickness of all pixels beneath the superior and inferior part of the ellipse respectively. **Superior maximum** and **Inferior maximum** is the average of the 1500 thickest pixels in the superior segment and the inferior segment respectively, reflecting the thickness of the superior and the inferior nerve fiber bundle. **Temporal median**, a non-standard parameter, reflects the mean value of the 1500 median pixels in the temporal segment and is obtained by dividing the superior maximum by the superior ratio. Likewise, **Nasal median**, also a non-standard parameter, reflects the mean value of the 1500 median pixels in the nasal segment and is obtained by dividing the superior maximum by the superior/nasal ratio.

Patients Amblyopia was defined as a difference in visual acuity between the two eyes of at least two Snellen lines with optimal correction. Initially, thirty-one patients met the inclusion criteria of strabismic amblyopia, no nystagmus, and no neurological disease. Of these, eleven patients were excluded because no high quality NFA image could be obtained. In these cases the amblyopic eye was unable to fix

when the sound eye was imaged. Our standard image qualitiy criteria were: centered optic disc, sharp focus, equal and total illumination in all segments and no eye movements during image acquisition. Of the remaining 20 patients (mean age 37.7 years; range 15-60) a high quality NFA image could be obtained in both eyes. Strabismic history, visual acuity, and refraction have been presented in table 5-1. Nine patients had amblyopia in the right eye and 11 patients in the left eye. All patients had intraocular pressures <21 mm Hg and normal looking optic discs. Visual acuity ranged from 20/32 to 20/16 in the sound eye, and from counting fingers to 20/25 in the amblyopic eye.

Scanning procedures Standard clinical procedures were adopted for this study. During all measurements, we saw to it that patients had their heads as upright as possible. Ambient lights were left on. Multiple scans were obtained in most cases and mean images from three images of high quality were created by the software. All measurements were carried out by the same operator.

Statistical procedures All data was entered into an SPSS statistical package. Differences (and 95% confidence interval) between sound eye and the amblyopic eye were calculated per parameter with a paired student's t-test. The level of statistical significance was set at $\alpha = 0.05$.

Results

All parameters except the nasal median showed a thinner retinal nerve fiber layer in the amblyopic eye as compared with the fellow eye (table 5-2). The nasal median parameter was on average 0.4μ higher in the amblyopic eye than in the sound eye. The differences were small for all parameters, and not statistically significant at the $\alpha = 0.05$ level.

Discussion

There was no statistically significant difference in NFL thickness between the two eyes in strabismic amblyopia, as measured with the Nerve Fiber Analyzer. Therefore, strabismic amblyopia does not appear to be associated with structural changes at the level of the retinal nerve fiber layer.

The NFA measures retardation, mainly originating from the structural arrangement of microtubules inside the retinal axons. Therefore, the conclusions of the present study probably apply to structure only, and not to function.

Considering the known changes in the cortex and the LGN, one could hypothesize transsynaptical changes in the retinal ganglion cells and thus in the NFL. Our results do not support this hypothesis. Neither is it supported by results by De Lint,[3] who found no evidence of retinal dysfunction at the level of cone photoreceptors in amblyopic eyes.

Conclusion

Measured with the Nerve Fiber Analyzer, there are no differences in nerve fiber layer thickness between the amblyopic and the sound eye, in patients with strabismic amblyopia. Therefore, when the NFA is used for monitoring amblyopic eyes, data can be compared with the built-in normative database, that was compiled without amblyopic eyes.

Table 5-1. Patient characteristics and strabismic history

Patient nr.	Age (yrs)	Strabismus History	Treatment	Strab. Actual	VA ODU	Refraction
1	28	ET	none	XT 5 Δ	20/20	S +0.25
					20/32	S +4.0
2	52	ET	none	ET 50 Δ	20/50	S +3.0
					20/20	S + 0.5
3	48	ET	none	XT 40 Δ	20/200	n/a
					20/20	
4	26	micro ET	none	ET 5 Δ	20/16	plano
					20/200	
5	56	ET	Str. surgery	XT 65 Δ	20/20	S +0.25 C-0.50 x 115°
					20/80	S 0 C-0.25 x 153°
6	30	ET	occlusion	XT 20 Δ	20/40	S + 4.5 C 0.5 x 5°
				RHT 10 Δ	20/20	S + 3.5
7	22	ET	occlusion	XT 30 Δ	20/20	S + 0.25
					20/125	S + 0.5
8	21	ET	Str. surgery	XT 20 Δ	20/100	S + 0.5 C -0.5 x 180°
				LHT 16 Δ	20/16	S plano
9	15	ET	Str. surgery	XT 16 Δ	20/50	S + 1.0
					20/20	S + 2.0 C -0.5 x 40°
10	41	ET	Str. surgery	XT 25 Δ	20/20	S + 4.5 C -0.75 x 135°
					cf	S + 5.0 C -0.75 x 180°
11	44	ET	none	XT 40 D	20/20	plano
					cf	S - 0.50 C +0.5 x 180°
12	23	ET	none	ET 20 D	20/125	S +5.0 C -1.5 x 90°
					20/25	S +4.25 C -2.0 x 90°
13	45	ET	Str. surgery	15 Δ	20/32	S +1.5 C -1.0 x 90°
				RHT 8 Δ	20/20	S -1.0 C -0.5 x 80°
14	60	ET	none	ET 10 Δ	20/200	S + 1.5
				RHT 5 Δ	20/20	S + 2.75 C - 0.5 x 180°
15	28	n/a	none	n/a	20/32	n/a
					20/20	
16	55	ET	n/a	n/a	20/20	S plano C -0.5 x 68°
					20/200	S + 0.75 C -0.75 x 81°
17	34	ET	Str. surgery	n/a	20/20	S + 2.0 C -0.5 x 152°
					20/200	S + 3.75 C -1.0 x 10°
18	51	n/a	n/a	n/a	20/16	n/a
					20/25	
19	30	ET	Str. surgery	10 Δ	20/200	S plano C + 1.0 x 90°
					20/20	S -1.5 C + 2.5 x 90°
20	44	ET	n/a	n/a	20/32	n/a
					20/20	

For every patient, age at NFA measurement and strabismic history are given where available. N/A stands for not available clinical data. In addition, visual acuity (VA; cf stands for counting fingers) and refraction are presented.

Table 5-2. NFL thickness parameters in the amblyopic and the sound eye.

parameter	sound eye (μ)	amblyopic eye (μ)	difference in thickness (μ)	95% CIN for difference (μ)	p-value
average thickness	67.4 (11.4)	66.4 (9.9)	1.0	-3.2 ; 5.2	0.627
superior average	82.6 (13.4)	82.6 (12.3)	0.0	-4.5 ; 4.5	1.000
inferior average	81.5 (10.7)	78.7 (12.0)	2.8	-2.6 ; 8.2	0.290
temporal median	43.5 (13.7)	41.6 (10.3)	1.9	-3.7 ; 7.4	0.492
nasal median	48.0 (12.6)	48.4 (9.2)	-0.4	-5.7 ; 5.0	0.883
superior maximum	100.2 (14.8)	99.4 (17.0)	0.8	-5.7 ; 7.3	0.799
inferior maximum	95.2 (11.7)	90.6 (13.7)	4.6	-0.9 ; 10.1	0.096

NFL thickness parameters are all in microns (μ). Presented is the average thickness in the sound eye and in the amblyopic eye, together with its standard deviation (in parentheses). Next, the mean differences in thickness between the sound eye and the amblyopic eye, as well as its 95% confidence interval (CIN) and p-values are given.

References

1. Von Noorden G, Crawford M. The lateral geniculate nucleus in human strabismic amblyopia. Investigative Ophthalmology & Visual Science 1992;33:2729-32.
2. Romano PE, Bird JJ. Fluorescein Retinal Angiographic Studies of Functional Amblyopia. Journal of Pediatric Ophthalmology and Strabismus 1980;17:318-9.
3. Delint P, Weissenbruch C, Berendschot T, Norren D. Photoreceptor function in unilateral amblyopia. Vision Research 1998;38:613-7.
4. Weinreb RN, Shakiba S, Sample PA, et al. Association between quantitative nerve fiber layer measurement and visual field loss in glaucoma. Am J Ophthalmol 1995;120:732-8.
5. Tjon-Fo-Sang MJ, Lemij HG. The sensitivity and specificity of nerve fiber layer measurements in glaucoma as determined with scanning laser polarimetry. Am J Ophthalmol 1997;123:62-9.
6. Xu L, Chen PP, Chen YY, et al. Quantitative nerve fiber layer measurements using scanning laser polarimetry and modulation parameters in the detection of glaucoma. J Glaucoma 1998;7:270-7.
7. Weinreb RN, Zangwill L, Berry CC, et al. Detection of glaucoma with scanning laser polarimetry. Arch Ophthalmol 1998;116:1583-9.
8. Choplin NT, Lundy DC, Dreher AW. Differentiating patients with glaucoma from glaucoma suspects and normal subjects by nerve fiber layer assessment with scanning laser polarimetry. Ophthalmology 1998;105:2068-76.
9. Tjon-Fo-Sang MJ, de Vries J, Lemij HG. Measurement by nerve fiber analyzer of retinal nerve fiber layer thickness in normal subjects and patients with ocular hypertension. Am J Ophthalmol 1996;122:220-7.
10. Dreher AW, Reiter K. Retinal laser ellipsometry: a new method for measuring the retinal nerve fiber layer thickness distribution. Clin Vis Sci 1992;7:481-8.
11. Weinreb RN, Shakiba S, Zangwill L. Scanning laser polarimetry to measure the nerve fiber layer of normal and glaucomatous eyes. Am J Ophthalmol 1995;119:627-36.
12. Weinreb RN, Dreher AW, Coleman A, et al. Histopathologic validation of Fourier-ellipsometry measurements of retinal nerve fiber layer thickness. Arch Ophthalmol 1990;108:557-60.
13. Tjon-Fo-Sang MJ, van Strik R, de Vries J, Lemij HG. Improved reproducibility of measurements with the nerve fiber analyzer. J Glaucoma 1997;6:203-11.