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Influence of optic disc size on parameters of retinal nerve fiber analysis as measured using GDx VCC and ECC in healthy subjects.

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Abstract

Background: To date there is no knowledge about the impact of optic disc size on the retinal nerve fiber layer (RNFL) as measured with GDx VCC or GDx ECC. Since the size of the optic disc is not taken into account for the analyses carried out by the GDx it might affect sensitivity in large optic discs and specificity in small ones.

Methods: Topographic optic disc measurements and RNFL thickness values of 80 healthy subjects were measured using HRT3 and GDx VCC and ECC. Subjects were divided into 3 equally large groups depending on optic disc area. We used ANOVA to test the differences between groups of the GDx VCC and ECC parameters TSNIT average, superior average, inferior average and NFI.

Results: The mean optic disc sizes of the three groups were 1.61, 2.00, and 2.61 μm^2 . The optic disc area significantly affected none of the tested RNFL parameters of the GDx and HRT3. For all HRT3 parameters of optic disc morphology, ANOVA showed statistical significance between the three groups of different optic disc areas.

Conclusion: We could not find a significant impact of optic disc size on any RNFL parameter measured with GDx VCC or ECC and with HRT3.

Introduction

The objective and quantitative measurement of peripapillary retinal nerve fiber layer (RNFL) is possible with scanning laser polarimetry (SLP; GDx Nerve Fiber Analyzer; Carl Zeiss Meditec, Dublin, CA) which relies on the assumption that the parallel arrangement of microtubules within the RNFL causes a change in the state of polarisation of an illuminating laser beam, known as retardation.[1, 2] The amount of retardation of the incident laser light is proportional to the thickness of the tissue.

Although retardation is considered to be proportional to the thickness of the RNFL it can be affected by other polarizing ocular structures, such as the cornea, which interferes with accurate assessment of RNFL thickness.[3] Accordingly, GDx with variable corneal compensation (GDx VCC) was developed for better compensation of corneal birefringence, which has better diagnostic ability than GDx with fixed corneal compensation (FCC).[4] Nevertheless, there is still some interference from the subretinal structures, and the image produced by GDx VCC sometimes shows an atypical retardation pattern (ARP). ARP is seen as alternating peripapillary circumferential bands of low and high retardation [5] and is thought to be common in eyes with a low signal-to-noise ratio resulting from thinning of the retinal pigment epithelium, such as in aged or myopic eyes.[6]

Enhanced corneal compensation (ECC) is based on the compensation method. This scheme has been developed to improve the signal-to-noise ratio and to eliminate the ARP associated artefacts.[7] Several studies have demonstrated the benefits of GDx ECC for the assessment of the RNFL.[7, 8]

Since a couple of years it is possible to have reliable sophisticated three dimensional analysis of the optic nerve head with the development of confocal scanning laser tomography.[9, 10] In the present study we used HRT3 for quantifying the morphological optic disc parameters.

Considerable variability in RNFL thickness values among normal subjects, and some overlap in thickness values between normal subjects and glaucoma patients have been reported in several studies conducted by SLP.[11, 12] The number of nerve fibers and the configuration of the optic nerve head vary considerably in the normal population. The size of the scleral canal, the tilt of the optic nerve head, and the number of the nerve fibers and their arrangement all contribute to this variation.[13]

Previous studies using SLP with fixed corneal compensation (GDx FCC) found out that the peripapillary RNFL thickness increases linearly as the area of the optic nerve head increases.[14, 15] Medeiros et al. found that the diagnostic performances of the GDx VCC, HRT2, and Stratus OCT in glaucoma patients were significantly influenced by the optic disc size.[16] Larger optic discs were associated with decreased sensitivity for the Stratus OCT parameter average thickness and the GDx VCC parameter nerve fiber indicator, whereas small optic discs were associated with increased sensitivity. For the Moorfields regression analysis classification measured by the HRT2, an inverse effect was observed. Also RNFL thickness measurements obtained by Stratus OCT increased significantly with an increase in optic disc size.[17]

To date there is no knowledge about the impact of optic disc size on the RNFL as measured with GDx VCC or GDx ECC. Since the size of the optic disc is not taken into account for the analyses carried out by the GDx it might affect sensitivity in large optic discs and specificity in small ones.

It was the aim of the present study to quantify the influence of optic disc size on the peripapillary RNFL thickness as measured with the current and upcoming SLP technologies in healthy subjects.

Subjects and Methods

Subjects

The study protocol was approved by the Ethics Committee of the Medical University of Vienna and followed the guidelines of Good Clinical Practice and the Declaration of Helsinki. 83 healthy, caucasian subjects of both sexes aged between 18 to 69 years were included.

Inclusion and exclusion criteria

Inclusion criteria were normal findings in medical history and normal ophthalmic findings, especially normal appearance of the optic disc and normal visual fields. An abnormal visual field was defined as a glaucoma hemifield test outside normal limits and/or a corrected pattern standard deviation with $P < 0.05$. [18] Any of the following excluded a subject from participation in the trial: Evidence of any eye disease except refractive error, history of ocular trauma or intraocular surgery within the last 6 months, ocular inflammation or infection within the last 3 months, pregnancy, astigmatism more than +2.0 diopters and ametropia of more than +/-5.0 diopters. Subject with tilted disc and peripapillary atrophy that extended into the measurement ring were excluded.

Experimental Paradigm

Initially a prestudy screening was carried out, where the medical and ocular history was taken. A complete ophthalmological examination was performed, including fundoscopy, visual acuity and the measurement of intraocular pressure (IOP) by Goldmann applanation tonometry. Standard automated perimetry (SAP) was performed with the Humphrey field analyser II (HFA II, program 30-2, SITA Standard, Carl Zeiss).

Subjects eligible for participating in the study according to the inclusion/exclusion criteria were included. One eye was randomly selected for the measurements.

The study was performed at the Department of Ophthalmology, Medical University of Vienna, Austria.

Methods

Automated visual field testing was performed with the Humphrey field analyzer II (program 30-2). Visual field eligibility criteria were less than 33% false-positive responses, less than 33% false-negative responses, and less than 33% fixation losses. Optic disc morphology was measured with the confocal scanning laser tomograph HRT3. Two series of 3 images each were acquired and the series with the smaller standard deviation was chosen for analysis. Scanning laser polarimetry GDx VCC was performed using a commercial GDx system Version 5.5.0 (NDB Version 1.05.00, Carl Zeiss Meditec, Dublin, CA, USA). Additionally we used the Gdx ECC method, which was run on the same commercial GDx system specified above. The general principles of GDx have already been described elsewhere.[4, 19] Briefly, SLP assesses the RNFL thickness in the peripapillary retina by measuring the RNFL birefringence with a near-infrared diode laser. With GDx ECC software, the corneal polarization compensator is automatically adjusted so that the combined retardation magnitude from the cornea and the compensating retarder is approximately 55 nm with a vertical slow axis of polarization. This adjustment bias serves to boost the signal to overcome low sensitivity that can make retardation measurements susceptible to optical and electronic noise. After image acquisition, the bias is subtracted to yield the RNFL retardation values. In our study, each subject with pupils undilated had scans on the same day performed by an experienced operator. The spherical equivalent refractive error was tested subjectively and entered into the software to allow the GDx to focus on the retina. To optimize image quality

the focus was adjusted manually in 0.25-diopter steps if necessary. All selected images were of high quality (quality scan score of 8 or greater), with a centred optic disc, were well focused and illuminated throughout the image, and were without any motion artefacts. A fixed concentric measurement band with 27 pixels (approximately 2.4 mm) inner and 35 pixels (approximately 3.2 mm) outer diameter was centred on the optic disc, after which the measurements of peripapillary retardation were conducted. Retardation was converted to an estimate of RNFL thickness by the software.

The GDx VCC and ECC parameters investigated in this study were the RNFL thickness values including temporal-superior-nasal-inferior-temporal (TSNIT) average, superior average, inferior average, ellipse standard deviation (TSNIT SD), as well as nerve fiber indicator (NFI) and typical scan score (TSS).[7]

Statistical methods

Subjects were divided into 3 equally large groups depending on the optic disc area as measured with HRT3 (group 1: <1.85 mm²; group 2: 1.85 mm² to 2.23 mm²; and group 3: >2.23 mm²). Parameters of HRT, GDx VCC and GDx ECC were compared between groups by ANOVA. All statistical analysis was done with the SPSS® software package (SPSS Inc., USA) release No. 16.0.2.

Results

44 males and 36 females were included into the study. Subjects' baseline characteristics, IOP and visual field mean deviation (MD) are given in table 1. We were only able to obtain a sufficient measurement quality of 80 out of 83 subjects. Hence all data on optic nerve head morphology are from 80 subjects.

Average optic disc area measured with HRT3 was within the expected range, and as expected, with a huge variability between subjects. The mean optic disc area was 2.07 mm^2 (SD ± 0.48) and ranged from 1.17 mm^2 to 3.62 mm^2 . Table 2 shows means and standard deviations of optic disc topographic parameters as measured with HRT3 and for comparison the ophthalmoscopically determined C/D ratios (examined with the Volk Superfield lens). For all parameters of optic disc morphology, ANOVA showed statistical significance between the three groups of different optic disc areas. In contrast the HRT3 measures of the RNFL did not reach the level of significance in ANOVA.

In table 3 the parameters of GDx VCC and ECC are shown and compared between the groups of different optic disc areas. None of the tested variables depended significantly on the optic disc area.

Discussion

Reference values for retinal RNFL thickness of different ages, ethnic groups, and refractive stages are needed to define normality of RNFL thickness. Several studies have been performed with SLP on variation of RNFL thickness measurements with age and ethnicity [1, 12] but the effect of axial length and optic disc size on RNFL thickness measurements is still not clear.

Our data showed a tendency towards an increase of RNFL thickness measurements with an increase in optic disc size but we could not find a significant impact of optic disc size on any retinal nerve fiber parameter measured with GDx VCC or ECC and with HRT3. While this possibly might be attributed to our sample size, the power was sufficient to detect a clinically

meaningful difference between groups of 10%.

Contrary to our results, previous studies using Gdx FCC, HRT II, and Stratus OCT showed a significant increase of RNFL thickness by the optic disc size.[15, 16, 18, 20] It might be that eyes with large ONHs show a thicker RNFL as a result of an increased amount of nerve fibers.[21] Medeiros et al. have demonstrated reduced sensitivity of GDx VCC and Stratus OCT in eyes with large optic discs and have discussed that this might be an effect of increased number of optic nerve fibers in those eyes.[17]

Nagai et al. found a significant positive correlation of RNFL and disc size both measured with Stratus OCT, but not with disc size as measured with HRT.[20] The possibly most important difference between these two instruments in determination of the disc size is that the Stratus OCT does not correct for ocular magnification error caused by refractive errors (even if they are corrected for by optical means, for example by focussing) while the HRT at least partially does. As a consequence in the Stratus, and also the GDx VCC the apparently fixed diameter of the measurement ring becomes dependent on refractive error. With OCTs and also the GDx VCC the optic discs of hyperopic individuals appear to be larger and the measurement rings unchanged, while on the retina the measurement ring is reduced in size and the optic disc may be just average. The opposite holds true for myopic subjects. While this effect may account for part of the correlation between disc size and RNFL thickness, the same holds true for our study where we did not find a correlation.

There is more than one possible interaction between apparent optic disc size in retinal imaging and refractive error. Due to different magnification the effective imaging area in myopic eyes gets larger resulting in apparently smaller disc size. Taking into account the range of refractive errors in the population studied, this may have resulted in errors of the image sizes of -7% to +7% without mathematical correction as in GDx, or -4% to +3% with the built-in correction method of the HRT.[22] Secondly there is a correlation between axial

length and the size of the optic disc, although it may be valid only for refractive errors outside -8 to +4 diopters.[23]

The technical aspects of the different measurement methods might explain the controversial study results. SLP determines RNFL thickness in the peripapillary region by measuring the total retardation in the light passing through the microtubules within the ganglion cell axons. This means that it does not directly measure the thickness of the RNFL but an optical property linked to the viability of the RNFL. OCT on the other hand measures the thickness of RNFL on cross sectional images of the retina generated by low coherence interferometry.

Also when comparing our results to studies using the GDx FCC [15, 16] some differences in methods should be noted. The GDx FCC used a measurement circle of variable diameter depending on disc diameter. Thus, the circumference of this ring increases together with disc size. While the spreading nerve fibers should have similar cross-sectional area at different radius of the circle, some artefacts such as effects of corneal retardation due to the fixed cornea compensation, or atypical pattern may add to the RNFL. This addition might increase the integral of RNFL with increasing size of the measuring ring.

At least for the small optic discs another factor might also influence results of the GDx FCC. It has been reported that the RNFL determined with GDx FCC increases from disc margin over 0.2 to 0.4 mm before reaching its maximum. In small discs the measurement ring might sometimes be too close to the disc and therefore miss the maximum RNFL thickness. However this finding has not been confirmed by others.[24]

As shown in a previous study with ECC [7], TSS was higher than with VCC, suggesting that atypical retardation patterns were less pronounced in images obtained with ECC. The origin of atypical scans is not yet fully understood. However, it appears to be associated with a low signal-to-noise ratio. In areas of low signal the measurements are influenced by electronic

noise. By introducing and later removing a preset measurement bias, the instrument's measurement sensitivity is enhanced, what reduces its susceptibility to such errors. This could explain why atypical retardation is reduced with ECC. In the present study our data confirm the previous finding of Toth et al.[7] Although the TSS scores were very high in both groups we were able to prove a significant difference between the VCC and the ECC derived values ($p < 0.001$, Wilcoxon test).

Concerning the influence of optic disc size on disc morphology our findings are consistent with literature. All HRT3 parameters of areas and volumes within the optic disc were highly significantly dependent on optic disc area as measured with HRT3.[25]

We could not find a significant impact of optic disc size on any retinal nerve fiber parameter measured with GDx VCC or ECC and with HRT3. In contrast, all parameters of optic disc morphology measured with HRT3, showed statistical significance between the three groups of different optic disc areas. The size of the optic disc does not appear to be an important factor to be taken into account, when analysing images of the GDx VCC or ECC.

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Competing Interest

None declared.

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Table 1 Subjects baseline characteristics, IOP and visual field MD *

3 groups depending on the optic disc area	Small (group 1: <1.85 mm ²)	Medium (group 2: 1.85 mm ² to 2.23 mm ²)	Large (group 3: >2.23 mm ²)
Age (years)	25.0 (±4.8)	28.6 (±10.9)	26.6 (±7.8)
Refractive error (diopters)	-0.06 (±2.03)	0.34 (±1.51)	-0.42 (±1.46)
IOP (mmHg)	14.8 (±2.2)	14.4 (±2.3)	15.2 (±2.8)
MD (db)	-0.18 (±0.81)	-0.17 (±0.1)	-0.04 (±0.87)

IOP = intraocular pressure, MD = mean deviation, dB = decibel

Table 2 Optic disc parameters and RNFL thickness as measured with HRT3*

3 groups depending on the optic disc area	Small (group 1: <1.85 mm ²)	Medium (group 2: 1.85 mm ² to 2.23 mm ²)	Large (group 3: >2.23 mm ²)	ANOVA (p values)
Optic Disc Area (mm ²)	1.61 (±0.19)	2.00 (±0.11)	2.61 (±0.37)	<0.001
Rim Area (mm ²)	1.38 (±0.22)	1.71 (±1.19)	2.00 (±0.41)	<0.001
Rim Volume (mm ³)	0.40 (±0.11)	0.48 (±0.13)	0.54 (±0.23)	0.008
C/D Area Ratio	0.14 (±0.09)	0.14 (±0.09)	0.23 (±0.12)	0.002
Linear C/D Ratio (HRT)	0.35 (±0.14)	0.34 (±0.18)	0.45 (±0.16)	0.019
Mean RNFL thickness (mm)	0.28 (±0.06)	0.27 (±0.06)	0.25 (±0.04)	0.180
RNFL Crosssectional Area (mm ²)	1.24 (±0.28)	1.36 (±0.30)	1.42 (±0.25)	0.053
C/D Ratio (Volk)	0.14 (±0.09)	0.21 (±0.12)	0.26 (±0.15)	0.003

C/D = cup/disc ratio, RNFL = retinal nerve fiber layer

Table 3 Comparison of GDx parameters between groups of different optic disc area (ANOVA) *

	3 groups depending on the optic disc area	Small (group 1: <1.85mm ²)	Medium (group 2: 1.85 mm ² to 2.23 mm ²)	Large (group 3: >2.23 mm ²)	ANOVA (p values)
VCC	NFI	18.6 (±10.7)	16.3 (±9.1)	14.1 (±6.0)	0.179
	TSNIT average	55.3 (±6.3)	55.6 (±5.8)	56.9 (±3.4)	0.506
	Superior average	66.5 (±8.5)	67.7 (±10.7)	70.0 (±6.0)	0.345
	Inferior average	66.5 (±10.3)	67.1 (±8.5)	67.7 (±6.5)	0.870
	TSNIT SD	24.5 (±5.4)	26.1 (±4.7)	25.7 (±4.6)	0.456
	TSS	96.4 (±6.7)	97.3 (±3.9)	94.5 (±7.0)	0.230
ECC	NFI	20.3 (±8.6)	17.1 (±10.1)	15.8 (±6.4)	0.157
	TSNIT average	51.6 (±5.1)	54.6 (±6.4)	54.5 (±4.3)	0.076
	Superior average	64.0 (±7.7)	66.3 (±11.0)	66.6 (±6.4)	0.500
	Inferior average	65.6 (±9.0)	69.3 (±8.5)	68.8 (±6.9)	0.208
	TSNIT SD	26.9 (±4.3)	28.3 (±4.4)	27.8 (±3.9)	0.498
	TSS	99.7 (±1.0)	98.6 (±4.2)	98.2 (±6.4)	0.420

* Results are presented as means ± SD (n=80).

VCC = variable corneal compensation, ECC = enhanced corneal compensation, NFI = nerve fiber indicator, TSNIT = temporal, superior, nasal, inferior, temporal average, TSNIT SD = ellipse standard deviation, TSS = typical scan score