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The Relationship Between Choroidal Abnormalities and Visual Outcomes in Pediatric Patients With NF1-Associated Optic Pathway Gliomas

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Background: Choroidal abnormalities (CAs) visualized on near-infrared reflectance (NIR) imaging are a new diagnostic criterion for neurofibromatosis type 1 (NF1), but the association between the presence of CAs and visual function remains unknown. This study evaluated the relationship between visual acuity (VA) with the presence, number, or total area of CAs visualized by NIR in children with NF1-associated optic pathway gliomas (NF1-OPGs).

Methods: Patients (<18 years) enrolled in a prospective longitudinal study of children with NF1-associated OPGs from 3 institutions were eligible if they had optical coherence tomography (OCT) of the macula (Heidelberg Spectralis) with ≥1 year of follow-up. The central 30° NIR images were reviewed by 2 neuro-ophthalmologists who manually calculated the number and total area of CAs. VA (logMAR) was measured using a standardized protocol. Cross-sectional associations of presence, number, and total area of CAs with VA, retinal nerve fiber layer thickness (RNFL), and ganglion cell–inner plexiform layer thickness were evaluated at the first and most recent visits using regression models. Intereye correlation was accounted for using generalized estimating equations.

Results: Eighty-two eyes of 41 children (56% female) were included. The mean ± SD age at the first OCT was 10.1 ± 3.3 years, with a mean follow-up of 20.4 ± 7.2 months. At study entry, CAs were present in 46% of eyes with a mean number of 2.1 ± 1.7 and a mean total area of 2.0 ± 1.7 mm^2 per eye. At the most recent follow-up, CAs were present in 48% of eyes with a mean number of 2.2 ± 1.8 lesions and a mean total area of 2.3 ± 2.1 mm^2 per eye. Neither VA nor OCT parameters at first and follow-up visits were associated with the presence, number, or total area of CAs (all $P > 0.05$).

Conclusions: CAs are prevalent but not ubiquitous, in children with NF1-OPGs. Although CAs are a diagnostic criterion for NF1, their presence and size do not appear to be associated with visual function.

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Neurofibromatosis type 1 (NF1) is an autosomal dominant disorder that occurs in approximately 1:3,500 births.[1,2] The disease can affect nearly all organ systems in the body and is particularly important for ophthalmologists because the eyes, ocular adnexa, and visual pathways may be involved with the potential for irreversible injury to visual function from compressive optic neuropathy and glaucoma. The diagnostic criteria for NF1 include ocular findings such as Lisch nodules, optic pathway gliomas, and, more recently added, choroidal abnormalities (CAs) visualized on near-infrared reflectance (NIR) imaging.[3,4]

CAs were first described in histopathologic reports as ovoid bodies in the choroid, consisting of hyperplastic Schwann cells, melanocytes, and ganglion cells.[5,6] In vivo, these lesions are undetectable by conventional ophthalmoscopy, but with advances in ophthalmic imaging and the widespread use of spectral-domain optical coherence tomography (SD-OCT) with near-infrared reflectance (NIR) imaging of the fundus, CAs were identified as an objective biomarker in NF1.[7–9] These lesions appear as bright patchy lesions on SD-OCT in NIR mode and are commonly identified in patients with NF1. Flores et al demonstrated that CAs are more prevalent and may be detected even earlier than Lisch nodules in pediatric patients with NF1.[7]

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Previous studies have shown that CAs can progress in number and size over time.[10,11] More recently, Godinho et al[12] reported a significant association between the number and area of CAs and the presence of optic pathway glioma (OPG). Interestingly, the impact of CAs on visual function in children with NF1-OPG has not been reported.

The purpose of this study was to evaluate the relationship between CAs and visual acuity (VA) in children with NF1-OPGs. We also aimed to investigate whether changes in the number and size of the lesions are associated with visual structure and function in a multicenter cohort of children with NF1-OPGs followed longitudinally.

METHODS

This was a prospective observational cohort study of pediatric patients with NF1-OPGs from Boston Children's Hospital, Children's Hospital of Philadelphia, and The Hospital for Sick Children. The institutional review board of each site approved the study. A parent or guardian of each participant gave written informed consent, and children also provided written assent when applicable as determined by the local institutional review board. The study adhered to the tenets of the Declaration of Helsinki for research involving human subjects and were conducted in accordance with the regulations of the Health Insurance Portability and Accountability Act.

The study included children 18 years and younger with formal diagnostic of NF1 based on National Institutes of Health diagnostic criteria[4] and with radiologic diagnosis of an OPG. The clinical information from baseline and the last follow-up visit, including age, sex, race, best-corrected visual acuity (BCVA), OPG location, spectral-domain optical coherence tomography (SD-OCT) scans, and NIR images (Heidelberg Engineering, GmbH, Dossenheim, Germany), was extracted. The VA assessment was performed using age-appropriate visual function tests[13] following a standardized protocol and BCVA was converted to logarithm of the minimum angle of resolution (logMAR). Eligible subjects for this study were required to have good-quality SD-OCT scans of the optic nerve and macula and at least 12 months follow-up.

Optical Coherence Tomography Parameters

At baseline and follow-up visits, patients underwent OCT scans centered on the optic nerve to measure the circumpapillary RNFL thickness and the macula to measure the ganglion cell layer (GCL) and inner plexiform layer (IPL) combined. After automated segmentation of each scan location, the same investigator (AG) would manually inspect and correct all segmentation errors. The retinal thickness map consists of 3 concentric rings with diameters of 1, 3, and 6 mm, with the 3- and 6-mm rings divided into quadrants. The ganglion cell–inner plexiform layer (GCIPL) thickness was calculated as an average of the four 3 mm subfields and four 6 mm subfields of combined GCL and IPL (the sum of GCL and IPL thickness, termed ganglion cell complex [GCC] hereafter) excluding the central area (1 mm radius) that corresponded to the foveola, as previously described.[14]

Assessment of Choroidal Abnormalities

The NIR images centered at the optic nerve and macula of each eye at the baseline visit and at the last follow-up were automatically recorded using HRA + OCT 5.1.2.0 (Heidelberg Engineering, Heidelberg, Germany; excitation light, 488 nm, barrier filter, 500 nm). Two trained neuro-ophthalmologists evaluated each image for presence, size, and number of Cas, and lesions were included by consensus. The borders of the hyperreflective lesions were manually delimitated using the OCT area tools (Fig. 1), and the area of each lesion was automatically calculated. The total area of CAs per eye was calculated as the sum of the area of the lesions.

Statistical Analysis

Demographic and clinical characteristics were summarized by standard descriptive summaries (e.g., mean and SDs for continuous variables such as age and percentages for

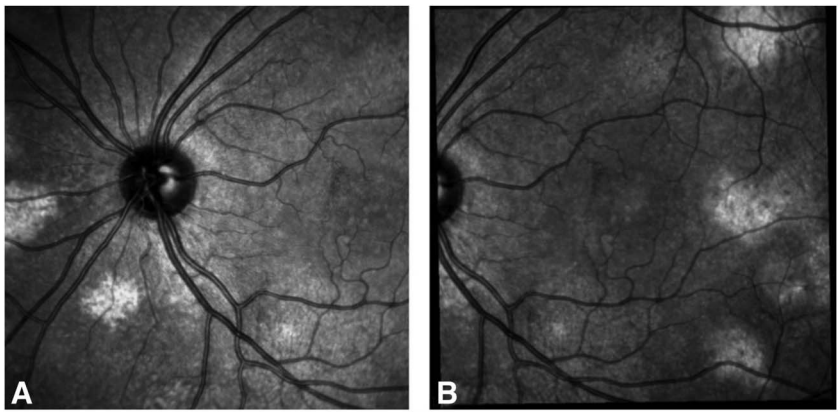

FIG. 1. Near-infrared reflectance images of the optic nerve (**A**) and macula (**B**) of children with neurofibromatosis type 1–associated optic pathway gliomas.

categorical variables such as sex). Cross-sectional associations of the presence, number, and total area of CAs with VA, RNFL, and GCC at baseline and the last follow-up were evaluated by generalized regression models with adjustment by age and sex, and intereye correlation was account for by using generalized estimating equations. All statistical analyses were performed using SAS v9.4 (SAS Institute Inc, Cary, NC), and two-sided P value <0.05 was considered statistically significant.

RESULTS

A total of 41 children (82 eyes) with NF1-OPGs from 3 centers were included in the study. The mean ± SD age of the children was 10.2 ± 3.3 years at baseline and 11.8 ± 3.3 years at the last visit with a mean follow-up period of 20.4 ± 7.2 months. Twenty-three children (56.1%) were female. The mean VA in logMAR was 0.08 ± 0.31 at baseline and 0.08 ± 0.29 at the last follow-up visit. The mean RNFL was 84.0 ± 24.0 μm (ranging from 34 to 160 μm) at baseline and 83.1 ± 24.1 (ranging from 30 to 157 μm) at the last follow-up. The demographic and clinical characteristics of the children included in the study are described in Table 1.

CAs were identified in 38 eyes (46.3%) at baseline. Among the eyes with CAs, the number of CA per eye ranged from 1 to 7 lesions with a mean of 2.1 ± 1.7 (median 1.0, [IQR] 1.0, 3.0) lesions and 22 (26.8%) eyes with only 1 CA. At the last follow-up visit, CAs were identified in 39 eyes (47.6%) with a mean of 2.2 ± 1.8 (median 1.0, [IQR] 1.0, 3.0) lesions per eye. The lesions had a mean total area of 2.0 ± 1.7 mm^2 (median 1.4, [IQR] 0.9, 2.4) at baseline and 2.3 ± 2.1 mm^2 (median 1.6, [IQR] 1.0, 2.7) at the last follow-up. Figure 2 shows a representative example of 1 eye with a single CA at baseline (a) and the last follow-up (b). In this eye, there was an increase of 0.20 mm^2 over the 14-month interval.

In the regression analyses, the VA was not significantly associated with the presence, total number, and area of CAs at baseline (P = 0.15, P = 0.27, and P = 0.34, respectively) or last follow-up (P = 0.18, P = 0.09, and P = 0.09, respectively). Neither RNFL nor GCIPL was associated with the presence, number, or total area of CAs ($P > 0.08$ in all comparisons, Table 2).

DISCUSSION

To the best of our knowledge, this is the first prospective study to examine the relationship between CAs and parameters of visual function and structure in children with NF1-OPGs. We did not find a statistically significant association of CAs (presence, number, or total area) with VA and OCT parameters at baseline nor at the last follow-up. These data suggest that CAs are not associated with structure and visual function in pediatric patients with NF1-OPGs.

CAs are a new ocular biomarker for NF1, but their relationship with structure and visual function still needs to be clarified. These nodules are located in the deep layers of the choroid below the choriocapillaris. It has been suggested that their presence may compress the overlying choroidal vessels causing some degree of choroidal ischemia.[15] Touzé et al[16] hypothesized that CAs could affect the electrical activity of the retinal pigment epithelium in subjects with NF1, but did not find a statistically significant difference in electrooculogram (EOG) value measures between patients

TABLE 1. Clinical features at baseline and last follow-up of the subjects included in the study

Demographic and Clinical Feature	Baseline	Last Follow-up
Subjects	41	41
Age (y), mean (SD)	10.1 (3.3)	11.8 (3.3)
Sex (female)	23 (56.1%)	
Follow-up (mo), mean (SD)	20.4 (7.2)	
Eyes	82	82
Presence of CAs, yes (%)	38 (46.3%)	39 (47.6%)
Number of CAs, median (IQR)*	1.0 (1.0, 3.0)	1.0 (1.0, 3.0)
CAs total area (mm^2), median (IQR)*	1.4 (0.9, 2.4)	1.6 (1.0, 2.7)
Visual acuity (logMAR)	N = 78	N = 80
Mean (SD)	0.08 (0.31)	0.08 (0.29)
Median (IQR)	0.00 [−0.10, 0.20]	0.00 [−0.10, 0.10]
RNFL (microns)	N = 77	N = 78
Mean (SD)	84.0 (24.0)	83.1 (24.1)
Median (IQR)	89.0 [67.0, 100.0]	87.0 [65.5, 98.0]
GCC global (6 mm + 3 mm) (microns)	N = 73	N = 70
Mean (SD)	70.8 (13.9)	69.6 (13.0)
Median (IQR)	74.5 [61.2, 79.0]	71.6 [58.6, 78.5]

*Among eyes with CAs.

CAs, choroidal abnormalities; GCC, ganglion cell complex; IQR, interquartile range; Max, maximum; Min, minimum; RNFL, retinal nerve fiver layer.

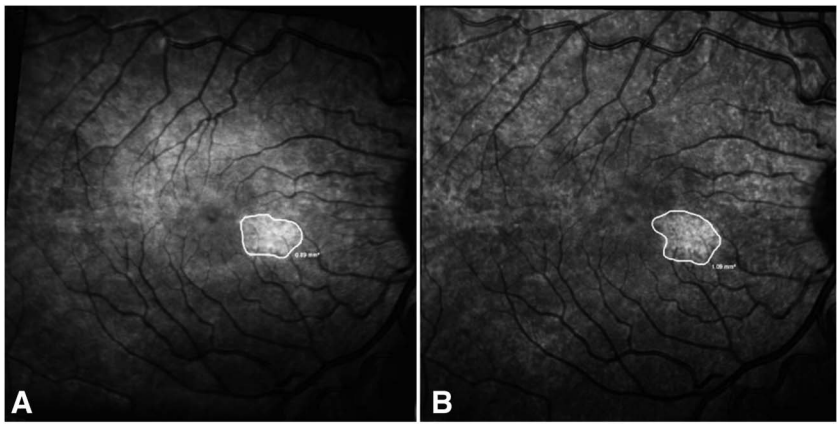

FIG. 2. Near-infrared reflectance images of a child with neurofibromatosis type 1–associated optic pathway glioma. **A**. Hyperreflective lesion with an area of 0.89 mm^2, and (**B**) the same lesion 14 months later, measuring 1.09 mm^2.

with NF1 with and without CAs, and no significant correlation between surface and number of CAs with measures of EOG. Godinho et al[12] also assessed the correlation between CAs and RNFL in a cohort of patients with neurofibromatosis type 1 and found no significant correlation between RNFL and CAs. In contrast to these studies that evaluated the patients at one point in time and included patients with NF1 with and without OPG, we evaluated the association of CA parameters with visual structure and function in patients with NF1-OPG who were evaluated at baseline and also after a mean follow-up of 20 months. We aimed to understand whether the presence, size, and number of these lesions were associated with VA, RNFL, and GCIPL. This is essential information for clinicians because a decline

TABLE 2. Regression analysis for evaluating the cross-sectional association between presence, number, and area of choroidal abnormalities with visual acuity and optic coherence tomography parameters at baseline and last follow-up, adjusted by age and sex

	Coefficient (SE)*	*P*	Coefficient (SE)*	*P*
CAs	Baseline VA N = 78		Last Follow-Up VA N = 82	
Presence		0.19		0.16
No	0.13 (0.06)		0.12 (0.06)	
Yes	0.04 (0.04)		0.04 (0.03)	
Number (per unit increase)†	−0.02 (0.02)	0.40	−0.02 (0.01)	0.12
Total area (per unit increase)†	−0.02 (0.02)	0.43	−0.02 (0.01)	0.08
CAs	Baseline RNFL N = 77		Last Follow-Up RNFL N = 78	
Presence		0.71		0.55
No	82.8 (5.2)		81.2 (5.2)	
Yes	84.9 (3.8)		84.6 (3.7)	
Number (per unit increase)†	0.21 (1.9)	0.91	0.83 (1.7)	0.62
Total area (per unit increase)†	0.89 (1.8)	0.61	1.13(1.3)	0.42
CAs	Baseline GCC (Global) N = 73		Last Follow-Up GCC (Global) N = 70	
Presence		0.87		0.56
No	70.5 (3.2)		67.8 (3.1)	
Yes	71.1 (2.0)		69.8 (1.9)	
Number (per unit increase)†	−0.09 (1.1)	0.94	−0.22 (1.2)	0.85
Total area (per unit increase)†	0.22 (1.1)	0.84	0.04 (1.0)	0.97

*Adjusted by age and sex.
†As a continuous value.
CAs, choroidal abnormalities; GCC, ganglion cell complex; IR, interquartile.

in visual parameters affects the management and treatment decisions in these patients. However, in agreement with previous studies, we also did not find a significant relationship of CA parameters with VA, RNFL, or GCIPL, suggesting that if there are any associated vascular changes, they may not be sufficient to affect retinal function.

In our study, CAs were present in 46% of eyes at baseline and 48% of the eyes at the last follow-up. Only 1 eye developed a new CA during the follow-up period. The total area of CAs was fairly stable with a median of 1.4 mm^2 at baseline and 1.6 mm^2 at the last follow-up. Interestingly, the prevalence of CAs in our cohort is smaller than reported in the pediatric population with NF1, which varies between 64% and 78.9%.[3,7,17,18] It may be partly explained by the lower age of the children included in our study, with a mean age of 11.9 ± 3.1 years at the last follow-up.

Our study had limitations. The CAs were manually measured, which may have interfered with the accuracy of the measurements of the CAs; however, this was unlikely to significantly affect the results. In addition, the NIR images were analyzed by a trained neuro-ophthalmologist and by a dual-trained pediatric and neuro-ophthalmologist to reduce observer bias. As another limitation, we only evaluated OCT NIR images 30° field posterior pole and around the optic nerve; thus, we did not account for CAs present in a more peripheral location. Nevertheless, there is no standardized protocol to identify CAs, and most choroidal nodules are located in the posterior pole.[3,19] Furthermore, it could be questioned whether enhanced depth imaging (EDI) OCT would be helpful in identifying CAs. Although EDI OCT visualizes the depth extent of the choroid, the identification of CAs is based on the NIR images.

This study found that although prevalent, CAs are not ubiquitous in children with NF1-OPGs. Our results suggest that the presence, number, and size of CAs are not associated with RNFL and GCC thickness or VA. However, further studies are necessary to evaluate the best imaging method to identify and determine the extent of these lesions, and longer follow-up may be required to confirm our findings.

REFERENCES

1. **Ferner RE**. Neurofibromatosis 1 and neurofibromatosis 2: a twenty first century perspective. Lancet Neurol. 2007;6:340–351.
2. **Ratner N**, Miller SJ. A RASopathy gene commonly mutated in cancer: the neurofibromatosis type 1 tumour suppressor. Nat Rev Cancer. 2015;15:290–301.
3. **Viola F**, Villani E, Natacci F, et al. Choroidal abnormalities detected by near-infrared reflectance imaging as a new diagnostic criterion for neurofibromatosis 1. Ophthalmology. 2012;119:369–375.
4. **Legius E**, Messiaen L, Wolkenstein P, et al. Revised diagnostic criteria for neurofibromatosis type 1 and Legius syndrome: an international consensus recommendation. Genet Med. 2021;23:1506–1513.
5. **Wolter JR**. Nerve fibrils in ovoid bodies. Arch Ophthalmol. 1965;73:696–699.
6. **Kurosawa A**, Kurosawa H. Ovoid bodies in choroidal neurofibromatosis. Arch Ophthalmol. 1982;100:1939–1941.
7. **Flores Pimentel M**, Heath A, Wan MJ, et al. Prevalence of choroidal abnormalities and Lisch nodules in children meeting clinical and molecular diagnosis of neurofibromatosis type 1. Transl Vis Sci Technol. 2022;11:10.
8. **Moramarco A**, Giustini S, Nofroni I, et al. Near-infrared imaging: an in vivo, non-invasive diagnostic tool in neurofibromatosis type 1. Graefes Arch Clin Exp Ophthalmol. 2018;256:307–311.
9. **Yasunari T**, Shiraki K, Hattori H, Miki T. Frequency of choroidal abnormalities in neurofibromatosis type 1. Lancet. 2000;356:988–992.
10. **Cosmo E**, Frizziero L, Miglionico G, et al. Choroidal abnormalities in pediatric NF1: a cohort natural history study. Cancers (Basel). 2022;14:1423.
11. **Chilibeck CM**, Shah S, Russell HC, Vincent AL. The presence and progression of choroidal neurofibromas in a predominantly pediatric population with neurofibromatosis type-1. Ophthalmic Genet. 2021;42:223–229.
12. **Godinho G**, Esteves-Leandro J, Alves G, et al. Correlation between ophthalmologic and neuroradiologic findings in type 1 neurofibromatosis. J Neuroophthalmol. 2022;42:101–107.
13. **Avery RA**, Bouffet E, Packer RJ, Reginald A. Feasibility and comparison of visual acuity testing methods in children with neurofibromatosis type 1 and/or optic pathway gliomas. Invest Ophthalmol Vis Sci. 2013;54:1034–1038.
14. **Kim EK**, Park HYL, Park CK. Segmented inner plexiform layer thickness as a potential biomarker to evaluate open-angle glaucoma: dendritic degeneration of retinal ganglion cell. PLoS One. 2017;12:e0182404.
15. **Rao RC**, Choudhry N. Enhanced depth imaging spectral-domain optical coherence tomography findings in choroidal neurofibromatosis. Ophthalmic Surg Lasers Imaging Retina. 2014;45:466–468.
16. **Touzé R**, Abitbol MM, Bremond-Gignac D, Robert MP. Function of the retinal pigment epithelium in patients with neurofibromatosis type 1. Invest Ophthalmol Vis Sci. 2022;63:6.
17. **Goktas S**, Sakarya Y, Ozcimen M, et al. Frequency of choroidal abnormalities in pediatric patients with neurofibromatosis type 1. J Pediatr Ophthalmol Strabismus. 2014;51:204–208.
18. **Vagge A**, Camicione P, Capris C, et al. Choroidal abnormalities in neurofibromatosis type 1 detected by near-infrared reflectance imaging in paediatric population. Acta Ophthalmol. 2015;93:e667–e671.
19. **Nakakura S**, Shiraki K, Yasunari T, Hayashi Y, Ataka S, Kohno T. Quantification and anatomic distribution of choroidal abnormalities in patients with type I neurofibromatosis. Graefes Arch Clin Exp Ophthalmol. 2005;243:980–984.