



# Conjunctival optical coherence tomography angiography imaging in sickle cell maculopathy<sup>☆</sup>

Glory E. Mgboji, Dennis Cain, Adrienne W. Scott<sup>\*</sup>

Retina Division, Wilmer Eye Institute, Johns Hopkins University School of Medicine, 600 North Wolfe Street, Baltimore, MD, 21287, USA

## ARTICLE INFO

### Keywords:

Sickle cell retinopathy  
Optical coherence tomography angiography  
Conjunctival sign

## ABSTRACT

**Purpose:** To compare conjunctival and macular manifestations of sickle cell anemia using optical coherence tomography angiography (OCTA).

**Observations:** OCTA imaging of the macula in two patients with HbSS and HbSC revealed areas of decreased vascular density, more prominent in the deep capillary plexus than in the superficial capillary plexus. Conjunctival OCTA of both affected patients revealed areas of reduced vascular density corresponding to the vascular abnormalities observed on slit lamp examination and prominent conjunctival flow voids when compared to an unaffected control.

**Conclusions and Importance:** OCTA allows for high resolution visualization of conjunctival findings present in sickle cell patients with macular vascular flow voids. Further studies are needed to explore the utility of conjunctival OCTA and the relationship between conjunctival and macular perfusion and systemic hemoglobinopathy.

## 1. Introduction

Sickle cell disease, the most common hemoglobinopathy in the world, affects an estimated 100,000 Americans,<sup>1</sup> with an additional three million Americans carrying the trait.<sup>2</sup> The pathophysiology of the disease involves a point mutation in the beta globin gene, which results in the substitution of glutamic acid for valine.<sup>3</sup> In homozygotes (HbSS), the abnormal hemoglobin molecules polymerize and red blood cells subsequently form rigid, crescent shaped structures that are prone to causing vaso-occlusive events and downstream hypoxia. Individuals with HbSC typically have less severe vaso-occlusive pain crises in comparison to those with HbSS; however, they are at increased risk for certain complications, such as proliferative retinopathy.<sup>4</sup>

Clinical features of sickle cell retinopathy have been well described with the help of retinal imaging tools like fluorescein angiography (FA) and spectral domain optical coherence tomography (OCT). Newer imaging modalities, such as optical coherence tomography angiography (OCTA), allow for the noninvasive, high resolution visualization of ocular vasculature (Fig. 1A–C display normal macular and conjunctival

vascular images obtained using this new technology). OCTA has already been used to reveal other clinical features of the disease, such as reduced macular vascular density in children with sickle cell disease.<sup>5</sup>

Though the posterior segment findings of sickle cell retinopathy currently dominate much of the literature, there is rich opportunity to explore anterior segment findings and potentially connect them to what is already known about systemic hemoglobinopathy. For example, the “conjunctival sickling sign” seen in patients with sickled cells was first observed in the 1950s. It was characterized by numerous short, comma shaped capillary segments and considered pathognomonic for sickle cell disease.<sup>6,7</sup> The severity of the conjunctival sign has been correlated to the percentage of hemoglobin S and the number of irreversibly sickled cells among patients carrying the hemoglobin S allele.<sup>7</sup> Paton described careful examination of the bulbar conjunctiva and observation of the conjunctival comma sign in SCD as a marker of the presence of systemic hemoglobinopathy, even when hemoglobin electrophoresis results are unknown.<sup>8</sup> Notably, conjunctival changes were described as the “most reliable evidence of clinically significant disease,” even more so than retinal changes.<sup>9,10</sup> Such a contention merits further exploration with

<sup>☆</sup> Reprinted from American Journal of Ophthalmology, Volume 209, Ong SS, Linz MO, Li X, Liu TYA, Han IC, Scott AW, Retinal thickness and microvascular changes in children with sickle cell disease evaluated by optical coherence tomography (OCT) and OCT angiography, pages 88–98, Copyright 2020, with permission from Elsevier.

<sup>\*</sup> Corresponding author. Retina Division, Wilmer Eye Institute, Johns Hopkins University School of Medicine, 600 North Wolfe Street Maumenee Building, USA. E-mail address: [ascott28@jhmi.edu](mailto:ascott28@jhmi.edu) (A.W. Scott).

<https://doi.org/10.1016/j.ajoc.2022.101428>

Received 7 July 2021; Received in revised form 5 February 2022; Accepted 14 February 2022

Available online 16 February 2022

2451-9936/© 2022 Published by Elsevier Inc. This is an open access article under the CC BY-NC-ND license (<http://creativecommons.org/licenses/by-nc-nd/4.0/>).

current imaging techniques.

Other anterior segment findings, like iris atrophy and depigmentation, have been associated with increased risk for proliferative sickle cell retinopathy.<sup>11</sup> The conjunctival sign could also be explored for possible relations to posterior segment features of disease. In fact, reduced conjunctival venular blood velocity has recently been described in patients with sickle cell disease and focal macular thinning.<sup>12</sup> Exploration of conjunctival features using novel imaging technologies is thus warranted to provide further insight into the clinical utility of conjunctival findings and associations with systemic hemoglobinopathy.

To our knowledge, there have been no reports of optical coherence tomography angiography (OCTA) to image the conjunctiva of patients with sickle cell disease. We sought to document two patients, one with HbSS, and one with HbSC disease using OCTA. Herein, we perform multi-modal imaging to describe conjunctival and macular OCTA findings.

## 2. Methods

This study is a single-institution, observational case series of two patients (four eyes). One patient with documented HbSS disease and one patient with documented HbSC disease were consented to participate verbally. Private health information was handled in accordance with HIPAA regulations.

Slit lamp and OCTA images of the macula and inferior bulbar conjunctiva were obtained for each eye 10 minutes after administration of 5% phenylephrine for pupillary dilation. Slit lamp images of 16 megapixels at 12X magnification were obtained using a Zeiss photo slit lamp and diffuse setting, flash 1 F44. Conjunctival and macular OCTA imaging was performed using Optovue RTVue XR (Optovue Inc, Fremont, California, USA), which has a speed of 70,000 scans/sec, 840 nm light source, and axial resolution of <math><5\ \mu\text{m}</math>. For the macula,  $3 \times 3\ \text{mm}$  and  $6 \times 6\ \text{mm}$  scans centered on the fovea were obtained.  $6 \times 6\ \text{mm}$  scans through the temporal macula, and at or near the fovea nasally, were obtained due to temporal paramacular involvement notably seen in sickle cell disease. For conjunctival imaging,  $3 \times 3\ \text{mm}$  scans were obtained using external fixation and a manual focus set at +20 diopters. Multiple scans were performed for each eye. Images with the best subjective quality (minimal motion artifact and adequate signal strength) were selected for analysis.

The control patient was similarly age-matched to the patients with SCD, and confirmed no history of systemic hemoglobinopathy or retinal vascular disease. No phenylephrine drops were used prior to imaging the control patient. Given the heterogeneous nature of ocular sickle cell findings among genotypes, two patients (one with Hgb SS and one with Hgb SC), were selected to represent the two most prevalent genotypes in sickle cell disease. We desired to compare posterior and anterior segment findings in patients with differing degrees of retinopathy in an exploratory and descriptive (rather than analytical) manner.

## 3. Findings

### 3.1. Case 1

A 27-year-old woman with sickle cell anemia (HbSS) on hydroxyurea presented for a new patient examination after being referred by her hematologist. The patient had a history of rare vaso-occlusive pain crises, which occurred once every three years, and two episodes of acute chest syndrome that both required intensive care unit level care. Otherwise, she managed intermittent pain at home and had no outstanding disease-related concerns. She had mild myopia and wore glasses for distance vision, and was otherwise visually asymptomatic.

Her uncorrected distance visual acuity was 20/50 in the right eye and 20/25 in the left eye. Slit lamp examination of the anterior segment revealed inferior, superior, nasal, and temporal corkscrew conjunctival vessels and short capillary segments, consistent with the conjunctival comma sign previously observed in SCD.<sup>6,7</sup> No sea-fan neovascularization was present. OCT of the macula revealed mild macular thinning in both eyes (Fig. 2A).

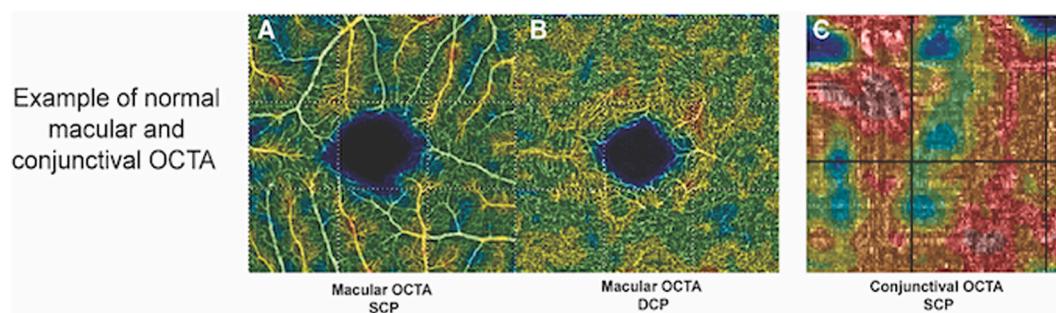
OCTA images of the macula (Fig. 2B and C) showed areas of patchy flow deficits, with greater severity in the right eye as compared to the left. The areas of decreased vascular density were more prominent in the deep capillary plexus than in the superficial capillary plexus. A slit lamp image of the patient's inferior bulbar conjunctiva was taken (Fig. 2D), along with corresponding OCTA images of the superficial and deep capillary plexuses (Fig. 2E and F, respectively). The OCTA images revealed vascular flow deficits that corresponded to conjunctival abnormalities seen on slit lamp examination (Fig. 2G and H).

### 3.2. Case 2

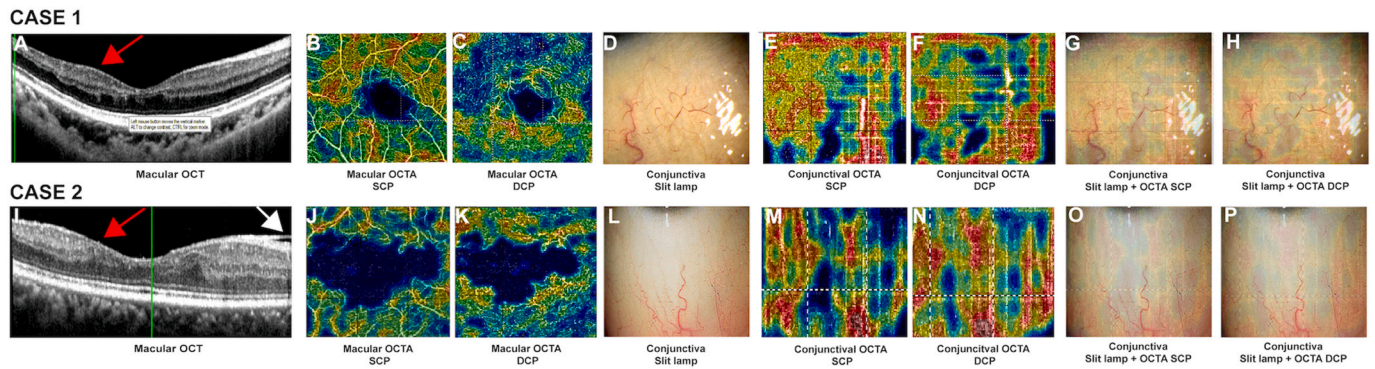
A 37-year-old male with HbSC (not on disease modifying therapy) presented for routine evaluation. He had a history of stage IV proliferative retinopathy as well as avascular necrosis of the hip, requiring total hip replacement. His pain crises were infrequent. His ocular history included recurrent vitreous hemorrhages, with previous scatter laser photocoagulation treatment with as needed intravitreal bevacizumab therapy for active sea-fan neovascularization in his right eye. He had phthisis bulbi in his left eye due to complications from Stage V proliferative sickle cell retinopathy in adolescence.

His distance visual acuity with glasses was 20/32 in the right eye with no light perception in the left eye. Slit lamp examination of the anterior segment showed a white and quiet conjunctiva in the right with a few short, comma shaped capillary segments. Partially-vascularized sea fan neovascular complexes were present through the retinal periphery. OCT revealed foveal splay with a mild epiretinal membrane (Fig. 2I).

OCTA images of the macula (Fig. 2J and K) showed significant flow deficits in both the superficial and deep capillary plexuses with an



**Fig. 1.** OCTA vascular density maps of the macula and conjunctiva in normal eyes. 1A and B display imaging of the macular superficial capillary plexus (SCP) and deep capillary plexus (DCP), respectively.<sup>8</sup> 1C displays a normal conjunctival superficial capillary plexus. Dark blue regions represent areas of flow deficits. The control patient did not receive phenylephrine prior to conjunctival imaging. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)



**Fig. 2. Macular and conjunctival imaging for Case 1 (top row) and 2 (bottom row).** 2A shows right eye macular OCT with the red arrow indicating inner retinal thinning in the temporal macula. 2B and 2C show imaging of the macular superficial capillary plexus (SCP) and deep capillary plexus (DCP). 2D displays a slit lamp image of the left eye's inferior bulbar conjunctiva, with short, comma shaped capillary segments in the center. 2E and 2F represent conjunctival OCTA imaging, with 2G and 2H displaying the vascular density maps for the superficial and deep capillary plexuses (respectively) superimposed on the area shown on slit lamp examination in 2D (Case 1). In a similar sequence, for Case 2, 2I shows macular OCT, with the red arrow indicating foveal splaying. A mild epiretinal membrane is present (white arrow). 2J and 2K show imaging of the right macular superficial capillary plexus (SCP) and deep capillary plexus (DCP). 2L displays a slit lamp image of the right inferior bulbar conjunctiva, with short capillary segments also apparent. 2M and 2N similarly display OCTA imaging, with 2O and 2P displaying the vascular density maps for the superficial and deep capillary plexuses (respectively) superimposed on the area shown on slit lamp examination in 2L (Case 2). Dark blue regions represent areas of flow deficits. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

irregular and enlarged foveal avascular zone. A slit lamp image of the patient's inferior bulbar conjunctiva was also taken (Fig. 2L), along with OCTA images of the superficial and deep capillary plexuses (Figures M and N, respectively). These showed vascular flow deficits corresponding to conjunctival abnormalities that were more prominent in the superficial capillary plexus (Figures O and P).

#### 4. Discussion

Though vascular conjunctival abnormalities have long been described in patients with sickle cell anemia and have been identified as potential targets for pharmacotherapeutic and surgical intervention, few studies have used advanced imaging techniques to explore the conjunctiva in these patients. Because isolated comma-shaped vascular segments have been previously described as the most reliable markers of disease,<sup>10</sup> they merit further characterization with contemporary imaging modalities. In this report, OCTA images of the macula and conjunctiva were compared in a HbSS and a HbSC patient with known sickle cell maculopathy demonstrated on OCT and OCTA. In both patients, macular OCTA imaging revealed bilateral reductions in vascular density that were more pronounced in the deep capillary plexus than in the superficial capillary plexus, as has been previously observed in the literature.<sup>13</sup> Additionally, we used conjunctival OCTA to reveal areas of reduced vascular density that corresponded to the "conjunctival sign" observed on slit lamp examination.

The conjunctival sign was present in both patients, and subjectively more notable in our HbSS patient, as previously described.<sup>7</sup> In comparison to a control, both patients had prominent flow voids in the conjunctiva on OCTA. It is notable that our control patient did not receive topical phenylephrine (which can cause conjunctival vessel vasoconstriction) prior to conjunctival imaging, which may limit the strength of our comparisons. However, given the exploratory, descriptive nature of the study (rather than analytical), we find the following observations worthy of discussion: When compared to each other, we observed that our HbSC patient had more flow deficits in the conjunctival superficial capillary plexus, while our HbSS patient had more flow voids in the deep capillary plexus. Future studies will be useful in reproducing and potentially linking these findings to other aspects of systemic anemia and SCD genotype.

Recent literature on the relationship between conjunctival changes and retinopathy is sparse but significant. Valeshabad et al.<sup>12</sup> were the

first to analyze conjunctival microcirculation in relation to the presence and absence of macular thinning using a prototypical optical imaging system. They found reduced conjunctival venular blood velocity in patients with sickle cell retinopathy and focal macular thinning. This supports our finding of reduced conjunctival vascular density in a HbSC patient with known advanced stage retinopathy.

New developments in our knowledge of conjunctival changes and systemic manifestations of sickle cell disease have surfaced within the last decade. Wanek et al.<sup>14</sup> were the first to explore differences in conjunctival microvascular flow dynamics between patients with the Hgb S allele. These authors reported a lower flow velocity in patients with Hgb SC disease compared to those with Hgb SS disease and a linear relationship between vessel diameter and blood velocity in those with AA and SS genotypes. These authors called for further attention to conjunctival microcirculation, as its features have been associated with other systemic findings, such as middle cerebral artery flow. Since then, examination of conjunctival microvasculature in this patient population has not only informed us of potential associations with macular thinning,<sup>12</sup> but also of other systemic features of disease, like kidney dysfunction,<sup>15</sup> stroke,<sup>16</sup> and vaso-occlusive pain crises.<sup>17</sup>

Conjunctival imaging could be a useful adjunct to posterior segment imaging in sickle cell retinopathy. Our case series is limited by description of two patients with sickle cell disease. Further study is needed to evaluate conjunctival OCTA and its relationship to the macular microvascular circulation and systemic hemoglobinopathy status such as degree of anemia, systemic oxygenation, or response to systemic disease-modifying therapies. Given the possibility of visualizing anterior segment changes in sickle cell disease with advanced imaging techniques, and the previously described associations of such changes with sickle cell retinopathy, larger scale studies are warranted to explore conjunctival abnormalities in sickle cell disease.

#### 5. Conclusions

OCTA permits the visualization of conjunctival findings present in sickle cell patients with macular changes. Further studies are needed to explore the utility of conjunctival OCTA and the relationship between conjunctival and retinal findings, and systemic hemoglobinopathy in sickle cell disease.

## Patient consent

The patients consented to publication of the cases verbally.

## Funding

No funding or grant support.

## Authorship

All authors attest that they meet the current ICJME criteria for authorship. All authors approve the order of authors listed in the manuscript. Contributions for each author are as follows:

- Glory Mgboji – Writing (original draft preparation), visualization
- Dennis Cain – Investigation (data collection)
- Adrienne Scott – Conceptualization, supervision, review and editing

## Declaration of competing interest

The authors have no financial disclosures.

## Acknowledgments

None.

## References

- Centers for Disease Control and Prevention. *Data and Statistics on Sickle Cell Disease*. Centers for Disease Control and Prevention; 2021. <https://www.cdc.gov/ncbddd/sicklecell/data.html>.
- Reeves SL, Jary HK, Gondhi JP, Kleyn M, Spector-Bagdady K, Dombkowski KJ. Incidence, demographic characteristics, and geographic distribution of sickle cell trait and sickle cell anemia births in Michigan, 1997–2014. *Molecular Gen Genomic Med*. 2019;7(8):e795.
- Sundt P, Gladwin MT, Novelli EM. Pathophysiology of sickle cell disease. *Annu Rev Pathol*. 2019;14:263–292.
- Scott AW. Ophthalmic manifestations of sickle cell disease. *South Med J*. 2016;109(9):542–548.
- Ong SS, Linz MO, Li X, Liu TYA, Han IC, Scott AW. Retinal thickness and microvascular changes in children with sickle cell disease evaluated by optical coherence tomography (OCT) and OCT angiography. *Am J Ophthalmol*. 2020;209:88–98.
- DeQuevedo A. *Microscopic Observation of the Circulation of Sickle Cell Anemia Patients Made in the Bulbar Conjunctival Vessels: Preliminary Note*. Library Medical College of South Carolina; 1952.
- Nagpal KC, Asdourian GK, Goldbaum MH, Raichand M, Goldberg MF. The conjunctival sickling sign, hemoglobin S, and irreversibly sickled erythrocytes. *Arch Ophthalmol*. 1977;95(5):808–811.
- Paton D. The conjunctival sign of sickle-cell disease. *Arch Ophthalmol*. 1961;66(1):90–94.
- Minatoya H, Acacio I, Goldberg M. Fluorescein angiography of the bulbar conjunctiva in sickle cell disease. *Ann Ophthalmol*. 1973;5(9):980–992.
- Paton D. The conjunctival sign of sickle-cell disease: further observations. *Arch Ophthalmol*. 1962;68(5):627–632.
- Osafo-Kwaako A, Kimani K, Ilako D, et al. Ocular manifestations of sickle cell disease at the Korlebu Hospital, Accra, Ghana. *Eur J Ophthalmol*. 2011;21(4):484–489.
- Kord Valeshabad A, Wanek J, Zelkha R, et al. Conjunctival microvascular haemodynamics in sickle cell retinopathy. *Acta Ophthalmol*. 2015;93(4):e275–e280.
- Han IC, Tadarati M, Scott AW. Macular vascular abnormalities identified by optical coherence tomographic angiography in patients with sickle cell disease. *JAMA ophthalmology*. 2015;133(11):1337–1340.
- Wanek J, Gaynes B, Lim JI, Molokie R, Shahidi M. Human bulbar conjunctival hemodynamics in hemoglobin SS and SC disease. *Am J Hematol*. 2013;88(8):661–664.
- Valeshabad AK, Wanek J, Saraf SL, et al. Changes in conjunctival hemodynamics predict albuminuria in sickle cell nephropathy. *Am J Nephrol*. 2015;41(6):487–493.
- Valeshabad AK, Wanek J, Mukarram F, Zelkha R, Testai FD, Shahidi M. Feasibility of assessment of conjunctival microvascular hemodynamics in unilateral ischemic stroke. *Microvasc Res*. 2015;100:4–8.
- Kord Valeshabad A, Wanek J, Gaynes B, Saraf SL, Molokie R, Shahidi M. Conjunctival microvascular hemodynamics following vaso-occlusive crisis in sickle cell disease. *Clin Hemorheol Microcirc*. 2016;62(4):359–367.