Quantitative and qualitative assessment of anterior segment optical coherence tomography capture of disease state in childhood anterior uveitis

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Synopsis

Anterior segment OCT imaging-based assessment of childhood anterior chamber inflammation is repeatable, responsive, correlates with clinical activity, makes the invisible visible for families, and may enable non-invasive capture of inflammatory cell type.

Abstract

Background/Aims

Anterior segment optical coherence tomography assessment of anterior chamber inflammation is an emerging tool. We describe the performance of AS-OCT in a paediatric population.

Methods

A mixed-methods prospective study, using routine clinical assessment as reference standard, and AS-OCT, with Tomey CASIA2 or Heidelberg Spectralis HS1, as index test, with data collected on patient perceptions of imaging. Repeatability, diagnostic indices, responsiveness to clinical change, and clinical correlations of imaging-based metrics (image cell count, size, density and brightness) were assessed, with construction of receiver operated characteristic (ROC) curves. Exploratory thematic analysis of responses from families was undertaken.

Results

A total of 90 children (180 eyes) underwent imaging. Bland Altman limits of agreement for CASIA2 repeatability ranged from +17 cells (95%CI 13.6 to 21.1) to -19 cells (95%CI -15.6 to -23.2) and HS1 from +1 (95%CI 0.9 to 1.2) to -1.0 (-1.2 to -0.8) cells. CASIA2 imaging had higher sensitivity of 0.92 (95%CI 0.78 to 0.97) versus HS1 imaging 0.17 (95%CI 0.07 to 0.34), with positive correlation between clinical grade and CASIA2 cell count (coefficient 12.8, p=0.02, 95%CI 2.2 to 23.4). Change in clinical grade at follow up examinations correlated with change in image based 'cell' count ($r^2$0.79, p<0.001). Patients reported a potential positive impact of seeing their disease activity.

Conclusion

Our findings suggest that OCT based imaging holds the promise of deeper understanding of disease, improved patient experience, and more granular monitoring of activity with resultant improved outcomes, but further work is needed to refine acquisition and analysis protocols.

What is already known on this topic

Anterior segment optical coherence tomography (AS-OCT) assessment of anterior chamber inflammation is an emerging tool.

What this study adds

We report that AS-OCT based quantification of disease state in childhood anterior uveitis is repeatable, responsive to clinical change, and welcomed by families and children with perceived benefits including the potential for community-based monitoring, and improved communication of disease state. AS-OCT metrics also hold some promise as a deeper diagnostic tool, through qualification of uveitis type.

How this study might affect research, practice or policy

Further work on refining acquisition and analysis protocols will support the use of AS-OCT for imaging-based surveillance and diagnosis, for remote management of disease, and for augmenting clinic decisions on therapeutic response or disease remission.

INTRODUCTION

Anterior uveitis, the most common manifestation of childhood onset uveitis,[1,2] is often asymptomatic, necessitating regular surveillance examinations for at risk children.[3] Increased anterior chamber cell count is associated with poorer outcomes,[4,5] but the systemic therapy often needed for disease control can be associated with negative impact on quality of life,[6] with consequent impact on family engagement with medical care. There are also challenges around defining disease control and disease remission.[7]

The current 'gold standard' clinical assessment for anterior uveitis is slit lamp biomicroscopy, with anterior chamber cell count (ACC) graded using the Standardization of Uveitis Nomenclature (SUN) scale, developed for use in adult disease through international consensus work.[8] Whilst the SUN scale brought standardisation into disease quantification, its basis in slit lamp examination leaves it open to intra- and interobserver variability,[9] with a 2-step improvement in ACC grade sometimes needed to evidence a 'true' clinical change.[10] An objective, sensitive disease assessment tool would aid disease management. It is also possible that images of asymptomatic active inflammation may support family engagement with disease, and concordance with treatment, by making an invisible disease visible. Recent studies have reported that anterior segment optical coherence tomography (AS-OCT) quantification of inflammation is feasible, with reasonable diagnostic indices,[11,12] but there is little evidence on the repeatability and responsiveness of imaging derived metrics. We aimed to address this evidence gap, and describe the performance of AS-OCT in a paediatric population.

METHODS

A prospective cross-sectional study. Study approvals were granted by the NHS Health Research Authority (19/SC/0283). Written informed consent and assent was secured. This research followed the tenets of the Declaration of Helsinki.

Eligibility criteria

Children and young people (aged under 18 years) diagnosed with all-cause uveitis with a history of previous anterior chamber inflammation were included. Recruitment took place at a specialist eye hospital (site 1), and a specialist children's hospital (site 2). Participants were approached consecutively on attendance to uveitis clinic, and re-approached wherever possible for repeated examinations at follow up appointments.

Reference and index tests

The reference standard test was routine undilated clinical assessment within a darkened room, carried out by the managing senior ophthalmologist (ALS, HP) with a desktop slit lamp (Haag Streit BM 900 site 1, BQ 900 site 2). The SUN anterior chamber cell count (ACC) grade was used to assess inflammation (0 ACC, no cells seen within a central 1x1mm long beam; +0·5, 1-5 cells seen within the beam; +1, 6-15; +2, 16-25; +3, 26-50; +4, more than 51 cells). Clinically active disease was defined as anterior chamber cell count grade ≥+0.5. Data were also collected on participant age, ethnicity, uveitis diagnosis and anterior chamber flare SUN grade. Laser flare photometry was not undertaken.

The index test was undilated anterior segment OCT (AS-OCT) within a darkened room, using the CASIA2 (Tomey Corporation, Japan; site 1) and Heidelberg Spectralis OCT1 (Heidelberg Engineering, Germany; site 2, acquisition protocols in supplemental data). One set of images was acquired from each eye consecutively, followed by a 1-2 minute rest and then repeated acquisition from each eye.

Following extraction from imaging systems as PNG files, pseudoanonymised cross sectional scans were analysed using ImageJ. Regions of interest (RoI), comprising the whole of the visible anterior chamber, were drawn manually by two authors (ALS, KE) with the senior author (ALS) reviewing all RoIs drawn. Default thresholding was used to binarise each image (figure 1). Automated 'cell' detection within the RoI was undertaken using the ImageJ Particle Analysis algorithm (http://imagej.net/Particle_Analysis, which provides a count and density of the hyper-reflective particles ('cells') within the RoI, particle size (in pixels), and relative brightness.

Children and their family were shown an image of hyper-reflective particles ('cells') on a cross sectional AS-OCT from a previous study[12] as part of the participant information procedure, asked to rate the acceptability of image acquisition on a 10cm visual analogue scale ("What did you think of the scan"), and invited to comment ("Please let us know if you have any other comments") on study aims.

Statistical analysis

Descriptive analysis of all measures was undertaken, with summary metrics where multiple B scans were clustered within one eye. Repeatability of the imaging-based

metrics (‘cell’ count, ‘cell’ size, ‘cell’ density and ‘cell’ brightness) was evaluated using measures from repeated image sets taken on the same eye at the same visit. Effectiveness of imaging-based diagnosis of inflammation was assessed using the AS-OCT performance versus the slit lamp assessment reference standard. Cases were AS-OCT ‘positive’ if ‘cells’ had been noted on any cross sectional image, and slit lamp ‘positive’ if SUN grade was >0. Correlations between imaging acquired ‘cell’ count and clinical assessment of inflammation was assessed using a multilevel linear regression model with correction for within-child correlation to account for the clustered structure of the eye level data for those children who had undergone assessment in both eyes. Receiver operated characteristic (ROC) curves were used to assess the impact of the different imaging metrics on the true positive and false positive rates, with adjusted areas under the ROC curve (AUC) presented. Exploratory thematic analysis[13] was undertaken in order to identify themes within free text responses from families. Coding was undertaken with an inductive approach to analysis, with frameworks developed by the investigators and then used to group data through an iterative process. Confidence intervals were reported at 95%, and a *p*-value threshold of 0.05 was used for statistical significance. Analyses were undertaken using Stata (version 15, StataCorp, College Station, Texas).

Patients and their families were involved in study conceptualisation, study design, and interpretation of study results. This study is supported by a ‘Generation R’ Young Persons Advisory Group (GOSH Y-PAG) and by a disease specific patient family advisory group (the Childhood Uveitis Study Steering group). Young people and patients co-designed the study to ensure minimisation of burden on study participants.

RESULTS

A total of 90 children (180 eyes) underwent imaging (supplemental figure 1). Median age at imaging was 11.5yrs (range 3 to 16, interquartile range 9 to 13). Of the 90 children, 58% were female (52 girls), and 55% (50 children) were from non-White ethnicity backgrounds (including Asian Indian or Pakistani origin n=28, Black African or Caribbean n=12). JIA associated uveitis was the most common disease entity (26 children, 29%), but idiopathic uveitis was the most common 'diagnosis' (40 children, 44%), (supplemental table 1). Of the 180 eyes, 22 eyes were aphakic, and 9 had peripheral (non-axial) band keratopathy. Clinical examination confirmed active disease in 66/180 eyes (37%) ranging from +0.5 to +3 ACC (+0.5 in 41, 23%, +1 in 11, 6%, +2 in 8, 5%, and +3 in 6, 3%). Anterior chamber flare was present in 103 eyes (range 0-3 SUN), including 30 clinically inactive (SUN=0) eyes.

The time interval between slit lamp examination (reference test) and OCT imaging (index test) ranged from 3 to 45 minutes (median 14 minutes), with no clinical interventions (eg mydriasis) occurring between procedures. There were no adverse events from performing the index test or the reference standard.

Diagnostic accuracy for AS-OCT

Across the individual images from 80 eyes acquired using the CASIA2 OCT, median 'cell' count per image was 1 'cell' (IQR 0 to 2 'cells', range 0 to 111 'cells'), and maximum 'cell' count per image per eye ranged from 0 to 135 (IQR 1 to 7), with 'cells' detected in scans from 35 of the 36 eyes with active disease, and 37 of the 44 eyes with inactive disease (supplemental table). Of note, amongst those 37 'false

positive' cases, 12 children had clinical observation of 'flare' in absence of cells (0 ACC) in the eye. The sensitivity of CASIA2 detection of clinically active disease was 0.92 (95% CI 0.78 to 0.97), specificity 0.16 (0.08 to 0.29), positive predictive value, PPV 0.47 (0.40 to 0.42), negative predictive value, NPV 1.0 (0.65 to 1.00), positive likelihood ratio, LR+, 1.10 (1.03 to 1.17), negative likelihood ratio, LR- 0.18 (0.02 to 1.35) (supplemental tables S2 and S3). Across images from 100 eyes imaged by the Spectralis OCT1 (HS1), median 'cell' count was 0, whilst maximum 'cell' count was 3, with cells detected in scans from only 5 of the 30 children with clinically active disease (SUN grades ≥+0.5), and 4 of the 70 children with inactive disease. Consequently, the sensitivity of HS1 detection was poor, at 0.17 (95% CI 0.07 to 0.34), with specificity of 0.93 (0.79 to 0.94), PPV 0.56 (0.27 to 0.81), NPV 0.73 (0.63 to 0.81), LR+ 2.39 (0.74 to 7.58), and LR- 0.88 (0.75 to 1.05).

Following adjustment for the clustering of data at eye level (multiple B-scans per eye) and child level (two eyes per child), 'cell' count was the imaging-based metric with the strongest association to the presence of clinical disease activity (regression coefficient, coeff, 0.15, p=0.002, 95% confidence interval 0.06 to 0.25), and to the presence of flare at the slit lamp (0.14, p=0.004, 0.04 to 0.23). 'Cell' size, 'cell' density and 'cell' brightness were not individually associated with these clinical states (supplemental tables S4 and S5).

ROC curves with covariate adjustment were constructed to explore the impact of 'cell' derived quantitative scores (median size and minimum size, supplemental tables S4) on diagnostic indices. Median and maximum detected 'cell' size (in pixels^2) both improved the AUC following covariate adjustment of ROC. AUC

improved from 0.58 (95% CI 0.44 to 0.72) to 0.86 (95% CI 0.53 to 1) with correction for maximum ‘cell’ size (figure 2). It further improved to 0.93 (95% CI 0.73 to 1.00) with correction for median ‘cell’ size. There was insufficient spread of ‘cell’ count or ‘cell’ size on HS1 imaging to allow construction of covariate adjusted ROC curves. Using a threshold for ‘cell’ size to define an OCT image as ‘positive’ for cells improved specificity (lowered the ‘false positive rate’ when compared to the reference test of slit lamp SUN grading) but reduced sensitivity (threshold of 2 pixel^2, sensitivity 0.5, 95% CI 0.35 to 0.66, specificity 0.82, 0.68 to 0.91, threshold of 3 pixel^2, sensitivity 0.33, 95% CI 0.20 to 0.50, specificity 0.91, 95% CI 0.79 to 0.96).

Repeatability of AS-OCT

Bland Altman limits of agreement (LoA) total ‘cell’ count across CASIA2 images taken 1 to 2 minutes apart ranged from 17 cells greater in the second image (95% CI 13.6 to 21.1) to 19 cells greater in the first (-15.6 to -23.2) (figure 3). There was evidence of a trend towards a lower ‘cell’ count in the second images, ie the images taken after the child had been sitting at the machine for the interval period. In CASIA2 images from 12/80 eyes (15%), there was discrepancy in ‘negative scans’ ie, cells detected on only one set of scans. Upper LoA for HS1 images was 1.1 (95% CI 0.9 to 1.2) and lower LoA was -1.0 (-1.2 to -0.8), with a discrepancy in ‘negative scans’ for 12/100 eyes (12%).

Quantification of inflammation

On multilevel regression modelling there was good positive correlation between clinical ACC score and image based ‘cell’ count (coeff 12.8, p=0.02, 95% CI 2.2 to 23.4). There was evidence of a wide range in ‘cell’ count at the more severe end of

clinical inflammation grading (figure 4), with median ‘cell’ count averaged across scans of 0 cells (range 0 – 9) in eyes at 0 SUN; 1 (range 0.5 – 7) in eyes at +0.5 SUN; 1.5 (range 0 – 3) at +1 SUN; 9.5 (range 1 – 17) at +2 SUN; and 58 (range 4 – 123) at +3 SUN.

Responsiveness

Repeat imaging was performed in 59 eyes, at between 2 weeks and 3 months from first image acquisition. All repeat imaging took place at site 1 (CASIA2 OCT). Clinical grading changed in 37 (62%) of these eyes, with worsening in 15 eyes (26%), and improvement in 24 (46%). Regression analysis with adjustment for clustering within individual and adjustment for baseline level of inflammation confirmed an association between change in clinical grade and change in image based ‘cell’ count ($r^2$0.79, p<0.001 change in ‘cell’ count with improving clinical grade). There was evidence of steeper change in ‘cell’ count with higher grades of baseline inflammation (baseline SUN 0.5+: *coeff* 0.3, *p*=0.1, 95% CI -0.1 to 0.6; SUN 1+: *coeff* 0.6, p<0.01, 95% CI 0.2 to 1.0; SUN 2+: *coeff* 2.0, p<0.001, 95% CI 1.6 to 2.4; SUN 3+: *coeff* 3.0, p<0.001, 95% CI 2.4 to 3.6).

Qualification of disease type

We observed an association between ‘cell’ size and diagnosis. Compared to a baseline of eyes from children with a diagnosis of idiopathic chronic anterior uveitis, eyes of children with active JIA associated uveitis and idiopathic panuvetis had a tendency towards populations of cells / particles with larger areas (figure 5). There was no association between ‘cell’ brightness and diagnosis, or between ethnicity and the imaging-based metrics.

Patient and family response to AS OCT imaging

Patient and / or parent acceptability of the process of OCT capture of disease state was high, with a median score of 9.3/10. Only 2 of the 53 respondents scored less than 8/10, with one respondent scoring 7 and another 0/10. Free text responses were returned by 11 families. The two themes which emerged (apart from supportive statements such as "*Just pleased that something is happening*", n=6, were around the potential impact of objective disease assessment in supporting community based monitoring of disease (n=7, illustrative quotes: "*As this is an ongoing problem, making life easier for families to be seen and checked locally would make a huge difference", "School would not be missed and work interrupted*") and the potential positive impact of seeing their / their child's disease activity, n=5 ("*I think that it is a good idea because we can see what is actually going on* ", "*people could…look and see what is happening with their eyes*").

DISCUSSION

From this study, we report that OCT derived metrics are dependent on OCT platform, and may be affected by patient movement. There is a high 'false positive' rate (when compared to a limited ground truth) but OCT based 'cell' count correlates with clinical grading of activity, is responsive to changes in clinical activity, and OCT derived 'cell' size may have some value in the diagnosis of uveitis type. Patients and families welcome objective disease metrics for childhood uveitis, with perceived benefits including potential for community based monitoring of disease, and enabling communication of disease state.

Laser flare photometry is an objective disease activity metric able to capture flare (the degree of light scatter causes by abnormal increases in protein content in the aqueous humor)[8] and has been used in clinical practice and clinical research.[14 15] This study is limited by the absence of LFP measures which would provide a 'gold standard' for comparison to OCT assessment of inflammation. However, flare levels show poor responsiveness, have uncertain prognostic importance, and fluctuate in relation to multiple child- and environment-specific factors.[15] Additionally, previous studies have suggested that OCT derived metrics which might indicate flare level (such as aqueous-to-air relative intensity, or ARI) are confounded by age, tear film and lighting levels.[16,17] Thus, the more robust cell markers have been used in this study. This study is also limited by the relatively small number of eyes at the higher grades of inflammation (+3 and +4 SUN ACC), a limitation typical of studies of anterior uveitis,[11,12,16,17] reflecting the 'real-world' distribution of disease severity. Further work on OCT metrics validation in severe inflammation should follow.

A significant finding is the apparent discordance at lower levels of inflammation. Our work shows that at the lower grades of inflammation, OCT is able to detect cells in apparently clinically inactive (as defined by SUN scoring criteria) eyes. SUN grading uses a repeatable clinical assessment (ie, visibility of cells within a central defined viewing field) to create a standardised score, but was not intended to be a comprehensive assessment of cell density within the 'whole' of the anterior chamber, and is unable to capture activity within the inferior anterior chamber, which has the highest aggregation of inflammatory cells.[18 19] It may be that the particles detected on OCT are not inflammatory cells, but are artefact created by pigment or protein. The same can be said of anterior chamber cells detected at the slit lamp. Our study

showed that ‘cell’ size did not correspond to ACC grading, suggesting that even the smaller particles on OCT corresponded to inflammatory cells.

Previous studies of OCT based quantification of anterior chamber inflammation have reported strong correlation of ‘cell’ count to clinical activity an inflammation grading, with differing performance by imaging platform.[11] There has been an absence of exploration of two key biometric aspects: responsiveness and repeatability. Our study suggests that imaging provides a responsive measure, but that patient movement may impact on ‘cell’ distribution within a mobile aqueous environment. Acquisition protocols with multiple cross sectional images will help to ensure comprehensive assessment of anterior chamber inflammation, and protocols may need to differ across different machines in line with machine performance. OCT based metrics provide greater information on disease state, but longitudinal validation, to enable assessment of these metrics as prognostic or predictive biomarkers able to inform on clinical outcomes and guide treatment.

Aqueous infiltration by polymorphonuclear neutrophils (approximately 9 micrometers/μm in diameter), lymphocytes (small, 7μm and large, 15μm) and macrophages (20 – 90μm) is a feature of uveitis,[19] with different cell populations seen in acute versus chronic, or granulomatous versus non granulomatous, or specific uveitides.[19] Despite its lower axial resolution, the greater AC depth and higher scanning speed achieved by CASIA2 appears to enable better capture of AC cells versus the (first generation) Spectralis HS1.[20] Newer machines with higher axial resolution and scanning speeds, such as the Spectralis HS2 or Anterion, may provide key additional imaging features in future.[20] Our findings, and findings from in

vitro, animal and human in vivo studies from other teams suggest that OCT 'cell' characteristics could indicate anterior uveitis subtype,[21] enabling non-invasive cellular phenotyping of disease, an essential and powerful progression in our understanding of pathogenesis and personalisation of uveitis management.[18,19] Caution should be exercised in the interpretation of our findings in the absence of tissue sampling (paracentesis of aqueous humour with cytological analysis).

Patient co-development of study methodology enabled capture of findings which suggest that OCT imaging may be a powerful aid in patient-clinician communication. The management of childhood chronic anterior uveitis, a potentially blinding disorder, can be complicated by poor family concordance with systemic therapy. Our findings suggest that families welcome an objective measure of disease. This is consistent with qualitative work and patient co-developed 'core outcome sets' in adult onset uveitis.[22,23] Future, more detailed qualitative analysis of patient and family perceptions of the positive or negative impact of 'seeing' uveitis activity state is warranted.

Standardised and validated imaging-based biomarkers would supporting future clinical and pathogenesis research,[24] precision treatment and asynchronous telemedicine management of disease. Future work should refine and standardise AS-OCT imaging for ocular inflammation across different platforms and develop methods to improve feasibility of image acquisition in very young children, or those with media opacities such as band keratopathy. A longitudinal approach with collection of clinical data will be needed to allow clinical validation of imaging-based biomarkers as tools for diagnosis, disease monitoring and prediction of key

outcomes such as long term disease remission. OCT based imaging holds the promise of improved understanding of disease, improved outcomes, and improved patient experience.

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Figure 1. Image acquisition and analysis process

Details of image acquisition on the Heidelberg Spectralis OCT1 and the CASIA2 AS-OCT are provided in the supplemental document

Figure 2. Receiver operated characteristics for CASIA2 OCT detection of clinically active inflammation

ROC curves showing improvement in AUC following adjustment for 'cell' size in pixels

Figure 3. Bland Altman plots showing limits of agreement between repeated scans

Plots showing agreement between scans acquired using CASIA2 (A and B) and HS1 (C and D) OCTs with regression between the difference and the average used to alter the limits of agreement (B and D)

Large circle at 0 in image (D) represents multiple HS1 scans with 0 'cells' noted.

Figure 4. Correlation of clinical examination of disease activity with OCT imaging cell count

(A) Distribution of OCT image cell count across the different grades of inflammation, and (B) correlation between changes in CASIA2 cell count and changes in clinical grading across consecutive visits.

SUN: standardised uveitis nomenclature; ACC: anterior chamber cells

Figure 5. Distribution of 'cell' size on CASIA2 OCT imaging by diagnosis

CAU: chronic anterior uveitis; JIA: juvenile idiopathic arthritis; Symp AAU: symptomatic onset (acute) anterior uveitis

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