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Diagnosis of Ocular Surface Lesions Using Ultra-High Resolution Optical Coherence Tomography

Mohamed Abou Shousha, MD^{1,2,3}, Carol L. Karp, MD¹, Ana Paula Canto, MD¹, Kelly Hodson, MD¹, Patrick Oellers, MD¹, Andrew A. Kao, MD¹, Brett Bielory, MD¹, Jared Matthews, MD¹, Sander R. Dubovy, MD^{1,4}, Victor L. Perez, MD¹, and Jianhua Wang, MD, PhD¹

¹Bascom Palmer Eye Institute, University of Miami, Miller School of Medicine, Miami, FL

²Saint Louis University Eye Institute, Saint Louis University, Saint Louis, MO

³Department of Ophthalmology, Faculty of Medicine, University of Alexandria, Egypt

⁴Florida Lions Ocular Pathology Laboratory, Miami, FL, USA

Abstract

Purpose—To assess the use of ultra high resolution optical coherence tomography (UHR-OCT) in the diagnosis of ocular surface lesions.

Design—Prospective, non-comparative, interventional case series.

Participants—Fifty four eyes of 53 consecutive patients with biopsy proven ocular surface lesions; 8 primary acquired melanosis, 5 amelanotic melanoma, 2 nevi, 19 ocular surface squamous neoplasia, 1 histiocytosis, 6 conjunctival lymphoma, 2 conjunctival amyloidosis, and 11 pterygia.

Intervention—UHR-OCT imaging of the ocular surface lesions.

Main Outcome Measures—Clinical course and photographs, UHR-OCT image and histopathological findings.

Results—UHR-OCT images of all examined ocular surface lesions showed close correlation with the obtained histopathological specimens. When clinical differential diagnosis of ocular surface lesions was broad, UHR-OCT images provided optical signs that guided towards a more specific diagnosis and management. In cases of amelanotic melanoma, conjunctival amyloidosis, and primary histiocytosis and in one case of ocular surface squamous neoplasia, UHR-OCT was instrumental in guiding the diagnosis. In those cases, UHR-OCT suggested that the presumed clinical diagnosis was incorrect and favored a diagnosis which was later confirmed by histopathological examination.

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Correspondence and Reprints: Carol L. Karp, MD, Bascom Palmer Eye Institute, University of Miami Miller School of Medicine, 900 NW 17th Street, Miami, FL 33136, ckarp@med.miami.edu.

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Conclusions—Correlations between UHR-OCT and histopathology confirm that UHR-OCT is an adjunctive diagnostic modality that can provide a non-invasive means to help and guide diagnosis and management of ocular surface lesions.

Introduction

Diagnosis and differentiation of ocular surface lesions are of critical importance. While many lesions are benign, some can carry significant morbidity as well as risk of mortality.¹ Clinical examination and history frequently yield a correct diagnosis but in some instances only a broad differential diagnosis is possible. Adjunctive tests are then necessary to manage the patient. The gold standard is obtaining a biopsy, either incisional or excisional. In some cases an incisional biopsy is sufficient but in other cases the whole lesion should be excised such as cases when melanoma is suspected.² Biopsy is not without its limitations and complications. It is an invasive diagnostic technique and it can simply miss lesions that were not included in the excised tissue.³ New diagnostic techniques; namely impression cytology and confocal microscopy, have been proposed and have proven to be helpful.^{4;5} Nevertheless, these techniques have their own limitations that affect their usefulness. Impression cytology assesses only superficial layers of cells, which are not always representative of deeper layers, while confocal microscopy does not provide a cross-sectional view of the lesion and thus is not useful to determine the vertical and horizontal extent of the lesion.^{6;7} The limitations of these diagnostic techniques mandated the search for new reliable, more sensitive, non-invasive techniques to diagnosis ocular surface lesions.

Anterior segment optical coherence tomography (AS-OCT) is a relatively new imaging modality that has proven to be of assistance in the diagnosis and management of many ophthalmic pathologies.⁸ The utility of time-domain AS-OCT in the diagnosis of ocular surface lesions was explored and had successfully disclosed optical diagnostic signs for some ocular lesions.^{8;9} However, owing to their limited resolution, details of tissue planes are not always visible, and the use of AS-OCT in ocular surface tumors has remained of questionable usefulness. The introduction of spectral domain prototype with ultra high resolution down to 3 μm has promise to be an excellent adjunct in the management of ocular surface lesions.^{10–15} Shousha et al¹³ have reported the use of UHR-OCT in managing conjunctival and corneal intraepithelial neoplasia (CCIN). Kieval et al¹⁴ have also reported the utility of UHR-OCT in differentiation of pterygia from ocular surface squamous neoplasia (OSSN). Both reports have shown excellent correlation to histopathological examination demonstrating that UHR-OCT can provide valuable non-invasive images that can guide the diagnosis and management of ocular surface lesions such as pterygia and OSSN. Nevertheless, for this to become a reality, various other ocular surface lesions need to be studied as well using UHR-OCT in order to identify optical diagnostic signs specific for various pathologies and evaluate if UHR-OCT can differentiate the different pathological entities.

Reporting optical signs of various ocular surface lesions is critical in order to evaluate UHR-OCT in the diagnosis and management of ocular surface lesions. We herein describe a case series of biopsy proven ocular surface lesions that were imaged using UHR-OCT and images correlated to the clinical picture and histopathology. Our study disclosed optical diagnostic criteria of studied ocular surface lesions, which allowed for narrowing the differential diagnosis, ruling in or out the clinical diagnosis and in some cases arriving at a definite diagnosis.

Methods

This prospective case series included 75 eyes of 74 consecutive patients who presented with ocular surface lesions. All patients underwent comprehensive ophthalmic examination by a cornea specialist. After preliminary clinical diagnosis was made, imaging of the lesion using custom built UHR-OCT was performed. If deemed clinically indicated, incisional biopsies at the slit-lamp under topical anesthesia or excisional biopsy in the operating room under local anesthesia were done to confirm the diagnosis. Cases that did not receive a biopsy were excluded from this study. This resulted in 54 eyes of 53 consecutive patients for inclusion in this analysis. Only one individual (CLK) graded all the UHR-OCT images and was masked to the histopathological findings. UHR-OCT images were correlated with the clinical picture and with histopathology of the obtained specimens.

Approval was obtained from the University of Miami Institutional Review Board and written consents were obtained from all patients. The methods adhered to the tenets of the Declaration of Helsinki and were compliant with the Health Insurance Portability and Accountability Act (HIPAA).

Instrumentation

In our study, we used a custom-built prototype high speed and ultra-high resolution spectral domain optical coherence tomography (UHR-OCT) to image various ocular surface lesions. Detailed description of UHR-OCT is available in a previous report.¹¹ Briefly, a three-module superluminescent diode (SLD) light source (Broadlighter, T840-HP, Superlumdiodes Ltd, Moscow Russia) with a center wavelength of 840nm and a full width at half maximum bandwidth of 100 nm was used. The A-line (depth scan) rate of the OCT system was set to be 24 kHz. The calibrated axial resolution of the system is 3 μm in tissue (the refractive index is ~ 1.39).¹⁶

The UHR-OCT scanning beam position was monitored using an infrared camera and centered on the studied lesion. Horizontal or vertical 12-mm images of the studied lesion were acquired at a rate of 32 frames per scan to capture the conjunctival and corneal lesions. The scanning probe was tilted in order to bring the scanning beam as perpendicular as possible on the lesion for the best image and to decrease tangential sampling.

Results

Our study included 54 eyes of 53 patients with ocular surface lesions; 8 primary acquired melanosis, 5 amelanotic melanoma, 2 nevi, 19 ocular surface squamous neoplasia, 1 histiocytosis, 6 conjunctival lymphoma, 2 conjunctival amyloidosis and 11 pterygia. Summary of UHR-OCT optical signs for each lesion are detailed in Table 1. For comparison, an UHR-OCT scan through the cornea and conjunctiva of a normal subject is demonstrated in Figure 1A.

Epibulbar Melanocytic Lesions

Conjunctival Primary Acquired Melanosis—Eight patients presented with acquired pigmented conjunctival and limbal lesions. On slit lamp examination (SLE), flat patchy acquired pigmented lesions were noted. (Fig 2A) A preliminary clinical diagnosis of primary acquired melanosis (PAM) was made. Prior to obtaining excisional biopsies, UHR-OCT images of the lesions were obtained. UHR-OCT images (Fig 2B) demonstrated normal thickness epithelium that was moderately hyper-reflective (Fig 2B-a) with strong hyper-reflectivity of basal epithelium (Fig 2B-b). This hyper-reflectivity was noted to be regular and seemed to be pronounced in the basal layer of the epithelium with no invasion towards

the subepithelial tissue layers. Interestingly, histopathological examination of the specimens (Fig 2C) indicated a high degree of correlation as it disclosed that conjunctiva contained melanocytes located within the basal epithelium with no invasion of underlying tissue; confirming the diagnosis of PAM. It is important to note that it was not possible to differentiate cases with atypia from cases with no atypia using the UHR-OCT.

Amelanotic Melanoma—Five patients were referred for evaluation of ocular surface squamous neoplasia (OSSN). SLE demonstrated raised, non-pigmented conjunctival lesions that were suggestive of OSSN. (Fig 3A) UHR-OCT images were obtained (Fig 3B) and interestingly it disclosed that lesions were present in the subepithelial tissue (Fig 3B-a) while epithelium was of normal thickness (Fig 3B-b) and thus clearly ruling out OSSN. The lesions did cast a significant shadow (Fig 3B-c). In addition, we noted epithelial cleavage which indicates some involvement of the epithelium with atypical melanocytes. UHR-OCT images thus ruled out OSSN and moved amelanotic melanoma higher on the differential diagnosis list, thus warranting excisional biopsies rather than incisional biopsies as the next step in the management plan to avoid possible melanoma seeding. Histopathological examination of specimens (Fig 3C) revealed the diagnosis of melanoma by disclosing that the lesions, which stained positive for Melan A, were composed of atypical melanocytic cells that were present within the epithelium and substantia propria.

Nevi—Two patients presented with pigmented conjunctival and limbal lesions (Fig 4A) that were clinically diagnosed as nevi. They underwent an excisional biopsy as one of them had family history of melanoma and the other had a history of recent growth. UHR-OCT images (Fig 4B) showed that lesion in one case was in the epithelium (Fig 4B-a) and substantia propria (Fig 4B-b), representing a compound nevus (Fig 4B-b). Other lesions were confined to the substantia propria without involvement of the epithelium. Thus, UHR-OCT images seem to be able to demonstrate which tissue layers are involved by nevus cells (compound versus subepithelial nevus). Lesions were highly hyper-reflective and the UHR-OCT was usually able to show that the lesion was well circumscribed. (Fig 4B-c) Cysts were noted on all lesions within the subepithelial tissue (Fig 4B-d). In one of the two cases the shadow casted by the pigmented lesion was intense so that it obscured the posterior border of the lesion. Histopathological examination of the biopsied lesions disclosed conjunctival tissue that contained nests of nevus cells with only minimal Ki76 staining and epithelial lined cysts confirming the diagnosis of conjunctival nevus. (Fig 4C)

Ocular Surface Squamous Neoplasia

Nineteen patients, presented with acquired corneal-conjunctival lesions. (Fig 5A) In all cases, the UHR-OCT images (Fig 4B) disclosed lesions that were located in the epithelium (Fig 5B-a). Images demonstrated a severely thickened, hyper-reflective epithelium (Fig 5B-a) with an abrupt transition between the normal and affected epithelium. (Fig 5B-b) A plane of cleavage between the lesion and underlying tissue was noted in most cases. (Fig 5B-c) In large lesions a shadow from the hyper-reflective epithelium sometimes obscured that plane of cleavage. Histopathological examination of all obtained specimens disclosed acanthotic conjunctival and/or corneal epithelium with faulty maturational sequencing extending either partial or full thickness (Fig 5C) without invasion to the underlying tissue confirming the diagnosis of conjunctival or corneal intraepithelial neoplasia (CCIN) and demonstrating excellent correlations to UHR-OCT images. Interestingly, the case in Figure 5 was referred as a conjunctival melanoma as the lesion was pigmented and slightly raised. The UHR-OCT image clearly disclosed that the epithelium was thickened and hyper-reflective and strongly pointed towards OSSN instead of melanoma. Histopathological examination of this case confirmed the diagnosis of pigmented CCIN. (Fig 5C)

Isolated Conjunctival Histiocytosis

A 56-year-old female patient presented with an elevated gelatinous corneal-conjunctival lesion that had tufts of blood vessels. (Fig 6A) Based on the clinical picture a diagnosis of CCIN was made and the patient had been treated with topical mitomycin-C. The lesion was not responding and the patient was referred for a second opinion. UHR-OCT images (Fig 6B) were not consistent with OSSN. UHR-OCT image disclosed that epithelial layer was uninvolved (Fig 6B-a) and that the lesion (Fig 6B-b) was confined to the substantia propria and was well circumscribed. (Fig 6B-c) The body of the lesion appeared to be stippled by small hyper-reflective dots that were suggestive of an infiltration of a homogenous cell population. (Fig 6B-b) Round and oval hypo-reflective areas were also noted and were suggestive of blood vessels within the lesion. (Fig 6B-d) Histopathological examination of the specimen confirmed that the lesion was conjunctival histiocytosis; not OSSN (Fig 6C) and demonstrated close correlations to UHR-OCT images.

Conjunctival Lymphoma

Six patients presented with pink conjunctival lesions that were clinically suspicious for lymphoma. (Fig 7A) UHR-OCT images (Fig 7B) showed a normal epithelium (Fig 7B-a) and a lesion in the substantia propria. (Fig 7B-b) The subepithelial lesions were overall quite homogenous, hypo-reflective and dark, but seemed to be formed of stippled hyper-reflective dots. Some shadow was casted by the mass on the underlying tissue. Histopathological examination, along with flow cytometry and gene rearrangement of specimens obtained from the 6 lesions (Fig 7C), disclosed a monoclonal lymphoid infiltrate located within the substantia propria confirming the diagnosis of extranodal marginal zone lymphoma. UHR-OCT images showed excellent correlations with histopathological specimens. Interestingly, one patient with conjunctival lymphoma (8A-C) was managed with a 20 day course of external beam radiation therapy (8D), and follow up UHR-OCT (8E) disclosed resolution of the lymphoma by demonstrating unremarkable tissue architecture and disappearance of the lesion.

Conjunctival Amyloidosis

Two patients presented with yellowish pink conjunctival lesions that were clinically diagnosed as conjunctival lymphoma. (Fig 9A) UHR-OCT images (Fig 9B) showed normal epithelium (Fig 9B-a) and a lesion (Fig 9B-b) present in the substantia propria. Lesions were well demarcated from the underlying tissue. Interestingly, UHR-OCT images were different from those of lymphoma. In these cases, no cellular-like dots were noted, and the area of the lesion had both hypo-reflective areas with interspaced areas/lines of hyper-reflectivity. Thus, UHR-OCT images questioned the presumptive diagnosis of conjunctival lymphoma. Histopathological examination of specimens disclosed conjunctiva that contained a moderate amount of paucicellular material located within the substantia propria that was PAS, congo-red positive and demonstrated apple-green birefringence with polarized light (Fig 9C). Gene rearrangement and flow cytometry studies were negative, confirming a diagnosis of conjunctival amyloidosis and excluding lymphoma.

Pterygia

Eleven eyes presented with pterygia. UHR-OCT images of those lesions revealed a normal thickness epithelium with moderate hyper-reflectivity. (Fig 10B-a) The lesions seen on UHR-OCT demonstrated a highly hyper-reflective fibrillary subepithelial demarcated layer (Fig 10B-b). Lesions were seen to grow over the cornea and remain between corneal epithelium and Bowman's layer. Histopathological examination (10C) demonstrated an unremarkable epithelium and solar elastosis in the substantia propria confirming the diagnosis of pterygium, and providing excellent correlation to the UHR-OCT images.

Discussion

Our study characterizes various optical signs of ocular surface lesions and discloses excellent correlations with histopathological examination of all studied lesions. This highlights the utility of UHR-OCT as an adjunctive diagnostic modality that can provide a non-invasive means to guide diagnosis of ocular surface lesions. It was possible to determine the exact location of lesions in relation to surface epithelium and this readily allowed for the differentiation between OSSN and lesions of the substantia propria or stroma. Moreover, UHR-OCT demonstrated optical signs that provided a clue to the diagnosis with remarkable correlations to the histopathological examination. At times, the presumed clinical diagnosis was questioned by UHR-OCT evaluation as seen in our amelanotic melanoma cases, conjunctival amyloidosis, and primary histiocytosis and in one case of ocular surface squamous neoplasia.

Our five cases of amelanotic melanoma were presumed to be OSSN clinically, but the UHR-OCT revealed that processes were subepithelial rejecting that clinical diagnosis. Moreover, it revealed specific signs that were consistent in the 5 cases namely the epithelial cleavage indicating some involvement of the epithelium with atypical melanocytes, and the presence of a subepithelial lesion. Incisional biopsy was not performed for our amelanotic melanoma cases, to avoid any possible risk of seeding of melanoma cells.^{17;18} While we expected perhaps some thickening of the epithelium, this was not found in these our cases.

In our ocular histiocytosis case, the clinical picture was very suspicious for OSSN. The patient was referred to our service after being started on topical mitomycin C for a presumed OSSN. Obtained UHR-OCT images were striking as they clearly indicated a normal thickness epithelium overlying a well confined subepithelial lesion, findings inconsistent with OSSN UHR-OCT findings. Biopsy was performed, and histopathological specimen showed close correlation to the UHR-OCT. In this case, UHR-OCT was critical in ruling out the diagnosis of OSSN and saved the patient of further inappropriate treatment with topical chemotherapy.

The role of UHR-OCT in guiding the clinical diagnosis was also highlighted in one of our OSSN cases, which was referred for conjunctival melanoma. The UHR-OCT suggested that the lesion was confined to the epithelium with classic optical signs for OSSN ruling out melanoma.

In cases of PAM, UHR-OCT disclosed that lesions were confined to the basal layer of the epithelium and showed optical signs that morphologically correlated with histopathological examination of obtained specimens, but details of atypia were not visualized at this time.

Shields et al¹⁹ had reported the use of AS-OCT, with 18 μm in the diagnosis of nevi, which were visualized as circumscribed lesions often with subepithelial cysts. Our study confirms their results and reveals that with an even higher resolution OCT, tissue planes will be better delineated, as seen in our case of a compound nevus. This may help in the future to study the natural evolution of nevi and address questions about the risk of development into melanoma.

The unique capabilities of UHR-OCT to *in vivo* demonstrate histological architecture non-invasively with 3 μm resolution allowed for ruling in or out OSSN. The presence of normal epithelium overlying a subepithelial lesion confidently rules out OSSN. The presence of a hyper-reflective thickened epithelium was a consistent specific optical sign for OSSN, which was demonstrated in all of our OSSN cases with excellent histopathological correlation. One pigmented conjunctival lesion was referred as a melanoma, but the UHR-OCT image clearly demonstrated the specific signs of CCIN including thickened hyper-

reflective epithelium. This was confirmed by histopathology. Our results confirm that the optical signs of OSSN reported by our previous studies (Shousha et al¹³ and Kieval et al¹⁴) are consistent.

In cases of conjunctival lymphoma, UHR-OCT demonstrated interesting optical signs that were consistent in all 6 patients. UHR-OCT image showed dark homogeneous lesions with excellent correlation to the histopathological examination. The stippled dots seen on the UHR-OCT likely represent the infiltrates of the lymphocytes. The UHR-OCT was also able to demonstrate the resolution of a conjunctival lymphoma after a 20 days course of external beam radiation. This demonstrates that UHR-OCT may be a valuable tool to monitor and guide treatment of ocular surface tumors in a novel and non-invasive fashion.

The importance of identifying the aforementioned optical signs of conjunctival lymphoma was highlighted when UHR-OCT images of conjunctival amyloidosis were captured. The two conjunctival amyloidosis cases had been assumed to be conjunctival lymphoma. With the knowledge of optical signs of lymphoma by UHR-OCT seen in our 6 biopsy proven conjunctival lymphoma cases, UHR-OCT here was inhomogeneous and clearly different. This led us to doubt the presumed clinical diagnosis. In histopathological examination confirmed the diagnosis of conjunctival amyloidosis and showed excellent correlation with the UHR-OCT images.

Kieval et al¹⁴ have reported UHR-OCT optical signs of pterygia and demonstrated close correlations with histopathological examination. UHR-OCT of OSSN demonstrates a thickened hyper-reflective epithelium while pterygia have a fibrillary lesion underneath an unremarkable epithelium. There is a great utility of UHR-OCT in differentiating pterygia from OSSN clinically and the optical signs are very specific and sensitive.¹³ Our study is not only confirming these results but also highlighting the larger picture of the use of this novel non-invasive technique in diagnosis of other ocular surface lesions.

Our study is not without its limitations. It seems that thick pigmented lesions tend to impede the penetration of light to deeper tissue and thus limiting the depth of our images. In such cases, it was sometimes difficult to determine the posterior limit of the lesion. Despite that resolution would be adequate to image single cells, optical information at this time is not sufficient to study signs for cellular atypia such as atypical nuclei or prominent nucleoli. While the UHR-OCT was not able to define atypia, it ruled out gross involvement of the substantia propria in melanocytic lesions, favoring PAM over melanoma. Future studies to report optical signs of atypia and the intralesional characteristics of ocular surface lesions are needed, and is likely to improve as this technology advances.

It is important to note that commercially available AS-OCT such as time domain AS-OCT have an axial resolution of 18 μ m and thus are not able to differentiate the epithelium from the substantia propria and unable to display the specific signs disclosed by our custom made UHR-OCT. New spectral domain OCT machines have now made it to the market which have resolution closer to our UHR-OCT, and may potentially be able to reproduce and confirm our results. A future study using the new high resolution commercially available spectral domain units would be helpful.

Diagnosis of ocular surface lesions can be challenging. While UHR-OCT is not a substitute for histopathological specimens, it can be a valuable adjuvant diagnostic tool. Our study found that UHR-OCT images correlated remarkably to histopathology in all studied lesions. This novel, non-invasive diagnostic technique can reveal the structure and location of the lesion and aid in guiding the diagnosis and management. This was especially helpful in the cases of amelanotic melanoma and pigmented OSSN, and to different degrees in all ocular surface lesions studied. Details of the histological tissue architecture are visible with UHR-

OCT in a non-invasive fashion, highlighting its role as an adjunctive technique for the diagnosis and management of ocular surface lesions. Reporting and understanding the optical signs that may be specific for various ocular surface lesions are crucial to set the stage for the continued evolution of UHR-OCT for anterior segment lesions. Future studies are warranted to better understand the role of UHR-OCT in the management of ocular surface lesions.

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Figure 1.

Ultra high resolution optical coherence tomography image of the limbus of a normal subject. Normal corneal epithelium (1A-a) appears hypo-reflective. Note Bowman's layer (1A-b), corneal stroma (1A-c) and Descemet's membrane (1A-d). Conjunctival epithelium appears hypo-reflective (1A-e). Note the underlying substantia propria (1A-f).

Figure 2.

Slit lamp photograph (2A) of primary acquired melanosis (PAM). Ultra high resolution optical coherence tomography (UHR-OCT) image (2B) in the area of the black dotted arrow disclosed normal thickness epithelium (2B-a) with regular, intense hyper-reflectivity of the basal epithelial layer (2B-b). No invasion of the underlying tissue was seen. Histopathology (2C) confirmed the diagnosis of PAM with moderate atypia (stain, hematoxylin–eosin; original magnification, X200).

Figure 3.

Slit lamp photograph (3A) of a raised, reddish, non-pigmented conjunctival lesion referred as a possible squamous neoplasia. Ultra high resolution optical coherence tomography (3B) in the area of the black dotted arrow disclosed a subepithelial lesion (3B-a) beneath a normal thickness epithelium (3B-b). The lesion was casting a significant shadow (3B-c) on the underlying tissue. Foci of epithelial cleavage (Fig 3B-d) and moderate hyper-reflectivity of the epithelium were noted. Histopathologic examination (3C) confirmed the diagnosis of melanoma. (stain, hematoxylin–eosin; original magnification, X200).

Figure 4.

Slit lamp photograph (4A) of a pigmented conjunctival lesion (4A). Ultra high resolution optical coherence tomography (4B) in the area of the black dotted arrow disclosed a well circumscribed hyper-reflective lesion in the epithelium (4B-a) and substantia propria (4B-b) with shadowing (4B-c). Cystic structures were noted (4B-d). Histopathologic examination (4C) revealed nevus cells within the epithelium and substantia propria. Epithelial lined cysts were present in the superficial substantia propria (4C-a). Pathologic diagnosis confirmed a compound cystic nevus. (stain, hematoxylin–eosin; original magnification, X100).

Figure 5.

Slit lamp photograph (5A) of a gelatinous pigmented conjunctival lesion referred as a possible melanoma. Ultra high resolution optical coherence tomography image (5B) in the area of the black dotted arrow revealed severely thickened hyper-reflective epithelium (5B-a). Abrupt transition between unremarkable and thickened hyper-reflective epithelium was noted (5B-b) as well as a plane of cleavage between the lesion and the underlying tissue (5B-c), all suggestive of an ocular surface squamous neoplasia (OSSN). Histopathological examination of the excised specimen (5C) confirmed the diagnosis of OSSN (stain, hematoxylin–eosin; original magnification, X200).

Figure 6.

Slit lamp photograph (6A) of an elevated gelatinous corneal-conjunctival lesion with tufts of blood vessels. Ultra high resolution optical coherence tomography (UHR-OCT) image (6B) in the area of the black dotted arrow disclosed an unremarkable epithelium (6B-a) and a lesion (6B-b) confined to the subepithelial layer (6B-c). The mass was composed of small dots; which may represent cellular infiltration (6B-b). Note oval hypo-reflective areas that represent blood vessels (6B-c). Histopathologic examination of the excised specimen (6C) confirmed UHR-OCT findings of cellular infiltration, in this case of CD68 positive histiocytes (6C-a) underneath an intact appearing epithelium (6C-b), suggesting a diagnosis of conjunctival histiocytosis. (stain, hematoxylin–eosin; original magnification, X200).

Figure 7.

Slit lamp photograph (7A) of a salmon pink conjunctival lesion. Ultra high resolution optical coherence tomography (7B) indicates a normal epithelium (7B-a) and a lesion in the substantia propria (7B-b) formed of stippled dots. The lesion was hyporeflexive and homogeneous in nature. Histopathologic examination (7C) discloses a monomorphous population of small to moderate sized blue cells located within the substantia propria. Flow cytometry and gene rearrangement results confirmed the diagnosis of extranodal marginal zone lymphoma. (stain, hematoxylin–eosin; original magnification, X200).

Figure 8.

Slit lamp photograph (8A) and ultra high resolution optical coherence tomography (UHR-OCT) in the area of the black dotted arrow demonstrate conjunctival lymphoma, a lesion that was formed by small cellular like dots (8B-a) on UHR-OCT. Histopathologic examination (8C) along with flow cytometry and gene rearrangement confirmed the diagnosis of extranodal marginal zone lymphoma. The lesion resolved after a 20 days course of external beam radiation (8D). A follow up UHR-OCT image (8E) confirmed resolution. (stain, hematoxylin–eosin; original magnification, X100).

Figure 9.

Slit lamp photograph (9A) of a yellow-pink conjunctival lesion that was thought to be conjunctival lymphoma. Ultra high resolution optical coherence tomography in area of black dotted arrow (9B) disclosed a normal epithelium (9B-a) and hypo-reflective lesion (9B-b) present diffusely in the substantia propria intermixed with hyper-reflective linear areas (9B-c). Histopathological examination (9C) revealed paucicellular material located diffusely within the substantia propria that was PAS, congo-red positive and demonstrates an apple green-birefringence (9C) with polarized light. Flow cytometry and gene studies were negative. A diagnosis of conjunctival amyloidosis was made. (stain, hematoxylin–eosin; original magnification, X100).

Figure 10.

Slit lamp photograph (10A) demonstrates a pterygium. Ultra high resolution optical coherence tomography (UHR-OCT) in the area of the black dotted arrow disclosed a moderately hyper-reflective epithelium (10B-a) and a highly hyper-reflective fibrillary subepithelial lesion (10B-b). Hypo-reflective areas may correspond to blood vessels (10B-c). Histopathological examination (10C) demonstrated an unremarkable epithelium with solar elastosis within the substantia propria confirming the diagnosis of pterygium (stain, hematoxylin–eosin; original magnification, X200).

Table 1
Summary of ultra high resolution optical coherence tomography optical signs of ocular surface lesions

	Epithelial layer		Subepithelial layers
	Thickness	Reflectivity	
PAM	Normal	Moderately hyper-reflective with strong regular hyper-reflectivity of basal epithelium	No invasion in the sub-epithelial tissue layers
Amelanotic Melanoma	Normal or mildly thickened	Epithelial cleavage which indicates some involvement of the epithelium with atypical melanocytes.	Severely hyper-reflective sup-epithelial lesion Significant shadow obscuring the end point of the lesion
Nevi	Normal or mildly thickened	In compound nevi, lesion is seen in the epithelium and substantia propria.	Well circumscribed highly hyper-reflective lesion Confined to the substantia propria in subepithelial nevi Cysts within the subepithelial tissue Shadow casted may sometimes obscure the posterior border of the lesion
OSSN	Severely thickened	Hyper-reflective epithelium Abrupt transition between normal and affected epithelium. Cleavage plane between the lesion and underlying tissue may be seen in some lesions In large lesions a shadow from the hyper-reflective epithelium may obscure inferior border	Uninvolved in CCIN;
Conjunctival Histiocytosis	Normal	Normal	Well circumscribed lesion Stippled by small hyper-reflective dots suggestive of an infiltration Intralesional round and oval hypo-reflective areas suggestive of blood vessels.
Conjunctival Lymphoma	Normal	Normal	Homogenous, hypo-reflective and dark lesion Seemed to be formed of small stippled hyper-reflective dots suggestive of a homogenous cell population. Some shadow is casted by the mass on the underlying tissue.
Conjunctival Amyloidosis	Normal	Normal	Well demarcated from the underlying tissue. Lesion has both hypo-reflective areas with interspaced areas/lines of hyper-reflectivity. Inhomogeneous.
Pterygia	Normal	Moderate hyper-reflectivity or normal	Highly hyper-reflective fibrillary sub-epithelial demarcated layer under the conjunctival epithelium and present between the corneal epithelium and Bowman's layer.

PAM: Conjunctival Primary Acquired Melanosis CCIN: Corneal and conjunctival intraepithelial neoplasia

OSSN: Ocular Surface Squamous Neoplasia