Visualising Middle Ear Biofilms in Otitis Media: a new benchmark for successful treatment

Stephen A Boppart, MD, PhD,
Bliss Professor of Engineering,
Director, Imaging at Illinois

Correspondence
Departments of Electrical and Computer Engineering, Bioengineering, and Medicine,
Beckman Institute for Advanced Science and Technology,
405 North Mathews Avenue,
Urbana, IL 61801, USA.
E: boppart@illinois.edu

Declaration of Competing Interests
SAB receives royalties from patents licensed by the Massachusetts Institute of Technology related to optical coherence tomography. He also conducts research sponsored by Welch Allyn, Inc. He is co-founder and Chief Medical Officer of Diagnostic Photonics, Inc., which is developing interferometric synthetic aperture microscopy for surgical guidance.

The accurate diagnosis of Otitis Media is one of the greatest challenges in primary care medicine, pediatrics, and otology. For this most common disease in children, the costs associated with patient discomfort and pain, parental care and missed work, repeated office visits and antibiotic treatments, and often surgery, are all high. While most expert physicians can differentiate subtle changes in the tympanic membrane and middle ear effusions via standard and pneumatic otoscopy, currently there is limited ability to visualise microscopic changes within the middle ear to indicate the presence of biofilms, and limited technologies that can quantitatively or algorithmically determine the state of the disease process to help in the diagnostic and treatment decision making process. Recently, a new optical biomedical imaging technology called optical coherence tomography (OCT)[1,2] has been developed and integrated with handheld scanners for primary care imaging (Figure 1)[3,4], functioning much like a standard otoscope or ophthalmoscope, but with the ability to acquire digital 3D images of tissue structures in real-time at micron-scale resolution and up to several millimetres deep, even through highly scattering tissues such as the tympanic membrane, turbid effusions, or skin.

We are discovering that biofilms are playing an increasingly important role in infectious disease. These thick, adherent, glue-like films formed by polymicrobial communities of bacteria act as protective microenvironments where bacteria can reside and colonise. Antibiotic treatments poorly eradicate the bacteria present within these films, leading to increased antibiotic resistance while providing a reservoir for re-seeding the recurrent infections seen in chronic Otitis Media. In 1996, a seminal paper established a clear link between the presence of middle ear biofilms and chronic Otitis Media in children [5]. This invasive study sampled middle ear mucosa from patients with chronic Otitis Media undergoing tympanostomy tube placement, as well as from control patients undergoing procedures to place cochlear implants. Based on this study, it was evident that the presence of a middle-ear biofilm in chronic Otitis Media could significantly impact the expected outcome of standard antibiotic treatments. It was also realised that if new non-invasive technology was available to detect the presence of a biofilm, characterise its features and dynamics, and monitor

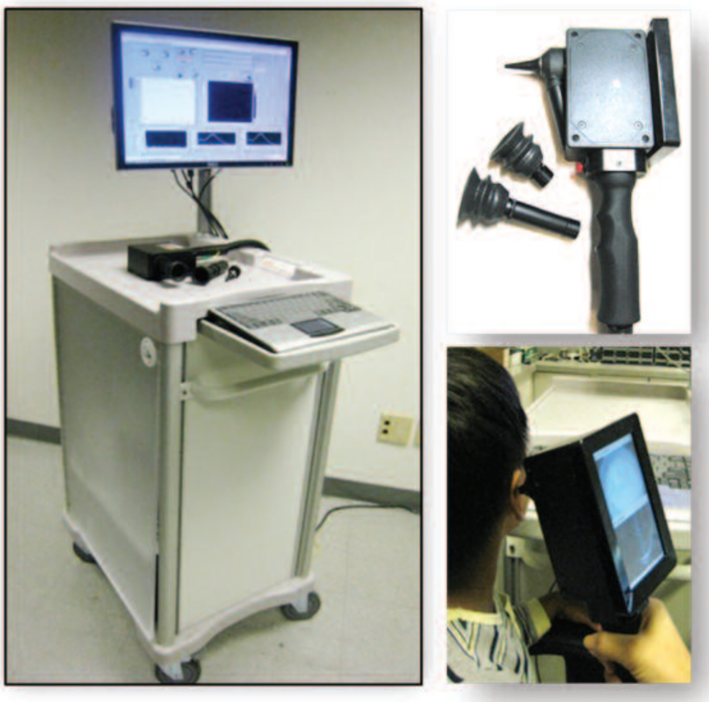

Figure 1: Primary care imaging system is contained in a portable cart along with a handheld scanner. The scanner has interchangeable tips for imaging the ear, eye, oral mucosa, and skin. The handheld scanner also has a video imaging system and display panel to show both the surface video image and the cross-sectional OCT image of the tissue, as shown while imaging the ear.

the middle ear for its progression or regression during antibiotic treatment, it would be possible to not only improve our prescription regimens, but also better understand the fundamental etiology and dynamic responses of these biofilms to our treatments. The detection and quantification of these biofilms could possibly become a new benchmark for treatment of chronic Otitis Media, with the end-goal being the complete eradication of the biofilm.

Optical coherence tomography is a non-invasive, high-resolution, real-time biomedical imaging technology that was developed in the early 1990s and has become the gold-standard for imaging the transparent and translucent structures of the human retina. OCT is also actively being developed for human coronary artery imaging to assess vulnerable plaques, and in oncology for intra-operative surgical guidance and the microscopic assessment of tumour margins and lymph nodes, enabling real-time optical biopsies of tissue rather than waiting for postoperative histological findings. OCT is the optical analogue to ultrasound imaging, except reflections of near-infrared light (~800-1,300nm) are detected rather than sound waves. The use of light waves instead of sound waves means that imaging resolutions can be 10-100 times higher, resulting in images that closely resemble histology, but are captured non-invasively without the need to physically resect tissue for processing, and without the need to use stains or dyes to identify features. Instead, OCT images are based on the inherent variations in the tissue optical indices of refraction, and are depth-resolved, up to several millimetres deep in highly-scattering tissues, much like a cross-sectional tissue section in histology. Because OCT performs depth-resolved optical ranging into tissue and detects only returning photons that remain coherent or correlated to photons in a reference-arm path, OCT can reject photons that would normally only contribute to noise, enabling this larger imaging penetration depth, and far-exceeding the depths achievable by light microscopy or confocal microscopy.

Several years ago, OCT was first shown to produce high-resolution 3D structural images of biofilms grown under laboratory conditions [6]. Subsequent *in vivo* imaging studies were performed in a rat model for Otitis Media [7], which showed excellent correlations between acquired OCT images and histological findings of biofilms. To enable human studies, OCT was integrated with a handheld scanner with interchangeable tips to image multiple tissue sites commonly examined in primary care medicine [3]. This system was then applied to the challenge of non-invasively detecting biofilms in the human middle ear (Figure 2)[4]. A total of 20 adult patients with clinically-diagnosed chronic Otitis Media along with normal human volunteers were imaged with OCT. In all patients with chronic Otitis Media, OCT was able to not only detect the presence of a biofilm, but also characterise its structural properties. No evidence of biofilms was evident in the normal control subjects. This study demonstrated the potential for using OCT to non-invasively detect and quantitatively monitor the progression or regression of biofilms during antibiotic treatment regimens, with the potential for more rapidly identifying which treatments are effective, and which should be stopped or changed.

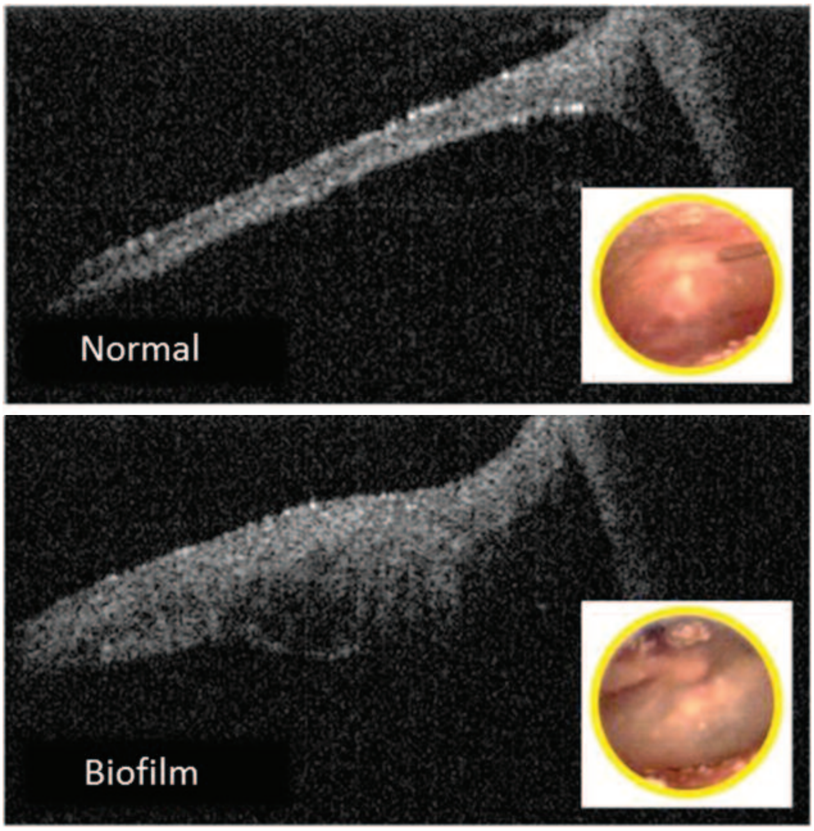

Figure 2: OCT and video images of a normal tympanic membrane (top) and one with a middle-ear biofilm (bottom). The biofilm appears as a highly-scattering structure adherent to the inner surface of the tympanic membrane. Three-dimensional OCT imaging of biofilm structure is possible.

The use of OCT also offers a new opportunity to better understand the dynamics of biofilms *in vivo* in our patients, and studies are underway to quantify temporal changes, to link biofilm structural features to specific bacterial species, and to evaluate the use of OCT in other ear pathologies including differences between acute and chronic Otitis Media. Technological advances such as OCT are likely to provide physicians and health care providers with a wealth of new information, data, and indicators which will enable better diagnostic and monitoring capabilities, and ultimately, improved patient care for one of the most common yet most challenging diseases of the ear.

Web: http://biophotonics.illinois.edu

References

1. Drexler W, Fujimoto JG. Optical Coherence Tomography: Technology and Applications. Springer; New York: 2008.
2. Zysk AM, Nguyen FT, Oldenburg AL, Marks DL, Boppart SA. Optical coherence tomography: a review of clinical development from bench to bedside. *J Biomedical Optics* 2007;**12(5)**:051403.
3. Jung W, Kim J, Jeon M, Chaney EJ, Stewart CN, Boppart SA. Handheld optical coherence tomography scanner for primary care diagnostics. *IEEE Trans Biomed Eng* 2011;**58(3)**:741-4.
4. Nguyen CT, Jung W, Kim J, Chaney EJ, Novak M, Stewart CN, Boppart SA. Noninvasive in vivo optical detection of biofilm in the human middle ear. *Proc Natl Acad Sci USA* 2012;**109(24)**:9529-34.
5. Hall-Stoodley L, Hu FZ, Gieseke A, Nistico L, Nguyen D, Hayes J, Forbes M, Greenberg DP, Dice B, Burrows A, Wackym PA, Stoodley P, Post JC, Ehrlich GD, Kerschner JE. Direct detection of bacterial biofilms on the middle-ear mucosa of children with chronic otitis media. *JAMA* 2006;**296(2)**:202-11.
6. Xi C, Marks DL, Schlachter S, Luo W, Boppart SA. High-resolution three-dimensional imaging of biofilm development using optical coherence tomography. *J Biomed Opt* 2006;**11(3)**:34001.
7. Chaney EJ, Nguyen CT, Boppart SA. Novel method for non-invasive induction of a middle-ear biofilm in the rat. *Vaccine* 2011;**29(8)**:1628-33.