# Chemical Imaging in Middle Ear Pathology: Quo Vadis?

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**Abstract**
Accurate diagnosis of most middle ear pathological conditions including otitis media remains challenging in a clinical setting. Standard ear evaluations are still performed primarily on the basis of human recognition of morphologic patterns *in vivo* using white light otoscope and suffer from significant observer variability providing minimal understanding of a disease's underlying biochemistry. Owing to the biochemical specificity and multiplexing capability, spectroscopy-based chemical imaging has emerged as a promising tool for middle ear disease diagnosis in a real-time, label-free and non-destructive manner. Here, we critically review the spectroscopy-based approaches employed to study middle ear pathological conditions.

**Keywords:** middle ear, otitis media, light otoscope, spectroscopy, middle ear pathology

## Introduction

Middle ear infection -also known as otitis media (OM)- is the most common childhood illness; and accounts one third of all pediatric visits in the United States. Otitis Media is responsible for over 25 million office visits annually with direct costs for treatment estimated at $3 billion. At present, both primary care physicians and specialists typically perform otitis media diagnosis on the basis of visual inspection of ear drum (tympanic membrane) using an otoscope that has remained virtually unchanged for over a century. While a translucent with pale grayish appearance of tympanic membrane is usually a sign of normal condition, an opaque red/yellow and bulging may be an indication of infectious condition. Clearly, the above diagnostic approach suffers from significant observer variability and the lack and observer-invariant diagnosis leads to erratic decision thereby affecting therapeutic intervention. Further, some of the middle ear conditions like chronic OM do not have noticeable symptoms, and standard otoscopy-based diagnosis is fairly inaccurate with 74% sensitivity and 60% specificity (1, 2). Few adjuvant methods for instance tympanometry and pneumatic otoscopy have been developed to aid the standard otoscopy towards objective diagnosis but these methods can only yield limited improvement in accuracy and precision(1). It is important to note that even these methods contain subjective elements and significantly depend on the examiners expertise. Most importantly these devices are often not readily available in many primary care practices. Currently, a combination of pneumatic otoscopy and tympanometry is recommended for chronic OM diagnosis, if necessary. Misdiagnosis in Acute Otitis Media (AOM) may result in severe intracranial complications, such as meningitis, brain abscesses and hearing loss. This can lead to a significant decrease in the quality of patient's life in both the near and long term. Therefore, it is imperative to develop new reliable tools that can provide non-invasive and real-time middle ear diagnosis without necessitating human interpretation.

There is another aspect of diagnostic

uncertainty in OM management. OM is one of the most common pediatric infections for which antibiotics are prescribed. Due to lack of objective diagnosis many patients are being over-treated under the worst-case assumption and this poses great clinical concern over emerging antibiotic resistance of the common pathogens that cause middle ear infection. There is an ongoing debate regarding the appropriate prescriptions of antibiotics in the treatment of middle ear infection, indicating a convincing unmet need for additional quantitative data or biomarkers information that can aid a physician to improve the diagnostic accuracy.

Exploiting the endogenous molecular contrast may provide sufficient information to aid in obtaining objective and reliable disease diagnoses and guidance for correct treatment. The definition of the middle ear pathology in chemical terms provides objective biomarkers for diagnoses and may permit disease detection prior to morphologic manifestations. In this milieu, spectroscopic investigations offer a novel approach to achieve an unmet need. Molecular spectroscopy in particular has emerged as an intriguing possibility owing to its molecular specificity and multiplexing capability.

**Spectroscopic based approaches**

The area of middle ear pathology is largely unexplored spectroscopically. Sundberg et al. have employed diffuse reflectance spectroscopy to record reflectance spectrum from human tympanic membrane and demonstrated its applicability in otitis media diagnostics (3). Recently, Zhang et al. have combined reflectance spectroscopy and gas in scattering media absorption spectroscopy and performed experiment on ear phantom; and suggested the potential of combined spectroscopic approach in improving the diagnostic accuracy of middle ear infections (4). Raman spectroscopy provides molecular fingerprinting information and Raman spectrum is sensitive to the substitution (5) and even to the chemical environments (6). We have most recently employed Raman spectroscopy to differentiate between cholesteatoma and another middle ear lesion, myringosclerosis, by analyzing the spectral patterns of differentially expressed molecules (7). Differentiation of these two lesions is particularly challenging using white light otoscopy due to the similarity in visual appearance. In addition to revealing signatures consistent with the known pathobiology of these middle ear lesions, we observed the first evidence of the presence of carbonate- and silicate-substitutions in the calcium phosphate plaques found in myringosclerosis. The scope of this mini-review is limited to emerging chemical imaging platforms used in middle ear disease diagnosis.

**Imaging Based approaches**

On the photonic imaging front, in a pioneering work, Boppart's group has employed optical coherence tomography (OCT) to successfully image the biofilm in human patients (8, 9). A compact structured light based three-dimensional imaging of the tympanic membrane was recently proposed (10). Although these approaches could aid OM diagnosis by providing better morphological information, they lack molecular information.

In order to exploit the endogenous biochemical contrast that may provide sufficient information to aid the process of middle ear disease diagnosis we had recently employed auto-fluorescence imaging in the middle ear diagnosis (11, 12). We have built a multi-wavelength, video-rate "fluoro-otoscope" – an otoscope with auto-fluorescence imaging option, the block diagram is illustrated in Figure 1 (A)- and our proof-of-concept study demonstrated

the feasibility of auto-fluorescence imaging to differentiate congenital cholesteatoma from uninvolved middle ear tissue based on the characteristic auto-fluorescence signals (Figure 1 (B)). This direct *in vivo* imaging would enable complete surgical removal of the cholesteatoma, thereby reducing the likelihood of residual disease and improving surgical outcome and patient prognosis. Importantly, this fluoro-otoscope design uses existing conventional otoscope architecture as a platform and incorporates readily available optical components as a self-contained, add-on feature to enable auto-fluorescence imaging.

We have also designed a modified otoscope that rapidly captures narrow-band reflectance image based on the underlying variances in absorption, scattering, and depth of penetration of different pathophysiological conditions (13). We have shown its initial applicability in middle ear pathology by adopting this approach and using in conjunction with a minimally modified otoscope (figure 2). Differential absorption at the multiple wavelengths provides a measure of biochemical and morphological information - without substantially increasing system complexity and instrumentation cost- and analysis of these images aids in objective and detailed middle ear pathological evaluation.

**Closing remarks and Future Outlook**

Chemical imaging presents a promising route towards real-time, objective and detailed diagnosis of middle ear pathological conditions. The authors envision seeing deployment of new and innovative chemical imaging based sensors in actual clinical setting in near future. Since most diagnoses made in the presence of significant diagnostic uncertainty, and despite this antibiotics are still being prescribed, a tool that can enable robust segregation of otitis media subtypes would be of immense clinical value. At this juncture efforts have been to employ those techniques that are relatively very simple, ergonomically compatible and easy to adopt in clinical practice. In this milieu, other more sensitive chemical imaging modalities for instance, Raman imaging may find application in middle ear pathology. Further, multi-modal approach for examples combining the exquisite chemical specificity of Raman spectroscopy with wide-field auto-fluorescence imaging, a multidimensional algorithm capable of differentiating a broader range of middle ear pathologies may be obtained. The chemical imaging domain especially in context of middle ear pathology is in its early infancy more advancement is expected in this important area that will push the current subjective diagnostic making process toward an observer-invariant and quantitative diagnosis.

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# Figures

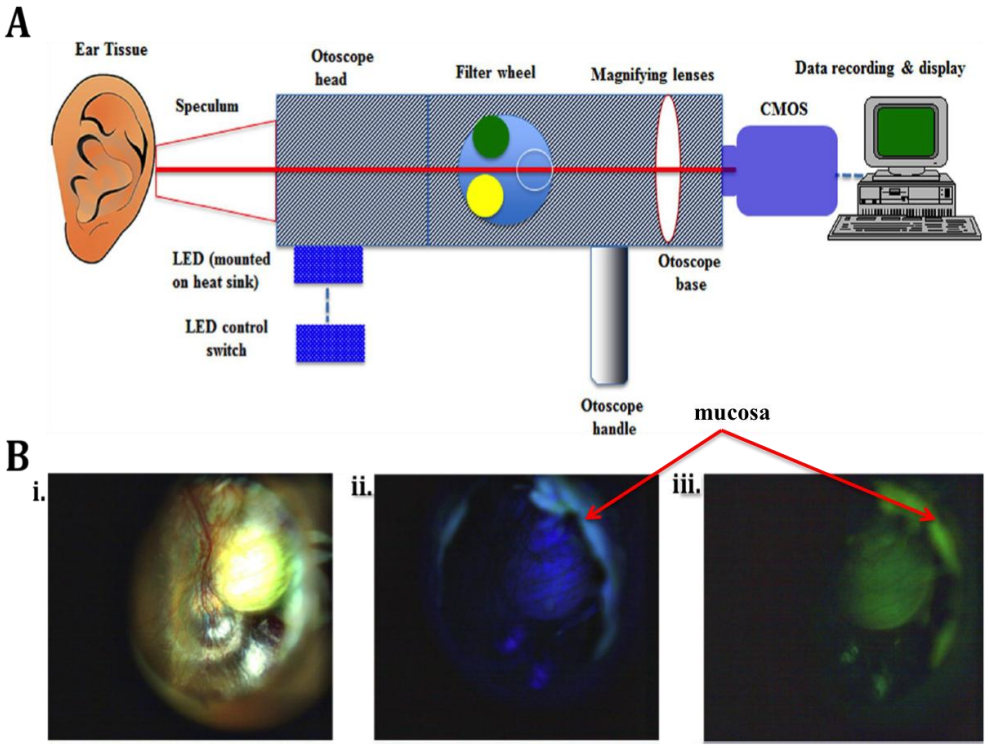


Figure 1. (A) Block diagram of fluoro-otoscope (B) White light (i.) and auto-fluorescence images (ii. & iii.) of a congenital cholesteatoma on the superior anterior quadrant of the tympanic membrane. The images ii and iii have been recorded at 405 and 450 nm excitation wavelengths respectively. The auto-fluorescence images provide clear differentiation between the cholesteatoma and the surrounding uninvolved mucosa. (Reproduced with permission from ref [11])

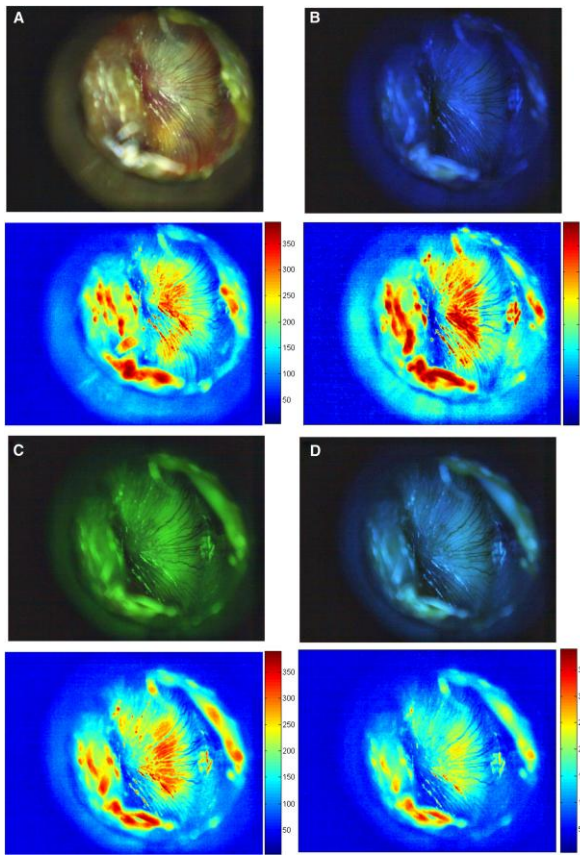


Figure 2: Representative images acquired from middle ear with acute otitis media. The images were recorded using A standard white-light illumination; B blue light illumination; C green light (523 nm) illumination; and D simultaneous blue and green light illumination. The corresponding CLAHE-derived spatial intensity maps are also provided to quantitatively assess the visualization possibilities under different illumination conditions. Evidently, the green and blue-green images show increased contrast of the purulent material behind the tympanic membrane, which can be difficult to visualize on the white-light images (Reproduced with permission from ref. [13]).