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Rapid Identification of Cryptococcus Organisms Easily Possible with Stimulated Raman Histology on a Pulmonary Tiny Biopsy Specimen: A Case Report

Howard D. Epstein[1], Cory D. Sessum[2], Christian Freudiger[2], Steven Pastore[2], Javier A. Longoria[3]

[1]Pathology Department, Hoag Memorial Hospital Presbyterian, Newport Beach, CA, USA
[2]Invenio Imaging Inc, Santa Clara, CA, USA
[3]Department of Pulmonary Medicine, Hoag Memorial Hospital Presbyterian, Newport Beach, CA, USA
Email: howard.epstein@hoag.org

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Abstract

Background: *Cryptococcus neoformans* is an opportunistic fungal pathogen that primarily affects immunocompromised individuals. While traditional histologic methods such as hematoxylin and eosin (H&E) staining can sometimes identify fungal organisms, definitive diagnosis typically requires microbiological culture or molecular testing. Stimulated Raman Histology (SRH) is an emerging imaging technology that enables rapid, label-free tissue analysis, potentially improving intraoperative diagnostic workflows. **Aim:** This case report explores the utility of SRH for the real-time identification of pulmonary cryptococcosis, highlighting its potential to enhance tissue triage and expedite diagnosis. **Case Presentation:** We report a 44-year-old man with a history of smoking and alcohol use who presented with a right lower lung mass. An ION robotic-assisted bronchoscopy was performed, and SRH was used intraoperatively for real-time tissue evaluation. Within approximately 90 seconds, SRH provided morphologic findings indicative of *Cryptococcus neoformans*, prompting additional microbiological testing, which confirmed the diagnosis. The patient required a six-week hospitalization with antifungal therapy. **Conclusion:** This case demonstrates the potential of SRH as a rapid, intraoperative diagnostic tool for detecting fungal infections in pulmonary specimens. By enabling real-time morphological assessment, SRH can optimize biopsy specimen triage, reduce the need for repeat procedures, and improve patient management. Integrating SRH into diagnostic workflows may be particularly beneficial in resource-limited settings, where timely cryptococcosis diagnosis is critical.

Keywords

Rapid OnSite Assessment (ROSE), FNA, Cryptococcus, Fungal Infection Diagnosis, SRH, Histology

1. Introduction

Cryptococcus neoformans is a globally significant fungal pathogen and a leading cause of opportunistic infections, particularly in immunocompromised populations. This environmental yeast is commonly found in soil, bird excreta (notably from pigeons), and decaying wood, with human infection typically occurring via inhalation of aerosolized spores or yeast cells. While primary pulmonary infections may be asymptomatic or cause mild respiratory symptoms, *C. neoformans* has a marked ability to disseminate hematogenously, often leading to life-threatening complications such as cryptococcal meningitis, especially in individuals with advanced HIV/AIDS, solid organ transplant recipients, or those undergoing immunosuppressive therapy [1] [2]. Globally, cryptococcal meningitis is estimated to cause over 220,000 cases annually, with a high mortality rate, particularly in resource-limited settings [3].

Despite advances in antifungal treatments, the significant morbidity and mortality associated with cryptococcosis highlight the need for improved diagnostic, prevention, and treatment strategies. Stimulated Raman Histology (SRH) is a form of histology that uses Stimulating Raman Scattering (SRS) to detect molecular vibrations in tissue representing CH2 bonds (in lipids) and CH3 bonds (in DNA/Proteins). The NIO laser imaging system applies a pseudo color mimicking traditional H&E, allowing for intra-procedural viewing of freshly scanned tissue. This case report aims to demonstrate the utility of SRH in the rapid diagnosis of pulmonary cryptococcosis, potentially transforming diagnostic workflows.

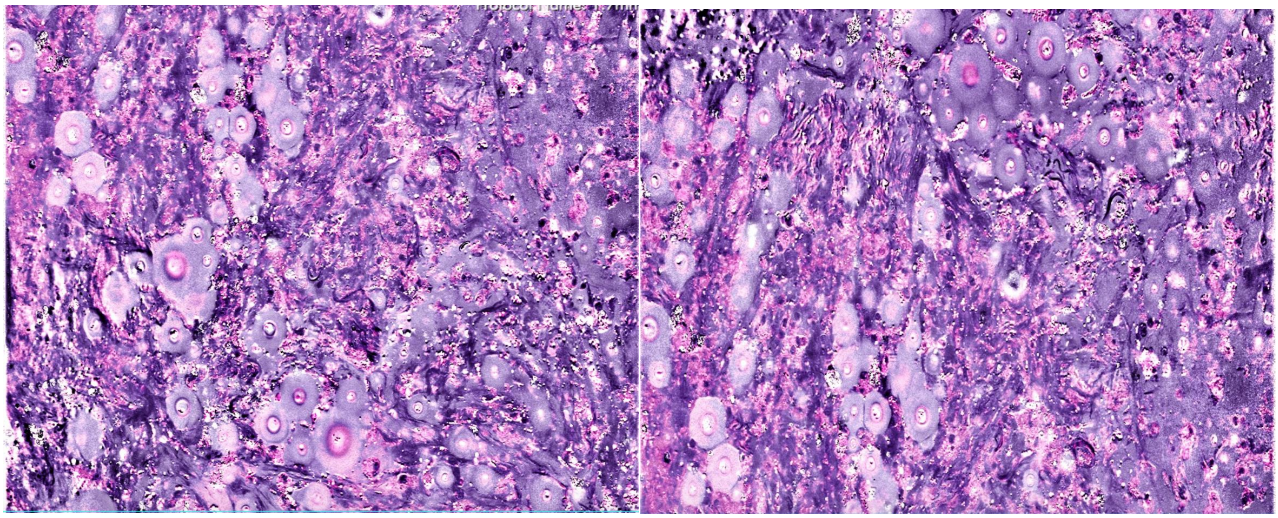
2. Case Report

A 44-year-old man with a history of smoking and alcohol-use presented to the hospital with a cough and right sided chest pain. His past medical history was otherwise negative for, and he denied history of, immunosuppressive conditions. He was HIV-negative and had no previous history of malignancy or chemotherapy. CT imaging showed a 3.3 cm spiculated right lower lung mass with a differential diagnosis of infection vs. neoplastic mass lesion. The patient was evaluated by the pulmonology team at Hoag Hospital Newport Beach and underwent an ION robotic-assisted bronchoscopy for biopsy. Hoag Hospital is a two-hospital system with a main 500 bed hospital and satellite 100 bed hospital with several large supporting outpatient centers in and around Orange County, CA. Our facility is evaluating a novel imaging known as Stimulated Raman Histology (SRH) in conjunction with standard of care Hematoxylin and Eosin (H&E) for histologic examination methods. During examination, and documented below, the biopsy

revealed a Cryptococcus fungal infection which eventually spread to various parts of the body and required a 6-week hospitalization with anti-fungal therapy under intensive care. SRH was able to identify a cryptococcal infection rapidly and accurately with diagnostic morphologic findings equivalent to the traditional H&E examination histologic standard of care. SRH was performed intraoperatively, taking approximately 90 seconds for acquisition, and provided rapid, accurate diagnostic findings equivalent to traditional H&E examination. Microbiological cultures were performed based on SRH findings, correlating with H&E results.

3. Materials and Methods (SRH)

The NIO laser imaging system by Invenio Imaging was used intraoperatively. Using Laser Raman Scattering the images in **Figure 1(a)** and **Figure 1(b)** were obtained in under 5 minutes during the procedure. For comparison, after biopsy completion, the sample was sent to pathology from which **Figure 2(a)** and **Figure 2(b)** were obtained from standard H&E-stained specimens. In both preparations, the characteristic polysaccharide capsules (the basis for cryptococcal antigen serologic tests) are easily identified.



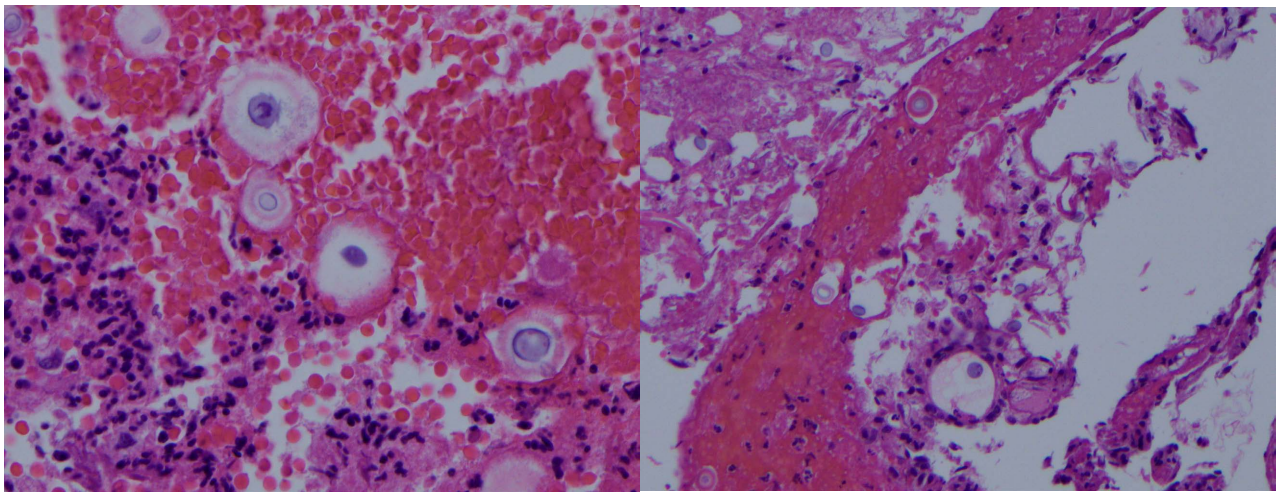
(a) (b)

Figure 1. (a) is a Stimulated Raman Histology image obtained from the NIO laser imaging system during bronchoscopy of lung mass, (a clinically suspected metastasis). (b) is a Stimulated Raman Histology image obtained from the NIO laser imaging system during bronchoscopy of lung mass, (a clinically suspected metastasis).

4. Discussion

The rapid pathological assessment of small tissue specimens from mass lesions often centers on excluding malignant and benign neoplasms for tissue triage. Pulmonary specimens, however, uniquely require prioritizing the exclusion of infectious etiologies, particularly in granulomatous diseases. Granulomas can mimic radiologically features of malignancy, displaying necrosis and rapid size changes that complicate initial diagnostic impressions [1] [3]. While molecular testing and culture remain the gold standards for microbial identification, histological examination

occasionally reveals fungal organisms if present in sufficient numbers or if their morphology is distinctive [2].



(a) (b)

Figure 2. (a) is a Hematoxylin and Eosin generated image obtained from the pathology department at Hoag Hospital in Newport Beach. (b) is a Hematoxylin and Eosin generated image obtained from the pathology department at Hoag Hospital in Newport Beach.

The expanding arsenal of imaging and biopsy technologies, such as endobronchial ultrasound (EBUS)-directed biopsies and supradimensional fine needle biopsies, has improved access to pulmonary tissue [4] [5]. However, the challenge persists in optimizing the triage of these often-limited specimens for both morphological and microbiological testing. Emerging technologies, such as Stimulated Raman Histology (SRH), provide a promising solution. First validated for rapid, label-free, high-resolution imaging of CNS lesions [6], SRH offers the advantage of preserving tissue integrity without compromising subsequent histological evaluation. Unlike traditional frozen sections that consume and alter tissue during processing, SRH enables non-destructive assessment, making it especially valuable for small specimens [7].

In this case report, we demonstrate the utility of SRH in diagnosing a *Cryptococcus* pulmonary infection presenting as a mass lesion. The ability of SRH to provide rapid, label-free morphological insights highlights its potential to transform tissue triage workflows. Importantly, SRH can inform whether additional sterile passes are necessary for microbiological testing in suspected infectious cases. Because SRH slides are not sterile, a separate sterile tissue sample is required for culture to ensure accurate microbiological diagnosis without the risk of contamination [8]. However, scanning an SRH slide that suggests fungal or bacterial presence provides immediate feedback to the proceduralist, allowing for targeted specimen collection and limiting the need for repeat procedures.

In large community and academic centers, interventional radiology (IR) and interventional pulmonology (IP) collect initial biopsy specimens, which occasionally may need to be repeated when sampling infectious etiologies. Incorporating

SRH into this workflow could streamline diagnosis by enabling rapid intra-procedural morphological assessment [9]. For example, if an SRH scan reveals fungal elements, proceduralists can immediately prioritize collecting additional samples for microbiological testing rather than sending initial samples to fixative or pathology without this critical context. This triage approach minimizes unnecessary delays, reduces repeat procedures, and benefits both patients and healthcare systems.

SRH offers an affordable, portable, and rapid diagnostic tool that does not require specialized cytotechnologists, making it suitable for resource-limited settings. Incorporating SRH into diagnostic workflows can reduce procedure times, minimize repeat biopsies, and improve patient outcomes. Limitations include initial setup costs and the need for training, but its ease of use by existing OR staff mitigates these challenges.

In resource-limited settings where cryptococcosis is prevalent, SRH can facilitate timely diagnosis, reducing morbidity and mortality. The rapid and non-destructive nature of SRH supports its integration into various clinical settings, enhancing diagnostic accuracy and efficiency.

Furthermore, SRH can support diagnostic decision-making in non-infectious cases by confirming malignancy or other neoplastic processes, potentially avoiding unnecessary microbiological testing and preserving specimen integrity [10]. This dual role rapidly identifying infectious or non-infectious pathology enhances the efficiency and accuracy of tissue triage. This non-consumptive system could lead to fewer biopsy passes, resulting in rapid procedure turnaround time with potentially less patient morbidity.

This case highlights an exciting application of SRH beyond its established role in CNS lesions, with implications for rapid, non-destructive pulmonary specimen assessment.

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Ethical Considerations

This case report adheres to all relevant ethical guidelines and standards. Informed

consent was obtained from the patient for the use of medical information and images in this report. All patient data have been de-identified to protect confidentiality.

Conflicts of Interest

The authors declare no conflicts of interest related to this case report or the innovative diagnostic techniques discussed herein.

References

[1] Mukhopadhyay, S. and Gal, A.A. (2010) Granulomatous Lung Disease: An Approach to the Differential Diagnosis. *Archives of Pathology & Laboratory Medicine*, **134**, 667-690. https://doi.org/10.5858/134.5.667

[2] Guarner, J. and Brandt, M.E. (2011) Histopathologic Diagnosis of Fungal Infections in the 21st Century. *Clinical Microbiology Reviews*, **24**, 247-280. https://doi.org/10.1128/cmr.00053-10

[3] Rosenbaum, L. and Rubin, E. (2016) Pulmonary Granulomas and Mimickers of Malignancy. *The American Journal of Pathology*, **186**, 3171-3182.

[4] Yasufuku, K., Chiyo, M., Koh, E., *et al.* (2004) Endobronchial Ultrasound Guided Transbronchial Needle Aspiration for Staging of Lung Cancer. *Thorax*, **59**, 794-796.

[5] Herth, F.J.F., Eberhardt, R., Sterman, D., *et al.* (2016) Navigational Bronchoscopy in the Diagnosis of Pulmonary Lesions. *Thoracic Surgery Clinics*, **26**, 255-262.

[6] Hollon, T., Lewis, S., Peng, L., *et al.* (2020) Rapid Intraoperative Diagnosis of Pediatric Brain Tumors Using Stimulated Raman Histology. *Cancer Research*, **80**, 4842-4850.

[7] Ji, M., Lewis, S., Camelo-Piragua, S., Ramkissoon, S.H., Snuderl, M., Venneti, S., *et al.* (2015) Detection of Human Brain Tumor Infiltration with Quantitative Stimulated Raman Scattering Microscopy. *Science Translational Medicine*, **7**, 309ra163. https://doi.org/10.1126/scitranslmed.aab0195

[8] Hollon, T.C., Pandian, B., Adapa, A.R., *et al.* (2020) Near Real-Time Intraoperative Brain Tumor Diagnosis Using Stimulated Raman Histology and Deep Neural Networks. *Nature Medicine*, **26**, 52-58.

[9] Chang, A. and Boiselle, P.M. (2016) Advances in Interventional Pulmonology. *Clinical Chest Medicine*, **37**, 299-311.

[10] Orringer, D.A., Pandian, B., Niknafs, Y.S., Hollon, T.C., Boyle, J., Lewis, S., *et al.* (2017) Rapid Intraoperative Histology of Unprocessed Surgical Specimens via Fibre-Laser-Based Stimulated Raman Scattering Microscopy. *Nature Biomedical Engineering*, **1**, Article No. 0027. https://doi.org/10.1038/s41551-016-0027