Laser Spectroscopy for in situ elemental imaging of lung tissue: a promising technology

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Introduction

Environmental and occupational exposures to **inhaled mineral particles, metals, and dust** are major health problems. Pathologists infrequently report the presence of metals in tissues. Some respiratory diseases such as interstitial lung diseases (ILDs) might even be considered "idiopathic" by lack of complementary investigations regarding to the elemental composition.

Objectives

To identify and localize several chemical elements within human formalin-fixed paraffin embedded (FFPE) lung specimens.

Methods

We used a laser spectrometry system (LIBS) on selected paraffin-embedded specimens with relevant idiopathic respiratory diseases such as sarcoidosis, fibrosis, etc. We worked either on endobronchial ultrasound (EBUS) or bronchial biopsies. The elemental findings were compared with the results of a mineralogical analysis performed by electron microscopy.

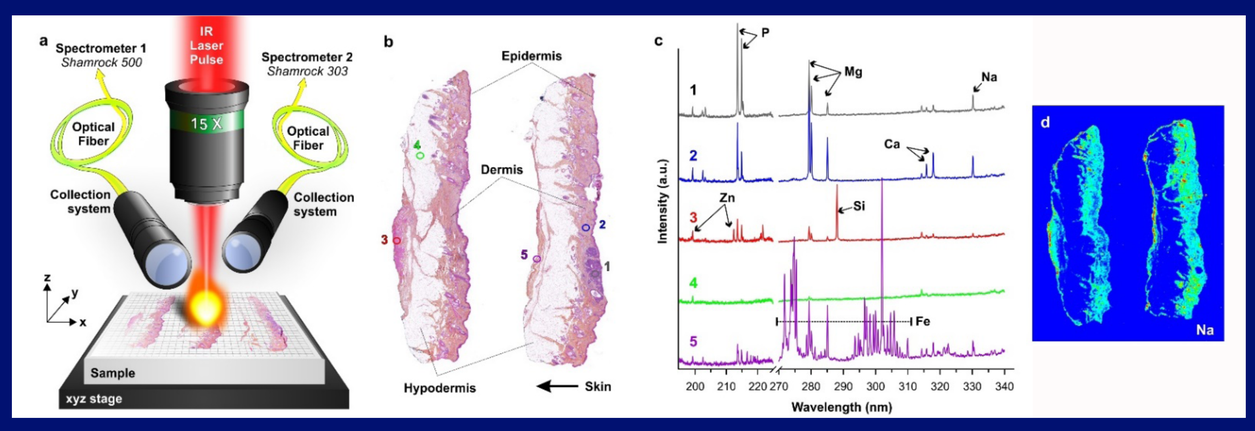

Fig. 1. Overview of LIBS imaging technology. (a) Schematic representation of the main components of the LIBS imaging instrument: a microscope objective to focus the laser pulse, a motorized sample stage and two optical detection systems coupled with two Czerny-Turner spectrometers. (b) High-resolution HES histological images of a healthy skin sample before LIBS analysis, (c) Typical single shot tissue spectrum covering the 270-340 nm for detecting Mg, Si, Fe, and Na, and the 185-225 nm detecting P and Zn on different areas of the tissue biopsy. (d) LIBS images for Sodium (Na) in the paraffin-embedded skin specimen.

Result 1 : Endobronchial ultrasound (EBUS)

The EBUS procedures allow physicians to perform a technique known as transbronchial needle aspiration (TBNA) to obtain tissue or fluid samples from the lungs and surrounding lymph nodes without conventional surgery.

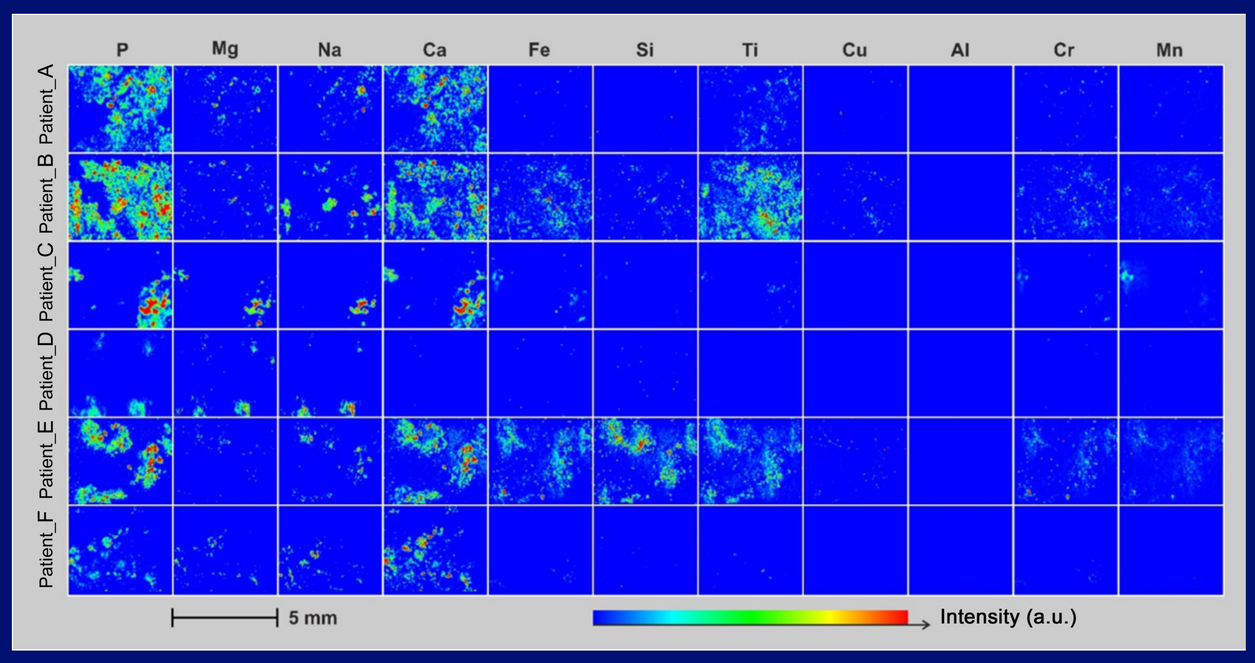

Fig. 2. Multi-elemental analysis of the EBUS samples from 6 patients with sarcoidosis (Patients A-F).

LIBS multi-elemental images shows the presence of high levels of several metal particles such as Si, Ti, Cu, Cr or Mn in the lymph nodes of patients with idiopathic sarcoidosis B and E. This is contrasting with other patients with sarcoidosis whose mediastinal lymph nodes contain very low levels of metals patients A,C,D, F). These data were confirmed by electronic microscopy.
We demonstrate that LIBS imaging of suspect lymph nodes reveals the presence of abnormally high levels of metals for some patients with sarcoidosis. This suggests to investigate occupational or environmental etiologies for such cases.

Result 2 : Lung parenchyma specimen

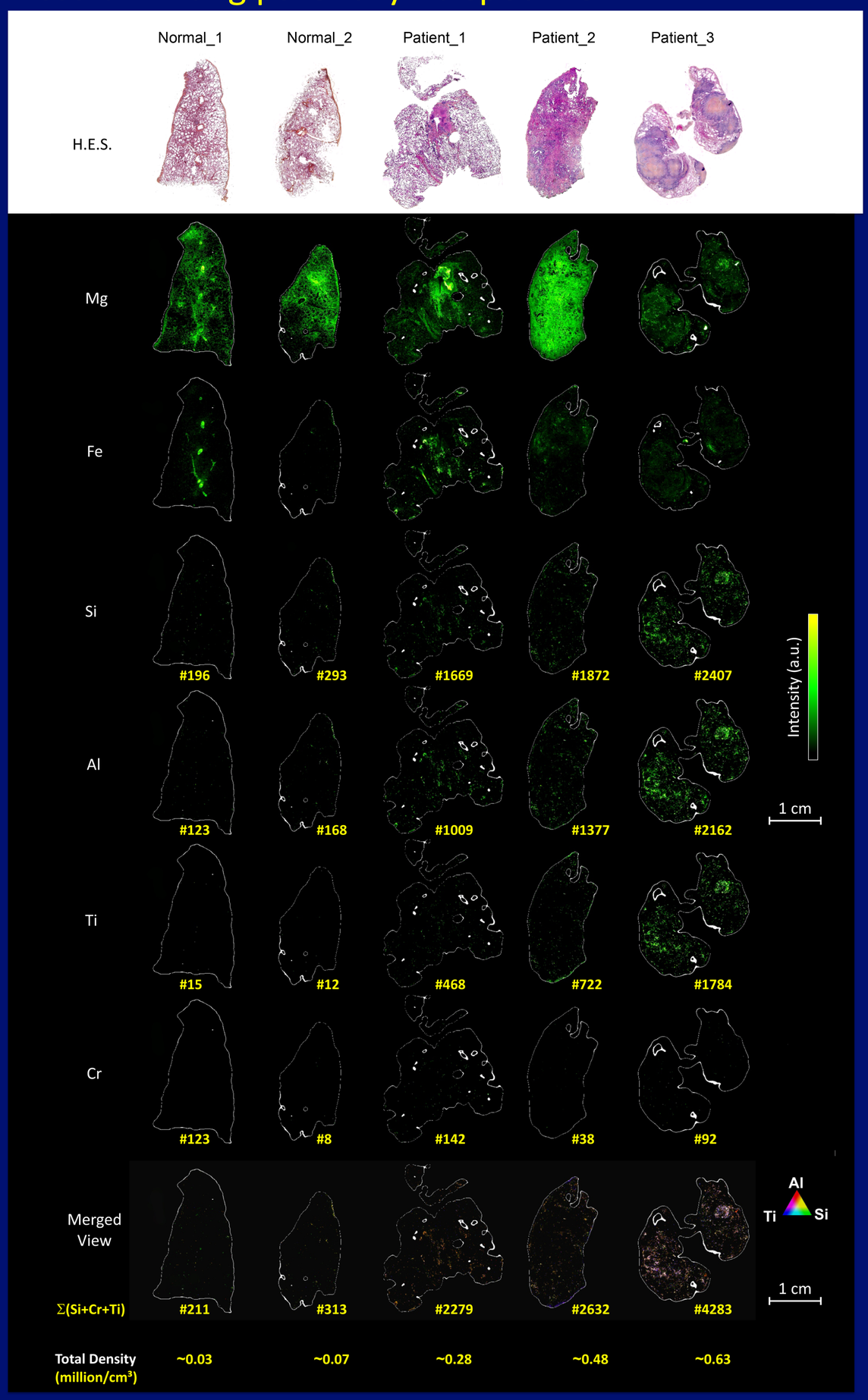

Fig. 3. Multi-elemental analysis of the EBUS samples from 3 patients with idiopathic ILDs (Patient 1-3).

LIBS multi-elemental images shows the presence of high levels of several metal particles such as Si, Ti, Al, or Cr in the lung parenchyma biopsies of patients with idiopathic ILDs (sarcoidosis/patient_1, idiopathic pulmonary fibrosis (IPF)/patient_2 and ILD with pneumoconiosis features/patient_3). These data were confirmed by electron microscopy.
We developed a method to quantify the total number of particles in the analyzed surface. We also displayed the total density of such particles per cm^3 of lungs.
We demonstrate that LIBS imaging of lung specimen from patients with idiopathic ILDs reveals the presence of abnormally high levels of metal particles, with a 10x increase compared to normal lung parenchyma. This suggests to investigate occupational or environmental etiologies for such cases.

Other results and ongoing studies:

- Achieving biological resolution in 2D (pixel size 10x10 mm^2) and in 3D ✓
- Compatibility with other imaging techniques (fluorescence, Raman) ✓
- Working fast, under ambient conditions (room pressure and temperature) ✓
- Working on frozen material
- Screening large cohorts (normal lungs and pathological lungs)

Conclusion:

LIBS imaging technique is highly versatile because almost any element (endogenous/exogenous) can be quantified with high sensitivity. Besides, this technique is **fully complementary with standard optical microscopy** used by the pathologists for diagnosis purposes. This approach could help the pathological analysis of FFPE specimen with lung diseases, especially "idiopathic" diseases.

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