

# LABEL-FREE DETECTION OF CARBONACEOUS PARTICLES USING FEMTOSECOND PULSED ILLUMINATION TO ASSESS THEIR TOXICOLOGICAL EFFECTS

Hannelore Bové<sup>1,2\*</sup>, Christian Steuwe<sup>2</sup>, Nelly Saenen<sup>3</sup>, Martin vandeVen<sup>1</sup>, Tim Nawrot<sup>3</sup>, Maarten Roeffaers<sup>2</sup>, Marcel Ameloot<sup>1</sup>

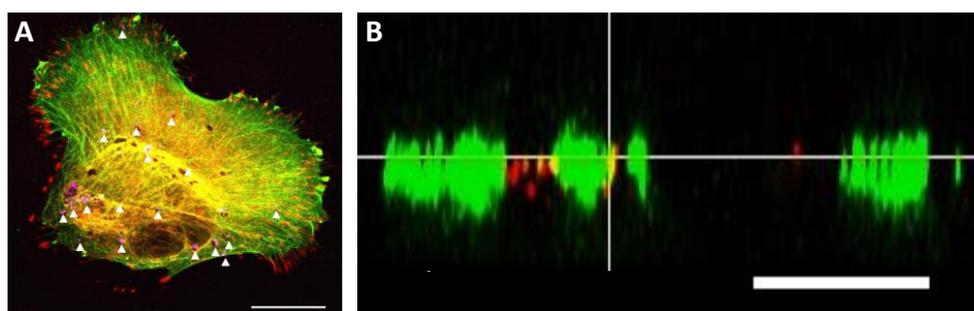
<sup>1</sup>Hasselt University, Biomedical Research Institute and <sup>3</sup>Centre for Environmental Sciences, Agoralaan Building C and D, 3590 Diepenbeek, Belgium

<sup>2</sup>KU Leuven, Centre for Surface Chemistry and Catalysis, Celestijnenlaan 200F, 3001 Leuven, Belgium

\*E-mail presenting author: [hannelore.bove@uhasselt.be](mailto:hannelore.bove@uhasselt.be)

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In 2012, 432,000 people across Europe died due to air pollution. No other type of environmental pollution imposes such a high health risk [1]. Fine dust is already known to provoke dementia [2], heart and lung diseases [3, 4]. The core of atmospheric pollutant particles is represented by carbonaceous particles (CPs), which are produced during the incomplete combustion of fuels. Despite the high, health related relevance of CP exposure, no direct, label-free approach for detecting such particles in fluids and at the cellular level exist. In this regard, we present a novel technique based on white-light (WL) generation by CPs under illumination with femtosecond pulsed near-infrared light [5]. In this study, we investigated various CPs representative for environmental exposure. The particles were illuminated with a femtosecond laser and the emitted signal was recorded using a commercial multiphoton laser-scanning microscope. Additionally, the technique was evaluated in various human fluids, tissues and cells (Fig. 1). To the best of our knowledge, we report for the first time non-incandescence related WL emission by CPs in aqueous environments and demonstrate the potential of this technique in biological context. It is a straightforward approach without the need of sample pretreatment. The nature of the signal makes it very versatile to combine it with conventional contrast-enhancing fluorophores used to visualize biological features (Fig. 1). In addition, the technique offers several advantages such as inherent 3D sectioning and high imaging depths owing to the multiphoton approach.



**Figure 1:** A) Imaging of cellular compartments of fixed human lung fibroblasts (MRC-5 cell line) stained with commonly utilized fluorophores and in combination with the detection of carbonaceous particles (4 h incubation of 5  $\mu\text{g}/\text{cm}^2$  at 37  $^{\circ}\text{C}$  prior to imaging). Arrow heads point out the very small engulfed particles. Scale bar: 25  $\mu\text{m}$ . B) Example XZ-cross section showing the uptake of the carbonaceous particles (red) between the tubulin cytoskeleton (green). Scale bar: 20  $\mu\text{m}$ .

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