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Analysis of sampled airborne particles by correlative microscopy

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While breathing, humans inhale a large variety and number of airborne objects that are part of the so-called exposome [1]. Nonetheless, origin, composition, shape and concentration of inhaled solid objects are mainly unknown as they depend on many details of the natural surroundings and the living or working environment and vary in the course of a day. They generally comprise natural and manufactured particles and fibres as well as bioaerosols, e.g., microorganisms, viruses or pollen. Depending on their aerodynamic properties, they may reach different compartments of the respiratory tract. Particular attention must be paid to objects that are toxic either due to chemical composition or biologic activity, or due to biodurability and respirability. It is therefore necessary to improve knowledge about the presence and nature of the respirable solid fraction of the exposome. Here, we describe our approach to an automated detection, classification and quantification methodology for solid airborne objects. It quantifies particles and fibres collected on track-etched membrane filters by first imaging filter areas representing a specific sampled air volume with scanning electron microscopy (SEM). High-resolution images allow localising and morphologically classifying objects based on secondary electron (SE) contrast. Elemental information is acquired by energy dispersive X-ray spectroscopy (EDS). Whenever elemental data is insufficient for identification, e.g., for carbon-based materials, complementing optical and Raman microscopic analyses are performed. All involved microscopes are operated with respect to stage control as well as images and spectra recording by our fully automatic in-house software [2]. Using constellation matching algorithms for the filter pores, objects of interest that are visible only in SEM can be located in the confocal Raman microscope with a spatial precision better than the optical diffraction limit [3]. This way, all optical and SE images as well as spectral mapping data are spatially correlated. Artificial neural network-based object segmentation allows to attribute compositional information to individual particles and fibres [4] aiming at classifying them by means of shape, elemental composition and chemical characteristics.

1. Wild, C.P. The exposome: from concept to utility. Int. J. Epidemiol. 2012, 41, 24–32, doi:10.1093/ije/dyr236
2. Schumann, J.; Bäger, D.; Dziurowitz, N.; Thim, C.; Meyer-Plath, A. Retrieving SEM sample positions under limited relocation accuracy using digital image processing. Proceedings of the MC2019 Conference 2019, Berlin; pp 391–392
3. Meyer-Plath, A. Correlating SEM and Raman Imaging of Nucleopore Filter Sampled Nanofibers: doi: 10.13140/RG.2.2.11130.72641. Virtual Raman Imaging Poster Summit 2020

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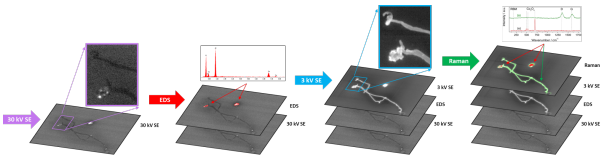

Fig. 1: Correlative microscopy with SEM-SE, EDS and Raman signals

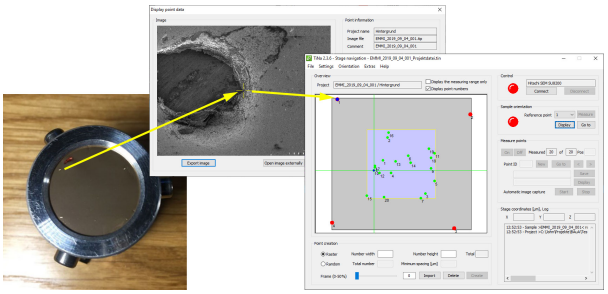

Fig. 2: Stage navigation software TiNa with sample orientation

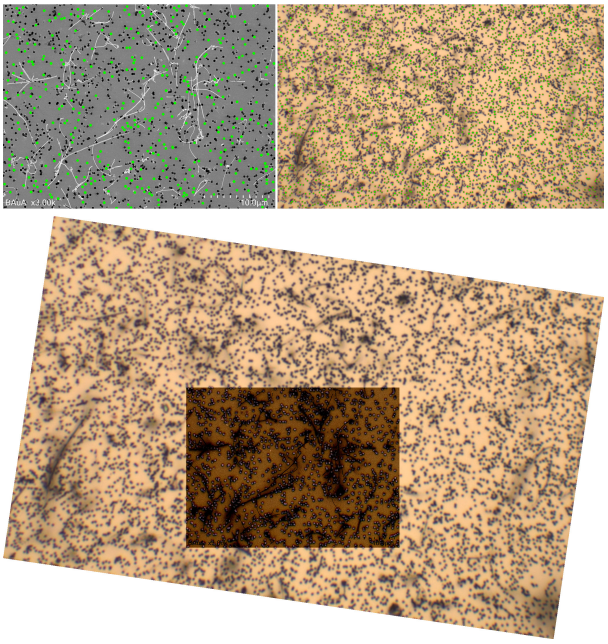

Fig. 3: Image matching between SEM-SE image and Raman optical image using a constellation matching algorithm

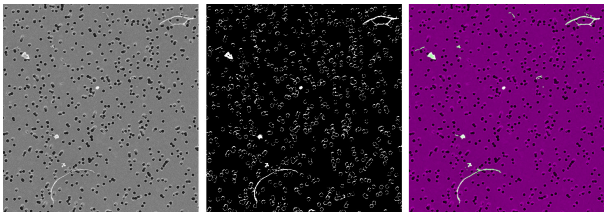

Fig. 4: Comparison of object segmantation, left: original SEM-SE image, middle: segmentation with classical image processing algorithms, right: segmentation with artificial neural network