

# Development of Rapid-E+ for real-time classification and quantification of airborne bacteria, fungi, and other bioaerosols



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## Introduction

The real-time identification of bacteria and fungi is difficult because they emit much weaker signals than pollen. In 2020, Plair developed Rapid-E+, which extends abilities of Rapid-E (Morrison et al., 2020; Šaulienė et al, 2019.; Tešendić et al., 2020) to detect smaller bioaerosols such as bacteria and fungal spores with diameters down to 0.3  $\mu\text{m}$ , while keeping the similar or even better capability for measurements of large bioaerosols like pollen. Rapid-E+ enables simultaneous measurements of (1) time-resolved, polarization and angle dependent Mie scattering patterns, (2) fluorescence spectra resolved in 16 channels, and (3) fluorescence lifetime of individual particles. Moreover, (4) it provides 2D Mie scattering images which give the full information on particle morphology (Figure 1). The parameters of every single bioaerosol aspirated into the instrument are subsequently analysed by machine learning.

## Method

Firstly, pure species of microbes, e.g., *Bacillus subtilis* (a species of bacteria), and *Penicillium chrysogenum* (a species of fungal spores), were aerosolized in a bioaerosol chamber for Rapid-E+ training. Afterwards, we tested microbes under different concentrations. We used several steps of data analysis to classify and identify microbes. All single particles were analysed by the parameters of light scattering and fluorescence in the following steps. (1) They were treated with a smart filter block to get rid of non-microbes. (2) By classification algorithm, we verified the filtered particles were microbes based on the calibration data. (3) The probability threshold (defined by the user) step provides the probability of being microbes ranging from 0 to 100%.

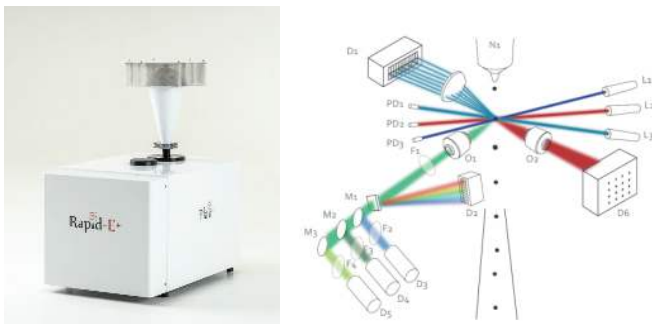


Figure 1. Rapid-E+ and the working principle

## Results and Discussion

We demonstrate how Rapid-E+ identified simultaneously microbes based on the results of *Bacillus subtilis* (bacteria) and *Penicillium chrysogenum* (fungal spores). Figure 2 shows that by using machine learning, Rapid-E+ achieved identification precision of 99% against the background. The further classification suggests the precision of 87% and 89% for *Bacillus subtilis* and *Penicillium chrysogenum*, respectively.

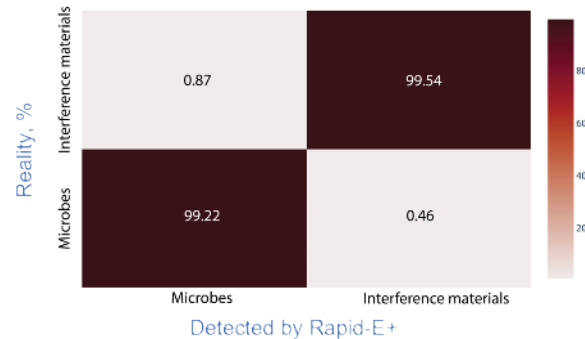


Figure 2. Rapid-E+ classifies microbes (bacteria and fungal spores) versus background by confusion matrix with high accuracy of more than 99%.

The developed algorithm was subsequently used to evaluate the performance of microbe classification and quantification in real-time. The bacteria and fungi were aerosolized again in the chamber with different concentrations. Figure 3 shows that Rapid-E+ can classify different types of microbes and then quantify them in real-time.

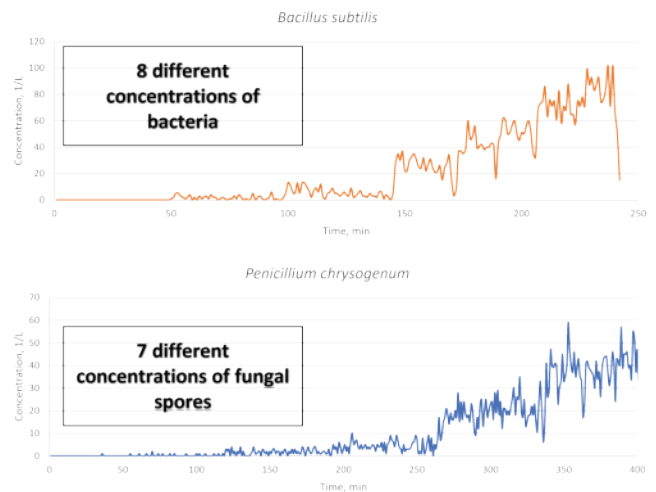


Figure 3. Rapid-E+ classifies and quantifies bacteria and fungi in real-time with high time-resolution. Rapid-E+ can identify them automatically based on the algorithms installed on the machine learning platform.

## Conclusions

Rapid-E+ enables classifying different types of microbes and quantifying them in real-time. Rapid-E+ is an all-in-one instrument which classifies and quantifies not only pollen, but also bacteria and fungi. Based on the machine learning platform, the user can further develop proprietary algorithms for specific microbes (e.g., virus aerosols) and other aerosols (e.g., combustion-related particles that contain polycyclic aromatic hydrocarbons).

<sup>1</sup> References: Morrison et al (2020) Atmosphere, 11, 944. Šaulienė et al (2019) Atmos. Meas. Tech., 12, 3435–3452. Tešendić et al (2020) Enterprise Information Systems.