First experiences with the newly-developed Swisens Pollen Monitor

Erny Niederberger*, Reto Abt*, Philipp Burch**, Yanick Zeder**

* Swisens AG and Lucerne University of Applied Sciences and Arts, ** Swisens AG, *** Lucerne University of Applied Sciences and Arts

HIGHLIGHTS

- Very promising results of the first pre-operational pollen monitor developed by Swisens
- The importance of real-time pollen identification is shown
- The holographic images taken by the device are very helpful for quality control
- The new device opens new perspectives for research and monitoring

ARTICLE INFORMATION

ABSTRACT

Extended abstract for oral presentation at technical conference on meteorological and environmental instruments and methods of observation (TECO) organized by the World Meteorological Organization (WMO) Commission for Instruments and Methods of Observation (CIMO) in Amsterdam, the Netherlands, 8-11 October 2018, Presentation No. O2_9 under topic: Emerging measurement technologies: from development to operation

Keywords: Pollen Optical methods Automatic monitoring Real-time Machine learning The new measuring device presented here called Swisens Poleno measures individual particles, especially pollen, in flight and is able to identify the pollen species automatically and in real time. It is possible to measure the local pollen concentration for the various pollen species related to allergies. The results from the first field test with Swisens Poleno together with MeteoSwiss are very promising and allow sensitized people to better cope with the allergy.

Pollen allergy causes high economic costs. Nowadays, targeted, effective preventive measures to minimize the contact with pollen can not be taken because of the insufficient accuracy of today's pollen prognosis. Exact information on the current pollen concentrations is not available today because the conventional pollen measurement method does not provide real-time data. With Swisens Poleno the timeconsuming and expensive manual identification and counting of pollen in the laboratory as well as the collecting and shipping of adhesive sampling stripes from the classic hirst-type pollen traps will be obsolete. Furthermore the value of pollen measurement data is increasing due to real-time availability. The accuracy of the pollen prognosis can be improved significantly.

With the availability of real-time data and accurate forecasts, allergy sufferers are empowered to take targeted preventative measures to minimize exposure to pollen and ultimately reduce symptoms. This strongly improves their quality of life and their work-performance so that the economic costs due to allergies can be reduced significantly. The latest results on the quality of the identification of pollen taxa with Swisens Poleno will be presented at the conference.

Introduction

Pollen allergy, economic impact and trends

The real-time measurement and identification of pollen is strongly related to allergies. This topic is very relevant because there are more and more people affected and the health costs due to allergies are increasing. In Switzerland, around 15 to 20% of the population is affected by a pollen allergy [1]. In Europe, costs are estimated at 55 to 151 billion Euros per vear [1]. Costs are that high because more than 60% of pollen allergy sufferers are not adequately medicated and the allergies subsequently lead to impaired productivity [2]. Pollen is one of the major allergens that cause asthma [3] because in untreated pollen allergies there is a risk of crossreactions [4] and a change of level to asthma, which causes on average 16.5 times more costs than normal hay fever [5].

According to European Federation of Allergy and Airways Diseases Patients Associations (EFA) studies, more than 80 million (over 24%) of adults in Europe suffer from allergies, and the prevalence among children, which is between 30 and 40%, is showing a rising trend. Asthma alone types of aerosols such as spores [15], diesel soot generates 17.7 billion Euros in direct costs (treatment, care, etc.) and 9.8 billion Euros in indirect costs (lost work, decrease in productivity, etc.) per year in Europe [6]. In a position paper, the EFA calls for an EU-wide legal anchoring of pollen measurements [7], [8]. There is evidence from scientific studies that there is an interaction between particulate matter and pollen that can aggravate the symptoms of the allergy [9]. According to World Health Organization there are 7 million premature deaths annually linked to air pollution every year worldwide [10]. In the future, a further increase

in pollen allergies due to global warming, urbanization, migration and the spread of invasive neophytes is expected. People in cities are more likely to be affected by allergies than those living in rural areas. Moving to a new biological environment increases the likelihood of allergies. Rising temperatures increase the allergic effect of the plants, which is why the allergy reactions are even stronger [11], [12], [13].

Need for real-time information

Many people affected by allergies do not know what the cause of the allergy is. One reason for this is that the connections between the causative particles and the symptoms of allergy are still not well known, because measuring the aerosol concentrations directly from the air and in realtime is not easy today. This makes it difficult to identify the causes. Therefore, the European Academy of Allergy and Clinical Immunology (EACCI) calls for new and standardized measurement methods for allergenic aerosols of biological origin [14]. The devices of Swisens AG are to close this gap, since they have the potential to identify besides pollen also other or mineral particles [16].

The conventional pollen measurement is very laborious and expensive, since it is based on manual identification in the laboratory. Today, pollen is collected using volumetric pollen traps of the Hirst type [17]. For this purpose, adhesive strips are mounted on a slowly rotating drum, to which pollen do settle. The pollen which stick to these strips are typically identified and counted once a week in the laboratory, by specially trained people in manual labor under a microscope. Due to the high costs, only a few measuring points per country are available today. For example, in Switzerland there are only 14 pollen measuring stations. In Europe, there are a total of 500 Hirst, Lanzoni or Burkard pollen traps in operation [18].

Need for more accurate pollen forecast

Due to the low spatial resolution and the large delay in time of up to one week until the pollen data is available, these measurement results are too late for a daily pollen warning and for accurate pollen forecast. This is one reason that today's pollen apps are very inaccurate and do suggest a pseudo-accuracy to allergy sufferers. Today, the values for pollen concentration are all based on model calculations in combination with weather forecasts and have relatively large uncertainty. Today allergic people do realize that there are pollen in the air because of their symptoms, due to the fact that they have already got into contact with the pollen. What is needed is an early warning so that preventive measures can be taken. This is possible in the future thanks to real-time pollen identification devices and networks.

Measured physical properties by the device

The first devices for the automation of pollen measurement are available now. Swisens AG has developed a device in close cooperation with Lucerne University of Applied Sciences and Arts, called Swisens Poleno. The identification of the different pollen species is based on the measurement of the following physical properties of the pollen:

Elastic light scattering / stray light

Figure 1 visualizes the principle of elastic light scattering. The large blue arrow on the left indicates the illumination light. This light hits the particle (black dot). The surface of a particle

typically reflects and absorbs part of the incident light. The small size of the particle also leads to diffraction, which in addition to reflection and absorption causes non-uniform stray light patterns characteristic for various particles.



Figure 1: Elastic light scattering

Autofluorescence

Autofluorescence is the natural emission of light by biological structures after an appropriate excitation. For example, pollen and spores are autofluorescent. The light emission depends on the autofluorescent molecules of the biological structure and the excitation wavelength. Measuring the fluorescence emission spectra and the fluorescence lifetime of different pollen-taxa makes it possible to distinguish between them.

Spectral fluorescence intensity

Figure 2 illustrates the fluorescence effect when a fluorescent particle is illuminated.



Figure 2: Fluorescence emission

Fluorescence occurs after a fluorescent particle has absorbed energy / photons. The large violet arrow on the left indicates the illumination, also called excitation light. A fluorescent particle emits light with a longer wavelength than the excitation light. In figure 2 the excitation is for example in the ultra-violet region and could be 280 nanometers. The fluorescent emission maybe in or beyond the visible spectrum, e.g. from 320 to 750 nanometers.

Fluorescence lifetime

The fluorescence lifetime refers to the average time the molecule stays in its excited state before emitting a photon. Typical lifetimes with excitation in the range of UV to near infrared are within 0.5 to 20 nanoseconds. The fluorescence lifetime values provide additional information on the sample and allow for improved quality of differentiation of biological particles like pollentaxa for example.

Description of air-flow cytometer

Swisens Poleno is designed for the precise and reliable identification and counting of airborne particles. It is specially optimized for the



Figure 3: Swisens Poleno inside weatherproof housing

identification of pollen taxa in real-time and for the long-term monitoring of the pollen concentration in the air. For monitoring in the field, the device is placed into an air-conditioned weatherproof housing. Poleno's dimensions are $30 \times 30 \times 50 \text{ cm}^3$ and the weight is 23 kg. Figure 3 shows the Swisens Poleno prototype inside a weatherproof housing for the field test. Figure 4 shows the schematic overview of the device.



Figure 4: Swisens Poleno schematic overview Swisens Poleno is an air-flow cytometer based on the analysis of light scattering, holographic images and UV-induced fluorescence. With these measurement methods, many independent features can be determined, allowing for excellent quality of particle identification.

Particle sampling

The whole system is designed to measure particles in the size range from 1µm to 300µm. The maximum recommended particle concentration is 30'000 particles per m³. The air including the aerosol particles are aspirated into the device through an inlet with a Sigma-2 geometry. An aerosol-concentrator with a concentration factor of 1000 enables a volume flow rate of 40 liters per minute to be analyzed, which enables high time resolution for the measurement of local pollen concentration down to minutes. Particles with a diameter larger than 10µm are concentrated up and are directed into the measurement chamber. The majority of the smaller particles are bypassed so that they do not pass the measurement chamber.

Morphological features

Information about the morphology of each individual pollen-grain is collected with the highresolution holography setup which delivers images of the particles.



Figure 5: Fagus sylvatica



Figure 7: Ambrosia



Figure 6: Corylus avellana



Figure 8: Quercus robur

Images of two cameras which are displaced by 90° are taken per particle. The pixel resolution is 0.56μ m/pixel. The camera features 2048 x 1536 pixels and a frame rate of up to 55 images/second.

The advantages of the holography setup includes a wide field of view in x, y and z-axis while maintaining a very high resolution.

Figures 5 to 8 show reconstructed images of pollen grains taken in flight with the holography measurement setup.

The images show an area of $115 \times 115 \mu m^2$. The time-resolved measurement of the vertical and horizontal polarized scattered light provides information about the surface structure, size and the polarization factor. The scattered light of a particle passing by the detector is measured with a time resolution of 4µs. Figure 9 visualizes the measured data.





Complementary information in addition to the morphological features is collected by spectrally resolved fluorescence intensity and fluorescence lifetime measurements. This characteristic values deliver an additional dimension of information

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and allow for the extremely accurate identification of the different pollen-taxa. There are three different modulated light sources to excite the particles while flying by. The fluorescence excitation is done with a 405nm laser diode as well as a 280nm and a 365nm LED. The fluorescence emission and its lifetime is measured with five carefully chosen spectral windows in the range of 320nm to 750nm. The measurement range for the fluorescence lifetime is 0.5 to 20ns. Figure 10 shows the fluorescence intensity measured at five different wavelength ranges between 320 and 750nm.





Figure 11 shows the correlation phase and correlation magnitude for a single particle measured at different modulation frequencies. By Measurement dataset, calibration dataset, means of curve fitting the different lifetime components can be extracted.

Integrated removable sample collector

To examine the system's performance and to validate its functions, a petri dish can be inserted into the measuring channel as an impaction filter. Aspirated particles accumulate on it and can then



Figure 11: Fluorescence lifetime measurement data visualization

be evaluated manually and compared with the data measured on the device.

Method and material

Calibration or training data of the different pollen-taxa to be determined are necessary for the identification. For this purpose, pollen is dispersed in the air near the inlet and then measured by the device. This procedure is performed sequentially for the different types of pollen. Training data sets for 15 different types of pollen were obtained in this way in the laboratory. The images allow visual inspection to clean the training data set. Various dry pollen from Allergon AB Sweden / Thermo Fisher and Bonapol AS Czechia were used.

training dataset and verification dataset

For the training data, a library was created. One measurement dataset consists of image data from the holography setup and numerical values from the other measurement methods. The raw image data is preprocessed to only contain the pollen in sharp focus. The measurement dataset has two images per pollen measurement which are

recorded by the two image sensors. The images have a 200x200 pixel resolution and are saved in greyscale format. Figure 5 to 8 are examples of some sample images from such a dataset. One calibration dataset consists of several hundred or thousand samples from the same pollen-taxa. Each calibration dataset is spit into a training dataset and a second verification dataset. The latter is used to qualify the identification algorithm.

Classification algorithms

The identification is based on machine learning algorithms and artificial neuronal networks. The calibration data is used to train the artificial neuronal network.

The outputs of the neuronal network are the labels and the confidence values for each measurement. A confusion matrix as shown in figure 12 is used for visualization of the results.



Figure 12: Identification quality shown by the confusion matrix

A self learning algorithms can recognize pollen features but also the details of the light conditions for example. To make sure the algorithm takes its decision just on features related to the particle and independent of the state of the device, a visualization of the regions most important for classification found by the algorithm was performed. Figures 13 to 16 show such a heatmap. This allows to check if the algorithm bases its decision on relevant features.







Figure 14: Corylus avellana





Figure 16: Quercus robur

Results

At the 11th international congress on Aerobiology held on beginning of September 2018 in Parma the results based on measurements from Swisens Poleno were presented by B. Crouzy [19]. Based on machine learning algorithms including artificial neural networks and cleaned training data sets B. Crouzy and Y. Zeder achieved an average **correct identification rate of over 90%** for the seven pollen species they trained. As improvements on the algorithms are ongoing the actual status of the achieved performance will be presented at the conference.

Conclusion

The already obtained results with Swisens Poleno are very promising. The holographic images are very helpful for quality control. Thanks to the images the verification is simple and intuitive. This allows to clean up the training dataset which increases the quality of the identification.

The new device opens new perspectives for research and monitoring.





Figure 18: Pollen with

Figure 17: Water droplet



Figure 19: Unknown object



Figure 21: Unknown object



Figure 20: Unknown object



Figure 22: Unknown object



Figure 23: Unknown object



Figure 24: Unknown object





Figure 25: Unknown object

Figure 26: Unknown object

As the figures 17 to 26 show, the device is also very valuable for the measurement and classification of other classes of aerosols. Swisens Poleno marks the beginning of a new era in the measurement of bioaerosols. It indicates a further step towards an early warning system for pollen which enables better managing of pollen allergy.

About Swisens AG

Founded in 2016 as a spin-off of Lucerne University of Applied Sciences and Arts, Swisens AG develops advanced sensing technologies to monitor environmental threats in our surrounding air. "We want to return some quality of life to allergy sufferers". Swisens offers solutions based on optical detectors for real-time airborne bioaerosol monitoring, especially allergenic particles. Visit our website https://swisens.ch for more information

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